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IDENTIFICATION OF MOSQUITO REPELLENT IN ETLINGERA ELATIOR (TORCH GINGER)

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ABSTRACT

The commercial repellent product and insecticide used to repel mosquitoes was reported to contain toxicity, caused skin irritant and has given rise to environmental concern. In consideration of this, the need to develop environmental friendly products is imperative to control the mosquito attack. The means of this study are to analyze the content of 2-undecanone (repellent agent) in the extracted Etlingera Elatior oil of its stem, midrib, leaves and flower and also to identify its prospect as a natural mosquito repellent. Steam distillation was chosen for extraction of Etlingera Elatior with 2 different solvent which are methanol and n-hexane. The characterization of the oil was analyzed using Gas Chromatography-Mass Spectrometry. The obtained result is 2-undecanone, where the desired component was not identified in the Etlingera Elatior plant. Despite of the absence of 2-undecanone, a great number of different components were detected and a few of other potential reported mosquito repellent were present such as limonene, hex anal and dodecanoic acid in small amount. In conclusion, Etlingera Elatior has the potential to be a natural mosquito repellent.

Keywords: steam distillation, extraction, essential oil, gas chromatography-mass spectrometry.

1. INTRODUCTION

In the tropical countries like Southeast Asia, more than two million people primarily are at risk caused by the worldwide health problems associated with mosquito-borne diseases [1]. Mosquitoes transmit deadly global illness, thus they are well known as important vectors of multifarious kind of diseases [1-5] which causing pathogens and is a nuisance pest. Mosquitoes carry many severe causal agents of diseases including Malaria (most serious) [4-7], dengue fever and yellow fever [3, 5, 6] from Aedesaegypti [4], the Chikungunya virus disease was spread in many countries by Aedesalbopictus which commonly known as the Asian tiger mosquito (one of the most obvious mosquito species in the world), several other alphaviruses, arboviruses and filariasis. Recently, a newly discovered disease known as Zika has been also mosquito-borne disease [6].

Protection for individual is one of the strategies to avert mosquito bites which refer to the use of repellent. Most widely used commercial repellents contained N,N-Diethyl-meta-toluamide (DEET), the "criterion standard" repellent. Previous study states that this compound is e□ective and possesses excellent repellency against a wide range of insects. Yet, it may bring to bear toxic reactions under some circumstances and age groups, and may damage plastic, synthetic fabric and painted surfaces [3]. In fact, the odor and sensation on human skin is unpleasant despite the long-lasting protection of up to 8 hours from time of application [2]. Besides that, DEET can damage brain cells, causes behavioral changes and hazardous reactions with some medications when frequently exposed to or after prolonged use of DEET [9]. Apart than that, recent research has also been centered on repellents of natural origins since industrial repellents and insecticides become ineffective to mosquitoes [10].

Commercial natural mosquito repellents have been renowned as natural alternatives, have good repellent properties and the application of it has been widened nowadays. Previous studies reported that these products are less harmful to the environment and health of humanity [11]. However, only recently this matter has become the subject of detailed research for the active chemicals present in such botanical sources. This valuable resource should be explored further in the development of new natural products that offer both reliable protection and adequate safety for consumers. Pertaining a natural option that is safer than DEET may be contributed by the application of most of natural plant oils used as repellents in food flavoring or the perfume industry, even though these oils may cause dermatitis. Table-1 shows the summary of natural plant tested as mosquito repellent.

Since natural repellents are proven to give benefits to mankind, more natural repellents should be discovered. A popular natural plant among tropics and no foreign to Malaysian is Etlingera Elatior. It is from the ginger family and produces large green shoots of rhizomes like ginger. In Malaysia, fruit of Etlingera Elatior is used traditionally to treat earache while leaves are applied for cleaning wounds whereas the leaves together with other aromatic herbs in water are used to remove body odor [13]. Thus, the safety of the flower is proven to be developed as drugs and medicine.

