Malaysian Journal of Chemistry, 2023, Vol. 25(3), 173-183
(Special Issue: *2nd Applied Science Research International Conference 2022*)

Diffuse Reflectance Spectroscopic and FTIR Analysis of Pure and Adulterated Tualang, Manuka, Royal and Kelulut Honey

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Recently, honey adulteration has become a global issue as this fraudulent activity has increased due to increasing consumer demand for honey. The adulterated honey may adversely affect consumers and lead to many chronic diseases due to its high sugar content. One simple and easy method to determine honey authenticity is by using diffuse reflectance spectroscopic (DRS) analysis. The instrumentation setup consists of a light source, fiber-optic contact probes, and a spectrometer connected to a computer. The reflectance value shows a linear relationship with the percentage of sucrose. Therefore, DRS analysis shows the potential to discriminate between pure, adulterated honey and different sucrose concentrations. The data set from DRS were further analysed by using principal component analysis (PCA) method. It shows that different types of honey at different locations in the PCA score plot proved that this technique could be used to determine honey types. The R^2 values obtained were between 0.824 and 0.988. Fourier transform infrared (FTIR) results show no significant difference between adulterated honey with different sucrose levels. However, FTIR results exhibit a difference between pure and adulterated honey.

Keywords: Diffuse reflectance spectroscopy; adulterated honey; principal component analysis; FTIR

Received: August 2022; Accepted: December 2022

Honey is one of the natural ingredients that have many benefits, especially for medical purposes, due to its biochemical composition. Generally, honey contains glucose, fructose, water, and many valuable nutrients such as vitamins (thiamine, riboflavin, niacin, pantothenic acid, pyridoxine, folic acid, ascorbic acid and phylloquinone), flavonoids and minerals (calcium, sodium, potassium, phosphorus, magnesium, selenium, copper, manganese, chromium, zinc and iron) [1]. There are many types of honey such as Tualang honey, Manuka honey, Kelulut honey and Royal honey; each is derived from different sources. All these types of honey have their own chemical composition and biological properties that benefit our bodies. Tualang honey is produced from a kind of bee named *Apis dorsata* or rock bee that can be found in many countries such as Malaysia, Sumatra, and Southern Thailand [2]. In terms of its chemical properties, Tualang honey contains high total phenolic acids and flavonoids and because of that, Tualang honey is good as an anti-oxidant [3]. Besides, study shows that Tualang honey has high radical scavenging activity because of its high ascorbic acid. Ascorbic acid is used to overcome the unbalance of reactive oxygen and nitrogen species that can cause many biologic complications [4]. Manuka honey is categorised as monofloral honey derived from *Leptospermum scoparium* or known as the Manuka tree. Manuka trees can be found in New Zealand and eastern Australia. The study on Manuka honey shows it contains many biological and chemical properties that are beneficial for medical purposes, especially for traumatic wounds, burns, and ulcers [5]. A recent study shows that Manuka honey can also be used for skin medicine which has been demonstrated by using Manuka honey as the primary ingredient in cosmetic and skin care products [6]. Manuka honey is also good for anti-oxidant activity as it contains flavonoid compounds which are isorhamnetin, kaempferol, and galangin [7]. Kelulut honey is produced by a stingless bee species known as *Heterotrigona itama*. Kelulut honey has high carbohydrate, protein, and vitamin C content. All this composition makes Kelulut honey important for medicinal purposes such as helping to reduce anxiety, blood glucose, and breast cancer. A study on breast cell cancer using Kelulut honey as a health supplement shows that the number of viable cells (cancer cell) decreased when the concentration of Kelulut honey increased [8]. Royal honey is produced by worker bees from their glands (mandibular and hypopharyngeal) and it is a creamy substance. Royal honey contains 13.9% total sugars which consist of glucose, fructose, and sucrose. A recent study shows that Royal honey has a lot of amino acids such as lysine, proline, and glutamic acids. Besides, Royal honey also contains beneficial vitamins for maintaining human health such as vitamin E, vitamin B6, thiamine,

†Paper presented at the 2nd Applied Science Research International Conference 2022 (2nd ASRIC 2022)

and 1.5% mineral salt (potassium, phosphorus, magnesium and calcium). A study on immune-regulatory function has proved that protein in Royal honey and antimicrobial peptides such as Jelleines can be used for immunoregulatory activity due to their effective reaction against yeast and gram-positive bacteria. Furthermore, Royal honey can also be used to prevent intestinal diseases by overcoming and reducing ulcerative erosion of colon tissue [9].

Recently, adulteration of honey has emerged as a global issue as this fraudulent activity has increased due to increasing honey demand from consumers. Adulteration of honey changes the composition of the natural structure of honey and diminishes the function of honey as a nutrient supplement for humans. Adulterated honey can affect all consumers because it not only can lead to many chronic diseases but is also being sold at a high price. Commercial and inexpensive syrups or sugars are frequently used to adulterate honey. The adulteration process has two ways which are direct adulteration and indirect adulteration. Direct adulteration is when the amount of foreign substance is added to the honey based on a certain ratio, while indirect adulteration is when the bees are fed with adulterants. Several studies have been published on the addition of sugar or syrup in honey production and the effects it has on the sugar profile, mineral content, phytochemicals, and viscosity. According to Riberio *et al*. (2014), physical and chemical properties of honey such as water activity, mineral content, pH, colour, ash content, and moisture content were varied due to the mixing of high fructose corn syrup (HFCS) with honey [10]. Yilmaz *et al.* (2014) discovered the effect fructose and syrup addition on the chemical, physical and rheological properties of honey [11]. Oraian *et al.* (2018) reported that water activity, pH and electrical conductivity were altered when honey was added with glucose, fructose and inulin syrup [12]. This problem leads to the introduction of many techniques to detect the authenticity of honey, such as near-infrared transflectance spectroscopy, attenuated total reflectance–Fourier transform infrared (ATR–FTIR) spectroscopy, nuclear magnetic resonance (NMR) and fluorescence spectroscopy. Kelly *et al*. (2006) used near-infrared (1100–2498 nm) transflectance spectroscopy to detect HFCS and beet invert (BI) in Irish honey [13]. Riswahyuli *et al*. (2020) reported that they employed ATR-FTIR to investigate the authenticity of the honey but they were unable to differentiate honey based on its origin, except for Bangka Belitung and Sumatera honey [14]. Schievano *et al*. (2020) claimed that they manage to provide an accurate sugar profile of Acacia honey by using NMR that can be utilised to determine its authenticity and quality [15]. Hao *et al*. (2021) reported that the authenticity of Acacia honey could be evaluated by using fluorescence spectroscopy. The three-dimensional fluorescence spectra, fluorescence intensities, and fluorescence lifetime of Acacia honey displayed a significant difference between syrups, concentrated Acacia honey, and pure Acacia honey [16].

