Indonesian Food Science and Technology Journal
IFSTJ : Vol 8 No 2, July 2025 (PP:201-207)
ISSN : 2615-367X, DOI. 10.22437/ifstj.v8i2.34374

INDONESIAN FOOD SCIENCE AND TECHNOLOGY JOURNAL (IFSTJ)

Journal homepage : online-journal.unja.ac.id/ifstj/issue/archive

Mitigating Anxiety Symptoms: Potential of Phenol Compounds in Organic Red Ginger from Simalungun Regency

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***Abstract*— According to WHO (World Health Organization), anxiety disorders affected 266 million people globally in 2019, or 5.3% of the population. As the second most common mental disorder after depression, anxiety impacts 284 million people and significantly contributes to the disease burden worldwide. Meanwhile, regional prevalence rates are 19.2%, 16.2%, 9.0%, and 11.8% in North America, Europe, Africa, and Southeast Asia, with particularly 57.5% and 68.8% rates among pregnant women in Bali (2020) and Semarang (2023), respectively. In this context, ginger (*Zingiber officinale*), traditionally used for various ailments, contains phenol such as shogaol, zingerone, gingerol, and 6-shogaol, which have antioxidant and anti-inflammatory properties for reducing anxiety. Therefore, this research aims to analyze the phenol contents of ginger using UV-Vis spectrophotometry in the Faculty of Pharmacy, University of North Sumatra. The results show that there is a strong correlation of 0.9794 between gallic acid and phenol content. Additionally, phenol levels are reported with different extraction methods including 43.2653 mgGAE/g (4.326%) with water, 62.7680 mgGAE/g (6.27%) with 70% ethanol, and 129.7456 mgGAE/g (12.97%) with 96% ethanol. The highest phenol content in organic red ginger using 96% ethanol suggests the potential to reduce anxiety by counteracting free radicals.**

***Keywords*— antioxidant; anxiety, free radicals; organic red ginger; phenol**

Manuscript received June 12, 2024; revised Nov 19, 2024; accepted May 10, 2025. Available online July 27, 2025

I. INTRODUCTION

Phenol aromatic organic compounds are found in a variety of foods and beverages, such as fruits, vegetables, tea, and coffee. Some research suggest that phenol has antioxidant and anti-inflammatory effects for relieving anxiety symptoms [1,2]. In this context, ginger (*Zingiber officinale* Roscoe) an herbaceous perennial plant, is native to Southeast Asia. The plant has been grown for ages in tropical and subtropical climates in the world [3]. Even though some direct sunshine may be tolerated, ginger is grown in green areas. Therefore, ginger is typically grown in shaded regions or beneath larger trees to avoid direct sunlight [4,5].

Based on the description above, the harvest period is approximately 8 to 10 months after planting when the leaves turn yellow. The countries cultivating the greatest quantities globally include India, China, Indonesia, Nepal, Thailand, and Nigeria [6,7]. The cultivation in Vietnam shows a wide geographical distribution, covering household gardens and other industries [8]. Ginger has several uses and advantages, such as lowering inflammation, enhancing digestion, preventing and combating skin aging, treating dandruff, promoting hair development, and acting as an aroma

therapeutic [9]. In addition, the plant possesses a fairly distinctive and pleasant aroma. Research was conducted in 2021 to determine the activity of total phenol, flavonoid, and mineral levels in the rhizomes of the 10 most widely used herbal plants [10]. The results showed that all herbal plant rhizomes had antioxidant activity and contained phenol, flavonoid, and mineral [11].

Rhizome has the highest antioxidant activity and total phenol content. In ginger, rhizome possess the highest flavonoid content [12]. Lidiane Diniz at, all showed that spices plant red ginger contained various phenol compounds, such as eugenol, carvacrol, and gallic acid [13]. These compounds have various important functions as a defense mechanism against stress in humans. Phenol helps plants cope with stress through high antioxidant activity by neutralizing free radicals [4]. In this context, free radicals can damage plant cells and cause a variety of problems to Deoxyribo Nucleic Acid (DNA) and proteins. Antioxidant activity of phenol protects plants from damage and reduces the effects of stress [14,15]. Phenol helps regulate levels of plant hormones in the stress response. Some hormones, such as abscisic acid (ABA) and ethylene can increase anxiety in plants. Phenol reduces levels of ABA and ethylene but increases auxin and gibberellin [16,17].

