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## Analysis Of Total Polyphenol Content Of Carrot Extract (*Daucus Carota L.*) Using The Folin-Ciocalteu Method

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### Abstract

*Carrots (*Daucus carota L.*) are known to contain bioactive compounds, particularly phenolic compounds, which act as antioxidants. Objective: This study aimed to determine the total polyphenol content in carrot ethanol extract and evaluate its potential as a natural antioxidant source. This study was a quantitative experimental laboratory study. Carrot *simplicia* was obtained through a process of sorting, washing, chopping, drying, and grinding, then extracted using 70% ethanol solvent. The total polyphenol content was determined using UV-Vis spectrophotometry with Folin-Ciocalteu reagent and gallic acid as a standard, and the results were expressed in mg Gallic acid equivalent per gram of extract (mg GAE/g). Results: Gallic acid solution was used to obtain a standard curve. The analysis results showed a linear regression equation of  $y = 0.0161x + 0.042$ ,  $R^2 = 0.9947$ , with a correlation coefficient ( $r$ ) value of 0.997. The total phenol content of each sample was determined with three replications. The results showed that carrot ethanol extract had a total polyphenol content of 42,205 mg GAE/g extract. This value indicates that carrot extract contains polyphenol compounds that have the potential to be a source of natural antioxidants.*

**Keywords:** Antioxidants, Carrot (*Daucus Carota L.*), Follin-Ciocalteu, Total Polyphenols, UV-Vis Spectrophotometry).

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## INTRODUCTION

Carrots (*Daucus carota L.*) contain  $\beta$ -carotene as the main component, which is converted in the digestive system after consumption (Lina et al., 2024). Carrots (*Daucus carota L.*) are known for their high  $\beta$ -carotene content, along with Vitamins B and E. Vitamin A in raw carrots contains 34.94%  $\beta$ -carotene.  $\beta$ -carotene has high antioxidant properties, thus inhibiting the action of free radicals that can potentially damage body cells. In addition, this compound also plays a role in boosting the immune system and maintaining eye health (Sagita et al., 2025). Polyphenols are bioactive compounds found naturally in plants. Several factors can affect the efficiency of the polyphenol extraction process (Imanda et al., 2025). The extraction method influences the polyphenol content. Factors that influence this include the particle size of the material, temperature, and the length of extraction time. Finer particle size, higher temperature, and longer extraction time can generally increase polyphenol yield (Perdana et al., 2024). In addition to the method, the right solvent also determines the final extraction result. The principle of solvent selection must comply with the principle of like dissolves like, where the solvent will dissolve compounds with the same polarity (polar with polar, non-polar with non-polar) (Samudra et al., 2022). Types of solvents frequently used in extraction analysis of polyphenol content are ethanol, methanol, and acetone. In this study, the solvent used was 70% ethanol, known to be good at extracting phenolic and flavonoid compounds, which are key components in antioxidant activity. This concentration can optimize the extraction of alcohol-soluble compounds, but may not be fully effective for compounds with higher polarity (Pramushinta et al., 2025).

Analysis of total polyphenol levels is very important because polyphenol compounds act as natural antioxidants that can ward off free radicals in the body. Several studies on various plants have shown that total polyphenol levels vary depending on the type of plant, the extraction process, and the environmental conditions of the plant. For example, research by (Rauf et al., 2023) on the ethanol extract of senggani fruit (*Melastoma malabathricum L.*), showed that the total polyphenol content in the ethanol extract of senggani fruit (*Melastoma malabathricum L.*) was 2.275 mg GAE/g extract and the IC<sub>50</sub> value of the ethanol extract of senggani fruit (*Melastoma malabathricum L.*) was 13.159 ppm with a very strong antioxidant category using the ABTS method.

The UV-Vis Spectrophotometry method is used to measure the intensity of light absorption by a solution at a certain wavelength that is proportional to the concentration of the substance (Ramadhani et al., 2023). In this study, the total polyphenol content was determined using the Folin-Ciocalteu method. Folin-Ciocalteu is an organic reagent that has the ability to oxidize phenolates (alkali salts), reducing heteropoly acids to a molybdenum-tungsten (Mo-W) complex in alkaline conditions (Sari & Zulfa, 2022). The principle of the Folin-Ciocalteu method is the formation of a blue complex compound that can be measured at the maximum wavelength of gallic acid (Nofita & Rahman, 2025).