However, all these techniques have many limitations such as complex and expensive instruments, and some of them require expertise to operate. DRS is a non-invasive, versatile and non-destructive technique, sensitivities to morphological composition [17] and capable of performing on-site analytical studies rapidly without incurring any damage on the under-investigation sample [18]. This optical measuring approach measures the amount of diffusely reflected light after absorption and scattering [17] and has been used to assess the quality attributes in black tea [19], detect the amount and level of fat and lipid in the liver during liver operation [20] and identify the pigments on Byzantine wall paintings [21]. Hence, this study used DRS, a portable and less time-consuming technique, to check the authenticity of Tualang, Manuka, Royal, and Kelulut honey.

EXPERIMENTAL METHODS AND CHARACTERISATION

Sample Collection

Pure Kelulut honey was collected from a farm in Klang, Selangor, Malaysia, while pure Tualang honey was originally from a forest in Raub, Pahang, Malaysia. Royal honey was collected from farm in Bintulu, Sarawak, Malaysia. Manuka honey was obtained from a farm in Aukland, New Zealand.

Sample Preparation

Pure honey was prepared at 10, 9.5, 9.0, 8.5, and 8.0 mL by using a measuring cylinder before being transferred into a beaker. For adulterated samples, 0.5, 1.0, 1.5, and 2.0 mL of sucrose were added to pure honey. After that, all the adulterated honey was stirred at room temperature to ensure well mixing of the sucrose with the honey. The samples' ID with their composition are tabulated in Table 1. The total number of samples in this study is 20.

Table 1. Sample ID with its composition

Sample	Content
0%	Pure honey (10 ml)
5%	9.5 ml pure honey + 0.5 ml sucrose
10%	9.0 ml pure honey + 1.0 ml sucrose
5%	8.5 ml pure honey + 1.5 ml sucrose
20%	8.0 ml pure honey + 2.0 ml sucrose

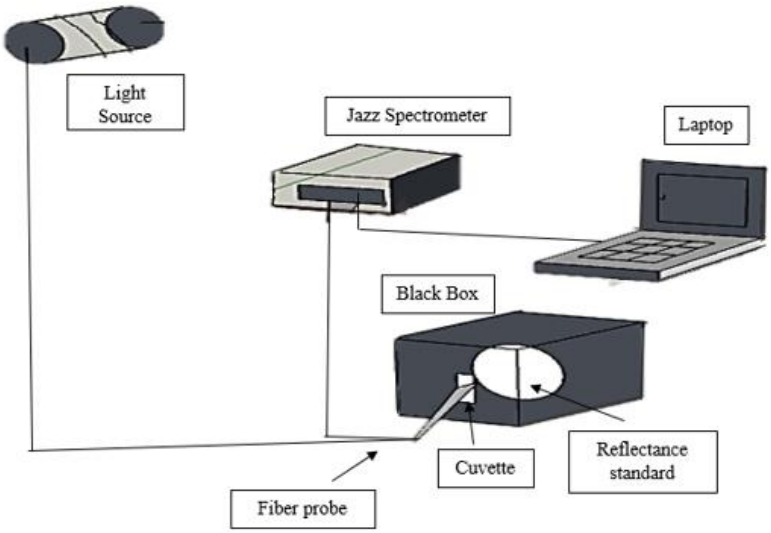


Figure 1. Schematic diagram of the diffuse reflectance spectroscopy instrumentation setup

Diffuse Reflectance Spectroscopic (DRS) Analysis

Honey authenticity was investigated by DRS in the visible range of 400–900 nm. The DRS instruments include the Jazz spectrometer, Ocean Optics (JAZ-EL200-XRI), fiber probe, reflectance standard (Ocean Optics, WS-1), and light source as shown in Figure 1. The fiber probes have two fiber ends where one of the end fibers is connected to the spectrometer while the other end is connected to the light source. The honey was placed in a cuvette for the measurement. The cuvette has precise dimensions (10 mm length) and is transparent to allow the wavelength of light required. The samples were placed in the cuvette in the black box to reduce ambient light from the surrounding which may affect the accuracy of the measurement. All samples were measured 5 times to ensure the data obtained was precise and accurate.

Multivariate Data Analysis

The DRS spectra obtained were analysed by using principal component analysis (PCA) method. The software used to perform PCA was Mathcad v.3. PCA was employed to reduce data redundancy while still maintaining the data accuracy and to help visualise the dataset. The input variables were from DRS data spectra. Normalisation was done through the calibration of the DRS instrument itself. Ocean Optics has a reference standard for calibration, and it was used during calibration process to obtain more accurate and precise results.

