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A COMPARISON OF CAFFEINE CONTENT IN SABAH COFFEES

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SCHOOL OF FOOD SCIENCE AND NUTRITION

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2009

DECLARATION

I hereby declare that the material in this thesis is my own except for quotations, excerpt, equations, summaries and references, which have been duly acknowledged.

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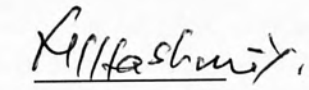

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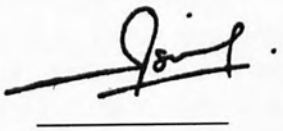

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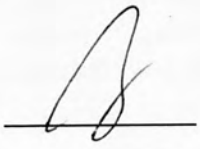

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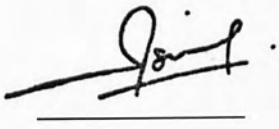

ABSTRACT

The analysis of Sabah coffees was a pioneer research in order to determine the caffeine content. The Sabah coffees used were Tenom Robusta (168 Entreprise), Tenom Arabica (Richwell Entreprise) and Tawau Arabica (Kopi Bintang Mas). Each sample was tested for its caffeine content by using methylene chloride to extract the caffeine. The caffeine content was analyzed by using UV-Vis Spectrophotometer at the absorbance of 272 nm. Among the three samples, Tenom Robusta sample yield the highest caffeine, followed by Tenom Arabica and Tawau Arabica. This proved that Robusta species coffee has higher caffeine content compared to Arabica species coffee and coffee from Tenom region has higher caffeine content compared to coffee from Tawau region. As further analysis, the four infusions added and four different heating durations used exhibited increase in caffeine values. Among the infusions, full cream milk powder yield the highest caffeine, followed by evaporated milk, then by non dairy creamer and the least caffeine by sugar. When the coffees are heated for 0 minute, one minute, five minutes, 10 minutes and 15 minutes, the yield of caffeine increased exhibiting the highest content when heated for 15 minutes and the least when not heated (0 minute). The varying caffeine content in coffee resulting from the parameters used should be further studied in order to determine whether the high caffeine content exhibited is available to human when consumed.

ABSTRAK

Perbandingan Kandungan Kafein dalam Kopi Sabah

Analisis dalam kopi Sabah adalah kajian utama untuk mengetahui kandungan kafein. Kopi-kopi Sabah yang telah digunakan adalah Tenom Robusta (168 Entreprise), Tenom Arabica (Richwell Entreprise) dan Tawau Arabica (Kopi Bintang Mas). Setiap sampel diuji untuk kandungan kafein dengan mengunakkan chlorometana untuk mengestrak kafein. Kandungan kafein dianalisis dengan menggunakan UV-Vis Spektrofotometer pada penyerapan 272 nm. Di antara tiga sampel yang digunakan, Tenom Robusta mempunyai kandungan kafein yang paling tinggi, diikuti oleh Tenom Arabica dan Tawau Arabica. Ini membuktikan bahawa kopi spesies Robusta mempunyai kandungan kafein yang lebih tinggi daripada kopi spesies Arabica dan kopi dari kawasan Tenom mempunyai kandungan kafein yang lebih tinggi daripada kopi dari kawasan Tawau. Sebagai lanjutan dalam analisis ini, empat jenis bahan campuran yang ditambah dan empat jangka masa pemanasan yang berbeza yang digunakan menunjukkan peningkatan dalam kandungan kafein. Antara bahan campuran yang digunakan, susu tepung penuh krim memberikan kandungan kafein yang tertinggi, diikuti dengan susu sejat, kemudiannya oleh krimer bukan tenusu dan yang paling kurang oleh gula. Apabila kopi dipanaskan untuk 0 minit, satu minit, lima minit, 10 minit dan 15 minit, kandungan kafein meningkat dengan memberikan kandungan paling tinggi apabila dipanaskan untuk 15 minit dan paling kurang apabila tidak dipanaskan (0 minit). Variasi kandungan kafein dalam kopi disebabkan oleh parameter-parameter yang digunakan perlu dikaji dengan lebih lanjut bagi menentukan samada kandungan kafein yang tinggi yang diperolehi adalah tersedia untuk manusia apabila diminum.

