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Physicochemical Properties, Phenolic Contents and Antioxidant Activity of Vietnamese Honey

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Abstract

Vietnam is located in the tropical monsoon climate area (Southeast Asia), along with the biodiversity of plant species, which has created many types of monofloral and polyfloral honey. However, there has not been a study that fully reports on the physicochemical composition, antioxidant activity and content of phytochemical compounds of honey in this country. Therefore, this study aimed to determine Vietnam's physicochemical properties, antioxidant activity, and phytochemical content of longan flower honey and natural polyfloral honey (from *Micrapis* honeycomb). The reference analytical methods were modified based on previous honey studies. Following proximate indicators were found: The moisture content varied from 21.9 to 30.5%, the ash content varied from 0.37 to 0.48%, the protein content varied from 0.25 to 0.41%, and the 5-(hydroxymethyl)-2-furaldehyde (HMF) varied from 33.4 to 75.4%. The total phenolic and flavonoid concentrations ranged from 0.890 to 1.110 mgGAE/g and 0.049 to 0.089 mgQE/g. Besides, the antioxidant activity of longan flower honey was recorded as the best, with the IC_{50} value of 68.49 mg/mL. In addition, Water content, free acidity, sugar content (glucose, fructose, and sucrose), and HMF concentration were all tested and presented.

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Introduction

Honey is a natural substance produced by honeybees from plant and insect nectar. Honey includes almost 200 distinct chemical constituents (including glucose and fructose, which account for 80-85%, water (15-17%), minerals (0.2%), protein and amino acids (0.1-0.4%), and other components such as phenolic chemicals (Idris, Mariod, & Hamad, 2011). The organoleptic, physicochemical and bioactive properties of honey are influenced by the plant source from which the bees get their honey (Kortesniemi *et al.*, 2018). The origin of honey is considered an essential factor for quality

validation, bioactivity potential and commercial value. Besides, each region and country will have a uniform standard developed (CA, EU, etc.). In general, current international standards establish procedures for the quality control of honeys based on their chemical and physical properties.

The chemical composition of monofloral and polyfloral honey depends on the plant source, geographical origin and time of honey collection. The main components of honey are sugar (mainly fructose and glucose) and water. The origin of flowers/plants plays an important role in forming the chemical composition of honey and determines their value or class (Hailu & Belay, 2020). However, monofloral honey is often valued as the more valuable variety because of its versatility in flavor selection. Honey whose botanical and geographical origin has designated is more likely to be accepted by consumers because of its quality and reliability.

Besides nutritional value, honey is also highly valued for its medicinal uses and has been used as an ingredient in traditional medicine remedies in many countries. There have been many studies demonstrating that the functional components of honey are very diverse such as: antioxidant, anti-inflammatory, anti-viral, anti-cancer and antibacterial (Al-Farsi, Al-Amri, Al-Hadhrami, & Al-Belushi, 2018). These effects are mainly due to the phenolic compounds present in honey, typical flavonoids with antioxidant properties and the ability to scavenge free radicals (Khalil *et al.*, 2012). An antioxidant is a molecule or set of substances found in plants or drugs. These compounds can reduce or prevent the overproduction of free radicals that the "endogenous antioxidant" system cannot destroy. Currently, scientists tend to look for sources of "exogenous antioxidants" (i.e. through foods and drugs into the body) for disease prevention, anti-aging and health promotion potent. Natural antioxidants boost plasma antioxidant capacity and lower the risk of various illnesses, including cancer, heart disease, and stroke (Marinova, Ribarova, & Atanassova, 2005; Pham *et al.*, 2020).