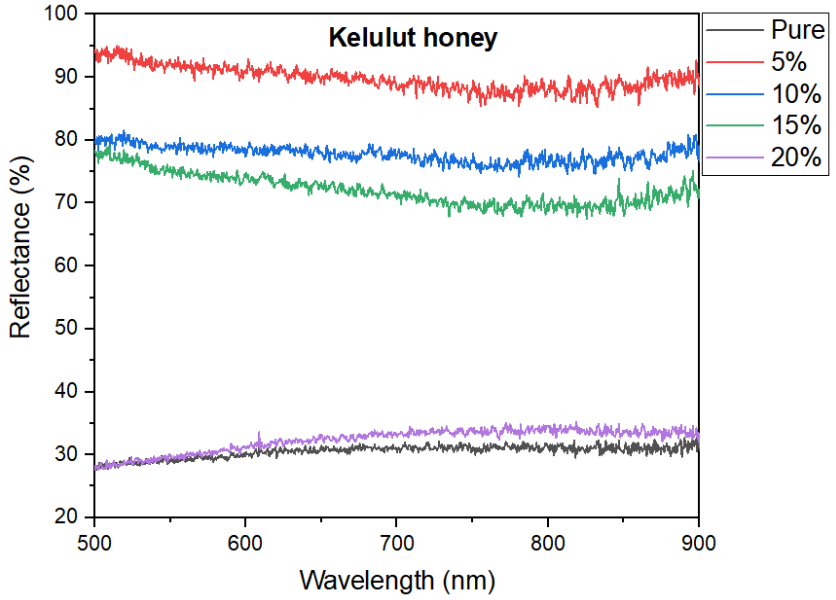
FTIR Analysis

The data obtained from DRS were compared with the data from Fourier transform infrared (FTIR) spectroscopy. It is well known that FTIR has been frequently used to study honey's authenticity [22,23]. A comparison was made to show that the DRS technique can also be employed to determine honey authenticity.

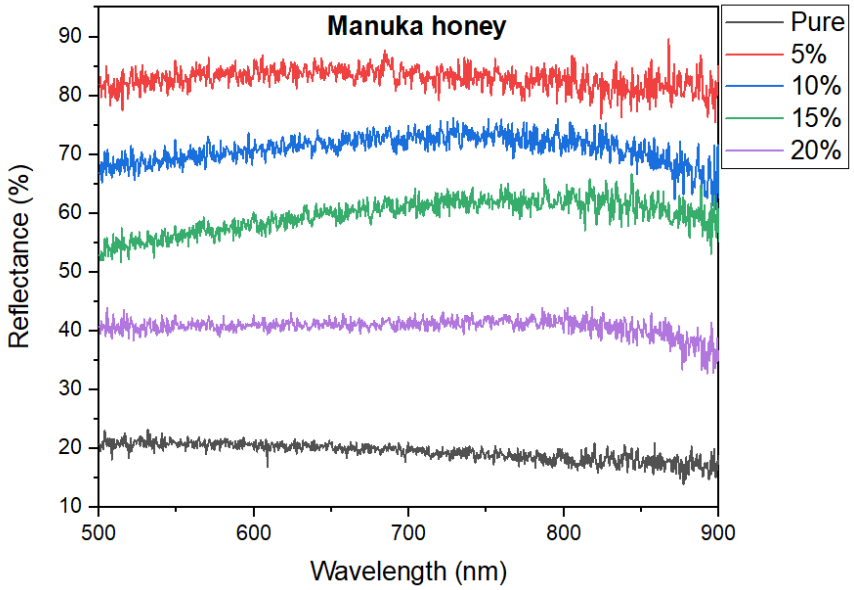
RESULTS AND DISCUSSION

Figure 2 (a)–(d) indicates the percentage of reflectance versus wavelength for pure and adulterated honey for each type. The spectra of pure and adulterated honey obtained were analysed in visible wavelengths between 500 and 900 nm. From the observation, there were different values of reflectance between pure honey and adulterated honey. The reflectance values decreased as the sucrose concentration increased. This is probably due to the changes in density when sucrose was added. Sucrose density (1.55 g/cm^3) is higher than pure honey density (1.11 g/cm^3) [24]. Therefore, as the percentage of sucrose concentration increased, the density of adulterated honey increased. The light source penetrated the sample and is scattered within the sample. The reflectance standard which was placed behind the sample was used for calibration purposes; it has high reflectivity of nearly 100% to obtain ideal reflection properties. The reflection light or outgoing wave known as diffuse reflectance would reveal the information regarding sample composition [25]. In general, as the density increases, it causes difficulty for the light to travel due to the arrangement of atoms that are close to each other. Hence, a lower reflectance value would be obtained for a higher density sample. Pure honey has its own reflectance value and this is probably due to pure honey has a different structure from adulterated honey. This shows that as sucrose was added to the pure honey, the original structure of the honey was altered and disrupted. Each type of pure and adulterated honey has different reflectance value because each of them has its own structural properties due to different levels of sugar crystal that can affect the light scattering. This light scattering effect due to honey adulteration has been observed by several previous studies [26,27]. This study proves that diffuse reflectance spectroscopy can be used as a technique to discriminate between pure and adulterated honey. Besides, this technique can also easily identify the concentration level of adulterants that were used in the adulteration process.

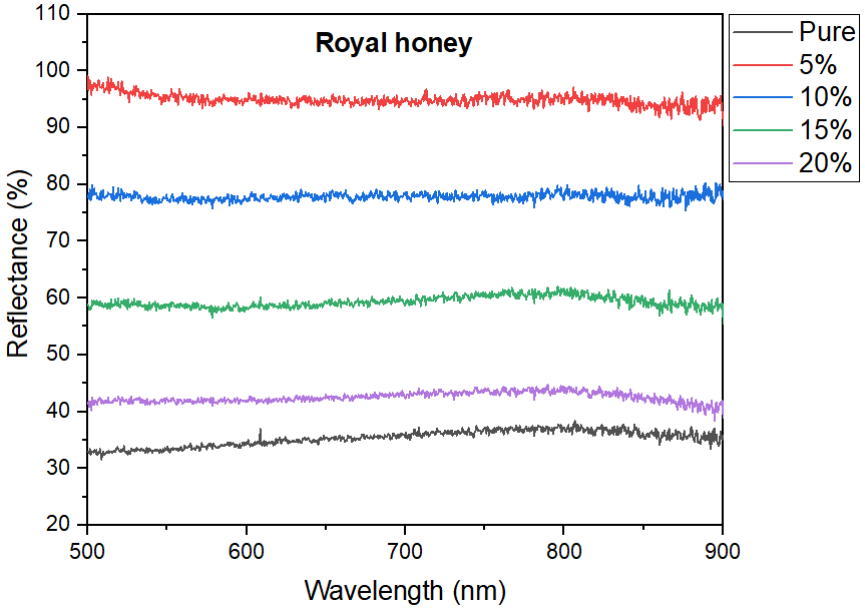
(a)


Kelulut honey
Pure
5%
10%
15%
20%
Reflectance (%)
Wavelength (nm)

