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Product Quality of Quercetin Extract From *Carica Papaya L* Flower by Microwave-Assisted Extraction (MAE)

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Abstract

These work aims are to determine the best condition extraction and product quality of quercetin from *Carica papaya L* flower extract. The extraction of quercetin from the *Carica papaya L* flower had done by microwave-assisted extraction (MAE) method. Dried flower prepared by cutting and grinding with a domestic mixer into 0,125 mm of particle size. Dried flower were extracted using methanol 80% (w/w) to determine influence of solid to liquid, S/L, ratio (1:10, 1:15, 1:20), microwave power (120 W, 200 W, 280 W, 400 W), and extraction time (1-5 minutes) on extraction process. Using the phytochemical test by Mg-HCl-amyl alcohol and UV-Vis spectrophotometer techniques, the quercetin detected by qualitative and quantitative analysis. As a result, the presence of flavonoids detected by the formation of a brownish red-colored flavylium compound. Increasing extraction time, power, and S/L ratio increased the extraction temperature. Consequently, yield quercetin decreased when the temperature extraction exceeds its temperature degradation. The highest quercetin yield, 0.214%, was detected with solid to liquid ratio (1:15), microwave power (400 Watt), and extraction time (4 minutes). FTIR spectrophotometer technique on the highest yield quercetin proved that have product quality with 91,17% similarity on group function like –OH, C=O, C=C, and C-H with quercetin standard spectrum.

Keywords: *Carica papaya L*. flower, Quercetin, Microwave-assisted extraction, Product Quality

1. Introduction

Carica papaya L. is one of many medicinal plants in Indonesia, which has several advantages, namely a lack of side effects, useful for chronical disease with chemical drugs, and low prices. The papaya plants with genus Carica of the family Caricaceae plants are widely studied because almost all parts of this plant can be used both leaves, juices, seeds, roots, stems, fruits, and flower. The bioactive compounds present in the papaya plants are papain enzymes, carotenoids, alkaloids (Mukhaimin, Latifahnya, and Puspitasari, 2018), monoterpenoids, flavonoids, minerals, vitamins, glucosinolates, carposides (Milind and Gurditta, 2011). Primarily flavonoids can be used as anticancer antioxidant (Am, Asmah and Fauziah, 2014), (Sadek, 2012), antidiabetic (Tradit and Altern, 2011), (Lakshman and Changamma, 2013), anti-inflammatory (Anaga and Onehi, 2010), (Lu *et al.*, 2010), antihelmintic (Akujobi, Ofodeme and Enweani, 2010), antibacterial (Nirosha and Mangalanayaki, 2013), antimalarial (Adedosu et al., 2014) (Patil *et al.*, 2013), (Tarkang *et al.*, 2013) ,and dengue fever treatment (Ahmad *et al.*, 2011). Rich in benefits makes many researchers tried to utilize all parts of the plants from roots to flower. However, the utilization of papaya plant flower until now it has not been maximized yet. In Indonesia, papaya flower commonly used as vegetable food ingredients. Therefore, the utilization of papaya flower in order to produce flavonoid extract will increase the value-added as medicinal plants with the proper extraction process of microwave-assisted extraction.

Quercetin is the most important flavonoid in papaya flower due to high antitumor effects (Ohshima *et al.*, 2018). Quercetin usually extracted by the conventional method such as maceration (Akujobi, Ofodeme, and Enweani, 2010) and heat reflux extraction (Nirosha and Mangalanayaki, 2013) which have several disadvantages, such as the lengthy process, large amounts of solvent volume, toxic residual in the product, low recovery and not economic. Therefore, a fast extraction method and preventing thermal degradation of quercetin is necessary.

Microwave-assisted extraction (MAE) becomes the solution in this process. MAE has many advantages, such as shorter extraction time, fewer samples, fewer solvent, and less high temperatures. These advantages make the MAE widely used in extraction processes such as Zingiberene extraction on ginger oil, alkaloid bioactive extraction compounds on Lotus plumule (Xiong *et al.*, 2016) and Stephania Sinica (Xie *et al.*, 2014), and extraction of flavonoids and alkaloids in Crotallia sesciliflora (Tang *et al.*, 2017). However, MAE needs the solvent which can absorb microwave, namely methanol, and ethanol. Furthermore, from solid to liquid ratios, extraction time, microwave power, and temperature are factors that can affect the MAE extraction process (Xiong *et al.*, 2016).