The key focus with antioxidants is phenol and flavonoid molecules, which have been demonstrated to have the potential to quench free radicals, preventing and curing numerous process-related disorders oxidize. They may be found in every part of the plant, including the leaves, fruits, seeds, roots, and bark (Pham *et al.*, 2020). The antioxidants present in honey include both enzymatic substances (glucose oxidase, catalase and peroxidase) and non-enzymatic substances, such as ascorbic acid tocopherol, carotenoids and more than 150 polyphenol compounds, including, flavonoids, phenolic acids, catechins and derivatives cinnamic acid (Al-Farsi *et al.*, 2018; Khalil *et al.*, 2012). Several studies reveal that the principal antioxidant components in various medicinal plants are phenol and flavonoid molecules (Le *et al.*, 2019; Liu, Qiu, Ding, & Yao, 2008; Machu *et al.*, 2015; Toan *et al.*, 2021). The phenolic content of honey was determined, and its correlation with the antioxidant content was recorded (Al-Farsi *et al.*, 2018). In addition, some reports have also noted that polyphenols are thought to influence the color and flavor of honey (Alvarez-Suarez *et al.*, 2010; Isla *et al.*, 2011). Along with that, a positive correlation between phenolic content, color and antioxidant activity of honey was also observed (Bertoncelj, Doberšek, Jamnik, & Golob, 2007).

However, at present, in Vietnam, the beekeeping industry for honey is only in its infancy because it is not possible to fully exploit the value of honey in each locality but only focuses on developing honey sources that have already been developed proven to be effective. At the same time, the published research data on the chemical composition, biological activity, and application of honey is still

very limited, and there is no reliable scientific data to get the full value of domestic honey. Therefore, the main objective of this study is to determine the data on physicochemical composition, content of secondary compounds and antioxidant capacity of common honey samples in Vietnam. From there, provide more information of Vietnamese honey to the world honey database.

Materials and methods

Materials

The materials used in the study were longan flower honey (*Dimocarpus longan*) and honey from the polyfloral (*Micrapis* honeycomb) collected at two natural locations in Ben Tre province in January 2021. Honey after being collected and centrifuged and then preserved. Store at 20 ^{o}C in the dark until use. All chemicals and reagents are for analytical use only.

MO_1: Natural longan flower honey

MO_2: Natural polyfloral honey

MO_3: Control honey collected at the local market.

Analytical procedures

Moisture

Water content was determined by a refractometer (REF-116, UK) reading at 20 ^{o}C, according to the relationship between honey water content and refractive index.

Total acidity

Free acidity was determined by the titrimetric method (AOAC, 2000). Use a solution containing 10 g of honey dissolved in 75 mL of distilled water and titrate with 0.1N NaOH until a pale pink color appears (Eco titrator, Mehtrom, Switzerland). Acidity is calculated as 10 times the volume of NaOH used for titration, expressed in milliequivalents of acid/kg honey (meq/kg).

Ash

Ash content was determined according to the method of AOAC (2000). 5 g of honey is placed in a crucible and then heated at 550 ^{o}C, in an oven for 5 h. After cooling at room temperature, the collected ash was weighed and the results recorded.

Hydroxymethylfurfural (HMF) analysis

The HMF content was determined using the standard AOAC procedure (AOAC, 2000). 5 g of honey dissolved in 25 mL of distilled water, then measure absorbance at wavelengths 284 and 336 (using Aligent Cary 60 UV, visible spectrometer, USA) with the control solution of 0.2% $NaHSO_3$ solution. Absorbance should be less than 0.6, if more significant, it must be diluted with 0.2% $NaHSO_3$ solution. The formula calculates HMF:

$$HMF(mg/kg) = (OD_{284} - OD_{336}) \times 179.7 \times 5 \times D/W \quad (1)$$

Where, D: is the dilution factor, and W: is the sample mass.

Protein analysis

The Kjeldahl method was used to determine protein concentrations.

Stage 1- sample inorganic: Weigh 100 mg of honey sample, 1 g of catalyst is put into the digestion tube + 5 mL of concentrated H_2SO_4, connect the gas collector and start the digestion. When the digestion time is over, allow the digestion tube to cool. Add 50 mL of distilled water, mix well and allow to cool, insert into the still.

Stage 2- distillation: Insert the digestion tube into the distiller, add 80 mL of 32% NaOH. Starting the distillation process, the distillate is transferred to a conical flask containing 20 mL of 4% boric acid solution with a color indicator. Stop distillation when the distillate is no longer NH_3 (test with litmus paper).

Titrate with 0.25N hydrochloric acid (HCl). Stop the titration when a reddish tint appears.

Stage 3- Calculation of results: Through the amount of 0.25N HCl, we know the amount of boric acid combined with NH_3 and therefore know the NH_3 released from

the sample. 1 mL of 0.25N HCl corresponds to 0.0035 g of organic nitrogen.