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LIST OF ABBREVIATIONS

var.	varietal
atm	atmosphere
UV	ultraviolet
Vis	visible

LIST OF SYMBOLS AND UNTS

g	gram
ml	milliliter
nm	nanometer
F	Farenheit
^{o}C	degree Celcius
M	molarity
ppm	part per million
mg	milligram
l	liter

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CHAPTER 1

INTRODUCTION

1.1 Background of the study

Coffee belongs to a large genus known as *Coffea* from the family of *Rubiaceae* (Rodrigues *et al.*, 2007). Coffee seeds referred to as coffee beans are used to produce the drinking beverage, coffee. The scientific names of the coffee beans vary according to their species such as Coffea *arabica*, Coffea *canephora* var. *robusta*, Coffea *liberica*, Coffea *fadenii* and many more (Beck, 2005).

Coffee trees are small shrubs with glossy leaves, fragrant white flowers and small red berries which are widely found in Southern Asia and subtropical Africa (Spiller, 1998b). Coffee trees are best grown in warm temperature and can normally live from 50 to 60 years although some possibly lives up to 100 years. In Malaysia, coffee plantations are found in Johore, Pahang, Sabah and Selangor. In Sabah, coffee plantations are widely found in Tenom, Tawau and Ranau.

Coffee mainly consists of groups of phenols, chlorogenic acids, alkaloids, amino acids and many other compounds. Alkaloids, structurally known as methylxanthines are biologically active components in coffee (Koshiro *et al.*, 2006). Caffeine, theobromine and theophylline belong to category of methylxanthine (Najafi *et al.*, 2003).

Caffeine, 1,3,7-trimethylxanthine a non volatile alkaloid is found in coffee, has intense bitter taste and can act as addictive stimulant to the central nervous system (Nehlig, 2004). Theobromine, 3,7-dimethylxanthine also present in coffee gives the same effects on human as caffeine but only in smaller dose. Theophylline, 1,3-methylxanthine is present in very small amount in coffee but is gradually reduced as the coffee beans are processed.

Coffee is the most widely consumed beverage due to its aromatic qualities and stimulating effects (Gupta, 1999). Although coffee is said to contain four times the amount of cancer fighting antioxidant as green tea and reduces risk of colon cancer yet it can lead to caffeinism, a chronic toxicity level due to excess caffeine intake (Nehlig, 2004).

Caffeine is famously known to be the world's most popular "drug" consumed. It is a powerful and consistent arousal agent that causes physiological, behavioral and emotional consequences when consumed (Margolis, 2002). Caffeine plays an important role in contributing to its anti oxidative activity and in the defense system where it acts as an antifungal agent which is a selective phytotoxin and a chemosterilant towards certain insects (Spiller, 1998b).

Freshly harvested coffee beans are processed using any of the three methods available (Chopra, 2005). Dry method, a naturally processing method is applied in hot countries where the coffee beans are sun dried or driers are used. This method is less expensive and gives a fuller flavour. Wet method which is the washing method, the coffee beans undergoes a fermentation stage, the dried under the sun or by mechanical driers. The result of this process is that the coffee beans are cleaner, brighter and fruitier. Semi-washed method, also known as pulped natural is a combination of both processes. It uses rapid drying method without fermenting the coffee beans. This method is able to retain some of the qualities of both the dry and wet method (Spiller, 1998b).

The raw coffee beans have to undergo another three major steps in order to be ready for consumption. The roasting step is the process where intense heat is given to remove almost 14 – 20 per cent of moisture, thus giving colour change according to the desired taste. Next step is the grinding step where the roasted coffee beans are cut and ground to obtain the powder form (Franca *et al.*, 2004). The final step is brewing, the beverage preparing step which can be done in four methods depending on how water is introduced to the ground coffee. The brewing types are boiling, pressure, gravity or steeping. A standard brew uses 5 g per 100 ml of water (Spiller, 1998c).

The study of visible spectrum is known as spectrophotometry which involves the visible light, near-ultraviolet and near infrared. The instrument used for light intensity measurement is the spectrophotometer (Hurst *et al.*, 1998). The instrument involves the spectroscopy of photons in the UV–Visible region and the normal visible region is 400 till 700 nm.

The UV-Vis Spectrophotometer is used in measuring the intensity of the light passing through the sample and compares it to the intensity of light before it passes through the sample (Donald *et al.*, 2001). It uses either the single beam or double beam mechanism where the samples are usually in the liquid form but solid and gaseous samples can also be measured.

The most important characteristic of a UV-Vis Spectrophotometer is its wide applicability to both organic and inorganic systems. It is also the most widely used of all quantitative analysis technique in chemical, environmental, forensic and clinical laboratories throughout the world (Feinstein, 1995). It has a moderate to high selectivity of detection, good accuracy and is best known for its ease and convenience of data acquisition (Skoog, 2007).