Based on the statement above, this study was conducted to determine the total polyphenol content in carrot (*Daucus carota* L.) extract using the Folin-Ciocalteu method with UV-Vis spectrophotometry. Through this study, it is hoped that information will be obtained regarding the level of natural antioxidant activity found in carrots as a source of bioactive compounds.

## RESEARCH METHODS

### Place and Time of Research

The research was conducted at the Chemistry Laboratory of the Faculty of Health Sciences, Duta Bangsa University, Surakarta, over a period of approximately three months, from November 2025 to January 2026.

### Types of research

This research was conducted experimentally with the aim of determining the total polyphenol content in carrot extract (*Daucus carota* L.) to show the potential of carrots as a source of antioxidants.

### Tools and materials

Research The equipment used in this study consisted of a UV-Vis Spectrophotometer (EMCLab®), cuvette, knife, blender (Philips®), 60 mesh sieve, 5 mL, 10 mL, 100 mL Pyrex® flask, Pyrex® measuring cylinder, Pyrex® beaker, oven (Mettler®), The materials used were carrot extract, Na<sub>2</sub>CO<sub>3</sub>, 70% ethanol, distilled water, Folin-Ciocalteu reagent, gallic acid, 2N HCl, Dragendorff, H<sub>2</sub>SO<sub>4</sub>, 3% FeCl<sub>3</sub>, Mg, concentrated HCl.

### Research Procedures

#### 1. Plant Determination

The carrot plants (*Daucus carota* L.) that were studied were determined.

#### 2. Making Simple Drugs

Fresh carrot samples were sorted, cleaned, and weighed. The samples were washed, cut into pieces, and air-dried indoors, away from direct sunlight. The dried carrots were then cut into small pieces and ground using a blender to obtain carrot leaf powder. The powder was weighed to determine its mass (Widiyantoro et al., 2023).

#### 3. Examination of Simplex Characteristics

### Powder Drying Shrinkage Test

In determining the drying shrinkage of simple substances, an oven is used, namely by inserting  $\pm 2$  grams of powder into the oven and then setting the temperature to 105°C for 30 minutes, after which the tool will produce the drying shrinkage value of the sample being tested (Wijaya, 2024).

### Total Ash Content Test of Powder

Determination of ash content was carried out by weighing 2 grams of carrot (*Daucus carota* L.) simplicia, put into a tared porcelain crucible, heated in a furnace slowly, then gradually increased to 600°C until carbon-free, then cooled in a desiccator and weighed the ash. Ash content formula (Krismayadi et al., 2024):

### Water Content Test

Using a moisture balance by inserting  $\pm 2$  grams of the simplicia then drying it at a temperature of 105°C for 5 minutes after which the tool will produce a water content value for the sample being tested (Ayun et al., 2025)

## **Maceration**

The extract was prepared using the maceration method. 300 grams of finely ground carrots were soaked in 1500 mL of 70% ethanol. The maceration process was carried out for 3 days with occasional stirring. The filtrate was then re-macerated for 2 days with occasional stirring. The resulting extract was evaporated using a rotary evaporator and recondensed using a water bath. The yield of the resulting thick extract was calculated using the formula (Purnama et al., 2021).

## **5. Extract Standardization**

### **Water Content Test**

Put approximately 2 grams of carrot extract (*Daucus carota* L.) into the moisture balance, dry it at a temperature of 105°C for 10 minutes after which the tool will produce a water content value for the sample being tested (Ayun et al., 2025).

### **Ethanol Free Test**

The ethanol-free test in carrot (*Daucus carota* L.) extract was performed using the following procedure. The extract was added with H<sub>2</sub>SO<sub>4</sub>, then CH<sub>3</sub>COOH, and heated. A negative test result was indicated if no characteristic ether odor was detected (Ardianti et al., 2024).

## **6. Phytochemical Test of Extracts**

### **Alkaloid Test**

A total of 0.5 grams of carrot extract was dissolved in 70% ethanol plus 1 mL of HCl and 3 mL of distilled water, heated in a water bath for 2 minutes, then cooled and filtered. Three drops of the filtrate were taken and 2 drops of Dragendorff's reagent were added. The formation of an orange color indicates a positive result for the presence of alkaloid compounds.

