DEVELOPMENT OF INTELLIGENT CLASSIFIER AND ESTIMATOR FOR TUALANG HONEY PURITY

by

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Thesis submitted in fulfilment of the requirements for the degree of Master of Science

August 2014

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LIST OF ABBREVIATIONS

AG	Agromas
ANN	Artificial Neural Network
AS	As-Shifa
ATR	Attenuated Total Reflectance
BR	Bayesian Regulation
BS	Beetroot Sugar
CS	Cane Sugar
DNA	Deoxyribonucleic Acid Analysis
E-Nose	Electronic Nose
EBP	Error Back Propagation
FTIR	Fourier Transform Infrared Spectroscopy
GCMS	Gas Chromatography Mass Spectrometry
HLF	High Level Fusion
HMF	Hyroxymethylfurfural
HPAED-PAD	High Performance Anion Exchange Chromatography Coupled with Pulsed Electrochemical Detection
HPLC	High Performance Liquid Chromatography

ILF	Intermediate Level Fusion
JDL	Joint Directors of Laboratories
LDA	Linear Discriminant Analysis
LLF	Low Level Fusion
LM	Levenberg-Marquardt
Logsig	Logarithmic Sigmoidal
MLP	Multi-Layer Perceptron
MLFF	Multi-Layer Feed Forward
MAE	Mean Absolute Error
NATO	North Atlantic Treaty Organization
NIR	Near Infrared Spectroscopy
PCA	Principal Component Analysis
Purelin	Pure Linear Sigmoidal
QN	Quasi-Newton
RBP	Resilient Backpropagation
SLFF	Single Layer Feed Forward
SLP	Single Layer Perceptron
SPME	Solid Space Microextraction
ST	Syair Timur

Tansig	Hyperbolic Tangent Sigmoidal
T	Tayyibah
T3	Tualang Tiga
TK	Tualang King
TLH	Tualang TLH
TN	Tualang N'Apis
UV-VIS	UV Visible
WT	Wild Tualang
YB	Yubalam Bahtera

PEMBANGUNAN PENGELAS DAN PENGANGGAR PINTAR UNTUK KETULENAN MADU TUALANG

ABSTRAK

Madu adalah bahan semulajadi yang terkenal sebagai makanan tambahan bagi mengekalkan kesihatan yang baik. Ia juga berguna sebagai ramuan dalam ubat-ubatan. Walau bagaimanapun, harga pasaran madu tulen yang mahal telah menyebabkan pihak yang tidak bertanggungjawab mencemari madu tulen dengan penambahan pelbagai jenis cecair gula. Adalah amat mencabar untuk menghasilkan kaedah yang sesuai untuk membuktikan kewujudan bahan dicemari di dalam madu tulen. Kebanyakan kaedah terdahulu melibatkan penelitian data daripada pakar lantas mengambil masa yang lama. Kajian ini mencadangkan pembangunan pengelas pintar bagi membantu kerja membezakan madu tulen daripada yang dicemari. Selain pengelas pintar, kajian ini juga telah membangunkan satu penganggar pintar bertujuan memberikan anggaran peratusan madu tulen yang wujud dalam sampel madu dicemari. Sistem pengelas dan penganggar madu tulen ini dibangunkan menggunakan kaedah Rangkaian Neural Buatan (ANN). Sebanyak sepuluh jenis madu tulen yang berbeza jenama dan sebatian gula telah digunakan bagi menyediakan sampel madu tulen dan madu dicemari (pada peratusan madu tulen yang berbeza). Data hidung elektronik (E-Nose) dan Fourier Transform Infrared Spectroscopy (FTIR) mentah telah diperolehi daripada pelbagai sampel madu. Data E-Nose, FTIR dan data gabungan E-Nose dan FTIR pada tahap rendah yang mentah dan ternormal telah digunakan untuk melatih beberapa ANN bagi

