Chemical Composition and Repellency Effect of Ferulago Angulate Plant Against Malaria Vector, Anopheles Stephensi

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Research

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Abstract

Background

Malaria is a health problem globally. There are several vector control measures. Using repellent to protect human from biting of vectors is one of the best ways. The aim of this study is to evaluate the efficacy of plant against biting of malaria vector, An.stephensi under laboratory conditions.

Methods

Chemical constituents of Ferulago angulate plant were determined by using gas chromatography-mass spectrometry. Efficacy and the protection time of plant were evaluated on Anopheles stephensi.

Results

A total of 40 compounds were identified in plant. Flourensiadiol (17.4%) dehydro-sabina keton (13.3%), β-maaliene (8.8%) was the highest in the plant. The mean assessed protection time and efficacy for plant was 60 and 100 minutes respectively. ED$_{50}$ and ED$_{90}$ values for this plant were 18.12 and 93.19 µl/cm$^2$ respectively.

Conclusion

Results showed that plant has an acceptable protection time, therefore, this plant could be considered as a good herbal repellent against anopheles mosquitoes.

1. Introduction

Malaria is the main vector borne diseases worldwide. According to the recent record of World Health Organization, 228 million cases have been reported in 2018 mainly in in African region [1]. According to the report of Ministry of Health of Iran, less than 89 locally-transmitted cases in 2017 have been reported. The aim of country is to eliminate the disease by 2025 [2]. Malaria continues to be a main vector-borne public health problem in Iran. Using insecticide caused resistant in the vectors. There are several reports on resistant status of malaria vectors including An.stephensi [3-5] An.maculipenis [6] , An.culicifacies ( [7-9] ) An. [10]). At the present, biopesticides are in the spotlight due to inflicting of chemical pesticide hazards to humans, livestock, environment as well as the occurrence of the inducing resistance to different groups of insecticides among human and animal disease vectors. A lot of attention is being paid to natural products in vector control as they are environmentally safe, degradable and target-specific. Recent studies have demonstrated that use of repellents is one of the effective ways to control [11]. There are 9 species of plant in Iran including: F. contracta, F. macrocarpa, F. phialocarpa, F. stellate, F. angulate, F. Bernardi, F. carduchorum, F. subvelutina, F. trifida

F. angulate is known as Chavil in the country. It is a perennial endemic aromatic herb from the nine Ferulago species growing particularly in Iran [12-13]. Its leaves have been traditionally used as antiseptic,
pain reliever, in digestive disorders, to treat intestinal worms, snake bites, hemorrhoids, chronic ulcers, and ailments of the spleen [14]. Furthermore, in Western Iran, this plant has been consumed as spice, and used as air freshener, decay preventer and flavoring oil [15]. In a study to assess the influence of geographical factors on essential oil composition, along with antiradical potential and phytochemical contents of *Ferulago angulata* (Apiaceae) extracts. Thirty-nine compounds were identified from the Eos of nine populations. α-Pinene was the predominant component. It can be considered as a perspective raw material in food and phytopharmaceutical industries [16]. Antioxidant and antibacterial activities of the essential oils of *F. angulata* collected from different natural habitats in the alpine regions of southwestern Iran showed positive results. The essential oil of *F. angulate* could be serving as a potential source for use in the food, cosmetic and pharmaceutical industries [17]. Cytotoxic activities of *Ferulago angulata* extract on human leukemia and lymphoma cells by induction of apoptosis were determined [18]. This study was conducted to measure the chemical contents and to evaluate the repellent properties of plant essential oil against *An. stephensi* under laboratory condition on animal model.

2. Materials And Methods

2.1. Collection, identification and extraction of plant, *Ferulago angulate*.

The plant of *Ferulago angulate* were collected in Chaharmahal and Bakhtiari Province in south-west of Iran (Fig.1) They were rapidly transported to the School of Public Health, Tehran University of Medical Sciences.

