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GC-MS Analysis of Ginger Rhizome with Various Extraction Methods

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Abstract

*Ginger (*Zingiber officinale*) is a rhizome plant well-known as a spice and medicinal material. Either one of the bioactive components is an essential oil, which is efficacious in preventing and treating various diseases. This research aims to determine the percentage yield of extracts and GC-MS analysis of the percentage of the chemical content of essential oils of ginger with various extraction methods. Firstly, compounds from ginger rhizomes were extracted using maceration, ultrasonic, and distillation techniques, and the extracts were analyzed using GC-MS Spectrophotometer. The highest percentage extract yield from ultrasonic extraction was 21.29%, then maceration extraction was 6.26%, and distillation extraction was 5.00%. The GC-MS results showed that the secondary metabolite compounds from maceration, ultrasonics, and distillation contained zingiberol, zingiberin, shogaol, borneol, linalool, citral, geraniol, and E-citral. The zingiberol group of compounds was obtained with the highest percentage from ultrasonic extraction at 1.07%, then maceration extraction at 1.03%, and distillation extraction at 0.27%.*

I. Introduction

Ginger (*Zingiber officinale*) is a rhizome known as a spice and medicinal ingredient. The rhizome is finger-shaped, getting more extensive in the center. Most of the intense flavor is due to the ketone compound zingiberone. Ginger belongs to the Zingiberaceae family. The characteristic properties of ginger are obtained from volatile oils and oleoresins. Ginger smells like essential oil, while oleoresin gives ginger a spicy flavor (Fuadi, 2012).

Ginger is used for spices, herbal medicines, and beverages. Ginger is also an herbal medicinal ingredient. It contains essential oils with active chemical compounds such as Zingiberin Lemonin, Borneol, Shogaol, Cineole, Fellandren, Zingiberol, Gingerol, and Zingerone which are efficacious in preventing and treating various diseases (Aryanta, 2019).

The secondary metabolite compounds of ginger plants mainly come from flavonoids, phenols, terpenoids, and essential oils (Handrianto, 2016). One of the compounds in ginger essential oil is sesquiterpenes, a group of terpenoid hydrocarbon compounds that are very dominant, namely gingerene (35%), curcumin (18%) and farnesene (10%). The essential oil also contains phenolic compounds such as 1,8-cineole, eugenol, zingerone, gingerdiols (Barki *et al*., 2017).

The main pharmacological activity of ginger is presumed to come from gingerol and shogaol. Phenylacetone or vanilyl ketone from ginger including 6-gingerol 8- gingerol and 10-gingerol, 6-shogaol, 8-shogaol, 10-shogaol and zingerone, 6-paradol, 6- and 10-dehydrogingerdione and 6-and 10 gingerdione have also been identified (Maulida, dan Indradi, 2019)

Ginger essential oil can be obtained by extraction or distillation. The extraction methods used are maceration and ultrasonic, in which maceration is used because it is suitable for extracting non-thermal compounds such as ginger essential oil (Rahmadani *et al*., 2018). Ultrasound can reduce extraction time and increase extraction yield (Fuadi, 2012).

Several researchers have reported the use of extraction methods to identify the type and percentage of chemical compounds in ginger essential oil using maceration, water distillation, and ultrasonic extraction methods. Extraction by water distillation yielded the most dominant compound, Ar-curumene, at 17.10%. Ultrasonic extraction revealed zingiberene compound which is the dominant compound in ethanol and ethyl acetate extracts with a percentage of 31.43 ± 14.88% (Nur *et al*., 2019). Maceration extraction using a solvent ratio of ethanol and methanol obtained more dominant zingiberene compounds with a percentage of 37.68% and 43.69% (Situmorang and Ricky, 2022).

In addition to using maceration, water distillation, and ultrasonic extraction methods to identify the type and percentage yield of chemical compound extracts in essential oils, several studies were conducted using other extraction methods, namely Sokhlet extraction. Phytol compounds with a concentration of 7.7-21.6% (Hidayati and Khaerunnisa, 2018).