### **Saponin Test**

To 1 mL of carrot extract dissolved in 70% ethanol, 10 mL of hot distilled water was added, cooled, and then shaken for 10 seconds. The formation of foam indicates the presence of saponins.

### **Tannin Test**

Weigh 20 mg of carrot ethanol extract and add 2 drops of 5% FeCl<sub>3</sub> solution. A positive reaction of tannin compounds is indicated by the presence of a blackish green color.

### **Flavonoid Test**

A small amount of Mg metal powder and a few drops of concentrated HCl were added to 1 mL of carrot extract dissolved in 70% ethanol. A reddish-orange color indicates the presence of flavonoids.

## **Determination of Total Polyphenol Content of Carrots**

### **Preparation of gallic acid stock solution (0.05%)**

A total of 50.0 mg of gallic acid was dissolved in 0.5 ml of ethanol pa, then diluted with distilled water to a volume of 100.0 ml.

### **Preparation of 7.5% Na<sub>2</sub>CO<sub>3</sub> solution**

Add 7.5 g of Na<sub>2</sub>CO<sub>3</sub> to 80 ml of distilled water, then boil until the Na<sub>2</sub>CO<sub>3</sub> powder dissolves completely. After that, let it stand for 24 hours, filter it, and dilute it with distilled water to a volume of 100.0 ml.

### **Determining Operating Time (OT)**

A total of 300 µl of gallic acid solution with a concentration of 30 ppm was added with 1.5 ml of Folin-Ciocalteu reagent (1:10), then shaken and left for 3 minutes. 1.2 ml of 7.5% Na<sub>2</sub>CO<sub>3</sub> solution was added to the solution, shaken until homogeneous, and the absorbance was measured every 5 minutes at a wavelength of 765 nm.

### **Determination of maximum wavelength (λ<sub>max</sub>)**

A total of 300 µl of gallic acid solution with a concentration of 30 ppm was added with 1.5 ml of Folin-Ciocalteu reagent (1:10), then shaken and left for 3 minutes. 1.2 ml of 7.5% Na<sub>2</sub>CO<sub>3</sub> solution was added to the solution, shaken until homogeneous, and left at room temperature for the operating time range, then the absorbance was measured at a wavelength of 600-850 nm.

### Determination of the standard curve of gallic acid

The determination of the standard curve was carried out by taking 300 µl of gallic acid solution with concentrations of 10, 20, 30, 40 and 50 ppm, each of which was put into a tube, then added 1.5 ml of Folin-Ciocalteu reagent (1:10) and shaken. After being left for 3 minutes, each solution was added with 1.2 ml of 7.5% Na<sub>2</sub>CO<sub>3</sub> solution, shaken homogeneously, and left in the operating time range at room temperature. All solutions were measured for absorbance at the maximum absorbance wavelength, then a calibration curve was made of the relationship between gallic acid concentration (ppm) and absorbance.

### Total polyphenol content testing

A total of 20.0 mg of carrot ethanol extract was dissolved to a volume of 10.0 ml with a mixture of ethanol: distilled water (1:1). The obtained extract solution was pipetted 300 µl and added 1.5 mL of Folin-Ciocalteu reagent and shaken. Let it stand for 3 minutes, added 1.2 ml of 7.5% Na<sub>2</sub>CO<sub>3</sub> solution and let it stand again at the operating time range at room temperature. The absorbance of the extract solution was measured with a UV-Vis Speccrophotometer at the maximum absorbance wavelength. This was repeated 3 times.

### Data analysis

Data analysis was first performed using the standard curve method. A linear regression equation using Microsoft Excel was used to calculate the absorbance versus the concentration of the standard solution. Then, from the obtained linear regression equation, the total polyphenol content was calculated using the formula (Wilijeng & Anggarani, 2021).

## RESULTS AND DISCUSSION

### Plant Determination

Determination of Carrot plants (*Daucus carota* L.) was carried out at the UPF Yankestrad Tawangmangu Laboratory-Dr. Sardjito General Hospital in Tawangmangu. Determination aims to determine the truth of the plants to be studied and avoid errors in collecting materials and avoiding the possibility of mixing the plants to be studied with other plants (Sandepogu & Somineni., 2024). Based on the results of the determination, it is proven that the carrot plant (*Daucus carota* L.) used in this study is a carrot plant (*Daucus carota* L.).