2.2. Plant identification: The plant was identified by experts in Department of Plant Sciences, Tehran University. (Fig.2)

2.3. Mosquitoes rearing

Susceptible strain of *An. stephensi* were reared and maintained at 28±2 °C and 65±5% relative humidity (RH) under a 16:8 (L: D) photoperiod. Under insectary situation, guinea pigs is used as blood feeding female mosquitoes for maturing the eggs.

2.4. Repellency test

Females of *An. stephensi* were used for the repellency tests. 12 hour before starting the experiments, the sucrose solution was picked up from the cage. Various repellency tests including protection time, failure time, effective dose and killing effects of EOs were carried out according ASTM E951-9 against 5-8 old female *An. stephensi*.

2.5. Extraction of essential oil of plant

All the extraction was carried out at Faculty of Pharmacology, Tehran University of Medical Sciences. Essential oils (EOs) of native medicinal plant of *Ferulago angulata*, were hydrodistilled in a Clevenger-type
apparatus for 4-6 h and dried over anhydrous sodium sulfate. The EOs were stored in the dark sealed vials at 4 °C until starting the repellency tests maximum after 2 days past of EO preparation.

2.6. Plant essential oils analysis

Chemical composition of plant was analyzed using an Agilent 7890–5975 gas chromatography mass spectrometer. With a HP- 5MS (5% Phenyl Methyl Silox) capillary column (30m×0.25mm, film thickness 0.25μm), split ratio, 1: 1, and using a flame ionization detector. The GC was programmed at 50 °C for 0.5 min and then increased at 5 °C/min to 280 °C, and finally held with an isothermal for 3min. The injector temperature was 280 °C. The flow rate of the carrier gas was 1ml/min. The identification of compounds was performed by comparing their retention times and mass spectra with mass spectra from Wiley library [19]

2.7. Plants essential oils analysis

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3. Results

3.1. GC-mass analysis

One microliter of each essential oil was injected to GC-mass. A total of 40 compounds were identified in plant. Flourensiadiol (17.4%) dehydro-sabina keton (13.3%), β-maaliene (8.5%) was the highest in the plant (Table 1).

Table 1. Chemical constituents of stem essential oil from Ferulago angulate
| NO | tR (Minutes) | compound                                    | %  | RI  |
|----|--------------|---------------------------------------------|----|-----|
| 1  | 1.80         | isovaleric acid                             | 0.3| 833 |
| 2  | 2.02         | (Z)-3-hexenol                               | 6.3| 856 |
| 3  | 5.27         | dehydro-sabina ketone                       | 13.3| 1120|
| 4  | 5.44         | α-campholenal                               | 0.6| 1135|
| 5  | 5.47         | cis-p-mentha-2,4-dien-1-ol                  | 0.3| 1137|
| 6  | 5.71         | camphene                                    | 0.5| 1157|
| 7  | 5.84         | borneol                                     | 1  | 1168|
| 8  | 5.92         | ethyl-benzoate                              | 1.4| 1174|
| 9  | 6.27         | n-decanal                                   | 1  | 1201|
| 10 | 6.52         | trans-carveol                               | 1  | 1221|
| 11 | 6.60         | nerol                                       | 3.2| 1228|
| 12 | 6.96         | geraniol                                    | 1  | 1256|
| 13 | 7.66         | undecanal                                   | 1.2| 1308|
| 14 | 8.04         | d-elemene                                   | 1.5| 1339|
| 15 | 8.58         | β-maaliene                                  | 8.5| 1381|
| 16 | 9.43         | geranyl acetone                             | 1.2| 1451|
| 17 | 9.60         | a-acoradiene                                | 1.4| 1465|
| 18 | 9.82         | germacrene D                                | 1.4| 1483|
| 19 | 10.48        | a-cadinene                                  | 11 | 1539|
| 20 | 10.56        | elemol                                      | 0.6| 1546|
| 21 | 11.62        | hinesol                                     | 0.4| 1639|
| 22 | 11.92        | bulnesol                                    | 3  | 1666|
| 23 | 12.21        | 8-cedren-13-ol                              | 0.36| 1692|
| 24 | 12.92        | xanthorrizol                                | 1.2| 1753|
| 25 | 13.15        | benzyl-benzoate                             | 1.1| 1772|
| 26 | 13.43        | nootkatone                                  | 1  | 1794|
| 27 | 13.77        | isopropyl tetradecanoate                    | 0.5| 1821|
| 28 | 13.90        | isopropyl tetradecanoate                    | 3  | 1831|
The ED\textsubscript{50} and ED\textsubscript{90} values of plant was 15.88 and 89.52 µl/cm\textsuperscript{2}, respectively (Table 2).

