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To cite this article: Misto et al 2021 IOP Conf. Ser.: Mater. Sci. Eng. 1173 012012

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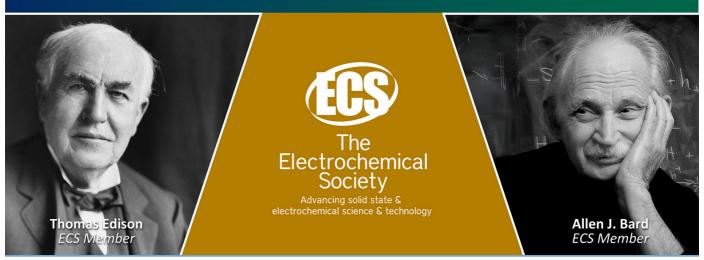
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doi:10.1088/1757-899X/1173/1/012012

### Spectrophotometric analysis of caffeine in local product of Arabica: observed at different roasted temperatures

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**Abstract.** Indonesia's geographical location supports the existence of various coffee commodities. As a plantation commodity, coffee has a high economic value. Coffee has a lot of substance that is beneficial to the body, one of is caffeine that serve to increase the stamina. Caffeine is naturally present in many types of plants as secondary metabolism. This research was done to know the content of caffeine in a local Arabica pure coffee with their variation of roasted temperatures. Coffee roasting is a stage of processing coffee beans into ready to grind, which can produce variations in the taste and content of coffee. The roasting process uses a roasting machine that automatically adjusts the roast temperature. Variations of roasted coffee are ground and used as a solution to determine the caffeine content. The determination of the coffee content is carried out using the spectrophotometric method, which measures the absorbance of caffeine in solution when it is passed by electromagnetic waves in the UV-Vis spectrum. The test results showed that the highest caffeine content was on samples of pure Arabica coffee with a roasted temperature of 195°C to 215°C with five levels of coffee roast. Caffeine content in sequential 3.217% and 2.597%.

Keywords: Arabica coffee, Caffeine, UV-Vis spectrophotometer

#### 1. Introduction

The coffee is one of Indonesia's plantation crops originating from Africa. In Indonesia, the existence of this plant has a very good impact on the country's economic growth [1]. One of the regions in East Java Province producing the second largest coffee commodity is Jember [2]. The type of coffee dominating plantations in Jember is Arabica coffee. Coffee grown at different heights and types of soil will produce different aromas and substances. Different qualities will also be obtained when the coffee beans are processed with certain roasting temperature variations [3]. The coffee content that has the most significant function is caffeine, which is believed to boost stamina for the body [4]. As an organic compound, caffeine contains nitrogen with a dual cyclic structure in the methylxanthine group (heterocyclic alkaloids) crystalline powder.

The chemical name of caffeine is 1,3,7-trimethyl xanthine, with the chemical formula C8H10N4O2 is shown in Figure 1 [5]. The purpose of this study was to determine the caffeine content of Arabica coffee, which is produced by the plantation in Jember. The caffeine content of Jember Arabica coffee was studied to identify the qualities of local Jember products analysed in various roasting conditions. The existence of pure Arabica coffee began to be modified by the addition of maize as to coffee blend

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1173 (2021) 012012

doi:10.1088/1757-899X/1173/1/012012

to obtain an economical profit by making the final product less expensive [6]. Most people have modified pure Arabica coffee products with maize ingredients to reduce the price of coffee which is still too expensive [7]. In this study, we will also analyse how the effect of maize with a composition of 10% and 20% of the total coffee on its caffeine content.

Figure 1. Chemical formula of caffeine [5]

Solvents such as Chloroform, Methyl Chloride, Ethanol, Acetone and Ethyl acetate are commonly used for the solvent extraction of caffeine. Chloroform (CHCl<sub>3</sub>) is a compound capable of dissolving caffeine and is assisted by CaCO<sub>3</sub> (lime). In the chloroform extraction process, caffeine can dissolve 94.53% [8]. The tool used to identify the caffeine content in Arabica coffee with variations in roasting temperature is the UV-Vis Spectrophotometer [9]. The UV-Vis spectrophotometry method, as shown in Figure 2, works based on the interactions between monochromatic rays and light sources. The result of the interaction in the form of energy absorbed by a specific value can cause the electron to be excited from the ground state to a higher energy state.