(2)

$$\%Nitrogen = \frac{0{,}0035 \times 100 \times V_{HCl}}{C}$$

V_{HCl} 0.25N: volume of 0.25N HCl used for titration (mL), and C: sample weight to be determined (g).

Crude Protein Content = Amount of Nitroge n×6.25

Total sugar content

Carbohydrates (simple sugars, oligosaccharides, polysaccharides and their derivatives) react under strong acid conditions and high temperature to give furan derivatives, condense with phenol to form yellow compounds stable and has a maximum absorption intensity at 490 nm. Determination of total sugar content via glucose standard curve by phenol-sulfuric acid method of DuBois, Gilles, Hamilton, Rebers, & Smith (1956). Express unit in g/kg or % (g/100g).

Reduce sugar content

Determination of reducing sugar content according to the method of Miller (1959). Honey solution (0.1 g/mL) was diluted 100 times with water. Take 1 mL of the diluted solution into a test tube, add 1 mL of DNS, and then incubate in a boiling water bath for 10 min. Allow the mixture to cool at room temperature for 10 min, then add 7.5 mL of water and measure the absorbance at 540 nm. The control sample used distilled water. Use glucose solution as standard. Note: The color of the mixture is only produced in alkaline media, so acidic samples must be neutralized before analysis. The reducing sugar content expressed in units of g/kg or % (g/100g), is determined according to the following formula.

(3)

$$X = a \times n \times V$$

Where, X: Amount of reducing sugar in the solution to be determined (g), a: Amount of reducing sugar in the measured sample (g), n: Solution dilution factor, and V: Number of measured fluid volumes (mL).

Reducing sugar content (%) = Total sugar content – reducing sugar

Determination of total phenolics content

Phenolics compounds are able to react with the Folin–Ciocalteu reagent to give a green color product, which has a maximum absorbance at 765 nm. The total phenolics content was determined according to the method of Folin-Ciocalteu, 1927 and referred to the procedure of Socha, Juszczak, Pietrzyk, & Fortuna (2009).

The total phenolics content of honey is determined by gallic acid equivalents (GAE) through the standard curve equation of gallic acid and determined by the following formula:

(4)

$$C = c \times V/m$$

Where, C: total phenolics content (mg/g) of GAE honey, c: concentration of gallic acid determined from the standard curve equation (mg/mL), V: volume of honey (mL), and m: mass of honey (g).

Determination of flavonoid content

Flavonoids in the sample form a yellow complex with $AlCl_3$ solution. The color intensity is proportional to the flavonoid content in the studied sample determined at 510 nm. The flavonoid content was determined according to the method of Zhishen, Mengcheng, & Jianming (1999).

The flavonoid content of honey was determined by the quercetin equivalence value through the quercetin standard curve equation and determined by the following formula:

(5)

$$F = f \times V/m$$

Inside, F: total flavonoid content, mg/g of honey QE, f: concentration of quercetin determined from the standard curve equation (mg/mL), V: volume of diluted

honey solution (mL), and m: mass of honey (g).

1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging method

The method was carried out according to the instructions of Hatano, Kagawa, Yasuhara, & Okuda (1988), with some adjustments. Dilute the sample to the appropriate concentration range, and draw 0.5 mL of the diluted sample into the test tube. The control sample replaced the ethanol extract (99.5%). Then, add 1.5 mL of DPPH solution (OD_{517} nm = 1.1± 0.02) into the test tube and leave in the dark for 30 min. Measure the optical absorbance at 517 nm on a UV-Vis spectrophotometer. Vitamin C (ascorbic acid) was used as the standard for comparison.

The DPPH free radical scavenging activity (IC %) was determined based on the formula:

$$IC\ (\%) = \frac{Abs_C - Abs_T}{Abs_C} \times 100 \quad (6)$$

Inside, Abs_C: Optical absorbance of the control sample and Abs_T: Optical absorbance of the specimen.

The results are recorded based on the IC_{50} value, the concentration at which the sample can reduce 50% of DPPH free radicals.

Statistical analyses

The results were statistically processed using the program Statghraphics Centurions 18.1.12 and Microsoft Excel 365. Analysis of variance (ANOVA) and LSD test was used to conclude about the difference between treatments. The results were reported as a mean ± standard deviation from the triplicate analysis.