(b)


Manuka honey
Pure
5%
10%
15%
20%
Reflectance (%)
Wavelength (nm)

(c)


Royal honey
Pure
5%
10%
15%
20%
Reflectance (%)
Wavelength (nm)

(d)

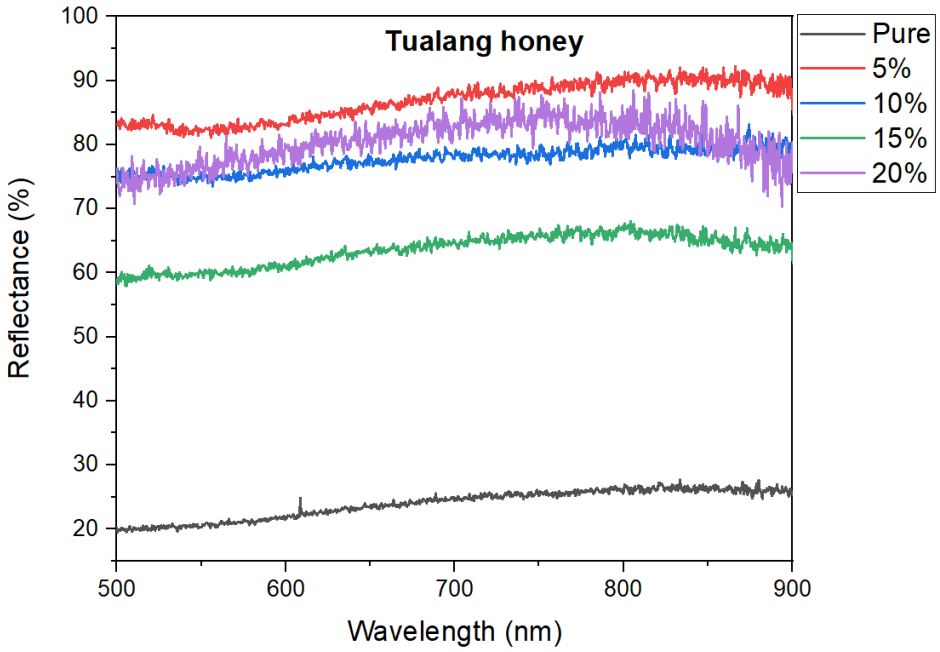


Figure 2. Diffuse reflectance spectra of (a) Kelulut, (b) Manuka, (c) Royal, and (d) Tualang honey.

Figure 3 shows the reflectance values for each type of pure honey: Royal, Kelulut, Tualang, and Manuka. Royal honey had the highest reflectance values, followed by Kelulut, Tualang, and Manuka honey. The difference in reflectance value is probably due to the moisture level. Manuka honey has the lowest moisture content (17.40 %), followed by Tualang honey (26.51 %), Kelulut honey (31.00 %) [28], and Royal honey (60.00%) [29]. Hence, the moisture content might affects the ability of light to pass through the sample. The sample with higher water content allows more light to pass through the sample and get reflected. Therefore, the highest reflectance value was obtained for Royal honey and the lowest reflectance value for Manuka honey. For Manuka honey, the structural network might have reduced the reflectance at a higher wavelength.

Figure 4 (a) – (d) shows the principal component 1 (PC1) versus the sucrose concentration for Kelulut, Manuka, Royal and Tualang honey. The R^2 values obtained were between 0.824–0.988. PC1 increased with the increment of honey concentration for all types of honey. Figure 5 shows the principal component 2 (PC2) versus PC1 for pure honey. All the pure honey are located at different locations, and this shows that PCA could be used to discriminate different types of pure honey.

The FTIR spectra for pure and adulterated honey are shown in Figure 6 (a) – (d). These spectra were analysed to discriminate between pure and adulterated honey for all types of honey. The absorption bands are observed in the region between 650 and 4000 cm^{-1}. These regions reveal the information about the molecular vibrations and chemical composition of the samples. It was observed that there are a spectral differences between pure honey and adulterated honey for Tualang, Manuka and Royal honey, but not for Kelulut honey. This spectral difference is due to the addition of sucrose, which can alter the structure and composition of pure honey. The peak that exists at certain wavelengths indicates the compositions and bonds inside the honey sample. For Kelulut honey, the spectra shown for both pure and adulterated honey are almost similar. Previous studies have shown that Kelulut honey contains the highest value of sucrose (0.025– 32.30 g/100 g) [28], followed by Manuka honey (0.62– 2.36 g/100 g), Royal honey (0.1–2.1 g/100 g) [9] and Tualang honey (0.01–1.66 g/100 g) [28]. Therefore, any addition of sucrose will not cause significant changes in the composition of Kelulut honey. Hence, there was no significant difference between pure and adulterated honey for Kelulut honey. The characteristic peaks at 3395, 3390, and 3400 show the presence of water (O–H stretching vibration). Besides, the weak stretching at 2932 cm^{-1} indicates C–H stretching [14,30]. Furthermore, the weak stretching at 1632 cm^{-1} is associated with C=O stretching in carboxylic acid.