Gas Chromatography-Mass Spectrometry (GC-MS) is an analytical method used to identify different components within experiment's sample(s) by using gas-liquid chromatography with mass spectrometry. Citronella oil, eucalyptus, lavender and peppermint oils are examples of essential oil that its constituents of the oil have been characterized through GC-MS in the existing research [1, 5, 14, and 15].

Table- 1. Summary of natural plant tested as mosquito repellent.

Natural Plants and Essential Oil	References
Cinnamon	[11]
Lemon eucalyptus	[11, 12]
Sandalwood	[11]
Turmeric	
Zingiber officinalis (Ginger)	[1]
Achyranthesaspera	
Citronella/Cymbopogon (lemongrass)	[5, 7, 12]
Achilleamillefolium (yarrow)	[12]
Birch/pine tar	
Clove	
Geranium	
Lavender	
1) Lily of the valley	
2) Peppermint	
3) Acantholippiaseriphioides	[3]
4) Achyroclinesatureioides	
5) Aloysiacitriodora	
6) Anemia tomentosa	
7) Baccharisspartioides	
8) Chenopodiumambrosioides	
9) Eucalyptus saligna	
10) Hyptismutabilis	
11) Minthostachysmollis	
12) Rosmarinus o□cinalis (rosemary)	
13) Tagetesminuta	
14) Tagetespusilla	

Hence, the purposes of this study are to extract the oil from each of EtlingeraElatior 4 plant parts (stem, midrib, leaves and flower), to compare the effect of two different solvent used to dilute the EtlingeraElatior powder and to analyze the content of 2-undecanone (mosquito repellent) in the extracted oil using GC-MS.

2. METHODOLOGY

Sample preparation

The dry Etlingera Elatior plant was collected in neighborhood area in Jerangau, Dungun, and Terengganu. The plant parts (stem, midrib, leaves and flower) were separated, rinsed, dried under the sun, ground into fine powder separately and dried in the oven at 100ºC until the material has a constant weight. Last but not least, powders were stored in 4 different labeled airtight plastic and kept until further use.

Extraction of etlingera elatior essential Oil

The prepared EtlingeraElatior ground materials as much as approximately 3g and 200mL of solvent was added and were exposed to steam distillation until all the solvent was evaporated. The distillate was collected. This extraction procedure was repeated using 8 different samples for which the manipulated variable would be different parts of the plant (stem, midrib, leaves and flower) and different solvent used (methanol and n-hexane).

The solvent was boiled off using a rotary evaporator at the boiling point of the solvent in which methanol and n-hexane are 65ºC and 68ºC respectively. Thus, the plant part essential oil was collected. Lastly, the oils were transferred into 8 different labelled airtight bottles and kept in sealer at 4ºC to prevent the oil from evaporating until further analysis via GC-MS.

Analysis by gas chromatography-mass spectrometry

The extracted oils in solvent were immersed in the solvent again using the ratio of 1:1. The specific conditions of the GC-MS were as stated in Table-2.

Table-2. Conditions of GC-MS.

Column	BP-X5 capillary column 30m x 0.25mm I.D 0.25 um film thickness
Injection	1uL injection volume Injection ratio 1:10 split mode Injector temperature maintained at 280°C
Carrier	Helium gas with flow rate of 0.7 ml/min
Oven program	Column maintained at 50°C for 6 min Programmed to 230°C at a rate of 10°C/min (held for 16 min) Ionization mode used to be electronic impact at 70eV
Detection	Scan rate-5 scans/s, scan mass range of 20 to 450 atomic mass unit

3. RESULTS AND DISCUSSION

GC-MS analysis

Chemical composition may vary due to the fact of the same plant species from different geographic areas. Some of the components of the analyzed essential oil were either previously reported as potential insect repellents, and were chemically similar to other components with repellent properties [16].

As expected, compounds that had previously shown repellent activity in most natural plants and plant based essential oil were detected and those are limonene, hexanal and dodecanoic acid. Repellents by this essential oil are not expected because their concentration is negligibly small in the sample. Table-3 shows the summary of area% of the main component in the extracted oil analyzed by GCMS.