Results and discussion

Physicochemical honey characteristics

Honey's water content is a crucial element influencing its quality since high humidity causes spoiling of the product during storage due to the growth of yeast that produces ethyl alcohol and carbon dioxide (Chirife, Zamora, & Motto, 2006). The alcohol will then be converted to acetic acid, resulting in the sour taste of honey. At the same time, numerous studies have found that honey's low moisture content plays a significant role in its resistance to fermentation and shelf life (Finola, Lasagno, & Marioli, 2007). Table (1) shows the findings of the quantitative analysis of longan and fly honey samples collected in Ben Tre province. The water content in the tested samples ranged from 21.2 to 30%. The moisture level of natural honey samples is greater (26.5±0.39 and 30.5±0,25%) than standard of 20%- Codex Alimentarius standard (Codex Alimentarius, 2001). This could be due to the influence of environmental circumstances, hive maturity, and the nectar quality from which the bees obtain their honey (Singh & Bath, 1997).

HMF is a critical factor in determining honey's purity and freshness (Khalil, Sulaiman, & Gan, 2010). HMF, which is typically present in negligible amounts of fresh honey, is derived from dehydration of glucose and fructose (the sugar with the highest percentage in honey). It is made gradually during storage and produced rapidly after honey heating or aging (Abdulkhaliq & Swaileh, 2017). The HMF value is also influenced by a variety of other parameters such as pH, temperature, period of heat exposure of honey, storage conditions, and floral source. Thus the HMF value can be used to determine the freshness of honey and the quality of storage conditions. The HMF level of honey samples was found to range from 33.4 to 75.4 mg/kg, according to the results of this analysis. Both natural honeys (MO_1 and MO_2) have low HMF values, with the exception of MO_3 processed honey, which has a value of 75.4 mg/kg. However, all HMF readings are within the Codex Alimentarius Committee-World Food Standards (WHO) recommended range (80 mg/kg-for nations with tropical climates).

Table 1. Physicochemical parameters of honey samples collected in Ben Tre province

Sample	Physicochemical parameters of honey						
	Moisture (%)	HMF (mg/kg)	Total sugar (%)	Reduce sugar (%)	Total acidity (mL/kg)	Ash (%)	Protein (%)
MO_1	26.5±0.39^{b}	33.4±1.12^{c}	88.52±1.14^{a}	82.24±1.21^{a}	31.75±1.36^{b}	0.37±0.02^{b}	0.38±0.03^{ab}
MO_2	30.5±0.25^{a}	41.9±0.11^{b}	84.58±0.52^{b}	79.42±1.15^{b}	39.13±0.46^{a}	0.38±0.01^{b}	0.25±0.01^{c}
MO_3	21.9±0.13^{c}	75.4±0.18^{a}	87.35±1.21^{a}	82.69±1.04^{a}	32.35±1.57^{b}	0.48±0.01^{a}	0.41±0.04^{a}

Letters in the same column indicate statistically significant difference (P<0.05)

In comparison to the results of Moniruzzaman, Khalil, Sulaiman, & Gan (2013), the HMF values of longan flower honey and polyfloral honey in Vietnam are lower when compared to Malaysian honey of longan flower, melaleuca flower, sorrel flower, and honey Pineapple bees (with HMF values ranging from 63 to 68.99 mg/kg) (Moniruzzaman *et al.*, 2013). This finding is comparable to the HMF level published before by the Chakir, Romane, Marcazzan, & Ferrazzi (2016) for honey originating from Morocco -a tropical nation- with values ranging from 53.38 mg/kg. As a result, it demonstrates that the quality of Vietnam's longan flower and polyfloral honey meets international standards and is more significant or equivalent to honey from other nations in the same region or environment.