menghasilkan pengelas (madu tulen atau dicemari) dan penganggar pintar (pecahan madu tulen). Keputusan kajian menunjukkan pengelas pintar madu tulen yang dibangunkan menggunakan data E-Nose, FTIR dan gabungan masing-masing memberikan ketepatan pengelasan sebanyak 100% dengan tempoh pembelajaran selama 0.390 saat, 99.72% dengan tempoh pembelajaran selama 0.359 saat and 100% dengan tempoh pembelajaran selama 0.094 saat. Perbandingan keputusan menunjukkan bahawa pengelas pintar madu tulen yang memberikan prestasi yang terbaik adalah yang dilatih dengan data gabungan. Sistem penganggar pintar pecahan madu tulen yang telah dibangunkan masing-masing memberikan purata ralat mutlak sebanyak 16.55%, 6.11% dan 4.88% untuk data E-Nose, FTIR dan gabungan. Keputusan kajian ini telah mendedahkan bahawa pengelasan dan penganggar pintar pecahan ketulenan madu yang dibangunkan berdasarkan data mentah gabungan E-Nose dan FTIR pada tahap rendah adalah lebih baik daripada data tunggal E-Nose atau FTIR. Namun, ketepatan anggaran pecahan madu tulen sebanyak 30.9% menunjukkan bahawa prestasi penganggar pintar pecahan ketulenan madu masih perlu dibaiki. Keseluruhannya, kajian ini telah menunjukkan bahawa ANN mempunyai potensi dalam membantu kerja membezakan madu tulen daripada madu dicemari dan menganggar pecahan madu tulen dalam sampel madu dicemari.

DEVELOPMENT OF INTELLIGENT CLASSIFIER AND ESTIMATOR FOR TUALANG HONEY PURITY

ABSTRACT

Honey is a natural substance well-known as supplement for maintaining good health. It is also useful as an ingredient in medicine. However, the market price of pure honey is expensive, causing irresponsible parties to adulterate pure honey by adding various sugar substances. It is very challenging to come out with a suitable method to prove the presence of adulterants in honey products. Most previous studies involved close data observation from experts that is time-consuming. This research proposes the development of intelligent classifier to aid the task of differentiating pure honey from adulterated ones. Besides intelligent classifier, this research has also developed an intelligent estimator for the purpose of giving a percentage estimation of pure honey that exists in adulterated honey sample. The pure honey classifier and estimator are developed using Artificial Neural Network (ANN) approach. Ten types of pure honey from different brands and sugar compounds have been used to prepare various pure and adulterated honey (at different percentages of pure honey) samples. Electronic nose (E-Nose) and Fourier Transform Infrared Spectroscopy (FTIR) raw data have been gathered from various honey samples. The E-Nose, FTIR low level Fusion data of E-Nose and FTIR of raw and normalized data have been used to train a number of ANNs to produce intelligent classifiers (pure or adulterated honey) and estimators (fraction of pure honey). The research results showed that intelligent pure honey clas-

sifiers developed using E-Nose, FTIR and Fusion data gives classification accuracies of 100% with 0.390 seconds training time, 99.72% with 0.359 seconds training time and 100% with 0.094 seconds training time, respectively. The result comparison show that the intelligent pure honey classifier gives the best performance is the one trained with Fusion data. The developed intelligent pure honey fraction estimator systems gave average absolute errors of 16.55%, 6.11% and 4.88% for E-Nose, FTIR and Fusion data, respectively. The research result has revealed that intelligent pure honey classifier and estimator developed based on raw E-Nose and FTIR low level Fusion data is better than those using single E-Nose or FTIR data. However, correct pure honey fraction estimation of 30.9% suggests that the performance of intelligent pure honey fraction estimator still needs to be improved. Overall, this research has shown that ANN has the potential to aid the tasks of differentiating pure honey from adulterated ones and estimate the fraction of pure honey in adulterated honey samples.

CHAPTER 1

INTRODUCTION

1.1 Background on Honey

South East Asia, particularly Malaysia is rich in natural forest resources such as honey. Honey is a natural sweet substance produced by honeybee, *Apis Dorsata* from the nectar plants or from secretions of living parts or excretions of plant sucking insects on the living parts of plants. Bees are the natural source in producing honey and the production of honey depends on the geographical areas and different varieties of plant (Molan, 1992).

Honey starts out as nectar that bees collect from flowers. Basically, nectar is a reward that plants produce to attract pollinating insects and birds. Sugar fluid which includes the aromatic oils that give flowers their scent, as well as other trace substances. Bees collect this nectar by drawing it through their proboscis and storing it in their honey stomach. Honeybees then carry it back to their hive in tiny, 40-milligram loads.

The foraging bees regurgitate the nectar and pass it to worker bees in the hive. These bees then gradually transform the nectar into honey by evaporating most of the water from it. Nectar is as much as 70% water, while honey is only about 20% water (Leelaw, 2012). Bees get rid of the extra water by swallowing and regurgitating the nectar over and over. They also fan their wings over the filled cells of the honeycomb. This process retains lots of sugar and the plant's aromatic oils while adding enzymes from the bees' mouths. The finished honey is thick, sticky and very sweet. It contains several types of sugar, including sucrose, laevulose and dextrose. Its flavor and color depends on the flowers from which the bees harvested their nectar (Leelaw, 2012).