The novelty of this study is the use of *Carica papaya L* flower as the primary raw material for quercetin extraction using the microwave-assisted extraction method. Dried papaya flower was extracted using methanol 80% (w/w) with several variables, namely solid to liquid ratio, extraction time, and microwave power, to study its influence extraction process condition. Quantitatively the extract was analyzed by UV-Vis spectrophotometer techniques, whereas Mg-HCl-amyl alcohol used for flavonoid phytochemical test and FT-IR spectrophotometer techniques for product quality analysis of quercetin in 80 % extract based on the structure and functional group.

2. Methods

2.1. Material

Carica papaya L flower collected from a local farmer in Cimahi, West Java, Indonesia, during periods August – October 2017. Quercetin (analytic grade) purchased from the Mandiri lab (Indonesia). Methanol, Mg-HCl-amyl alcohol reagents, KBr (analytic grade) purchased from Brataco. Co (Indonesia).

2.2. Sample Preparation

To adjust the moisture content of *Carica papaya L* flower, *Carica papaya L flower* was dried under sunlight and in an oven (mammert) at 50^{o}C for 24 hours. The flower was ground in a domestic mixer to enlarge the surface area of particle and was sieved screen into 0.125 cm. 10-gram flower powder was stored in each plastic sample at room temperature until used in the extraction experiment

2.3. Microwave-Assisted Extraction

The extraction process conducted in a modified microwave (Sharp R-230RS) with output power ranging from 40 to 400 W and the frequency 2450 MHz. The microwave had designed in a closed system with condenser, 250 ml volume of the extraction flask, and magnetic stirrer to utilize the effect of differences of condition process**.** Figure 1 describes a schematic of the MAE system.

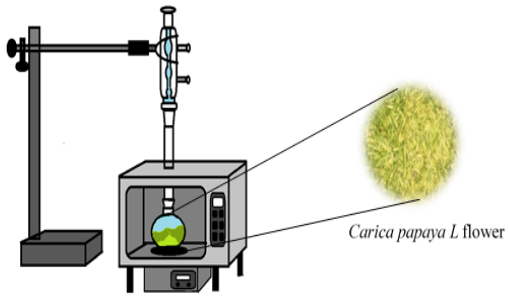


Figure 1. Experimental set-up of the microwave-assisted extraction

The extraction was run by dissolving a Carica papaya L. flower powder in 80% aqueous methanol solution on the extraction flask with 200 rpm stirring speed. The variables studied were solid to solvent, S/L ratio (1:10, 1:15, 1:20), power microwave (120 W, 200 W, 280 W, 400 W), and extraction time (1-5 minutes).

2.4. The Phytochemical Analysis

To prove the presence of flavonoid in *Carica papaya L* flower and in 80% methanol extract, the phytochemical screening analysis used. It tested using a porcelain cup by adding magnesium, hydrochloride acid, and amyl alcohol. The transformation from brownish yellow to brownish red on color liquid proved the presence of flavonoid (Seniwaty *et al.*, 2017)

2.5. The determination of Quercetin Yield

Determination of the percentage of quercetin yield in the Carica papaya flower using the UV-Vis spectrophotometer (Shimadzu-1800). A standard solution was obtained by dissolving quercetin in 80% aqueous methanol solution. The solution measured by absorbance at a wavelength of 373 nm. The quercetin extract was calculated using a linear equation from the standard quercetin curve and solved with Equation 2

$$y = 0{,}0444x + -0{,}0143 \quad (1)$$

$$Yield = \frac{Concentration\ extract\ (ppm)}{initial\ concentration\ (ppm)} \times 100\% \quad (2)$$

2.6. Quercetin Product Quality Analysis

Analyzation product quality of quercetin identified through IR spectrophotometer (IR-Tracer 100) transmittance of solid from the highest yield of quercetin on *Carica papaya L* flower extract. Spectroscopy of quercetin was taken in KBr. Finally, the recorded characteristic IR spectra of quercetin extract were compared with quercetin standard and Carica papaya flower powder to assign quercetin product quality (Churasiya, Upadhayay and Shukla, 2012)

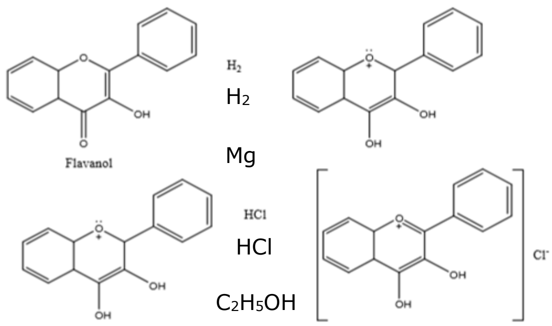
3. Results and Discussion

3.1 The Phytochemical Test

Phytochemical test on 80% methanol extract of *Carica papaya L* flower with Mg-HCl-amyl alcohol reagents that make the color changed from brownish-yellow into brownish red liquid. Magnesium acts as a reducing agent, whereas the reduction is carried out in acids in the presence of HCl. The results of the reduction of magnesium and hydrochloric acid produce a brownish-red liquid color. It is suggested that brownish-red color showed a definite presence of flavonoid by the formation of flavylium salt compounds like in Figure 2. (Seniwaty *et al.*, 2017).