Honey contains simple carbohydrates, specifically glucose and fructose, which are known as monosaccharides (simple sugars -reducing sugars) and account for 65-80% of total soluble solids, with the remaining 25% made up of oligosaccharides (disaccharides, trisaccharides, tetrasaccharides- poly sugars) (Da Costa Leite *et al.*, 2000). The higher the simple sugar content, the better the bile. In this investigation, the total sugar costs of natural and processed longan flower honey samples were 88.52 and 87.35%, respectively. This finding exceeds the data collected by the report for longan honey in Malaysia, which had a total sugar content of 55.33-56.67% (Moniruzzaman *et al.*, 2013). When comparing polyfloral honey to Malaysian polyfloral honey, the overall sugar value was 1.5-2 times higher (Moniruzzaman, Sulaiman, Khalil, & Gan, 2013). At the same time, the total sugar content of longan and polyfloral honey is significantly larger than that of honey collected in countries with similar tropical climates, such as Bangladesh (45.3-66.7%), Palestine (79-84%), and Pakistan (69.46-81.63%) (Abdulkhaliq & Swaileh, 2017; Conti, Stripeikis, Campanella, Cucina, & Tudino, 2007; Islam *et al.*, 2012). However, the ratio of total and reducing sugar content in the MO_1 and MO_2 samples in the study was rather high, resulting in an unreduced sugar (sucrose) value that exceeded the 5% mark (the maximum regulatory limit according to the standard Codex Alimentarius) are 6.28 and 5.16%, respectively. The decreasing sugar content of honey changes during honey ripening due to the enzyme invertase from bee saliva or microbial metabolism and enzymes from the stomach of insects present on flowers (White & Maher, 1953). The greater sucrose concentration in these samples could be attributed to nectar collecting during the flowering season -January- when the honey in the nest did not have enough time to convert sucrose to glucose or fructose completely. This also demonstrates that the sugar concentration of honey varies based on regional conditions, the time of collecting, and the source.

The greatest total acidity level was 39.13 meq/kg, according to the values shown in Table (1). This result is lower than the acceptable level (50 meq/kg) of the international honey standard (Codex Alimentarius, 2001) and the allowable

amount (40 meq/kg) of European Union law (Council, 2002). Acidity denotes the presence of organic acids and other substances found in honey (lactones, esters, and inorganic ions) (da Silva, Gauche, Gonzaga, Costa, & Fett, 2016). This characteristic also suggests the presence of proteins, phenolics, and vitamin C, which are chemical components capable of donating H+ atoms found in honey (Halliwell, 2007). Furthermore, it is a characteristic that may be used to assess the quality and freshness of honey because the increase in this index is related to the time when carbohydrates ferment into organic acids. Previous reports on honey from around the world reported free acid values ranging from 17.6 to 41.55 meq/kg (Thailand), 16 to 32 meq/kg (Australia), and 9 to 19 meq/kg (Serbia) (Ajlouni & Sujirapinyokul, 2010; Sakač *et al.*, 2019; Wanjai, Sringarm, Santasup, Pak-Uthai, & Chantawannakul, 2012). As a result, the findings of this study are consistent with the climate conditions in Ben Tre province, Vietnam -a place where it is hot and sunny most of the year, and exposure to high temperatures is a factor that induces an increase in the acid index, HMF in honey.

In terms of ash concentration, the trial findings revealed that the MO_3 sample had the highest ash level of 0.48%, while the natural honey samples had values ranging from 0.37 to 0.38 %. This outcome is consistent with international standards (no more than 0.6%). The ash percentage of the polyfloral honey sample used in this investigation was more significant than the ash content of polyfloral honey in Algeria (0.09-0.21%) (Ouchemoukh, Louaileche, & Schweitzer, 2007). Furthermore, Wanjai *et al.* (2012) showed that Thailand's longan honey sample was lower when compared to Vietnam's longan honey sample, with a recorded value of 0.23%. Furthermore, other honey samples tested in the same study by this group of authors revealed low concentration (0.16-0.31%) (Wanjai *et al.*, 2012). Although conductivity analyses have largely replaced the determination of ash mass fraction in conventional honey quality control settings, ash mass fraction is still regarded as a useful parameter in determining the botanical origin of the honey and distinguishing between honeydew nectar and honey (Anklam, 1998).

Besides, it was discovered that the tested honey samples included a significant level of protein, with the highest value being 0.41%. Natural honey and processed honey had a statistically significant difference (P<0.05). This result is comparable to Abdulkhaliq & Swaileh (2017) report on honey in Palestine, which found values ranging from 0.2 to 0.49%. Simultaneously, the report before measured the protein content of 45 distinct species of polyfloral honey in various regions, with values ranging from 0.1 to 0.5% (Tewari & Irudayaraj, 2004). Although most of the proteins in honey are bee enzymes, high protein content in honey can indicate high pollen levels, indicating that the honey is entirely created from natural, high-quality pollen (Abdulkhaliq & Swaileh, 2017).