The type of essential oil compound can be determined by the gas chromatography method. Gas chromatography is a dynamic method for separating and detecting volatile compounds in a mixture. A mass spectrophotometer is a device that can select gas molecules that have a mass or weight charge (Amin *et al*., 2014).

II. Research Method

II.1 Sample Collection and Processing

The sampling of ginger (*Zingiber officinale*) was carried out on ginger that grows in the South Sulawesi region. Ginger (*Zingiber officinale*) of as much as 8 kg has been collected then cleaned from dirt, chopped, and dried in the dryer. Ginger that has been dried as much as 640 grams was ground with a blender. The powder has been finely weighed and stored in a clean place and free from water for the extraction process.

II.2 Sample Extraction

II.2.1 Maceration

A total of 200 grams sample was put into a maceration container then the sample was soaked with 70% ethanol (1; 7.5) for 3 days with occasional stirring after which it was filtered. The macerate was collected and evaporated with a rotary vacuum evaporator at 60°C and concentrated on a waterbath at 60-70°C until a thick extract was obtained.

II.2.2 Ultrasonic

50 grams of ginger powder was weighed and put into a 500 mL Erlenmeyer and 375 mL of 70% ethanol solvent (1:7.5) was added. Extraction was carried out ultrasonically at 50°C for 2 hours. The extract was filtered with filter paper, then evaporated using a rotary vacuum evaporator at 60°C until a thick extract was obtained.

II.2.3 Distillation

A total of 50 grams of ginger powder was weighed and put into a distillation flask, then 375 mL of distilled water (1:7.5) was added. After that, the distillation process was carried out for 4-5 hours with temperatures ranging from 100°C-105°C.

II.3 Identification of Compound Components using Gas Chromatography–Mass Spectrometry (GC-MS)

II.3.1 Sample preparation

0.1 mL of sample was added to a mixture of chlorophom and methanol (1:1) as much as 5 mL. extraction using a sonicator for 20 minutes at 40°C. the top layer formed was pipetted into a vial and GC-MS test.

II.3.2 Operating GC-MS Ultra QP 2010 Shimadzu

Pipette 0.5 mL of isolate into a 50 mL volumetric flask and dilute with acetone and dilute to the limit. Pipette as much as 3 mL and put it into a vial. GC-MS instrument conditions Injector temperature 250°C with Splitless mode, pressure 76.9 kPa and flow rate 14 mL/min and ratio 1:10. Ion source and interface temperatures 200°C and 280°C, solvent cut time 3 minutes, 400-700 m/z. Column type SH-Rxi-5Sil MS column length 30 m with an inner diameter of 0.25 mm. The initial column temperature was 70°C with a holding time of 2 minutes and the temperature was increased to 200°C at a rate of 100°C/min and a final temperature of 280°C with a holding time of 9 minutes at a rate of 50°C/min for a total analysis time of 36 minutes. The chromatogram data obtained was read using NIST and Wiley 9 libraries.

III.4 Data analisys

The percentage yield value of the extract can be calculated by the Formula 1.

$$\text{Percentage Yield} = \frac{\text{Lemongrass extract weight}}{\text{Lemongrass sample weight}} \times 100\% \quad (1)$$

III. Results and Discussion

The samples used in this study were ginger rhizomes (*Zingiber officinale*) from South Sulawesi. 8 kg of ginger was dried into simplisia. In the manufacturing process, fresh ginger was washed thoroughly with running water to remove impurities such as soil and other materials. Ginger was sliced thinly to facilitate drying, then dried in a drying cabinet for a week until 640 grams of ginger simplisia was obtained, after which the sample was pollinated to reduce the size and increase the contact area between the powdered simplisia and the liquid so that the active substance content has been much extracted. The ginger rhizome powder was then weighed as much as 200g for maceration extraction, 50g for water distillation extraction and 50g for ultrasonic extraction. The three methods were used to determine which extraction method gave the highest percentage yield of extracts.

