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Effect of Heating Time and Temperature on the Extraction of Bioactive Compounds from Turmeric (*Curcuma longa* L.) in vegetable oil

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Abstract. Herbal oil of turmeric *(Curcuma longa L.)* vegetable oil extract was identified to contain secondary metabolite compounds such as curcumin and total phenol content which has the potential as an antioxidant and antibacterial. This study aimed to determine the total phenol, carotenoid, and β-carotene content in turmeric rhizome extract in Extra Virgin Olive Oil (EVOO) and Virgin Coconut Oil (VCO). The method used for extracting was hot maceration, with variations in heating times of 4, 5, and 6 hours and variations in temperature of 50, 60, and 70 °C. Using a UV-Vis spectrophotometer, the extracted herbal oil was determined for total phenol, carotenoid, and β-carotene content. The study found that the amounts of phenols, carotenoids, and β-carotene were higher in turmeric VCO extract compared to turmeric EVOO extract. The top amount of phenol in 40% turmeric extract in VCO was 11.4% b/b GAE at 70°C and extraction time of 70°C and 4 hours. The most carotenoids found in turmeric extract in VCO were 342.18 ppm when extracted at 50°C for 5 hours. The turmeric extract in EVOO had the most β-carotene content at 9.83 ppm when extracted at 70 °C for 6 hours.

Keywords: Carotenoids, Maceration, Turmeric, Vegetable oil, β-carotene

1. Introduction

Curcuma longa Included in the *Zingiberaceae* family, has many health benefits because it consists of primary and secondary metabolites, namely carbohydrates, proteins, saponins, tannins, flavonoids (including curcumin), phenols, and sterols [1]. Turmeric, also called Curcuma longa, has strong antimicrobial properties [2]. Turmeric rhizome extract (*Curcuma domestica Val.*) in vegetable oil is an *herbal oil* that contains phenolic compounds, carotenoids, and β-carotene which have antibacterial, anti-inflammatory, anti-acne, and antioxidant pharmacological activities. Herbal oil is a traditional medicine obtained from nutritious plants and extracted with vegetable oil solvents [3]. The VCO contains MCT (*Medium Chain Triglycerides*) especially lauric acid which is the most dominant fatty acid at 48.24% and phenolic compounds α-tocopherols as natural antioxidants [4]. EVOO contains the compound oleuropein which is the main polyphenol in olives [5][6]. Turmeric olive oil extract has the ability as an antibacterial to suppress bacteria *Propionibacterium acne* and *Staphylococcus aureus* [7]. Curcumin levels in turmeric VCO extracted using the maceration method are at 31 ppm [8].

Carotenoids are chemical compounds that give a yellow color where the carotenoids contained in turmeric, EVOO, and VCO are 0.51%; 11.5 – 25.2 mg/kg and 323 μg/mL [9][10]. Carotenoids, total phenols , and β-carotene in turmeric can be extracted using EVOO and VCO into herbal oils that have benefits as herbal medicines. The method used to extract carotenoids and β-carotene in herbal oil preparations is hot maceration because it can simply extract phytoconstituents, produces high yields, uses low costs, and does not damage the active substances extracted. Maceration time greatly affects yield, carotenoids, total phenols, β-carotene levels, and turmeric VCO extract [11]. Carotenoids, total phenols, and β-carotene are non-polar and soluble in non-polar solvents such as EVOO and VCO oils. Research on moringa herbal oil vegetable oil extract with a hot maceration method at a temperature of 50 °C for 2 hours obtained a total phenol level of 15.78% GAE. The process of maceration extraction is the immersion of the sample into a solvent, so that the cell wall and membrane break due to pressure outside the cell, and finally the secondary metabolites in the cytoplasm will be dissolved in oil [12].