3.2 The Effect of Extraction Time

Extraction time is one of the factors affecting the extraction process. To determine the effect of extraction time on yield quercetin, *Carica papaya L* flower dried particles extracted in solid to the solvent ratio 1:10, 1:15, 1:20 w/w and 400 w for 1 – 5 minutes. Figure 3. shows the percentage of quercetin yield increased with an increase of extraction time until reaches maximum yield, 0.214%, in the 4th minute, 1:15 S/L ratio and 400 W microwave power While in the 5th minute there was a decrease in the percentage of quercetin yield to 0.209%. It occurs due to the actual extraction temperature exceeded the degradation temperature of quercetin at 75^oC.



(a)

(b)

Figure. 2. (a) the formation reaction of flavilium salt and (b) the result of the phytochemical test

Thermal degradation of quercetin occurs because the heating process with a microwave is different from conventional heating like on soxhlet extraction. The heating process in a microwave is localized heating in the solvent with dipole rotation and ionic conduction mechanism. Besides, the ability of *Carica papaya L* flower matrix to absorb microwave irradiation and produce heat is dependent on dielectric loss of its solvent and moisture content which Achieving into 86,27%. The evaporation occurs, and build intense pressure on a cell wall, which ruptures cause the release of the quercetin (Ekezie, Sun and Cheng, 2017). Even though moisture content had noticed could help to diffuse quercetin in cell-matrix flower (Franco-vega *et al.*, 2015), moisture content could accelerate increasing temperature in a matrix cell. It occurs because of water more polar and has a higher dissipation factor than other solvents to absorb microwave energy. This process makes degradation temperature of quercetin more rapidly achieved in a matrix cell and not give enough time to diffuse quercetin from matrix cell flower to solvent.

The extraction process was also aided by stirring to increase the intensity of collisions between dispersed particles and the distribution of heat in the solution. This action increased the rate of flavanoid diffusion to the solvent. However, the increasing time extraction rose temperature extraction because Figure 4 indicates that extraction temperature keeps going up until 5th minute extraction time. This because increasing reaction time allowed the solvent to absorb microwave energy, to increase the temperature, and to contact with few traces of moisture content in cell-matrix flower.