### Sample Preparation

A total of 2 kg of fresh carrots were wet sorted, washed, chopped, and air-dried at room temperature for 15 days. The dried raw material was then blended and sieved using a 60-mesh sieve. The resulting raw material powder was 157 grams. The yield of the raw material is shown in Table 1.

**Table 1. Yield of carrot (*Daucus carota* L.) simplicia**

Initial simplex(g)	Final simple (g)	Yield of simple drug (%)
2000 grams	157 grams	7.85%

### Standardization of Simple Drugs

#### Drying Loss

**Table 2. Drying loss of carrot (*Daucus carota* L.) simplex**

Replication	Results	Average
1	7.6%	
2	8.9%	8.0167%
3	7.55%	

In this study, the results of the drying shrinkage of carrot simplex obtained a drying shrinkage value of 8.0167%, slightly higher than the study by Ayun et al (2025), in carrot leaves, namely 7.3%. This difference is caused by the difference in sample types between tubers and leaves.

**Determination of Water Content**

**Table 3. Determination of water content of carrot (*Daucus carota* L.) simplex**

Replication	Results	Average
1	5.63%	
2	5.79%	5.77%
3	5.89%	

The water content results obtained for carrot (*Daucus carota* L.) simplicia were 5.77% on average. The water content requirement is no more than 10%, so the results obtained from the water content test are in line with (Tasya et al., 2025).

**Determination of Ash Content**

**Table 4. Results of determining the ash content of carrot samples (*Daucus carota* L.)**

Replication	Results	Average
1	5.1%	
2	6.3%	6.117%
3	6.95%	

**Extraction**

Extraction was carried out using the maceration method using 70% ethanol in a 1:10 ratio. From 130 grams of powdered simplicia, 43 grams of thick extract was obtained. The extraction results produced a yield as shown in Table 5.

**Table 5. Yield of carrot extract (*Daucus carota* L.)**

Simple Powder (g)	Thick Extract (g)	Extract Yield (%)
130 grams	43 grams	33.076%

**Extract Standardization**

**Determination of Water Content**

**Table 6. Results of determining the water content of carrot extract (*Daucus carota* L.)**

Replication	Results	Average
1	5.23%	
2	5.77%	5.683%
3	6.05%	

In this study, the results of the drying shrinkage of carrot simplex obtained a drying shrinkage value of 8.0167%, slightly higher than the study by Ayun et al (2025), in carrot leaves, namely 7.3%. This difference is caused by the difference in sample types between tubers and leaves.

**Determination of Water Content**

**Table 7. Results of determining the ash content of carrot extract (*Daucus carota* L.)**

Replication	Results	Average
1	7.7%	
2	7.85%	7.967%
3	8.35%	

The water content results obtained for carrot (*Daucus carota* L.) simplicia were 5.77% on average. The water content requirement is no more than 10%, so the results obtained from the water content test are in line with (Tasya et al., 2025).

**Ethanol Test Free Inspection**

**The test results showed that the carrot extract did not have an ester odor.**

**Phytochemical Screening**

The test tube phytochemical screening test includes the examination of alkaloids, saponins, tannins, and flavonoids. Identification of chemical compounds, or phytochemical screening, is performed to determine the chemical compounds contained in carrot extract. The results of the carrot phytochemical screening test are shown in Table 8.

**Table 8. Results of phytochemical screening of carrot extract (*Daucus carota* L.)**

Contents	Reagent	Color	Information
Alkaloid	<i>Drangendorff</i>	Orange	Positive
Saponin	Hot distilled water	There is foam	Positive
Tannin	FeCl <sub>3</sub> 5%	Blackish green color	Positive
Flavonoid	Concentrated HCl + Mg Powder	Reddish orange	Positive

**Determination of Total Polyphenol Content of Carrots**

**Determining Operating Time (OT)**

From the results obtained, the absorbance was stable at the 30th minute so it can be concluded that the Operating Time for determining the total polyphenol content of carrot extract was 30 minutes, as can be seen in table 9.