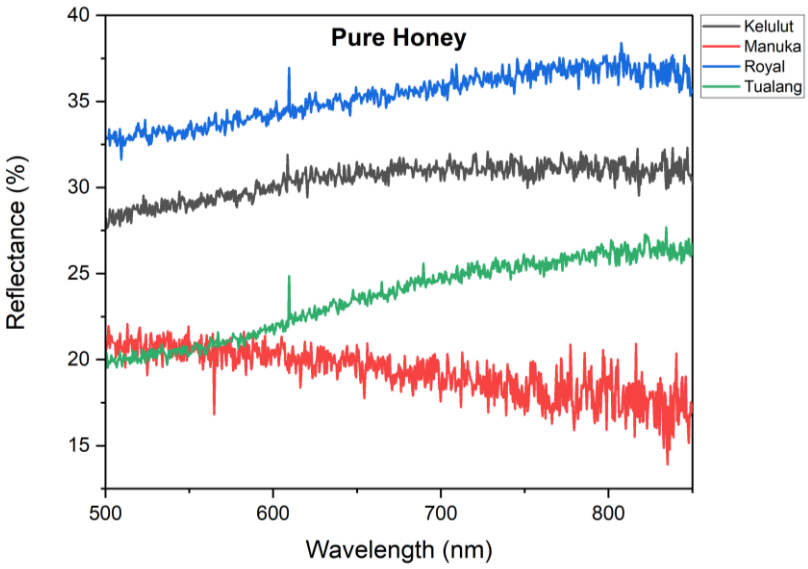
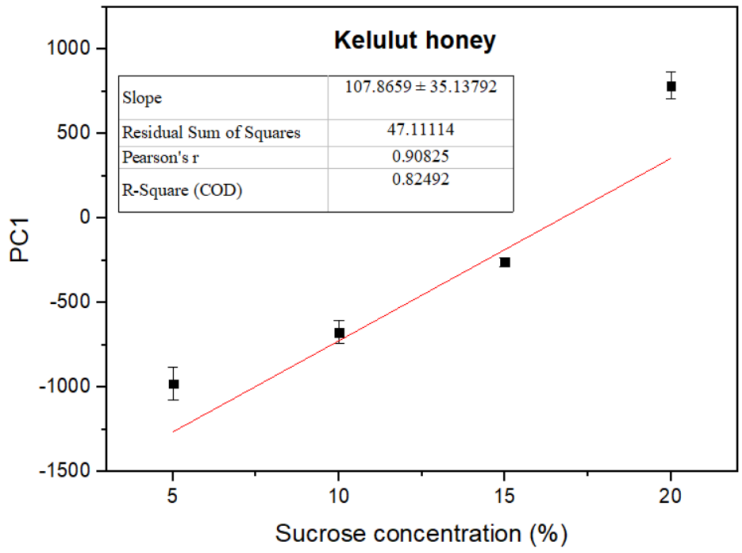
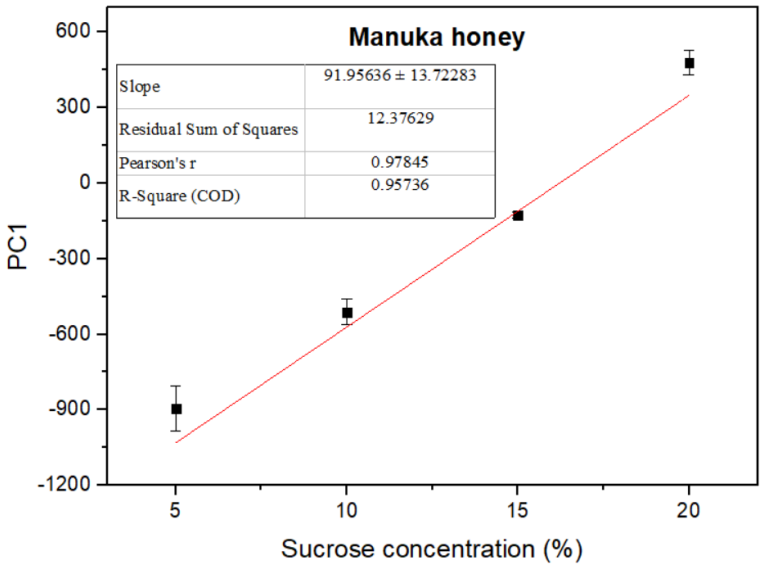


Figure 3. Diffuse reflectance spectra of pure Manuka, Royal, Kelulut and Tualang honey.

(a)



(b)



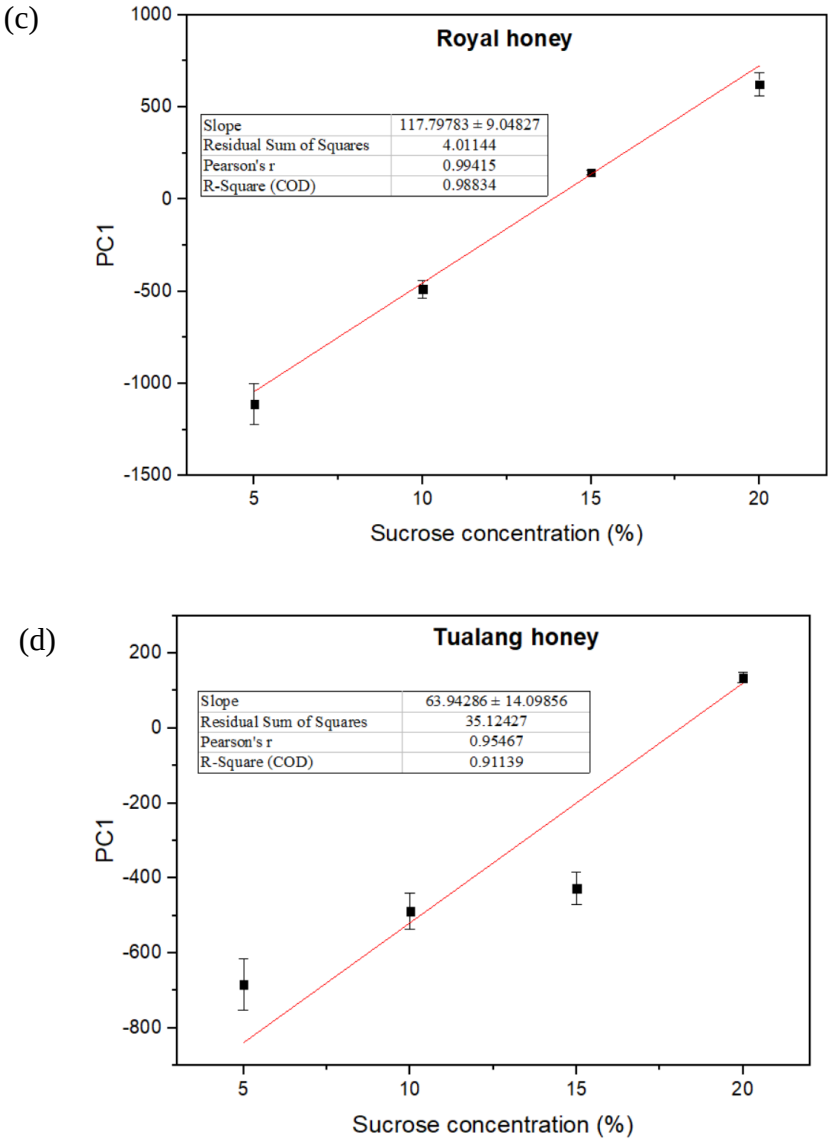


Figure 4. Principal component analysis of DRS for (a) Kelulut (b) Manuka (c) Royal and (d) Tualang honey with different sucrose concentrations