Honey naturally contains at least 181 substances which is a complex mixture of water, sugar contents (fructose, sucrose, glucose, and maltose), proteins and amino acids, vitamins, minerals and special enzymes (White, 1957). Table 1.1 shows a typical list of honey composition. The composition of honey depends on the geographical and source of nectar such as monofloral (one type of flower), polyfloral (various types of flower) or blended honey (mixed of various type of honey).

Table 1.1: List of typical honey composition

Nutrient	Average amount in 100g of honey	Allowable Range
Water	17.1 g	12.2-22.9 g
Carbohydrate (total)	82.4 g	
Fructose	38.5 g	25.2-44.4 g
Glucose	31.0 g	24.6-36.9 g
Maltose	7.2 g	0.50-2.90 g
Protein,Amino acid, Vitamin and mineral	50.0 g	
Energy	304 Kcal	

Honey is a common sweetener for food and it can be consumed by all level of ages from infant to the elderly. As a natural multivitamin, honey has many benefits for expectant mothers to increase their stamina and maintain good health during pregnancy. Due to its high carbohydrate content and functional properties, honey is an excellent source of energy or supplement for athletes, adult and children (Blasa et al., 2006).

Since ancient times, honey has been used as medicine. Doctors and surgeon have used honey in their medical practice and even recommended its use. This is affirmed in verses 68-69 of surah al-Nahl in the Quran which state:

> "*And your Lord inspired the bee: Build your homes in the mountain and in the trees and in the hives made by mankind. Then (he taught the bee) to feed on every kind of fruit (of the earth) and to follow the ways of your Lord made smooth. There comes from inside their bellies a drink of diverse colours in which is healing for mankind. Surely in this is a sign for those people who reflect*"
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> (al-Nahl, 16, 68-69)

In many cultures, honey has been proven to be very effective in the treatment of burns, ulcers and wounds (Postmes et al., 1993; Coulston, 2000). Low water contents while high sugar contents in honey prevent the growth of microorganisms in wounds and effectively helps to dry the wounds. It has been observed to kill bacteria at the site of the wounds, clean, rapidly replace dead cells and allow formation of tissues (scar).

1.2 Overview of Honey Purity Research

Due to the advantages of honey for human health, many honey-related research works have been carried out, particularly on its quality. Quality assessment of honey is often related to its purity, which is identified through its flavor, chemical contents and nutritional values. These qualities are usually the prime criteria in distinguishing pure and adulterated honey. In relation to this, many research works have been carried out to distinguish pure honey, adulterated honey and sugar solution (Bogdanov et al., 1999; White and Winters, 1989). In 1978, White et. al found that proline, potassium and sodium contents in honey can be used to distinguish pure honey samples from adulterated ones, particularly those adulterated using sucrose. Guler et al. (2007) found that the biochemical properties of honey such as moisture, ash, acidity, hydroxymethylfurfural (HMF) and electrical conductivity could be evaluated to successfully discriminate various pure honey.

Morales et al. (2008) reported that High-Performance Anion Exchange Chromatography Coupled with Pulsed Electrochemical Detection (HPAEC-PAD) technique had been successful at detecting and classifying adulteration in honey. The results have proved that this method is able to detect honey adulteration with as low as 5% of corn syrup.

In addition, Chua et al. (2012) had studied on multi-element composition and physical properties of honey. A total of five physical properties of honey (color, ash, pH, moisture and electrical conductivity) and thirty elements composition of honey (potassium, sodium and etc) have been investigated and the results have shown that both mineral profiles and physical properties could be used to successfully identify pure honey.

Nonetheless, the study on the detection of pure honey has started to decrease after the year 2008. Most researchers have been more interested to study the geographical and origin of honey. For instance, analytical method such as one using head-space solid phase microextraction (SPME) combined with comprehensive two-dimensional gas chromatography time-of-flight mass spectrometry (Stanimirova et al., 2010), and another work using near infrared spectroscopy (NIR) Chen et al. (2012) has been carried out to analyze the volatiles in honeys of various geographical and floral origins.

From the literature works, it is clear that most of the studies have employed various analytical procedures to determine honey product authenticity based on chemical contents. The data employed in the studies were based on Liquid Chromatography Mass Spectrometry (LCMS), High Performance Liquid Chromatography (HPLC), Gas Chromatography Mass Spectrometry (GCMS), UV-Visible (UV-VIS) Spectrometry, isotopic analysis and deoxyribonucleic acid (DNA)-based analysis. Not many studies have used electronic or electrical properties of honey in their works.