Total phenols, carotenoids, and β-carotene were measured in turmeric rhizome extracts in EVOO and VCO using UV-Vis spectrophotometers. UV-Vis spectrophotometers have several advantages in selectivity, namely, they do not take a long time and the cost required is much cheaper [13]. The measurement of carotenoid and β-carotene levels in VCO extract carrots using a UV-Vis spectrophotometer obtained results of 1272.0 – 2425.9 mg/L, and 113.87 – 123.39 mg/mL [14]. This study aims to measure the amounts of carotenoids and β-carotene in turmeric extract in EVOO and VCO by maceration method using variations in maceration time and temperature. The product produced is in the form of herbal oil which is expected to be able to become an alternative drug preparation for skin care because of its antioxidant content. They offer an effective method for harnessing the active compounds found in the herbs and the oil, giving them potential as an herbal medicine.

2. Material and Methods

Research materials include turmeric powder (Materia Medika Batu), virgin coconut oil (A&D), extra virgin olive oil (Borges), β-carotene (Merck), n-hexane pa (Merck), Follin-Ciocalteu (Merck) and aquadest. The research equipment includes a UV-Vis spectrophotometer called Cary 50 Conc UV-VIS Photometer variant.

2.1 Turmeric Rhizome Extraction Process in Vegetable Oil with Dosage Variations

Turmeric rhizome samples with a dosage variation of 0%-40% (gr/ml) in the form of powder were put into a 100 ml measuring flask and oil was added to the limit mark. Then stirred and heated for 2 hours at 50°C. Next, strain the solution through a cheesecloth. Next, transfer the filtered liquid into a glass bottle and keep it in a dark room until additional testing is conducted. The results of herbal oil are carried out by phytochemical screening.

2.2 Turmeric Rhizome Extraction Process in Vegetable Oil with Temperature Variation and Heating Time

40 grams of turmeric powder is put into a 100 ml measuring flask and oil is added until the limit mark is followed by the extraction process using a heating temperature variation of 50-70°C and a heating time of 1-6 hours. The mixture is filtered through cheesecloth and stored in a dark glass bottle.

2.3 Analysis of total phenol levels of turmeric vegetable oil extract

An aliquot of 2.5 mL turmeric vegetable oil extract was placed in a measuring flask and then mixed with methanol solvent. A 0.5 mL portion of this solution was combined with 5 mL of Folin-Ciocalteu reagent and left to stand for 3 minutes, after which 4 mL of 10% Na_2CO_3 solution was added. The mixture was incubated in the dark at room temperature for 30 minutes. Absorbance was measured at the maximum wavelength, and the oil solution concentration was applied to the regression equation for the gallic acid standard to determine the total phenolic content, expressed as milligrams of gallic acid equivalent per gram of oil (mg GAE/g oil). Calculations followed the equation $y = ax + b$

2.4 Determination of Total Carotenoids and Total β-carotene

50 mg of herbal oil is dissolved with 5 mL of n-hexane, and stirred until the carotenoids are dissolved. Then filtered with filter paper, the filtrate was marked limited to 5 mL, and the carotenoid content was analyzed with a UV-Vis spectrophotometer at wavelengths of 480, 645, and 663 nm.

1 mg of β-carotene is dissolved in a 20 mL measuring flask with n-hexane added to the cut-off mark. After that, the maximum wavelength was determined by pipetting 0.5 mL of the parent solution dissolved with n-hexane in a 5 mL measuring flask, where the absorption was measured by UV-Vis spectrophotometer at λ 380-780 nm. 10 mg of herbal oil was diluted with 5 mL of n-hexane, then filtered with filter paper and marked with 5 mL of n-hexane. Absorption is measured with a UV-Vis spectrophotometer at λmax with n-hexane as the blank. The β-carotene content in the sample was determined by the linear regression equation $y = ax + b$.

3. Results and discussion

3.1 Phytochemical analysis and total phenolic content of turmeric extract in VCO and EVOO

The results of phytochemical tests in Table 1 show that hot maceration extract of turmeric in VCO and EVOO got positive results for phenolics, terpenoids, alkaloids, and flavonoids and negative results for steroid compounds, saponins, and tannins. In turmeric extracts of EVOO and VCO, the color of the test results was darker, and more foam was formed than in VCO and EVOO. This shows that the content of secondary metabolites of phenolic compounds, terpenoids, alkaloids, and flavonoids in turmeric extracts of VCO and EVOO is more than that of VCO and EVOO.