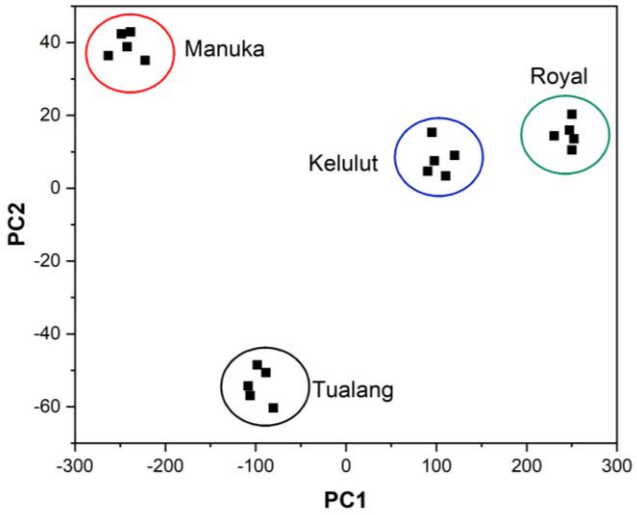
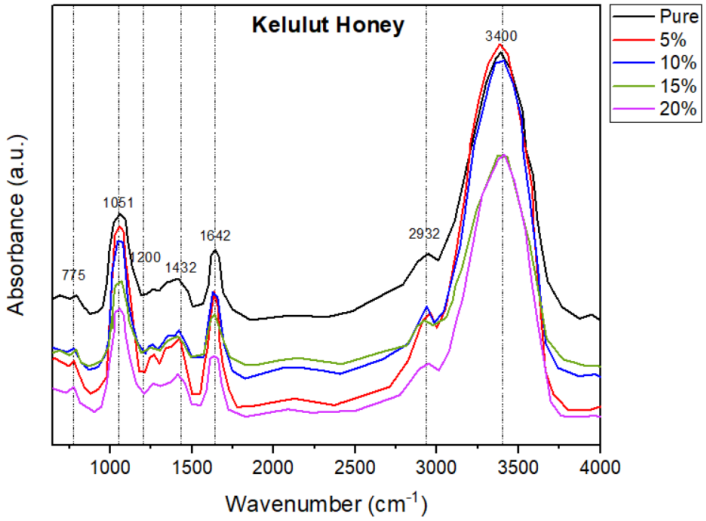


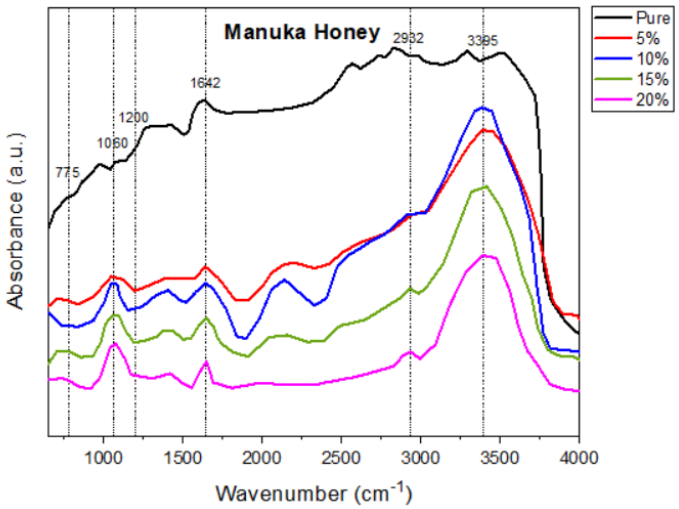
Figure 5. Principal component 2 (PC2) versus principal component 1 (PC1) of DRS for pure honey.

The absorptions bands around 1500–1200 cm^{-1} are attributed to the deformation of an angular deformation of C–C–H and H–C–O linkage [31]. The most important spectra are located between 1500–650 cm^{-1} because of major absorptions of monosaccharides such as fructose and glucose and disaccharides such as sucrose in honey. The region 1180–750 cm^{-1} indicates the anomeric region, which contains anomalous peaks that correspond to the spectra analysis of carbo-hydrates in IR spectroscopy and also indicates the presence of sucrose [31]. These regions are utilised to discriminate between pure and adulterated honey. The absorption peaks observed at 1051 cm^{-1} (Kelulut), 1060 cm^{-1} (Manuka), 1050 cm^{-1} (Royal) and 1049 cm^{-1} (Tualang) are C–O stretching in the C–OH group and C–C stretching in the carbohydrate structure [32]. These peaks are less intense in the pure honey samples except for Kelulut honey, which has high sucrose content. Furthermore, the weak absorption peaks at 921 cm^{-1} (Tualang) and 775 cm^{-1} (Manuka, Royal, and Kelulut) correspond to C–H bending in the carbohydrate [32,33]. For overall observation, the peak observed between 1180 –750 cm^{-1} does not show any significant difference with increasing percentage of sucrose.

