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(Research Article)

Comparative Studies on Physicochemical Properties and GC-MS Analysis of Essential Oil of the Two Varieties of Ginger (*Zingiber officinale*)

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ABSTRACT
Ginger or ginger root is the rhizome of the plant Zingiber officinale consumed as a delicacy, spice and medicine. It lends its name to its genus and family (Zingiberaceae). In this study essential oil of the two varieties of ginger from Bangladesh and China were investigated by GC-MS. Total 11chemical constituents were found by gas chromatography and mass spectrometry (GC-MS) analysis from the essential oil of Bangladeshi ginger. The oil rich in zingiberene, (38.10%), beta phellandrene (12.00%), β-sesquiphellandrene (9.546%), α-curcumene (9.224%), camphene (5.94%), α-Farnesene (4.573%), β-bisabolene (4.39%), citral (3.91%), α-pinene(2.33%), eucalyptol (1.27%), germacene D(1.14%) respectively i.e. total monoterpene(25.45%) and sesquiterpene (66.97%). On the other hand total 10 chemical constituents were found from the China zinger .The oil contains α-curcumene(26.54%),camphene(20.60%),citral(17.90%),α-pinene(6.63%),borneol(5.40%),β-isabolene(4.57%),eucalyptol(2.44%), 1,2disisopropenylcyclobutane (2.84%), hexadecanoic acid (1.55%), L-alloaromadendren(0.86%) i.e total monoterpene (56.24%) and sesquiterpene (33.97%). Quantification of active principles through analytical tools is essential for establishing the authenticity and credibility. Steam distillation extraction combined with GC-MS has been shown to be a valuable tool for the analysis of ginger constituents and can provide a useful guide to component variation. The main objective of the present study was focused on identification and quantification of chemical constituents present in the essential oil of ginger by GC-MS methods.

Key Words: Gas chromatography and mass spectrometry (GC-MS), *Zingiber officinale*, Rhizome quantification, Essential oil, Sesquiterpene, Chemical constituents.

INTRODUCTION
Zingiber officinale is the most important of all the spices obtained from the underground plant part. It was also one of the first oriental spices to be grown to the Europeans. But now it is found to grow extensively in the tropical and sub-tropical regions of the world particularly in Bangladesh, India, Taiwan, Jamaica, Africa, Mexico, China and Japan. It is a household remedy for dyspepsia, flatulence, colic and diarrhea[1]. Ginger rhizomes contain both aromatic and pungent compounds [2]. Ginger contains from 0.25 to 3% of volatile oil of light yellow color [3]. The major pungent compounds found in ginger are the gingerols [4]. 6-gingerol [5-hydroxy-1-(4`-hydroxy-3`-methoxyphenyl)-3-decanone] has been shown to have an antipyretic, hypotensive, cardiotonic, antiplatelet, antiangiogenic, anti-inflammatory and analgesic, cytotoxic, apoptotic and antitumor activities [5-9]. The oil is sparingly soluble in 95 percent alcohol but is generally soluble in 90 percent alcohol. Ginger oil is used primarily as food flavoring agent in soft drinks, as spices in bakery products, in confectionary items, pickles, sauces and as a preservatives etc. It is used for treating nausea caused by sickness, morning sickness and chemotherapy [10, 11].
However many researches have been carried out on *Zingiber officinale,* but no systemic research on comparative studies has been reported on the essential oil of ginger in Bangladesh and China. Some disagreement about the presence of its constituents was observed. Therefore the present work was undertaken to carry out a complete investigation of the essential oil of *Zingiber officinale* of two varieties (Bangladeshi and Chinese) including its physical and chemical properties along with by GC-MS analysis.

MATERIALS AND METHODS
The fresh Bangladeshi ginger is available in the local markets of Dhaka city (collected from Chittagong). And the China ginger is also collected from local markets as China ginger is being imported in Bangladesh in bulk quantities. The collected samples were washed thoroughly with water to remove earthy matter. Then they were dried. Finally the dried ginger was cut to 2-5 cm flat chips with heavy knives.