**Table 9. Determination of Operating Time**

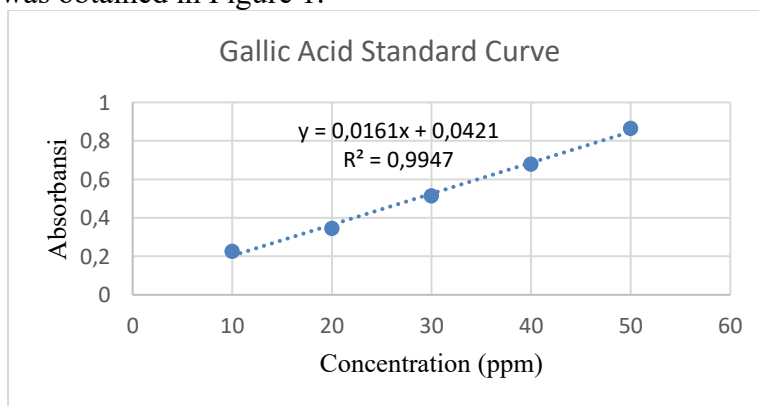
Time (s)	Abs
1440.0 s	0.372
1500.0 s	0.372
1560.0 s	0.373
1620.0 s	0.373
1680.0 s	0.374
1740.0 s	0.374
1800.0 s	0.374

**Determination of Maximum Wavelength (λmax)**

Based on the results of the scanning that has been carried out, the maximum wavelength used for reading the standard curve of gallic acid and samples is 752 nm.

**Determination of the Standard Curve of Gallic Acid**

The results of the standard solution measurements were then made into a curve and the regression equation was obtained in Figure 1.



**Figure 1. Gallic acid standard curve**

**Total Polyphenol Level Testing**

**Table 10. Absorbance measurement values for carrot extract samples (*Daucus carota* L.)**

Replication	Wavelength	Absorbance	Average
1	752 nm	0.178	0.178
2		0.177	
3		0.179	

In this study, the total polyphenol content in carrot extract was 42.205 mg GAE/g, indicating that every gram of carrot ethanol extract contains phenolic compounds with activity equivalent to 42.205 mg gallic acid.

### **Data analysis**

The results of the total polyphenol content in plants are expressed in GAE (Gallic Acid Equivalent) units, namely mg of extract concentration per gram of sample (mg/g). Measurement of total polyphenol compounds in this study showed a correlation coefficient (r) value of 0.997, which is in the range of 0.996-1, thus meeting the linearity requirements of the analytical method. Thus, the regression equation of the gallic acid standard curve obtained can be used as a comparison in determining the levels of total polyphenol compounds in carrots (*Daucus carota* L.). These results are in accordance with research conducted by Maryam et al (2024), which also reported that the gallic acid standard curve has a correlation coefficient (r) value in the range that meets the linearity requirements, even though the samples used are different. The similarity of the linearity values indicates that the Folin-Ciocalteu method used has good accuracy in determining total polyphenol levels in various types of plant samples.

In this study, the total polyphenol content in carrot extract was 42.205 mg GAE/g, indicating that each gram of carrot ethanol extract contains phenolic compounds with activity equivalent to 42.205 mg gallic acid. These results are consistent with a study conducted by Sari & Zulfa (2022), which reported that carrot tuber extract had a total phenolic content of 43.3 mg GAE/g. The closeness of the total polyphenol content values in the two studies indicates that carrot extract is a potential source of phenolic compounds and has antioxidant activity. However, the total polyphenol content in this study was slightly lower than the study by Sari & Zulfa (2022), which is thought to be influenced by differences in solvent concentrations used, where this study used 70% ethanol while the study by Sari & Zulfa (2022), used 96% ethanol. In addition, differences in extraction conditions, particle size of the crude drug, and characteristics of the raw materials can also affect the total polyphenol content produced. Nevertheless, the results of this study indicate that carrot ethanol extract still contains significant phenolic compounds and has the potential to be a source of natural antioxidants.

## **CONCLUSION**

Determination of total polyphenol content of carrot extract (*Daucus carota* L.) can be done using the Folin-Ciocalteu method, which is based on the oxidation reaction of phenolic compounds by Folin-Ciocalteu reagent in alkaline conditions to produce a blue complex that can be measured using a UV-Vis spectrophotometer. The total polyphenol content in carrot extract (*Daucus carota* L.) was obtained at 42.205 mg GAE/g extract. Based on the total polyphenol content value, it shows that carrot extract (*Daucus carota* L.) contains phenolic compounds so it has the potential as a source of natural antioxidants that can play a role in warding off free radicals.

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