1.2 Objectives of study

The objectives of this study are

a) To extract, determine and compare the caffeine content in three types of coffees from Sabah by using UV-Vis Spectrophotometer
b) To investigate the caffeine content in coffees added with various types of infusions
c) To study the caffeine content in coffees heated for various time durations

1.3 Scope of study

This study will focus on the comparison of caffeine level in three types of coffees from Sabah. Coffee extracts will be used to determine the caffeine level by using the UV-VIS spectrophotometer. Coffee is being operated in many ways according to individual preference to enhance flavour and taste by addition of evaporated milk, creamer, full cream milk powder and sugar. There might be chemical reactions between substances like milk and sugar that might affect the overall content of caffeine in coffees.

The heating duration of coffee is also said to influence the overall caffeine content (Albanese *et al.*, 2009). Therefore various heating time is used to determine whether the time duration has influence on the caffeine content and the best heating duration to yield the extract with optimum caffeine content.

The aim is that the samples chosen to undergo this analysis are coffee powders which are freshly grounded at the point of purchase by the vendors of the Sunday Pasar Tamu in Kota Kinabalu, Sabah. Therefore, the coffee powders are not subjected to any prior proximate analyses or chemical tests. As a result, a part of this analysis will give a deeper insight on various factors that influence the caffeine content in the caffeine extraction of these untested coffee powders. The main focus of this study is to do the comparison of caffeine content in Sabah coffees.

CHAPTER 2

LITERATURE REVIEW

2.1 Origin of Coffee

The first coffee consumed was said to be in the 9th century. It was first discovered in the highlands of Ethiopia (Sánchez-González *et al.*, 2005) which eventually spread to Egypt and Yemen. By the end of 15th century coffee reached Armenia, Persia, Turkey and Northern Africa and from these Muslim world, it spread to Italy, then to the rest of Europe and America. Coffee was an important export commodity in 2004 and it tops the agricultural export for 12 countries in 2005 (Fujioka & Shibamoto, 2008). Coffee is the world's seventh largest legal agricultural export by value.

The English word coffee came to use in the early to mid 1600s but the early forms of word dated to last decade of 1500s (Spiller, 1998b). The term was introduced to Europe via the Ottoman Turkish 'Kahve' which was in turn derived from the Arabic, 'Qahweh' the origin of the Arabic terms remain uncertain. It was either derived from the name of the Kaffa region in Western Ethiopia where coffee were cultivate or by the Arabic term 'qahwat al-būnn' which means wine of the bean.

Coffee is the world's largest, most valuable, legally traded commodities after oil (Fujioka & Shibamoto, 2008). Coffee is the second important raw material within the international trade, the most important foreign exchange supplier for many agricultural oriented countries, an attractive source for tax yield and the most popular

drink (Huck *et al.*, 2005). This commodity is a cash crop for most the third world country which is the primary source of income for African countries like Uganda, Burundi, Rwanda and Ethiopia as well as the other Central American countries.

The two mainly cultivated coffee species are Coffee *arabica* and Coffea *canephora* var. *robusta* (Prance & Nesbitt, 2005). Arabica coffee tastes better then Robusta coffee because Robusta has more bitter taste and is less flavour. Arabica is cultivated almost three quarter worldwide. Robusta is less susceptible to disease than Arabica and can be cultivated in an environment that Arabica cannot thrive. Robusta contains more caffeine then Arabica (Rodrigues *et al.*, 2007).

Arabica is cultivated widely in Latin America, Eastern Africa, Arab and Asia whereas Robusta is cultivated widely in Western and Central Africa, throughout Southeast Asia and Brazil. The coffee beans from different countries or region has distinctive characteristic such as difference in flavour, aroma, body and acidity. The factors not influenced by the region are the genetic subspecies known as the varietals and the processing methods (Baumann *et al.*, 1998). The varietals are known by the region it is grown depending whether it is grown in Colombian, Java or Kono.

2.2 Classifications of Coffee

Coffee belongs to the botanical family of Rubiaceae and the genus Coffea (Rodrigues *et al.*, 2007). The most commonly identified commercial species of coffees in the genus Coffea are Coffea *arabica*, Coffea *canephora* var. *robusta* and Coffea *liberica* which are usually known as Arabica, Robusta and Liberica respectively (Sánchez-González *et al.*, 2005). All three varies from their shrub size to the coffee beans. Many other species of coffees are available but not cultivated commercially namely Coffea *bengalensis*, Coffea *racemosa*, Coffea *stenophylla*, Coffea *zanguebariae* and many others (Beck, 2005).