Table 2. Average content of phytochemical compounds and antioxidant capacity of honey samples

Sample	Phenolic (mgGAE/g)	Flavonoid (mgQE/g)	Antioxidant activity (IC_{50}, mg/mL)
MO_1	1.020 ± 0.011^b	0.073 ± 0.002^b	85.49 ± 1.25^b
MO_2	1.110 ± 0.025^a	0.089 ± 0.004^a	68.31 ± 0.08^c
MO_3	0.890 ± 0.012^c	0.049 ± 0.002^c	135.45 ± 1.15^a

Letters in the same column indicate statistically significant difference (P<0.05).

Total phenolics, flavonoids content and antioxidant activity

The bioactive chemicals in honey with antioxidant, antibacterial, anti-inflammatory, and tyrosinase inhibitory properties are mostly polyphenol compounds (Machado De-Melo, Almeida-Muradian, Sancho, & Pascual-Maté, 2018). Table (2) displays the average secondary chemical composition and antioxidant capacity of experimental honey samples as measured by the IC_{50} value. The phenolic content (TPC) is a value used to calculate the total phenol content of a mixture of several substances, such as honey. According to Al *et al.* (2009) study, this approach is sensitive and reliable enough for determining the total phenol content of honey. Table (2) shows that the TPC of longan and polyfloral honeys range from 0.890 to 1.110 mgGAE/g. All three samples utilized in the investigation had a statistically significant difference (P<0.05). Recognizing the study's phenolic content's resemblance to that reported by other research groups such as India (0.47-0.98 mgGAE/g), Bangladesh (0.15-0.68 mgGAE/g), Romania (0.23-1.250 mgGAE/g), or Manuka honey (0.89 mgGAE/g), (Al *et al.*, 2009; Alzahrani *et al.*, 2012; Islam *et al.*, 2012; Saxena, Gautam, & Sharma, 2010). Furthermore, the TPC levels in polyfloral honey samples were greater than in Omani (0.78-0.93 mgGAE/g) and substantially higher than in China (0.047 mgGAE/g) (Al-Farsi *et al.*, 2018; Cheung, Meenu, Yu, & Xu, 2019). It was discovered that polyfloral honey contains more TPC than longan flower honey, which can be explained by the combination of different types of honey from different flowers, resulting in anti-inflammatory characteristics. The polyfloral honey samples had superior oxidation. In general, TPC content varied significantly (0.005-1.3 mgGAE/g), (Cheung *et al.*, 2019). The composition and content of these compounds are affected by beekeeping conditions, flower species, harvest time, climatic and soil conditions where the bees are maintained, and honey preservation and processing procedures.

The MO_2 sample had the highest flavonoid concentration (0.053 mgQE/g), while the MO_3 sample had the lowest (0.049 mgQE/g). Furthermore, the concentration of phytochemicals in raw honey samples is higher than in processed honey samples. This value is larger than that of polyfloral honey samples from Nigeria and Spain, which have flavonoid contents ranging from 0.025 to 0.016-0.059 mg/g (Combarros-Fuertes *et al.*, 2019; Ita, 2011). Based on past investigations and study findings, it was discovered that polyfloral honey had higher phenolic and flavonoid content than monofloral honey. Furthermore, both of these factors are related to antioxidant capacity; the more significant the ratio, the better the oxidation resistance of the sample. Flavonoids are plant pigments made up of polyphenols that are generated from the amino acid phenylalanine. Anthocyanins, catechins, flavanone glycosides, flavanones, flavones, flavonol glycosides, flavonols, and isoflavones are among the subclasses. Honey contains flavonoids generated from nectar, pollen, or propolis (Hamdy, Ismail, Al-Ahwal Ael, & Gomaa, 2009). The flavonoid content was assessed by coloring the mixture yellow based on the complex formation of aluminum ions with carbonyl and hydroxyl groups. Simultaneously, flavonoids are thought to be the category of chemicals that accounts for the most significant proportion of polyphenols and are thought to be one of the potential bioactive molecules with health advantages.