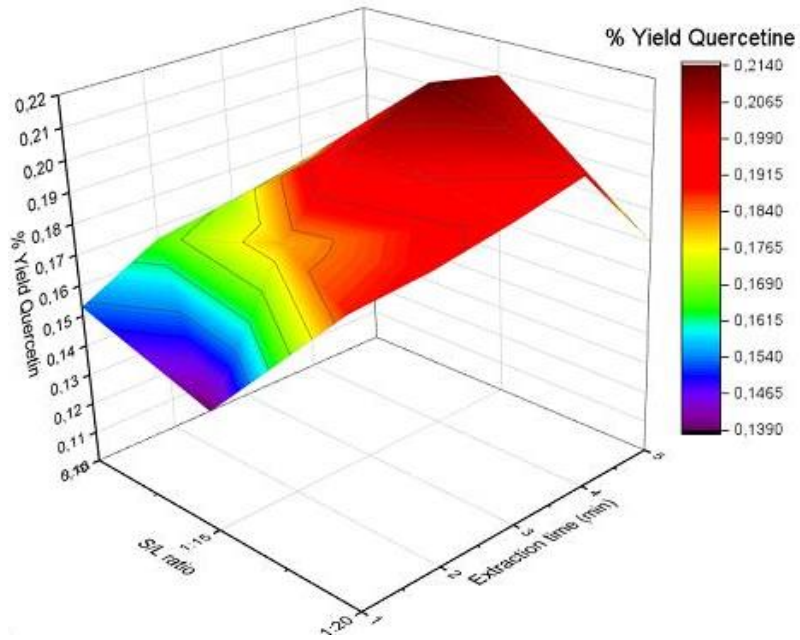


Figure 3. Effect of extraction time on the quercetin yield

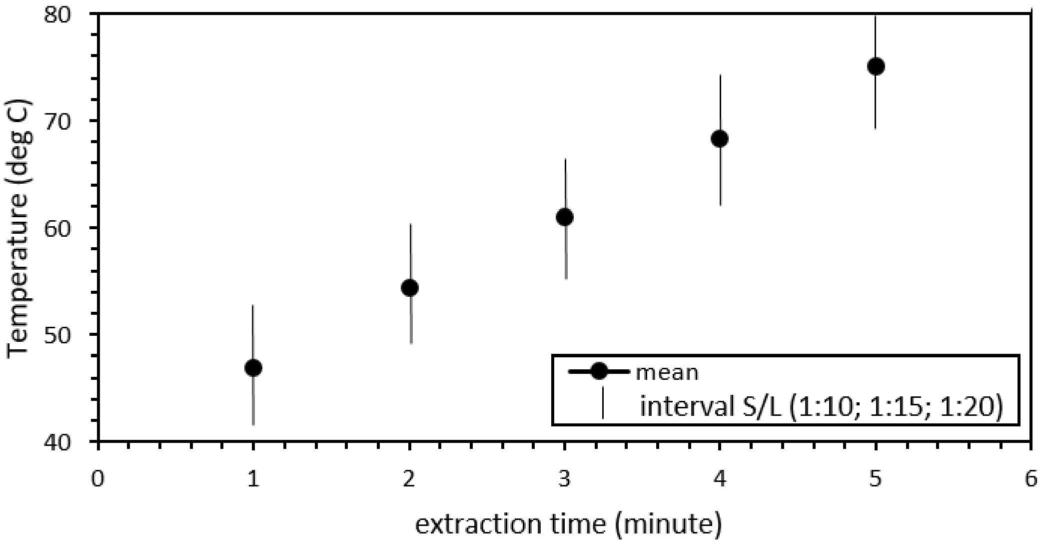


Figure 4. Effect of extraction time on temperature extraction

3.3. The Effect of Microwave Power

Power setting on MAE operations implicated in the energy and operating temperature in the extraction process. To know the influence variables in power microwave on yield quercetin, yield quercetin in 80 % extract compared in stratified microwave power with solid to the solvent ratio 1:15 for four minutes extraction time. Figure 5 shows that increasing microwave power increased temperature extraction. This appropriate with the theory of the total Absorbed Power Density (APD), which proportional to the total heat absorbed by the solvent (Q) with the calorimetric method (Chan, Yusoff, and

Ngoh, 2014). At 400 watt, extraction temperature was 68 ° C, under the range optimum temperature that was described previously, thus yield quercetin got higher at 400 watts than other microwave powers. The other microwave powers need more time to absorb microwave energy and to increase temperature extraction.

3.4. The Effect of solid to liquid ratio

The particle size of *Carica papaya L* flower was cutting and grinding into 0.0125 cm^2 to enlarge the surface area of the particle and to increase the contact area between solute and solvent so that the solvent will be faster to diffuse quercetin than the larger ones. In the extraction process, the use of the right solvent and solvent composition can maximize the acquisition of the target compound.

Figure 3 represents the effect of S/L ratio to quercetin yield. The maximum condition has conducted with 1:15 S/L ratio. Generally, in conventional extraction techniques, higher volumes of solvent increased recovery target, but in MAE, the higher solvent volume can provide lower recovery (Maran and Prakash, 2015). It was due to the excessive solvent in the extraction process at 1:20 S/L ratio. Excessive solvent leads to excessive swelling of the material that causes excessive thermal stress. Excessive thermal stress adversely affects on phytochemical compounds.