Another role of phenol is to strengthen plant cell walls and become more resistant to damage caused by stress [18]. Some phenol acts as defense signals that can trigger other responses. Stressed plants produce certain phenol to trigger the production of other defense compounds, such as protective proteins and antioxidant enzymes [19].

Phenol can interact with microorganisms in and around plants. Some of the microorganisms help plants cope with stress [20]. Phenol reduces anxiety by increasing serotonin and dopamine levels. An important role of neurotransmitters is the modulation of mood and emotions [21] to reduce chronic inflammation with the capacity to increase the risk of anxiety. This substance increases antioxidant activity, affording the body protection from the damaging effects of free radicals [22,23]. Previous research have been carried out to determine the comparison of phenol compounds with the highest anti-free radical and flavonoid effects in red ginger using water, 70% ethanol, and 96% ethanol solvents.

II. MATERIAL AND METHODS

This pre-experimental research was conducted in the phytopharmacology laboratory of the Faculty of Pharmacy, University of North Sumatra. Approximately 8 kg of fresh ginger from local farmers in Simalungun Regency, North Sumatera Province, Indonesia which obtained 2000 g of simplicia, resulting in 133 g of concentrated extract.

The total phenol content was initially determined, followed by the preparation of a standard solution of gallic acid. The maximum wavelength of gallic acid was determined using a UV-Vis spectrophotometer in the range of 400 nm to 800 nm. A calibration curve of gallic acid was constructed, and the total phenol content of the red ginger extract was finally determined using UV-Vis spectrophotometry at a maximum wavelength of 742 nm. In this process, 5 repetition samples were used with different concentrations at 10, 20, 30, 40, and 50 ppm. Therefore, simple linear regression calculations were used to analyze phenol content in red ginger extract.

The calculation of phenol content in each type of red ginger extract used 3 repetition samples. The calculation of phenol content was conducted using the linear regression equation obtained at gallic acid calibration curve stage (stage C). The results of each sample in the extraction method were averaged to show phenol content.

Research Stages

A. Preparation of Gallic Acid Raw Master Solution

A 25 mg solution of gallic acid was prepared by weighing the compound and dissolving in 1 mL of methanol. Subsequently, distilled water was added to a final volume of 50 mL, resulting in a concentration of 500 ppm.

B. Determination of Maximum Wavelength of Gallic Acid

In obtaining a 30 ppm concentration, 0.3 mL of the volume was added to a 5 mL flask with 500 ppm gallic acid standard solution. After adding 0.5 mL, vortexing was performed for about a minute. Subsequently, 1 mL of Na2CO3 10% was added, and the mixture was allowed to incubate for 35 minutes. UV-Vis spectrophotometer was used to measure the absorbance in the 400–800 nm wavelength range to determine the maximum wavelength.

C. Preparation of Gallic Acid Calibration Curve

A volume of 0.1 mL, 0.2 mL, 0.3 mL, 0.4 mL, and 0.5 mL of the standard solution of gallic acid (500 ppm) was transferred to each 5 mL flask, leading to a solution of 10 ppm, 20 ppm, 30 ppm, 40 ppm, and 50 ppm, respectively. A volume of 0.5 mL was pipetted and 2 mL of distilled water was added. Subsequently, 0.5 mL of Folin-Ciocalteau 10% was added and vortexed for approximately 1 minute. The mixture was incubated for 35 minutes after adding 1 mL of Na2CO3 10%. The maximum absorbance was observed at 742 nm wavelength to obtain a calibration curve and linear regression equation for gallic acid ($y = ax + b$).