70% ethanol solvent was used in this study because ethanol is a universal solvent that can attract compounds that are soluble in non-polar and polar solvents (Padmasari *et al*., 2013). The essential oil component of ginger is terpenes, with the essential oil contained in the oleoresin being polar. Because the polar components of essential oils such as zingiberol dissolve in solvents that are also polar, while non-polar essential oil components such as zingiberene will also be partially extracted (Anam, 2010).

The percentage yield of extracts of ginger rhizomes extracted using maceration, ultrasonic, and water distillation can be seen in Table 1.

Table 1. Effect of extraction method comparison on Lemongrass stem extract yield (*Cymbopogon citratus*)

Extraction	Solvent	Fresh Sample Weight (kg)	Dried Sample Weight (g)	Total Solvent (mL)	Extract Weight (g)	Yield Value (%)
Maceration	Ethanol	8	200	3000	12.52	6.26
Ultrasonic	70%		50	375	10.64	21.29
Destillate	Aquadest		50	375	2.50	5.00

The extraction of chemical compounds from ginger rhizomes in this study was carried out using various extraction methods, namely maceration, ultrasonic and water distillation. Maceration extraction method is used to extract compounds that are unstable to heat. Ultrasonic extraction is used because some ultrasonic extractions give better and faster extraction results than conventional methods (Jos *et al*., 2011). Distillation extraction is used because it is commonly used to extract essential oils from plants containing essential oils (Warsito, 2018). The percentage yield of extracts obtained by maceration extraction process is different from the percentage yield of extracts obtained by ultrasonic extraction and water distillation. The extraction yield rate in ultrasonic extraction is higher than in maceration extraction and water distillation. With ultrasonic extraction, the extraction yield was 21.29%. This is due to the presence of ultrasonic waves and assisted by a little heating resulting in a higher extraction percentage compared to maceration extraction of only 6.26% and water distillation of 5.00%. The extraction method affects the yield of the extract, the higher the yield value produced shows the value of the extract produced more. Maceration extraction has the disadvantage that it takes a long time and requires a lot of solvent, while ultrasonic extraction utilises ultrasonic waves to accelerate the extraction process so that ultrasonic extraction has a short extraction time, more extracts and less solvent required (Nofita *et al*., 2021).

The measurement of ginger rhizome extract was then analysed using a GC-MS spectrophotometer which showed the presence of several chemical compounds contained in the ginger rhizome extract seen based on peak height, retention time (RT), and percent area. The results of identification of essential oil compounds in ginger rhizome extract (*Zingiber officinale*) can be seen in Table 2.

Table 2. Chemical compounds of essential oil extracted from ginger rhizome (*Zingiber officinale*)with the three highest percentage areas

Extration method	No	Peak	RT (minute)	Chemical compound	% area	Boiling point (°C)
Ethanol 70% extract of Ginger with maceration	1	50	16,013	2H-3,9a-Methano-1-benzoxepin, octahydro-2,2,5a,9-tetramethyl-, [3R-(3.alpha.,5a.alpha., (Zingiberol)	10,68	287°C - 288°C
	2	30	13,291	Selin-6-en-4.alpha.-ol (Zingiberol)	5,27	287°C - 288°C
	3	33	13,703	2-(4a,8-Dimethyl-2,3,4,5,6,8a-hexahydro-1H-naphthalen-2-yl)propan-2-ol (Zingiberol)	3,88	287°C - 288°C
	4	28	13,067	2-Naphthalenemethanol, 1,2,3,4,4a,5,6,8a-octahydro-.alpha.,.alpha.,4a,8-tetramethyl-, (2.a (Zingiberol)	2,63	287°C - 288°C
	5	31	13,43	2-((2R,8R,8aS)-8,8a-Dimethyl-1,2,3,4,6,7,8,8a-octahydronaphthalen-2-yl)propan-2-ol (Zingiberol)	2,26	287°C - 288°C
Ethanol	1	85	25,864	1-(4-Hydroxy-3-	14,8	427,5°C