Table 1. Results of the phytochemical analysis of turmeric extract in vegetable oil.

Phytochemical analysis	Volume of turmeric (%)			
	EVOO		VCO	
	0	40	0	40
Alkaloids (Mayer)	+	+++	+	+++
Flavonoids	+	+++	+	+++
Steroids	-	-	-	-
Phenolic	+	+++	+	+++
Terpenoids	+	+++	+	+++
Tannins	-	-	-	-
Saponins	-	-	-	-

Note: Sign – : no color change or foam observed
Sign + : light color or little foam
Sign ++ : quite concentrated or quite a lot of foam
Sign +++ : very concentrated or very much foam.

The higher the dose of turmeric used in the extraction, the more its secondary metabolite content is indicated by the more intense yellow color produced in the herbal oil. Based on Table 2, it is shown that at a dose of 40%, most phenols are found in turmeric extracts mixed with EVOO and VCO. The more doses of turmeric are added, the higher the total phenol levels so that the content of secondary metabolite compounds in turmeric is also extracted, which causes the potential of phenol as an antioxidant to increase. Adding turmeric rhizome extract in EVOO and VCO can produce a higher total phenol content than oil without adding turmeric. VCO can extract phenol compounds better than EVOO because it produces a higher total phenol level of turmeric VCO extract than turmeric extract EVOO. Coconut oil like other vegetable oils contains saturated fatty acids which are dominated by medium-chain triglycerides (MCTs) which have a higher polarity than EVOO which contains long-chain triglycerides (LCT) so VCO has a better ability to extract phenolic compounds in turmeric [15].

Table 2. Total phenol content of turmeric extract in EVOO and VCO with varying doses

Turmeric (%)	Total phenol levels (mg GAE/g)	
	Turmeric extract EVOO	Turmeric extract VCO
0	1.39	1.22
20	2.09	2.22
30	2.31	2.56
40	2.59	2.89

In Figure 1, most phenols found in turmeric extracts mixed with EVOO of 7.48% b/b GAE were obtained at 70°C and extraction time of 70°C and 6 hours. In turmeric EVOO extract, the increasing temperature and the length of heating in the extraction process will result in a higher total phenol content because at high temperatures it can increase the release of phenol compounds on the cell wall. However, at 6 hours of heating, the temperature of 60°C of EVOO samples and 60°C to 70°C of VCO samples tend to begin to decrease. Most phenols found in turmeric extracts mixed with VCO was 11.4% b/b GAE at 70°C and 4 hours. The longer the extraction from 1-4 hours, the greater the total phenol content is determined, the longer the extraction time is 6 hours and the decrease because the longer the extraction time can cause an oxidation reaction so that the compounds contained in the extract will also be reduced.