D. Determination of Total Phenol Content of Extract

In obtaining a concentration of 1000 parts per million (ppm), 10 mg of red ginger extract were dissolved in 1 ml of methanol and mixed with 10 ml of distilled water. Meanwhile, 0.5 mL of 1000 ppm concentration solution was pipetted with 2 mL of distilled water, 0.5 mL of Folin-Ciocalteau 10%, and 1 mL of 10% Na2CO3. The solution was incubated for 35 minutes. The absorbance of each concentration was measured at a maximum wavelength of 742 nm against the blank reagent using UV-Vis spectrophotometry.

III. RESULT AND DISCUSSION

Gallic acid value of the correlation coefficient of r=0,988 (p=0,001, p<0,05) was found in the simple regression linear calculation of the curve. Therefore, there was a variable

relationship between gallic acid and phenol content with antioxidant levels and high flavonoids.

TABLE I
REGRESSION EQUATION CALCULATION OF GALLIC ACID UPTAKE CURVE OF ORGANIC RED GINGER EXTRACT

No	X	Y	XY	X^2	Y^2	R	*p*
1	10	0,293	2,93	100	0,0858		
2	20	0,487	9,74	400	0,2372		
3	30	0,623	18,69	900	0,3881	0,988	0,001
4	40	0,783	31,32	1600	0,6131		
5	50	0,849	42,45	2500	0,7208		
Σ	150	3,035	105,13	5500	2,045		
$\bar{X}$	**30**	**0,607**					

Regression equation: Y = 0,014X + 0,185

The r value on gallic acid refers to the correlation coefficient (r) as a measure describing the linear relationship between two variables. The r value ranges from -1 to 1 when close to 1 where the two factors typically have a significant and positive association. The connection between the two variables is strong and negative when r is near -1. There is no linear relationship or a weak connection between the variables when r approaches 0.

TABLE II
DETERMINATION OF TOTAL PHENOL CONTENT OF ORGANIC RED GINGER WATER EXTRACT

Sample (g)	Sample Volume (ml)	TF	Absor-ban Avera-ge	Con-centra-tion (µg/ml)	Fenol Level Average (mgGAE /g extract)	Average
0,0103	10	1	0,833	46,287	44,9376	44,9376 mgGAE/ g extract or (4,494%)
0,0103	10	1	0,833	46,287	44,9376	
0,0103	10	1	0,833	46,287	44,9376	

TF= Thinning Factor

The phenol content of each red ginger extract sample was calculated using the regression equation. The results of repetitions 1, II, and III obtained total and average phenol content in red ginger water extract of 44,9376 mgGAE/g extract or 4,494 %. A high concentration of phenol at 4,494% shows good antioxidant properties. The results of repetitions 1, II, and III obtained total and average phenol content in 70% ethanol extract of 62.7860 mgGAE/g ginger or 6.27%.

The results of repetitions 1, II, and III obtained a total average phenol content in ethanol 96% red ginger water extract of 129.7456 mgGAE/g or about 12.974%. The r value can be used to measure the relationship between gallic acid concentration and other variables, such as antioxidant activity or biological effects in plants. Gallic acid absorption curve refers to the relationship between the concentration and light absorption rate at a specific wavelength. Plants in the Verbenaceae family are well-known for possessing bioactive chemicals, such as gallic acid, which is measured as an antioxidant [24]. Additionally, gallic acid is used as a standard in the analysis of total phenol content calculated as the concentration in the extracted plant sample [25].