Extraction of Essential Oil

There are a number of methods employed for the extraction of essential oil or volatile oil from the plants. In the present study, steam distillation method was used. This extraction procedure was carried out at mild conditions so it could retain its volatile constituents and characteristic_flavor. In the process, definite amount of sample (dirt free fresh ginger rhizome) were taken in a distillation flask (Clevenger's apparatus). Then distilled water was added up to two third volumes of the flask capacity. Then the flask was heated by electric heating mental for 4 hours. Volatile substances of ginger and generated steam in the flask were condensed by water condenser. The essential oil was lighter than water and so could be separated out. The steam distilled essential oil layer which was collected over water, was extracted and washed with analytical grade ether or chloroform. The ether extract of the oil was dried over anhydrous Na_2SO_4 and then filtered. It was collected in vial. The ether or chloroform was removed in vacuum condition. Thus the essential oil of fresh ginger was collected.

GC-MS Analysis

The essential oil of *Zingiber officinale* (Ginger) of two varieties were analysed by Electron Impact Ionization (EI) method on GC-17A gas chromatograph, coupled to a GC-Ms 2010 plus mass spectrometer; fused silica capillary column temperature of 40˚ C (was held 2 min) was maintained with carrier gas helium at a constant pressure of 90kPa.Samples were injected by spilliting with the split ratio 10. Essential oil sample was dissolved in chloroform. The operating condition were as follows: name of column- RTS-5MS, diameter 30 cm, length 0.25mm, temperature of the column- initial temperature 40˚C (was held 2 min) , injector temperature- 220 ˚C, helding time 5 min, column packing- column packing was done with 10% diethylene glycol succinate on 100-120 mesh diatomic CAW, splitting- samples were injected by splitting with the spilt ratio 10, carrier gas- helium gas at constant pressure 90 kPa, sample dissolved in chloroform, range of linear temperature increase 10˚C per min.

Preparation of Essential Oil Samples for GC-MS Analysis

Essential oil was diluted to 7% by chloroform. An inert gas (i.e. nitrogen) was introduced, from a large gas cylinder through the injection part, the column and the detector. The flow rate of the carrier gas was adjusted to ensure reproducible retention time and to minimize detector dirt. The sample was then injected by a micro syringe through a heated injection part when it was vaporized and carried into the column. The long tube of the column was tightly packed with solid particles. The solid support was uniformly covered with a thin film of a high boiling liquid (the stationary phase). The mobile and stationary phases were then partitioned by the samples and it was separated into the individual components. Then the carrier gas and sample component were emerging from the column and passed through a detector. The amount of each component at a particular concentration is recorded by the device and generates a signal which was registered electrically. The signal passed to a detector.

Identification of the Components

The physicochemical properties of the essential oil of *Zingiber officinale* (Ginger) of two varieties are presented in table-1. Interpretation of mass spectroscopy (GC-MS) was conducted using data base of National Institute Standard and Technology (NIST) having more than 62000 patterns. The spectrum of the unknown component was compared with the spectrum of the known component stored in the NIST library. The retention time, molecular weight, molecular formula and composition percentage of the sample material was recorded and presented in table 2(Essential oil of ginger from Bangladesh and China).

RESULTS AND DISCUSSION

The physical characteristics such as color, appearance, specific gravity, optical rotation, solubility, refractive index of the essential oil were determined by conventional methods. The result of the physical properties of *Zingiber officinale* (Ginger) rhizome essential oil of Bangladesh and China are shown in Table-1. Chemical characteristics of the oil such as acid value, ester value were determined by the conventional methods. The comparative results are shown in Table-2.

The slight variation of this oil content and the composition of the essential oil depend on several factors such genotype, stage of maturity, cultivation peculiarities, soil composition and climate differences in various geographical locations. Fluctuation of the oil composition can impart change in the organoleptic properties of the plant belonging to the botanical spices and variety. So far we aware that systemic investigation on the essential oil of *Zingiber officinale* (Ginger) rhizome have not been investigated in Bangladesh by using modern analytical techniques.

GC-MS analyzed results which include the active principles with their retention time, molecular formula, molecular weight and composition of the essential oil of *Zingiber officinale* (Ginger) of two varieties are presented in Table-2. From essential oil of Bangladeshi variety, *Zingiber officinale* (Ginger), 11 chemical constituents were found and the oil rich in zingiberene, (38.10%), beta phellandrene (12.00%),β-sesquiphellandrene(9.546%), α-curcumene(9.224%), Camphene (5.94%), α-Farnesene (4.573%), β-bisabolene (4.39%), citral (3.91%), α-pinene(2.33%),eucalyptol (1.27%), germacene D (1.14%) respectively i.e. total monoterpene (25.45%) and sesquiterpene (66.97%). On the other hand total 10 chemical constituents were found from the China zinger essential oil.The oil contains α-curcumene(26.54%),camphene (20.60%),citral(17.90%),αpinene(6.63%),borneol(5.40%),βb isabolene(4.57%),eucalyptol(2.44%),1,2disisopropenylcyclo butane (2.84%), hexadecanoic acid (1.55%), L-alloaromadendren(0.86%) i.e. total monoterpene (56.24%) and sesquiterpene (33.97%).