2.2.1 Coffee *arabica*

The tree of Coffee *arabica* is a small tree that has glossy leaves, white fragrant flowers and bears red berry fruits. It was first introduced to Arabia, Yemen, then to Ethopia and sooner into the tropical colonies of the Dutch, French and English (Prance & Nesbitt, 2005). Brazil has been the world's major supplier of coffee since the first seed was obtained and cultivate in 1723.

The leaves of Coffea *arabica* are small in size and have a darker green colour. The fruit of the coffee plant changes colour from green to yellow and finally to red. The fruits of Coffea *arabica* will fall of by itself once ripen and is not plucked (Beck, 2005).

The ideal growth condition for Coffea *arabica* is between 64 to 72F with 40 to 60 inches of rainfall all round the year. A warmer or cooler condition decreases it disease-resisting ability, thus becoming prone to Hemileia *vastatrix* attack which is a type of fungal disease causing a leaf spot disease (Beck, 2005).

2.2.2 Coffea *canephora* var. *robusta*

The leaves of Coffea *canephora* var. *robusta* have are lighter green colour and have slightly bigger size then Coffea *arabica*. The coffee fruit of Coffea *canephora* var. *robusta* will shrink and turns into black in colour once ripen and not plucked but it remains on the stalk (Beck, 2005).

Coffea *canephora* var. *robusta* was identified in 1895 in the African Congo. Coffea *canephora* var. *robusta* has distinct tolerance to more extreme growth condition of 60 to 80 F and 75 inches of rainfall. It tolerates humid condition without the Hemileia *vastatrix* attack (Beck, 2005).

2.3 Chemistry of Coffee

2.3.1 Acidity

The coffee acidity is correlated with the coffees grown at very high altitude and in mineral rich volcanic soil. The coffee beans that undergo washing process are perceived to have higher acidity level than those coffee beans that undergo natural or dry process (Chopra & Potter, 2005). The coffee body is preserved in the naturally processed coffee and since body masks the acidity, therefore naturally processed coffee have lower acidity level (Franca, *et al.*, 2005).

Acidity varies depending on brewing time and temperature where higher temperature increases the acidity. Acidity also shows variation according to particle size whereby coarse coffee is less acidic than medium coffee which is less acidic than fine coffee (Rodrigues *et al.*, 2007).

2.3.2 Aroma

Coffee aroma is the most important attribute in specialty coffee whereby the aroma determines the quality and acceptable level of the coffee (Franca *et al.*, 2005). Instant coffee also have aroma but the lack of most of the volatile compound causes dramatic decrease in the overall coffee flavour.

2.3.3 Bitterness

Bitterness is capable of taming the coffee acidity and adding favourable dimension to the brew at low levels (Perrone *et al.*, 2008). But bitterness at high levels has the tendency of overpowering other compounds and causing undesirable effects (Fujioka & Shibamoto, 2008). A shorter roasting duration, decaffeination and soaking of coffee in fresh water after fermentation can reduce the bitterness of the coffee. Proper brewing method and coarser ground of coffee are also ways to reduce the coffee bitterness.

2.4 Coffee Processing Methods

2.4.1 Picking

A coffee plant produces its first flower after three to four years after planting. Eventually the fruit (known as the coffee berries) are developed from the flowers. The berries takes about eight months time to fully-ripen, where it changes colour from green to red indicating its ripeness and will be the best time for harvesting (Spiller, 1998b).

For countries with uneven landscape, the berries are all hand picked but for countries with flat landscape, machines are used. However, both the harvesting techniques are based on two harvesting methods (Chopra & Potter, 2005). The first method is the strip picking method which is either done by machine or by hand. All the cherries are stripped off the branch at one go. The second method is selectively picked. But this method is done only by hand technique. Only the fully ripened berries are harvested. As this method is labour intensive, so it needs a higher production cost and primarily the finer arabica coffee beans are harvested by using this method.

Figure 2.1 Ripening berries of Coffea *arabica* (Ashihara *et al.*, 2001)

2.4.2 Processing

Freshly harvested coffee beans are processed using three widely applied methods available namely the wet-processed, dry-processed and pulp-natural methods (Spiller, 1998b).

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