70% extract of Ginger with ultrasonic				methoxyphenyl)dec-4-en-3-one (6-shogaol)	0	
	2	88	27,802	5-Hydroxy-1-(4-hydroxy-3-methoxyphenyl)decan-3-one (6-gingerol)	6,47	265,0°C
	3	21	11,516	1,3-Cyclohexadiene, 5-(1,5-dimethyl-4-hexenyl)-2-methyl- (Zingiberene)	5,32	34°C
	4	25	11,915	Cyclohexene, 3-(1,5-dimethyl-4-hexenyl)-6-methylene-, (Zingiberene)	4,88	34°C
	5	23	11,688	beta.-Bisabolene (CAS) (Bisabolene)	3,01	34°C
Aquadest extract of ginger with water-destillation	1	16	7,260	3-CYCLOHEXENE-1-METHANOL, .ALPHA.,.ALPHA.,4-TRIMETHYL-, (S)- (Geraniol)	35,02	212°C
	2	14	6,926	BICYCLO[2.2.1]HEPTAN-2-OL, 1,7,7-TRIMETHYL-, (1S-ENDO)- (Geraniol)	28,27	212°C
	3	8	5,731	Linalool	9,42	198°C
	4	17	7,675	2,6-OCTADIEN-1-OL, 3,7-DIMETHYL-, (Z)- (Geraniol)	4,35	212°C
	5	19	8,055	Fenchol, exo-	3,68	230°C

Maceration extraction of ginger rhizomes contains various types of chemical compounds, namely zingiberene, zingiberol and bisabolene which based on the results of the analysis that has been analysed from the GC results detected there are 25 peaks as chemical compounds of essential oil groups, After being analysed into the form of essential oil grouping based on GC and MS information, 5 peak chemical compounds were obtained which had the highest percentage area in the content of ginger essential oil where the components of essential oil chemical compounds are 2H-3,9a-Methano-1-benzoxepin, octahydro-2,2,5a,9-tetramethyl-, [3R-(3. alpha.,5 a.alpha., zingiberol group which has the highest peak with a percentage of 10.68% at retention time (16.01 minutes) where this compound is not found in ultrasonic extraction and water distillation methods. The main components of ginger essential oil that cause the fragrant odour are zingiberene and zingeberol. Zingiberene is a sesquiterpene hydrocarbon with the formula $C_{15}H_{24}$, while zingiberol is a sesquiterpene alcohol with the formula $C_{15}H_{26}O$ (Handayani *et al*., 2015).

Ultrasonic extraction of ginger rhizomes contains various types of chemical compounds i.e geraniol, E-citral, 6-shogaol, 6-gingerol, zingiberene, bisabolene, farnesen and zingiberol which based on the results of the analysis that has been analysed from GC results detected there are 23 peaks as chemical compounds of essential oil groups, After being analysed into the form of essential oil grouping based on GC and MS information, 5 peaks of chemical compounds were obtained which had the highest percentage of area in the content of ginger essential oil with the chemical compound component of essential oil 1-(4-Hydroxy-3-methoxyphenyl)dec-4-en-3-one group 6-shogaol which has the highest peak with a percentage of 14.80% at retention time (25.86 minutes) where this compound is not found in maceration and water distillation extraction methods. Shogaol compounds are found in ultrasonic extraction because extraction temperatures above 45°C will cause changes in gingerol to shogaol components (Hartuti and Supardan, 2013). 6-shogaol is a dehydration of gingerol, changes in the form of gingerol during the heating process will form shogaol so that in the ultrasonic extraction method the compound with the highest peak is 6-shogaol because it is decomposed (Pathiassana *et a*l., 2020).

Water distillation extraction of ginger rhizomes contains various types of chemical compounds, namely linalool, citral, geraniol, E-citral, and zingiberol which based on the results of the analysis that has been analyzed from the GC results detected there are 11 peaks as chemical compounds of essential oil groups, after being analysed into the form of essential oil grouping based on GC and MS information obtained 5 peak chemical compounds that have the highest percentage area in the content of ginger essential oil with the components of essential oil chemical compounds 3-cyclohexene-1-methanol,.alpha.,.alpha.,4-trimethyl-, (s)-geraniol group which has the highest peak with a percentage of 35.02% at retention time (7.26 minutes) where this compound is not found in maceration and ultrasonic extraction methods.