Results show that essential oil from both of the two countries is a complex mixture of numerous compounds, many of which are found in trace amounts. It is worth monitoring that there is a great variation in the chemical composition of these two regions oil of *Zingiber officinale* (Ginger). This confirms that the reported variation in oil is due to geographic divergence and ecological conditions.

Table 1: Comparative studies on physical and chemical properties of essential oil of ginger from Bangladesh and China.

Physical Properties		**Bangladeshi Ginger**	**China Ginger**
Oil yield (%) g/1000g		0.204	0.203
Organoleptic	Taste	Pungent	Pungent
	Odor	Spicy	Spicy
	Color	Golden Yellow	Golden Yellow
	Appearance at room temperature (30˚ C)	Homogeneous, transparent liquid, lighter than water	Homogeneous, transparent liquid, lighter than water
Specific gravity at 30˚ C		0.918	0.901
Refractive index [$\eta^{t^\circ C}$]		1.4875	1.4873
Optical rotation $[\alpha]^{26}$		-36˚	-34˚
Solubily in	60% alcohol	Not soluble	Not soluble
	70%alcohol	Cloudy up to 9.8 volume	Cloudy up to 9.8 volume
	80% alcohol	Soluble in 5.0 Volume	Soluble in 5.0 Volume
	90% alcohol	Soluble in 0.1 Volume	Soluble in 0.1 Volume
	100% alcohol	Soluble at any Volume	Soluble at any Volume
Chemical properties			
Acid value		8.7	6.5
Ester value		33.56	31.06

Table-2: Comparative analysis of chemical constituents of essential oil of *Zingiber officinale*.

No.	**Retention time**	**Name of the compound**	**Molecular weight**	**Molecular formula**	**Composition (%)**	
					Bangladeshi ginger	**China ginger**
1.	934	Cyclobutane,1,2-bis(1-methyllethenyl)-trans (1,2disisopropenylcyclobutane)	136	$C_{10}H_{16}$	---	2.838
2.	943	Bicyclo[2,2,1]heptane,2,2dimethyl-3-methylene (IS) (Camphene)	136	$C_{10}H_{16}$	5.94	20.60
3.	948	Alpha-pinene	136	$C_{10}H_{16}$	2.337	6.60
4.	964	beta-Phellandrene	136	$C_{10}H_{16}$	12.00	---
5.	1059	Eucalyptol	154	$C_{10}H_{18}O$	1.265	2.881
6.	1138	Borneol	154	$C_{10}H_{18}O$	---	5.395
7.	1174	2,6-Octadienal,3,7-dimethyl-, (E) (Citral)	152	$C_{10}H_{16}O$	3.91	17.90
8.	1369	Cyclohexene,1,1,2tri-methyl-3,5bis(1-methylethynyl)-,(2 alpha,3beta,5beta) (Hexadecanoic acid)	206	$C_{15}H_{26}$	---	1.55
9.	1386	Cycloprop[e]azulene,decahydro-1,1,7-trimethyl-4-methylene,[1aR-(1a.alpha,4a.alpha,7alpha,7a.beta,7b.alpha)] (L-Alloaromadendrene)	204	$C_{15}H_{24}$	---	0.854
10.	1446	Cyclohexene,3-(1,5-dimethyl-4-hexenyl)-6-methelene- [S-R,S] (β-Sesquiphellandrene)	204	$C_{15}H_{24}$	9.546	---
11.	1451	Zingiberene	204	$C_{15}H_{24}$	38.10	---
12.	1458	Alpha-farnesene	204	$C_{15}H_{24}$	4.573	---
13.	1500	Cyclohexene,1-methyl-4-(5-methyl-1-methylene-4-hexenyl)-(S)- (β-Bisabolene)	204	$C_{15}H_{24}$	4.39	4.573
13.	1515	1,6-Cyclodecadiene,1-methy-5-methylene-8-(1-methylethyl)-,[s-(E,E] (Germacene D)	204	$C_{15}H_{24}$	1.137	---
14.	1524	Benzene,1-(1,5-dimethyl-4-hexenyl)-4-methyl (α-curcumene)	202	$C_{15}H_{22}$	9.224	26.58
Total %					92.42	89.80

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