TABLE III
DETERMINATION OF TOTAL PHENOL CONTENT OF 70% ETHANOL EXTRACT OF ORGANIC RED GINGER

Sample (g)	Sample Volume (ml)	TF	Absor-ban Avera-ge	Con-centra-tion (µg/ml)	Fenol Level Average (mgGAE/g extract)	Average
0,0105	10	1	1,186	71,5000	68,0952	68,3363 mgGAE/g extract or (6,834%)
0,0104	10	1	1,187	71,5714	68,8187	
0,0105	10	1	1,186	71,5000	68,0952	

TF= Thinning Factor

Gallic acid uptake curves are used to compare total phenol content in different types of plants and assess the effects of environmental stress. This is because total phenol content can change in response to environmental stress such as drought, soil salinity, and pest attacks. Gallic acid uptake curves are used to monitor changes and assess the effects of environmental stress on plants [26]. The absorption curve provides an estimate of total phenol content [25].

TABLE IV
DETERMINATION OF TOTAL PHENOL CONTENT OF 96% ETHANOL EXTRACT OF ORGANIC RED GINGER

Sample (g)	Sample Volume (ml)	TF	Absor-ban Avera-ge	Con-centra-tion (µg/ml)	Fenol Level Average (mgGAE/g extract)	Average
0,0105	10	1	2,349	154,571	147,2109	147,1533 mgGAE/g extract or (14,715%)
0,0104	10	1	2,351	154,714	148,7637	
0,0106	10	1	2,344	154,214	145,4852	

TF= Thinning Factor

Other phenol compounds in the plant may have a different light absorption to gallic acid, which may affect the analysis [27, 52]. Therefore, additional analytical methods, such as high-performance liquid chromatography (HPLC), are crucial to obtain a complete phenol profile [28]. Total phenol content in aqueous solvents varies depending on the different phenol content. Some plants with high phenol content generally include fruits, vegetables, and nuts. Cultivation conditions including environmental stress, pests, diseases, and fertilization affect phenol levels [29].

Total phenol content and ginger extracted with 70% ethanol may differ based on several variables, including Type of ginger:

Red ginger has a higher phenol content than white. Extraction method: Extraction duration, temperature, and ginger-ethanol ratio affect the amount of phenol extracted. Analytical methods: Different methods for measuring phenol content may give slightly different results [30].

Several research have shown that phenol content extracted with 70% ethanol ranges from 400 to 1100 mg gallate equivalent per 100 g of dried ginger [31]. The journal 'Molecules' found that total phenol content in red ginger extracted with 70% ethanol was 1062 mg gallic equivalent per 100 g [32]. Total phenol is stated in mgGAE/g and dry extract produced by high-pressure CO_2 extraction possesses 871 mg/g [33].

Maulana et,all in the found that total phenol content in ginger extracted with 70% ethanol was 785 mgGAE/g [34]. In comparison to ascorbic acid with an IC50 of 3.90 µg/mL, the 96% ethanol extract reported the greatest DPPH inhibition concentration of 50% (IC50), measuring 3.38 µg/mL. The activity of antioxidants is strongly correlated with total flavonoid content [35]. The method of extracting phenol from plants also affects the analysis results. Different extraction methods produce extracts with varying phenol levels [36]. Methods of analysis: Several methods are used to measure the total phenol content in aqueous extracts, such as Folin-Ciocalteu and Folin-Denis. Each method has advantages and disadvantages, hence, analytical results may differ based on the approach taken [37].

Total phenol content in aqueous extracts can range from 100 mg/g to 1000 mg/g dry sample. In this research, phenol content was below 100 mg/g. Total phenol content of ginger extracts in ethanol and water baked for 24-48 hours ranged from 504 to 31 µg/mL [38,39]. Many factors influence the difference and the value may vary. Total phenol content is an indicator of antioxidant activity of a plant. Other phenol compounds in the plant, such as flavonoids and anthocyanins, may also contribute to antioxidant activity. Therefore, comprehensive analyses should be conducted to ascertain the overall antioxidant activity and the spectrum of phenol compounds in a plant [40]. There are several markers of antioxidant capacity besides the total phenol concentration [41].