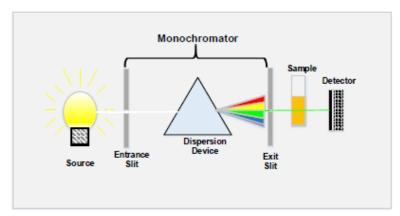


Figure 2. Schematic diagram of Spektrofotometer UV-Vis [10]

Quantitatively, the rays that are absorbed by matter can be arranged according to the Lambert-Beer law. This law states that the amount of visible light absorbed or transmitted by a solution is an exponential function of the concentration of the substance and the thickness of the solution. The Lambert-Beer law has the following equation [11],

$$T = \frac{I_t}{I_0} = I_0^{-acl} \tag{1}$$

$$\log(T) = \log \frac{I_0}{I} = -acl \tag{2}$$

$$T = \frac{I_t}{I_0} = I_0^{-acl}$$

$$\log(T) = \log \frac{I_0}{I_t} = -acl$$

$$\log\left(\frac{1}{T}\right) = \log \frac{I_0}{I_t} = acl = A$$
(1)
(2)

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#### 2. Materials and Methods

The material used was a local coffee Arabica pure and mixed Arabica-maize. We used CaCO<sub>3</sub> and CHCl<sub>3</sub> as standard caffeine and the solvents of caffeine. The instrument used for analysing caffeine absorbance is a UV-Vis Spectrophotometer with the wavelength range 250-300 nm. The five samples of Arabica coffee were prepared using a roaster, which is equipped with a roasting temperature controller. They were set at 194°C, 215°C, 225°C, 230°C and 240°C respectively to represent the variation types of roasting process.

#### 2.1. Manufacture of standard solution caffeine

A standard solution of 250 mg of caffeine is put into a beaker glass and then added 90°C-95°C distilled water. This solution is diluted again in a 100 mL volumetric flask. This standard of caffeine produces a concentration of 2500 ppm. This solution was piped with a size of 1 ml, 0.8 ml, 0.6 ml, 0.4 ml, and 0.2 ml and then put in a 50 ml volumetric flask. The concentration results after being homogenized are 50 ppm, 40 ppm, 30 ppm, 20 ppm, and 10 ppm, respectively.

#### 2.2. Determination of the maximum absorbance wavelength

Absorbance measurements were carried out in standard caffeine solutions with 10 ppm, 20 ppm, 30 ppm, 40 ppm, and 50 ppm in the wavelength range of 250-300 nm. The results of the wavelength scanning showed that the maximum absorbance for all samples was 273 nm. This wavelength was applied then to observe the caffeine content of various Arabica coffee roasted in various temperature.

#### 2.3. Sample Preparation of pure Arabica and adulteration of 10% and 20% maize

Caffeine extraction was proposed to separate the caffeine from coffee grounds. A 2-grams sample of coffee was put into a beaker, and 150 ml of hot water was added. The coffee solution obtained is then filtered through a funnel and filter paper into the Erlenmeyer. A total of 1.5 g of Calcium Carbonate (CaCO<sub>3</sub>) was added to the filtrate that had been put into a separating funnel, and then extracted three times with the addition of 25 ml of chloroform. The function of the addition of chloroform is to bind caffeine compounds to form caffeine extraction.

The extraction of caffeine at the bottom of the solution will then be evaporated with a rotary evaporator so that the chloroform can evaporate completely. The caffeine extract from each solvent-free coffee sample was put into a 100 ml volumetric flask, then diluted with distilled water. Besides pure Arabica coffee, this research will also analyze the type of Arabica coffee in the form of a mixture of pure Arabica and maize. The amount of maize that is mixed is as much as 10% and 20% of the total mixed coffee made [12].

#### 2.4. Determination of the caffeine content

Calculation of caffeine content in ppm was obtained through linear regression from the standard caffeine concentration and the maximum absorbance. The conversion of ppm units into mg/g was calculated using the following equation [9].

Caffeine content = 
$$\frac{\text{Concentration }(X) \times \text{The weight of extract} \times \text{Fp}}{\text{Sample weight}}$$
(4)

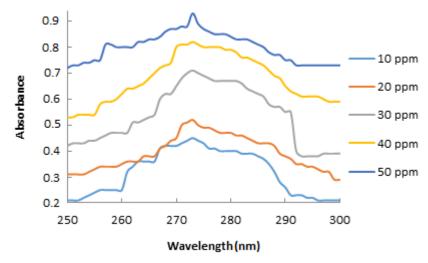
Where X is the concentration in mg/ml, the weight of the extract is the caffeine extract from each solvent-free coffee sample which is diluted with 100 ml distilled water and measured in litres, Fp is the dilution factor that is carried out several times dilution, and the sample weight is the sample weight. Ground coffee that has not been brewed in grams. The caffeine content then compared with Standart Nasional Indonesia (SNI).

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doi:10.1088/1757-899X/1173/1/012012

#### 3. Result and Discussion

The absorbance of caffeine standard solution with a concentration of 50-10 ppm at a wavelength of 250-300 nm is shown in Figure 3. The wavelength with the maximum absorbance occurs at a wavelength of 273 nm.



**Figure 3**. The absorbance of caffeine standards solution on the variation of the concentration in the range of the wavelength spectrum of 250 - 300 nm.

The absorbance measurement of the caffeine standard was carried out three times, so that the mean absorbance value and its uncertainty value are shown in Table 1.

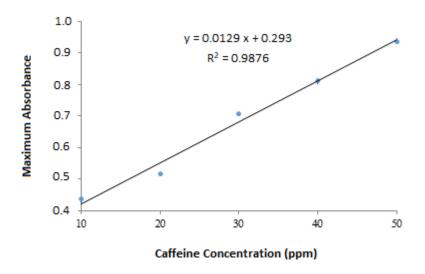
**Table 1.** The maximum absorbance of caffeine standart using Spectrophotometer UV-Vis.