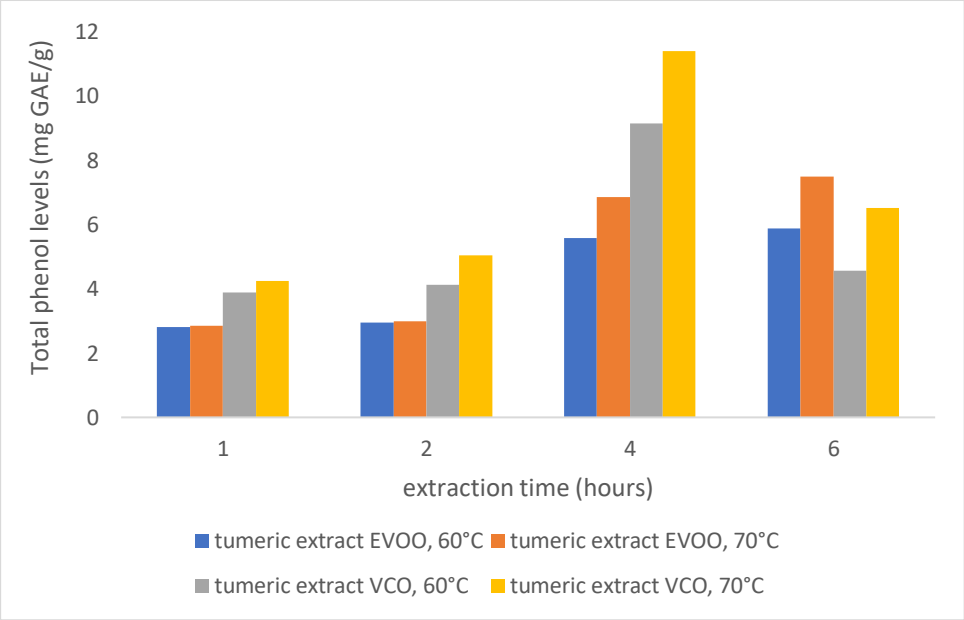

Figure 1. Total phenol levels of turmeric extract VCO and EVOO with temperature variation and heating duration.

3.2 Total Carotenoids of Turmeric Extracts EVOO and VCO

Total carotenoid levels increase with the length of maceration time of 4-5 hours and decrease at the maceration time of 6 hours. In Figure 2, the highest carotenoid content in turmeric VCO extract was 342.18 ppm at a temperature and maceration time of 50°C and 5 hours. The carotenoid levels of turmeric extract VCO showed a significant decrease in carotenoid concentration along with the length of extraction time because the carotenoids were easily degraded by the warming effect.

The highest carotenoid content in turmeric EVOO extract with a temperature and maceration time of 60°C and 6 hours. 60 C is the optimum temperature to obtain carotenoids with relatively high levels when compared to the temperatures of 50°C and 70°C. The decrease in carotenoid levels at 70 C is because carotenoids have begun to oxidize due to high temperatures, which is one of the main causes of carotenoid loss. However, the longer the extraction time, the higher the carotenoid level because the longer the contact time between turmeric and EVOO, the greater the solubility of carotenoids in EVOO.