Table 2. Effective dose of stem essential oil of \textit{F. angulate} against \textit{Anopheles stephensi} on animal subject under laboratory condition

| p-Value | \(\chi^2\) table (df) | \(\chi^2\) (heterogeneity) | ED\textsubscript{90} (µl /cm\textsuperscript{2}) ± 95\%C.L. | ED\textsubscript{50} (µl /cm\textsuperscript{2}) ± 95\%C.L. | b ± SE | a | plant |
|---------|-------------------|---------------------|----------------------|----------------------|-----|---|--------|
| 0.01    | 2                 | 0.29                | 93.19 (61.70-190.21) | 18.12 (14.42-22.67) | 1.8±0.26 | -2.2 | 0.01 |

The protection time of essential oil of plant was 60 minutes. The failure time of 100 minutes was observed against \textit{An. stephensi} on animal (table 3).

Table 3. Protection time and failure time of \textit{Ferulago angulate} against \textit{An. stephensi} on animal subject under laboratory condition

| Failure time (hour) | Protection time (hour) | Elavation (m) | Locality of collection | District |
|---------------------|------------------------|---------------|------------------------|----------|
| Mean                | Range                  | Mean          | Range                  |          |
| 2.06 h = 100 min    | 1.5 – 2.0              | 1.0 h = 60.0 min | 1.0 – 1.0              | 1502     |
|                     |                        |               | Bazoft                 | Koohrang |
Discussion

In this research a total of 40 compounds were identified in plant. Flourensiadiol (17.4%) dehydro-sabina keton (13.3%), β-maaliene (8.8%) was the highest in the plant. The mean assessed protection time and efficacy for plant was 60 and 100 minutes respectively. ED$_{50}$ and ED$_{90}$ values for this plant were 18.12 and 93.19 µl/cm$^2$ respectively.

The major constituents of the essential oils from *F. angulate* were a-pinene, and cis-b-ocimene [17]. There are several report on larvicidal activities of *Ferulago* species against malaria vectors worldwide. *Ferulago carduchorum* was effective against *Anopheles stephensi* with LC$_{50}$ and LC$_{90}$ values of 12.78 and 47.43 ppm, respectively [20]. LC$_{50}$ of stem, root, aerial parts, fruits, and flowers essential oils of *Ferulago trifida* against larvae of An. stephensi were equal with 10.46, 22.27, 20.50, 31.93 and 79.87ppm respectively [21].

The LC$_{50}$ of the total extract, chloroform, ethyl acetate and methanol fractions of *Ferulago carduchorum* were 0.4799, 0.2361, 0.7437 and 3.7017 ppm, respectively. The LC$_{90}$ of the total extract, chloroform, ethyl acetate and methanol fractions were 1.5090, 0.4547, 1.8918 and 10.8857 ppm, respectively [22]. Results of efficacy of different Iranian native plants against malaria vector *An. stephensi* at the LC$_{50}$ and LC$_{90}$ levels are presented. From these results it can be concluded that *Mentha spicata* and *Eucalyptus camaldulensis*, had the lowest and highest LC$_{50}$ respectively [23].

Conclusion

The plant of have been used for medicinal purposes. It repellency and killing effect also evaluated against medically important arthropods and agricultural pests. The appropriate formulation of plant should be prepared for vector and pest control. The results indicated that this plant could be used as larvicida and repellent against mosquitoes.

Declarations

**Ethics approval and consent to participate:** There is not applicable

**Consent for publication:** applicable

**Availability of data and material:** applicable

**Competing interests:** *The author declare that there is no conflict of interest*

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