In this study only discusses the 5 highest peaks because the 5 peaks represent the main components of the gologan of ginger essential oil chemical compounds based on the extraction method, this can indicate that each extraction method has different types of chemical compounds obtained based on data from GC and MS results seen in Table 3.

Table 3. GC-MS identification results of essential oil compounds of ginger rhizome extract (*Zingiber officinale*) with the same chemical compound components.

Essential oil	Peak	RT (minute)	Name of chemical compound	% area	Molecular weight	Compound group
Maceration	21	12,285	Cyclohexanemethanol, 4-ethenyl-.alpha.,.alpha.,4-trimethyl-3-(1-methylethenyl)-, [1R-(1.	1,03	222	Zingiberol
Ultrasonic	27	12,283	Cyclohexanemethanol, 4-ethenyl-.alpha.,.alpha.,4-trimethyl-3-(1-methylethenyl)-, [1R-(1.	1,07	222	Zingiberol
Water-destillation	26	12,297	Cyclohexanemethanol, 4-ethenyl-.alpha.,.alpha.,4-trimethyl-3-(1-methylethenyl)-, [1R-(1.	0,27	222	Zingiberol

The identification results of GC-MS method of ginger extract (*Zingiber officinale*) obtained various essential oil compounds consisting of zingiberin, zingiberol, geraniol, citral, linalool, shogaol and borneol. Cyclohexanemethanol, 4-ethenyl-.alpha.,.alpha.,4-trimethyl-3-(1-methylethenyl)-, [1R-(1.a chemical compound components of zingiberol group are compounds that exist from all three extraction methods namely maceration, ultrasonic and water distillation. Ultrasonic extraction obtained the highest percentage area of 1.07% at a retention time of 12.28 minutes compared to the maceration extraction method with a percentage of 1.03% at a retention time of 12.28 minutes and water distillation with a percentage of 0.27% at a retention time of 12.29 minutes. This is due to the influence of the solvent used, namely 70% ethanol, which is able to dissolve chemical compounds optimally, because 70% ethanol still contains quite a lot of water (30%) which helps the extraction process so that some of these compounds can be atomised in ethanol and some are attracted to water (Sani *et al.*, 2014) and influenced by the extraction method used, where maceration does not use the heating process, while distillation uses heating and ultrasonic uses ultrasonic waves and temperature.

Based on the data obtained, it appears that to extract Zingiberol compounds from essential oils of ginger should use maceration extraction. For 6-Shogaol compounds, ultrasonic extraction should be used, because ultrasonic extraction is affected by heat, so that hot gingerol compounds break down into shogaol compounds, while for geraniol compounds, water distillation should be used.

IV. Conclusions

Based on the results of the research that has been carried out, it can be concluded that The percentage yield of ginger extract from the three extraction methods used, i.e maceration, ultrasonic, and water distillation, showed different values, while the ultrasonic extraction method achieved a high extract yield of 21.29% compared to maceration extraction of 6.26% and water distillation of 5.00%.

The results of identification of essential oil compounds using GC-MS spectrophotometer of ginger extract obtained various types of chemical compounds of ginger essential oil (*Zingiber officinale*) consisting of zingiberene, zingiberol, geraniol, citral, linalool, shogaol and borneol. The compound cyclohexanemethanol, 4-ethenyl .alpha, alpha, 4-trimethyl 3(1methylethenyl)-,[1R-(1.a chemical compound component of the zingiberol compound group is a compound obtained from all three maceration, ultrasonic and water distillation extraction methods. The compound has the highest percentage area of 1.07% at a retention time of 12.28 minutes in ultrasonic extraction compared to the maceration extraction method with a percentage of 1.03% at a retention time of 12.28 minutes and water distillation with a percentage of 0.27% at a retention time of 12.29 minutes.

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