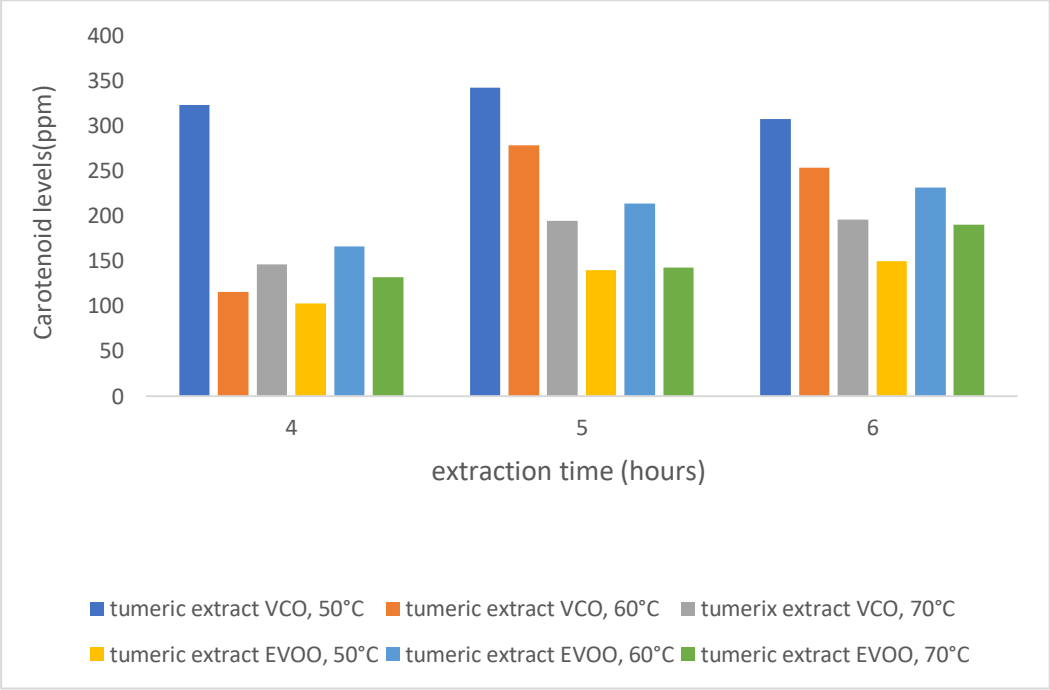

Figure 2. Carotenoid levels in turmeric extract VCO with variations in temperature and extraction time.

3.3 Total β-carotene turmeric extracts EVOO and VCO

The concentration of β-carotene is directly proportional to the extraction time across all temperature variations. The highest concentration of β-carotene in EVOO turmeric extract was 9.8 ppm at 70 °C with an extraction time of 6 hours. β-carotene is a type of carotenoid compound that is stable to heat so that extraction can occur optimally at a maceration temperature of 70°C. In turmeric VCO extract, the concentration of β-carotene increases with duration and maceration temperature. The highest β-carotene levels in VCO turmeric extract were obtained at a temperature maceration variation of 70 °C during a 6-hour extraction time. The increase in temperature has a significant impact on the movement of solvent molecules to be faster, resulting in expansion in the pores of the material solids.

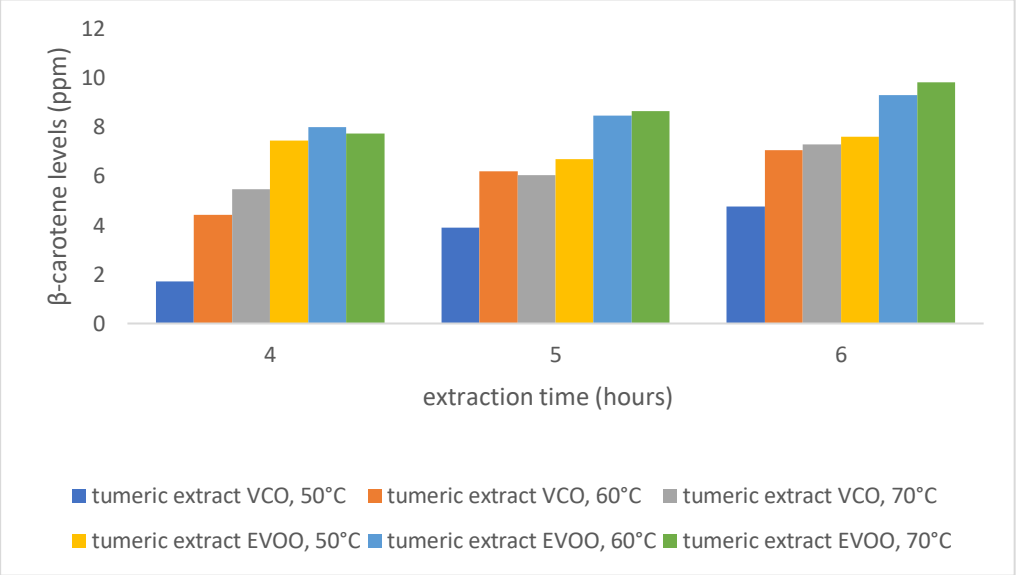

Figure 3. β-carotene levels in turmeric VCO extract with variations in temperature and extraction time.

4. Conclusion

The results of the total phenol test show that the higher the dose of turmeric in vegetable oil, the higher the total phenol level obtained. Temperature and heating time in maceration extraction also affect the total phenol value. The higher the temperature and the longer the heating, the more the total phenol level will increase, but it will decrease after reaching its optimum point. Likewise, the total value of carotenoids and β-carotene turmeric in the oil showed a significant increase with temperature variations and extraction time.

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