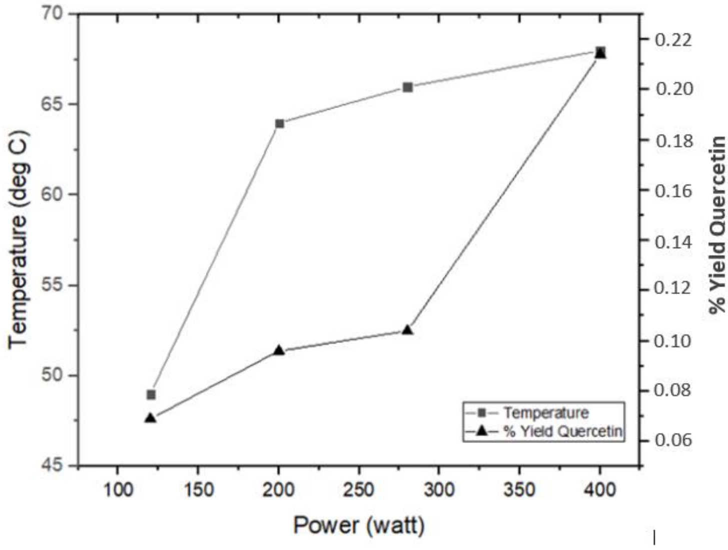


Figure 5. Effect of microwave power on temperature and yield quercetin

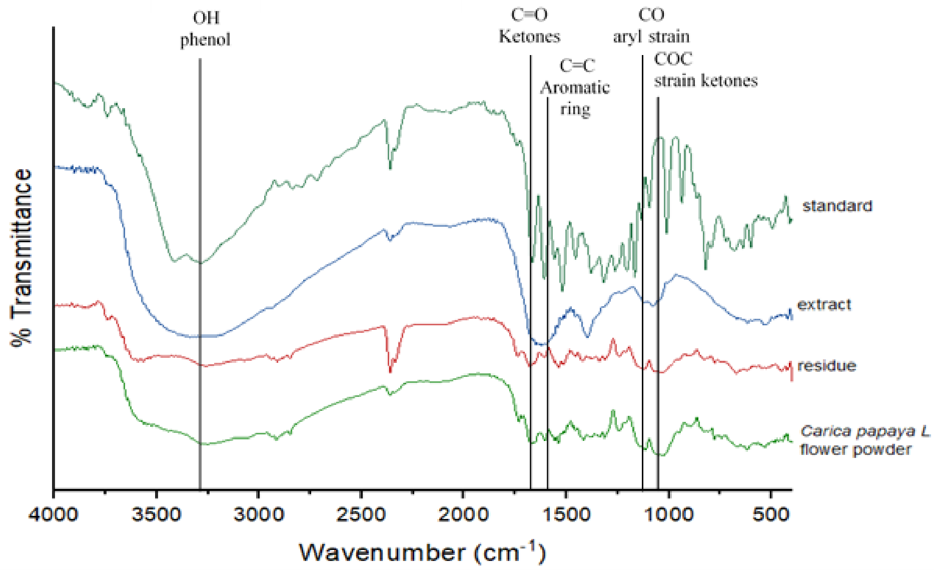


Figure 6.Interpretation of Vibration data of Quercetin and *Carica papaya L* flower

Figure 4 and Figure 5 indicate that the highest yield quercetin of *Carica papaya L*. flower, 2.14%, was produced at Solid to the solvent ratio 1:15, 400 W extraction time, and 4 minutes extraction time.

3.5. The Product Quality of Quercetin

Fourier Transform Infrared (FTIR) were tested to determine product quality by structure analysis on quercetin extract. According to research done by (Churasiya, Upadhayay and Shukla, 2012), the functional groups in quercetin compounds are composed of OH group bonds on phenol (3411 cm^{-1}), C = O ketones (1663.8 cm^{-1}), C = C aromatic ring (1608.8 cm^{-1}), aryl strain CO (1265 cm^{-1}), COC strain on ketones (1167 cm^{-1}) and aromatic ring CH (940.6; 821.4; 677; 602 , 3 cm^{-1}). Figure 6 showed that three kinds of spectra have the same similar spectrum. It also had shown the transformation spectrum on *Carica papaya L* powder, residue, and extract. It proved that microwave-assisted extraction was successful in extracting quercetin, and The extracted spectrum has product quality with 91,17% similarity on vibration data when compared to the quercetin standard spectrum. The differences detected in the spectrum that describes OH group at 3500 cm^{-1} – 3000 cm^{-1} of quercetin extract wider than standard spectrum.

Quercetin was known to have five OH group on arena structure, but the broader OH group spectrum was increasing the possibility of other bioactive compounds that can give a contribution to OH group spectra like tannin, saponin, and alkaloid. Higher OH group decreased the stability of quercetin on the microwave heating process that makes quercetin decomposed (Biesaga, 2011). Thus, the product quality of quercetin extract decreased. It also makes quercetin concentration of Carica flower L in this experimental slightly lower than (Maran and Prakash, 2015) do. Further research was proposed to make pure quercetin extract to increase product quality.

4. Conclusions

This research concludes that the quercetin was successfully extracted from *Carica papaya L* Flower by microwave-assisted extraction. The highest quercetin yield, 0.214%, was detected in 80% methanol extract with solid to the solvent ratio (1:15), microwave power (400 W), and extraction time (4 min). The Product quality of quercetine extract from the Carica Papaya L flower was successfully determined by the FTIR spectrophotometry method with an accuracy of the data vibration reaching 91.17% on group function vibration like –OH, C=O, C=C, and C-H when compared with the spectrum of the standard quercetin solution.

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