Table-3. Summary of area% of the main components of extracted oil analyzed by GCMS.

Sample	Main Components	Area%
Leaves-Methanol	Isophytol, acetate	16.33
Leaves-Hexane	2-Hexanone	5.68
Stem-Methanol	6-Octadecenoic acid, (Z)-	48.10
Stem-Hexane	Silane, cyclohexyldimethoxymethyl-	13.37
Midrib-Methanol	17-Pentatriacontene	36.65
Midrib-Hexane	Oleic acid, 3-(octadecyloxy)propyl ester	28.02
Flower-Methanol	Oxirane, hexadecyl-	18.66
Flower-Hexane	Oxirane, hexadecyl-	34.5

Effects of solvents

Non-polar solvent dissolved a large portion of non-polar compound and the solubility of polar compounds is lower in non-polar solvent than that of polar solvent. The selection of solvent affects the result as for each plant part that use different solvent during the steam distillation, they contained different components. Sample that use n-hexane as solvent has detected more components than that of methanol except for stem part.

Effect of plant parts

Components in each plant parts differ when compare with sample that use the same solvent, but different plant part. For every sample that use methanol, different parts contain different components, same goes to hexane. Comparing between 4 different plant parts, flower has detected the most components both in flower-methanol and flower-hexane samples. Table-4 shows a summary of the main components for each oil of plant parts.

Table-4. Summary of main components for each oil of plant parts.

Plant Parts	Main Components	Area%
Leaves	Isophytol, acetate	16.33
Stem	6-Octadecenoic acid, (Z)-	48.10
Midrib	17-Pentatriacontene	36.65
Flower	Oxirane, hexadecyl-	53.16

Undecanone

The analysis made did not detect exactly the desired component of 2-undecanone in all of Etlingera Elatior essential oils. But, it can only detect the components that share the same functional group or sub-structure as 2-undecanone which is a ketone. Those components are 2-nonadecanone, 2-hexanone and other ketones based on the result obtained.

The absence of 2-undecanone in the Etlingera Elatior extracted essential oil may be due to fewer possibilities. Those probabilities are the experimented Etlingera Elatior plants parts does not contain 2-undecanone or due to some problems that influenced the result during the research experiment. By comparing to [16] result, it most probably due to unsuitable solvent, the amount of raw sample and solvent not enough, particle size not fine enough and short duration of extraction.

The results suggest that EtlingeraElatior'send product may have the possibility to be the alternatives to other mosquito repellent, even without 2-undecanone due to the presence of its ketone family. Hence, the particular main components of essentials may indicate its potential use, but do not guarantee the application without the proven by bioassay.

4. CONCLUSIONS

The objectives to extract the oil from each of EtlingeraElatior 4 plant parts and to compare the effect of two different solvent used to dilute the EtlingeraElatior powder has been achieved. Next, the objective of analyzing the content of 2-undecanone (mosquito repellent) in the extracted oil using GC-MS is failing to be fulfilled as none of 2-undecanone was identified.

The most essential oil contained a great number of constituents when characterized by GC-MS. The results in this study revealed that the presence of mosquito repellent in EtlingeraElatior extract cannot be strongly proven since the percentage of potential mosquito repellent compounds like limonene, hex anal and dodecanoic acid are small and the absence of 2-undecanone. In conclusion, the chemical composition of every sample is varying for different plant part.

Nevertheless, the result suggesting that the main components of essential oil should be further tested as mosquito repellent. In addition, Etlingera Elatior flower is not toxic to human. Moreover, this plant extract has been widely used as the main ingredient in traditional medicine. Hence, Etlingera Elatior extract could be of considerable interest in encouraging the new development in this field of new repellent. It can be one of the alternatives to other commercial repellent of mosquito if further studied. The use of Etlingera Elatior's essential oil as the mosquito control measure may provide essential defenses from these mosquitoes.

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