(a)



(b)



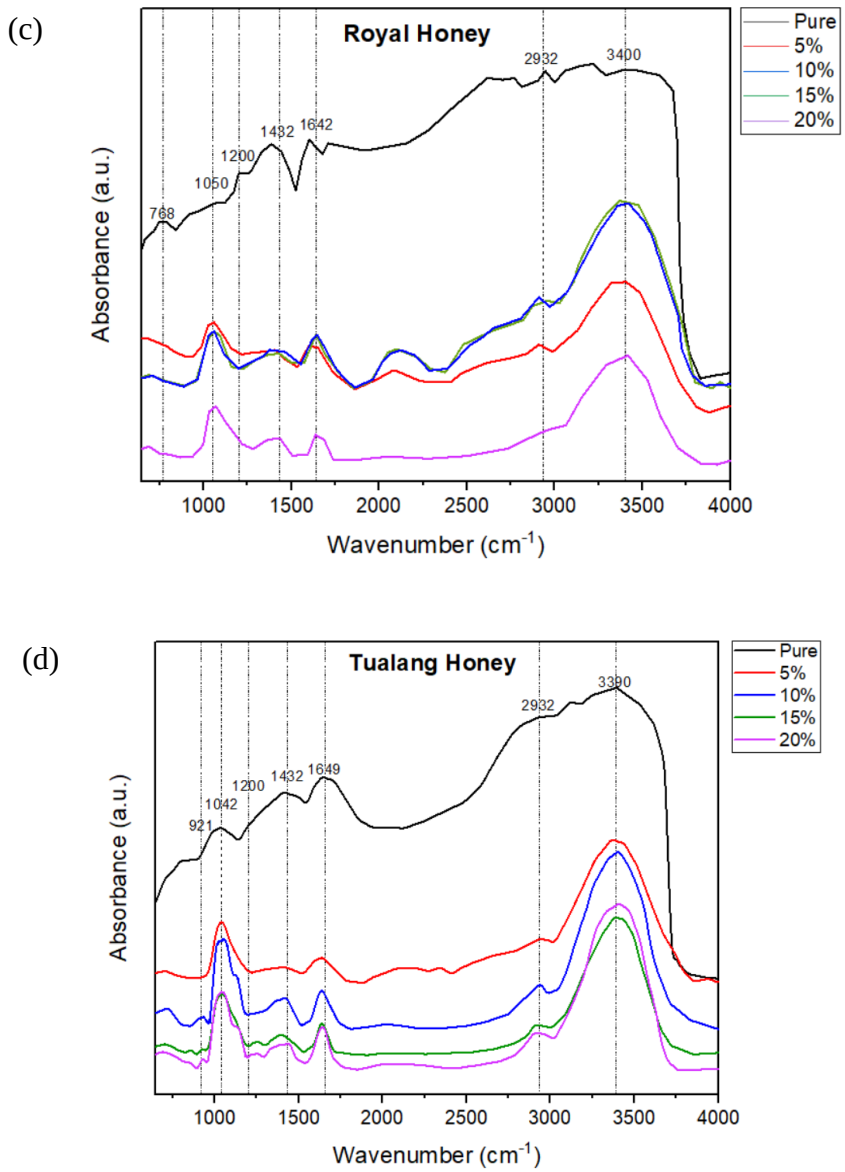


Figure 6. Fourier transform infrared spectra of (a) Kelulut, (b) Manuka, (c) Royal, and (d) Tualang honey.

CONCLUSION

DRS is a suitable technique that has the potential to be used in the identification of honey adulteration activity. Therefore, it can be utilised to discriminate between pure, adulterated honey, and different sucrose concentrations. This study also proved that the DRS technique is less time-consuming and it is also portable. PCA successfully showed the different types of honey used. The different types of honey at different locations in the PCA score plot proved that this technique could be used to determine honey types. The sucrose concentrations were quite difficult to be determined using FTIR because they showed the same pattern for adulterated honey with different concentrations. For Kelulut honey, the authenticity was unable to be determined by FTIR. However, FTIR is an efficient technique to determine the chemical bonding present in the sample. In conclusion, DRS provides more advantages as an instrument, as it is easy to handle, portable, and less time consuming compared to FTIR, which requires a complex instrument and is difficult to bring everywhere.

ACKNOWLEDGEMENTS

The work is financially supported by the Universiti Teknologi MARA through the MyRA grant (No. 600-RMC/MYRA 5/3/LESTARI (080/2020)). The authors also acknowledge the Faculty of Applied Sciences for the experimental and characterization facilities.

REFERENCES

1. Ajibola, A., Chamunorwa, J. P., Erlwanger, K. H. (2013) Dietary supplementation with natural honey promotes growth and health of male and female rats compared to cane syrup. *Scientific Research and Essays*, **8(14)**, 543–553.

2. Mohamed, Z. B. H., Alfarisi, H. A. H., Abdullah, N. Z., Harun, N., Muhammad, N. and Rahim, R. A. (2017) Renoprotective role of tualang honey against high cholesterol diet induced acute kidney diseases in an animal model. *Journal of Applied Pharmaceutical Science*, **7(12)**, 97–101.

3. Ahmed, S. and Othman, N. H. (2013) Review of the medicinal effects of tualang honey and a comparison with Manuka honey. *Malaysian Journal of Medical Sciences*, **20(3)**, 6–13.

4. Kishore, R. K., Halim, A. S., Syazana, M. S. N. and Sirajudeen, K. N. S. (2011) Tualang honey has higher phenolic content and greater radical scavenging activity compared with other honey sources. *Nutrition Research*, **31**, 322–325.

5. Alvarez-Suarez, J., Giampieri, F. and Battino, M. (2013) Honey as a source of dietary antioxidants: structures, bioavailability and evidence of protective effects against human chronic diseases. *Current Medicinal Chemistry*, **20**, 621–638.

6. Woodward, S. (2019) Moisture-associated skin damage: Use of a skin protectant containing manuka honey. *British Journal of Nursing*, **28(6)**, 329–335.

7. Alvarez-Suarez, J. M., Gasparrini, M., Forbes-Hernández, T. Y., Mazzoni, L. and Giampieri F. (2014) The composition and biological activity of honey: A focus on manuka honey. *Foods*, **3**, 420–432.

8. Ramli, N. Z., Chin, K. Y., Zarkasi, K. A. and Ahmad, F. (2019) The beneficial effects of stingless bee honey from Heterotrigona itama against metabolic changes in rats fed with high-carbohydrate and high-fat diet. *International Journal of Environmental Research and Public Health*, **16**, 4987.

9. Guo, J, Wang, Z, Chen, Y, Cao, J., Tian, W., Ma, B. and Dong, Y. (2021) Active components and biological functions of royal jelly. *Journal of Functional Foods*, **82**, 104514.

10. Ribeiro, R. D. O. R., Mársico, E. T., Carneiro, C. D. S., Monteiro, M. L. G., Júnior, C. C., Jesus, E. F. O. de. Detection of honey adulteration of high fructose corn syrup by low field nuclear magnetic resonance (LF ^{1}H NMR). *Journal of Food Engineering*, **135**, 39-43.