No.	Caffeine Consentrarion (ppm)	Maximum Absorbance		
1	10	$0.437 \pm 0.008$		
2	20	$0.517 \pm 0.004$		
3	30	$0.707 \pm 0.004$		
4	40	$0.810 \pm 0.007$		
5	50	$0.937 \pm 0.008$		

Based on this table, an increase in the concentration of caffeine solution causes the maximum light intensity absorbed in  $\lambda = 273$  also increase. Calibration curves for caffeine concentration and maximum absorbance produce a linear regression equation used to determine caffeine content (refer to Fig. 4).

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doi:10.1088/1757-899X/1173/1/012012



**Figure 4.** Calibration curve of caffeine standard solution of caffeine.

The calibration curve used to calculate the caffeine content of coffee powder sample. Furthermore, the measurement value of the maximum absorbance on ground coffee pure Arabica and a mixture of Arabica-maize 10% and 20%. Table 2 has shown the maximum absorbance of each coffee.

**Table 2.** The measurement of maximum absorbance of ground coffee pure

Arabica at a wavelength of  $\lambda = 273$  nm.

Coffee	Roasting Temperature	Absorbance Maximum
	195°C	$1.123 \pm 0.004$
Pure arabica	215°C	$0.963 \pm 0.011$
rule alabica	225°C	$0.637 \pm 0.008$
	230°C	$0.473 \pm 0.004$
	240°C	$0.387 \pm 0.004$
	195°C	$0.707 \pm 0.008$
Mintuna anahira	215°C	$0.640 \pm 0.007$
Mixture arabica-	225°C	$0.427 \pm 0.029$
maize 10%	230°C	$0.373 \pm 0.004$
	240°C	$0.347 \pm 0.004$
	195°C	$0.517 \pm 0.004$
M:	215°C	$0.467 \pm 0.004$
Mixture arabica-	225°C	$0.327 \pm 0.004$
maize 20%	230°C	$0.237 \pm 0.004$
	240°C	$0.300 \pm 0.000$

According to Table 2, the higher the roasting temperature used, the smaller the absorbance value measured. The decrease in maximum absorbance when the roasting temperature is increased is closely related to caffeine (caffeine content) in pure coffee. The high roasting temperature, past its melting point, causes the atomic bonds in caffeine to change. The melting point of caffeine is at a temperature of 228°C [5]. This property influences the change of caffeine absorbance to electromagnetic waves applied. We have also calculated the caffeine content on a 2 mg serving three times of coffee consumption per day and presented by Table 3.

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doi:10.1088/1757-899X/1173/1/012012

**Table 3.** The Total caffeine content of coffee consumed three times a day with the weight of the sample every dish as much as 2 g

Coffee	Roasting Temperature	Caffeine (mg/day)
	195°C	193.0
	215°C	155.8
Pure Arabica	225°C	79.9
	230°C	41.9
	240°C	21.7
	195°C	96.1
	215°C	80.6
Mix Arabica/Maize with Maize content of 10 wt %	225°C	31.0
waize content of 10 wt 70	230°C	18.6
	240°C	12.4
	195°C	51.9
	215°C	40.3
Mix Arabica/Maize with Maize content of 20 wt %	225°C	7.8
Marze content of 20 wt 70	230°C	4.7
	240°C	1.6

When referring to the FDA (Food Drug Administration) standards mentioned by Liska [13], the dose of caffeine consumed per day is 100-200 mg/day. Also, according to SNI 01-7152-2006, the threshold limit for caffeine in food and beverages is 150 mg/day [14]. Based on the fifteen samples that have been tested, according to both the FDA and SNI, this type of local Arabica coffee in Jember at 195°C roasting temperature and 215°C in pure coffee exceeds the limit of caffeine consumption for each serving three times a day. Arabica coffee provides acidic, sharp aromas and little bitter. The number of caffeine as its substance makes it popular to be consumed. Coffee has various benefits for the body, that it can be useful as an antioxidant and also for stimulating brain performance [15]. However, consuming a substance in excess amounts will also have a negative impact on the body.

#### 4. Conclusion

The caffeine content of Arabica coffee in Jember plantations has been identified based on variations in roasting temperatures. It was obtained based on the results of the analysis of the maximum absorbance changes at each roasting temperature variation. Increasing the roasting temperature causes the maximum absorbance value at a wavelength of 273 nm to decrease. A decrease in the maximum absorbance indicates a decrease in the caffeine content in coffee. Local Arabica coffee in the plantation area in Jember has caffeine content according to SNI for daily consumption, except for the pure Arabica types, which are roasted at 195°C until 215°C. The addition of maize as a mixing ingredient of up to 20% still shows sufficient caffeine levels for daily consumption.

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doi:10.1088/1757-899X/1173/1/012012

#### Acknowledgments

This research was funded by the University of Jember under community based research project. The authors were thanks to University of Jember for funding the research by Hibah Keris 2020.

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