As for the free radical scavenging ability, in this study, the DPPH method was chosen to measure the antioxidants in honey because it is an easy, precise and accurate method. In this method, parameter IC_{50} was used as a proxy for the concentration required of the sample to inhibit 50% of DPPH free radicals. Therefore, the low IC_{50} value in honey indicates more effective free radical neutralization. The antioxidant values of the

honey samples in this study ranged from 68.3 to 135.4 mg/mL. The MO_2 sample was found to have more effective antioxidant capacity than the remaining samples with IC_{50} value = 68.49±1.25 mg/mL. At the same time, the flavonoid/phenolic ratio in the MO_2 sample was 8%, which was higher than that of 7.15% in the MO_1 sample and 5.5% in the control sample. This result shows that the antioxidant capacity of polyfloral honey is more effective than that of monofloral honey. This is consistent with previous reports that the antioxidant capacity of polyfloral honeys (with a flavonoid/phenolic ratio from 3%) is better than that of monofloral nectar (usually less than 3%) (Combarros-Fuertes *et al.*, 2019). The difference in antioxidant efficacy of honey also depends on the source of the flower, the time of nectar collection and the geographical location where the flower source grows (Khalil *et al.*, 2010).

Conclusions

By the results obtained based on the evaluation methods used, we can conclude that polyfloral honey has the highest antioxidant activity. Honey with lower antioxidant activity than polyfloral honey is longan flower honey. Processed honey samples also exhibited comparable biological values to honey from other regions of the region. No correlation was observed between the antioxidant activity of honey and its total phenolic content, suggesting that components other than phenolic compounds may influence antioxidant activity. This study has shown that Vietnamese honey contains many natural antioxidants, although the physicochemical values are still some differences compared to international standards. However, in an effort to find a solution to combat the activity of free radicals, honey is a food with potential in the treatment of free radicals addition to the human daily diet.

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خواص فیزیکوشیمیایی، محتویات فنولی و فعالیت آنتی‌اکسیدانی عسل ویتنامی

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چکیده

ویتنام در منطقهٔ آب‌وهوای موسمی گرمسیری (آسیای جنوب شرقی) با تنوع زیستی گونه‌های گیاهی قرار دارد که باعث ایجاد بسیاری از عسل‌های تک‌گل و چندگل شده است. بااین‌حال، تاکنون مطالعه‌ای جامع پیرامون ترکیبات فیزیکوشیمیایی، فعالیت آنتی‌اکسیدانی و محتوای ترکیبات فیتوشیمیایی عسل در این کشور گزارش نشده است. بنابراین، این مطالعه با هدف تعیین خواص فیزیکوشیمیایی، فعالیت آنتی‌اکسیدانی و محتوای فیتوشیمیایی عسل ویتنامی برگرفته‌شده از گل لانگان و عسل طبیعی چندگل (از کندوی زنبور *Micrapis*) انجام شد. روش‌های تحلیلی مرجع براساس مطالعه‌های قبلی روی عسل، اصلاح شد. شاخص‌های تقریبی شامل میزان رطوبت از 21/9 تا 30/5 درصد، میزان خاکستر از 0/37 تا 0/48 درصد، محتوای پروتئین از 0/25 تا 0/41 درصد و غلظت 5-(هیدروکسی‌متیل)-2-فورالدئید (HMF) از 33/4 تا 75/4 درصد متغیر بود. غلظت کل فنول و فلاونوئید از 0/890 تا 1/110 میلی‌گرم گالیک اسید بر گرم و 0/049 تا 0/089 میلی‌گرم کوئرستین بر گرم متغیر بود. همچنین، فعالیت آنتی‌اکسیدانی عسل گل لانگان با مقدار IC_{50} 68/49 میلی‌گرم بر میلی‌لیتر به‌عنوان بهترین فعالیت ثبت شد. علاوه‌بر این، محتوای آب، اسیدیتهٔ آزاد، محتوای قند (گلوکز، فروکتوز و ساکارز)، و HMF آزمایش و ارائه شدند.

واژه‌های کلیدی: ترکیبات فنولی، عسل چندگل، عسل لونگان، عسل ویتنامی، فیزیکوشیمیایی