Scientists are still examining the mechanism of phenol in helping plants cope with stress and as a common mental disorder characterized by excessive feelings of worry, anxiety and fear. Chronic inflammation contributes to various diseases, including depression, anxiety and cognitive impairment [42]. Some research suggest that anti-inflammatory effects may help relieve symptoms of anxiety. A decrease in oxidative stress defined by an imbalance between free radicals and antioxidants is a process underpinning the association between anti-inflammatory effects and anxiety. The body may be shielded from free radical damage by anti-inflammatory effects to reduce anxiety [43]. Neurotransmitter modulation plays a role in chronic inflammation, contributing to a disturbance in the balance of serotonin and dopamine. Anti-inflammatory effects also restore the balance of neurotransmitters and relieve anxiety symptoms [44].

Chronic inflammation occurs in the brain and contributes to anxiety symptoms. Anti-inflammatory effects also reduce inflammation in the brain and relieve anxiety symptoms [1]. The use of ethanol solvents with different levels in the determination of phenol has several reasons. The effectiveness of ethanol extraction with different levels has different capabilities for phenol compounds [36]. In this context, ethanol with high levels of 96% is more effective in extracting polar phenol compounds compared to 70% ethanol. This is because ethanol with high levels has stronger polar properties, thereby attracting and dissolving polar phenol compounds better [45]. Phenol content using water extraction obtained an average of 43.2653 mgGAE/g extract or 4.326% using 70% ethanol. The average phenol content obtained was 62.7680 mgGAE/g extract or 6.27% and the use of 96% ethanol extract reported 129.7456 mgGAE/g extract or 12.97%.

Based on the description above, ethanol content affects the selectivity of extraction. Low levels of ethanol (50%) can extract other compounds besides phenol, such as sugars and pigments. Meanwhile, high levels of 96% ethanol are more selective in extracting phenol and minimizing contamination from other compounds [46].

The sample solubility of ethanol content needs to be adjusted. This is because the solubility of samples with high water content may require lower levels [41]. The analysis aims to obtain high levels of total phenol using ethanol with high levels of 96%. Ethanol with varying levels of 50%, 70%, and 96% can be used to extract phenol compounds with different polarities to obtain a more detailed phenol compound. In this context, 70% to 96% is often used in the determination of phenol [47,48,49]. However, ethanol is not the only solvent used for phenol determination [50,51,52]. Other solvents such as methanol, acetone, and water can also be adopted with different advantages and disadvantages [51, 53].

LIMITATIONS OF THE RESEARCH

The results cannot be generalized to organic ginger in other regions since this research uses samples from one location. Additionally, the measurement results of phenol content with the method used may differ from other methods. This research analyzes phenol content without conducting clinical trials to strengthen the result. The optimal dose used to reduce anxiety symptoms has not been explored.

SUGGESTIONS FOR FURTHER RESEARCH

The sample should be expanded beyond organic ginger in Simalungun using different measurement methods. Furthermore, clinical trials must be conducted on animal subjects followed by humans to determine the optimal dosage.

IV. CONCLUSION

The results show that there is a strong correlation of 0.9794 between gallic acid and phenol content. Additionally, phenol levels are reported with different extraction methods including 43.2653 mgGAE/g (4.326%) with water, 62.7680 mgGAE/g (6.27%) with 70% ethanol, and 129.7456 mgGAE/g (12.97%) with 96% ethanol. The highest phenol content in organic red

ginger using 96% ethanol suggests the potential to reduce anxiety by counteracting free radicals.

ACKNOWLEDGMENT

This research is independent and unfunded by any organization. The authors are grateful to Nanda Novziransyah, Dewi Pangestuti, Surya Akbar, Adi Raja Brando Lubis, and Mayang Sari Ayu, Donalry Agus Saputra for rendering assistance and support in the entire process. Ethical clearance was obtained from the Faculty of Medicine of the Islamic University of North Sumatra Number: 407/EC/KEPK/FKUISU/VII/2024.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

USE OF ARTIFICIAL INTELLIGENCE (AI) TOOLS STATEMENT

We used Grammarly (Grammarly Inc., 2025) to improve the clarity and grammar of the manuscript. The authors reviewed and approved all changes.

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