1.3 Problem Statements and Motivation

Honey is well-known as a source of high nutritional supplement as it has traces of minerals and vitamins. Most importantly, honey has been found to contain an antioxidant that is capable of destroying free radicals and slows down aging process. The European Union (EU) regulations (EU-Council Council Directive, 2002), the food Codex Alimentarius (CODEX STAN 12 Revised, Codex Standard for Honey, 2002) and various other international honey standards state that,

"Honey is a pure product that does not allow for the addition of any other substance"

Currently, there is high market demand for pure honey and hence, the price offered by manufacturers becomes expensive. Today's consumers are very conscious about healthy diet and are willing to pay high price for the benefits of honey. However, most medium to low-income consumers cannot afford to buy pure honey due to its expensive price. Hence, for them even just a little originality in honey still suffice. This has resulted in increased sales of adulterated honey in the market, claimed to be pure honey. Many manufacturers have started to add variants of sugar in pure honey so that the adulterated product resembles pure product to reap more profit (Utusan, 2008; Kosmo, 2009).

Until today, a few methods based on analytical methodologies as stated in the previous section have been used in determining honey product authenticity. These methodologies have shown good precision, accuracy and reliability. However, they are destructive in nature, time-consuming, require expensive equipment and are unsuitable for in-situ monitoring (Escriche et al., 2012). This is because the chemical compounds present in honey can undergo modifications through time and storage conditions and thus, reducing the dependability of those methods based on quantification of physico-chemical parameters (Escriche et al., 2012).

Also, many research works in the literature studied honey samples adulteration with different sugars (cane or corn) based on carbon stable isotopic ratio analysis (SIRA) method (White et al., 1998; Padovan et al., 2003; Martin et al., 1998). One lacking factor in such studies is the unspecified amount of sugar used for adulterating the honey samples. Hence, future comparison works are non-repeatable.

Another limitation of most honey studies is the detection of pure honey based on chemical procedures, requiring skilled personnel to operate the equipment and interpret the analytical results (Elizabeth et al., 1998). Traditionally, sensory evaluation and chromatographic techniques have been used to determine food quality. The quality assessments are essentially still best carried out by human panels due to the subjectivity involved as sensory evaluation provides immediate flavor information (Hong and

Wang, 2014). However, the human-sensory method suffers from some disadvantages such as the requirement for correctness of training, standardization of measurements, reproducibility, high expert costs and taste saturation of the panelist (Beullens et al., 2007).

Besides distinguishing pure honey from adulterated ones, to the best of author's knowledge, there has been no research conducted to estimate the percentages of purity in honey samples classified as adulterated. This information could help medium to low income honey consumers decide on compromising (a percentage) of honey purity for lower price of honey product. Thus, it is beneficial to have a device which is able to approximate the percentage of purity in adulterated honey products to help consumers decide on a favourable purchase comparable to what they could afford to buy.

1.4 Research Objectives

Based on the problems and limitations discussed in the previous section, the development of intelligent pure honey classifier and estimator is proposed in this research work. This work aims to classify pure honey from adulterated ones and also estimate the level of honey purity in adulterated honey samples. The objectives that need to be accomplished in this work are:

(i) to develop intelligent Tualang honey purity classifiers based on E-Nose, FTIR and Fusion data to classify either a honey product is pure or adulterated.

(ii) to compare the performance of developed intelligent Tualang honey purity classifiers with common statistical classification method.

(iii) to develop intelligent Tualang honey purity estimators based on E-Nose, FTIR and Fusion data to estimate the percentage of honey purity in adulterated honey samples.

(iv) to compare the performance of developed intelligent Tualang honey purity estimators to conclude the best honey purity estimator.

In this work, simple ANN models were employed as intelligent systems of Tualang honey purity classifiers and estimators.

1.5 Scope of Research

This research work only focuses on Tualang honey available in Malaysia. This is because the Federal Agricultural Marketing Authority (FAMA) only checks and certifies pure honey from the range of Tualang honey (Zaid et al., 2012). As only Tualang honey product authenticity is certified as pure, it is used as the source of pure honey in this work.

For honey adulteration in this work, sugar solution used were beetroot sugar and cane sugar from Germany and the United Kingdom, respectively. These sugar solutions were used because of their low prices. In addition, these sugar solutions have properties resembling pure honey (Sivakesava and Irudayaraj, 2001b). Furthermore, cane sugar has been commonly used as additional substance to adulterate honey (Rios-Corripio et al., 2010; Irudayaraj et al., 2003; Sivakesava and Irudayaraj, 2001a)

This research work also employs a commercial electronic nose (E-Nose) and Fourier Transform Infrared (FTIR) Spectroscopy to gather data from various honey samples.