11. Yilmaz, M. T., Tatlisu, N. B., Toker, O. S, Karaman, S. Dertli, E., Sagdic, O. and Arici, M. (2014) Steady, dynamic and creep rheological analysis as a novel approach to detect honey adulteration by fructose and saccharose syrups: correlations with HPLC-RID results. *Food Research International*, **64**, 634–646.

12. Oroian, M., Ropciuc, S. and Paduret, S. (2018) Honey adulteration detection using Raman spectroscopy. *Food Analytical Methods*, **11**, 959–968.

13. Kelly, J. D., Petisco, C. and Downey, G. (2006) Potential of near infrared transflectance spectroscopy to detect adulteration of Irish honey by beet invert syrup and high fructose corn syrup. *Journal of Near Infrared Spectroscopy*, **14**, 139–146.

14. Riswahyuli, Y,. Rohman, A., Setyabudi, F. M. C. S. and Raharjo, S. (2020) Indonesian wild honey authenticity analysis using attenuated total reflectance-fourier transform infrared (ATR-FTIR) spectroscopy combined with multivariate statistical techniques. *Heliyon*, **6(4)**, e03662.

15. Schievano, E., Sbrizza, M., Zuccato, V., Piana, L. and Tessari, M. (2020) NMR carbohydrate profile in tracing acacia honey authenticity. *Food Chemistry*, **309**, 125788.

16. Hao, S., Li, J., Liu, X., Yuan, J., Yuan, W., Tian, Y. and Xuan, H. (2021) Authentication of acacia honey using fluorescence spectroscopy. *Food Control*, **130**, 108327.

17. Yu, B., Fu, H. L. and Ramanujam, N. (2011) Instrument independent diffuse reflectance spectroscopy. *Journal of Biomedical Optics*, **16(1)**, 011010.

18. Cheilakou, E., Troullinos, M. and Koui, M. (2014) Identification of pigments on Byzantine wall paintings from Crete (14th century AD) using non-invasive fiber optics diffuse reflectance spectroscopy (FORS). *Journal of Archaeological Science*, **41**, 541–555.

19. Panigrahi, N., Bhol, C. S., Das, B. S. (2016) Rapid assessment of black tea quality using diffuse reflectance spectroscopy. *Journal of Food Engineering*, **190**, 101–108.

20. Reistad, N., Nilsson, J. H., Bergenfeldt, M., Rissler, P. and Sturesson, C. (2019) Intraoperative liver steatosis characterization using diffuse reflectance spectroscopy. *HPB*, **21(2)**, 175-180.

21. Cheilakou, E., Troullinos, M., Koui, M. (2014)

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22. Elhamdaoui, O., El Orche, A., Cheikh, A., Mojemmi, B., Nejjari, R. and Bouatia, M. (2020) Development of fast analytical method for the detection and quantification of honey adulteration using vibrational spectroscopy and chemometrics tools. *Journal of Analytical Methods in Chemistry*, 2020, 8816249.

23. Singh, B. and Barman, S. (2021) Rapid and precise discrimination between pure and adulterated commercial Indian honey brands using FTIR spectroscopy and principal component analysis. *Current Nutrition and Food Science*, **18(9)**, 780–784.

24. Kinoo, M. S., Mahomoodally, M. F. and Puchooa, D. (2012) Anti-microbial and physico-chemical properties of processed and raw honeys of Mauritius. *Advances in Infectious Diseases*, **2**, 25–36.

25. Li, S., Ardabilian, M. and Zine, A. (2021) Quantitative analysis of skin using diffuse reflectance for non-invasive pigments detection. In: *VISIGRAPP 2021 - Proceedings of the 16th International Joint Conference on Computer Vision, Imaging and Computer Graphics Theory and Applications*, **4**, 604–614.

26. Raypah, M. E., Omar, A. F., Muncan, J., Zulkurnain, M. and Abdul Najib, A. R. (2022) Identification of stingless bee honey adulteration using visible-near infrared spectroscopy combined with aquaphotomics. *Molecules*, **27**, 2324.

27. Rust, A., Marini, F., Allsopp, M., Williams, P. J. and Manley, M. (2021) Application of ANOVA-simultaneous component analysis to quantify and characterise effects of age, temperature, syrup adulteration and irradiation on near-infrared (NIR) spectral data of honey. *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, **253**, 119546.

28. Kamal, D. A. M., Ibrahim, S. F., Kamal, H., Kashim, M. I. A. M., Mokhtar, M. H. (2021) Physicochemical and medicinal properties of Tualang, Gelam and Kelulut honeys: A comprehensive review. *Nutrients*, **13**, 197.

29. Ab Hamid, N., Bakar, A. B. A., Mohamed, M. (2018) Phytochemical analysis and GC-MS profile of royal jelly from selected areas in Malaysia. *Malaysian Applied Biology*, **47(3)**, 101–107.

30. Huang, F., Song, H., Guo, L., Guang, P., Yang, X., Li, L., Zhao, H. and Yang, M. (2020) Detection of adulteration in Chinese honey using NIR and ATR-FTIR spectral data fusion. *Spectrochimica Acta - Part A: Molecular and Biomolecular Spectroscopy*, **235**, 118297.

31. Se, K. W., Ghoshal, S. K, Wahab, R. A., Ibrahim, R. K. R. and Lani, M. N. (2018) A simple approach for rapid detection and quantification of adulterants in stingless bees (Heterotrigona itama) honey. *Food Research International*, **105**, 453–460.

32. Cengiz, M. F. and Durak, M. Z. (2019) Rapid detection of sucrose adulteration in honey using Fourier transform infrared spectroscopy. *Spectroscopy Letters*, **52(5)**, 267–273.

33. Riswahyuli, Y., Rohman, A., Setyabudi, F. M. C. S. and Raharjo, S. (2020) Indonesian wild honey authenticity analysis using attenuated total reflectance-fourier transform infrared (ATR-FTIR) spectroscopy combined with multivariate statistical techniques. *Heliyon*, **6**, e03662.