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ESSENTIAL OIL PHYSICOCHEMICAL COMPOSITION OF THE Thymus fontanesii COLLECTED FROM TIARET A SEMI-ARID REGION OF ALGERIA

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Original Research Article

ABSTRACT

The aim of this work was to determine the exact composition of the essential oil extracted from *Thymus fontanesii* collected from Tiaret at the western region of Algeria by gas chromatography method. The fresh aerial parts of *T. fontanesii* were collected at the flowering stage on May 2017 from Tiaret, at the western region of Algeria, characterised by a semi-arid climate. The essential oil extracted was dried with anhydrous sodium sulfate and kept at a cool temperature of $+4^{\circ}$ C in glass bottles away from light. The essential oil from leaves of Thymus fontanesii was analysed by gas chromatography (GC-FID) and gas chromatography–mass spectrometry (GC/MS) realized by Pyrenessences Analysis, France. The oil was obtained from the aerial parts of *T. fontanesii* with an average yield of 2,256±0,094%. The chromatographic analyses identified ninety-one (91) compounds which represents approximately 99,92% of the total detected oil components mainly represented by carvacrol with 62,25% being by far the main component the other appreciable constituents identified were γ-terpinene with 12,62%, β-cymene with 7,43%, linalool with 4,26%, β-myrcene with 1,29% and α-terpinene with 1,31%. and thymol represented only 1,76% of the composition.

Keywords: Thym; Carvacrol; Thymol; γ-terpinene; β-cymene.

INTRODUCTION

Historically, natural products have provided an infinite source of medicine [1]. In the cancer's and infectious diseases between 60 and 75% of new drugs came from natural sources [2]. Generally, it is the essential oils of plants rather than their extracts who have had the greatest use in the treatment of infectious diseases, diseases of the respiratory system, urinary tract, gastrointestinal tract and bile ducts, as well as on the skin [3].

Essential oils can contain about 20-60 components to very different concentrations.

They are characterized by two or three main components to rather high concentrations (20-70%) compared to other components present in minute amounts [4]. The composition and functional structure of the oils play an important role in determination of their antimicrobial activity. Usually the compounds of the groups phenolic are the most effective. Among these, clove oils, oregano, rosemary, thyme, sage and vanillin have generally been shown to be effective against microorganisms [5].

Thyme extracts are widely used in traditional medicine in Algeria as astringent, expectorant and healing agents [6]. There

are nearly 350 species of thyme spread between Europe, West Asia and the Mediterranean, in Algeria, 12 species of Thymus colonize the territory of the country [7]. Among some of them are endemic to Algeria such as Thymus fontanesii grows spontaneously on the scrubland, giving over white flowers [8]. The aim of this work was to determine the exact composition of the essential oil extracted from Thvmus fontanesii collected from Tiaret at the western region of Algeria by gas chromatography method.

MATERIALS AND METHODS

The fresh aerial parts of *T. fontanesii* were collected at the flowering stage on May 2017 from Tiaret at the western region of Algeria characterised by a semi-arid climate. The identification of the plant was made by a botanist from Nature and life science faculty, Ibn Khaldoun university of Tiaret.

The leaves were washed, protected from light and dried in the laboratory, for 10 days, in open space. Extraction of essential oil was performed through a steam distillation process. The steam charged with essential oil was condensed in the refrigerant and the distillate collected in a separator funnel. The essential oil extracted was dried with anhydrous sodium sulfate and kept at a cool temperature of +4°C in glass bottles away from light. The yield of essential oil is estimated according to the total mass of the dry plant, according to the following formula: Yield of essential oil = Weight of essential oil (g) / (Weight of the sample (g) x100.

Essential Oil Analysis

The essential oil from leaves of *Thymus* fontanesii was analysed by gas chromatography (GC-FID) and gas chromatography-mass spectrometry realized by Pyrenessences (GC/MS) Analysis France. Briefly, the oil was analysed using a Hewlett Packard 5973 instrument, with HP INNOWAX polar column (60 m×0,25 mm×0,25 µm). One microliter of essential oil solution diluted in ethanol (10%) was injected and analysed, was the carrier gas with a flow rate of 30 psi/FID, 23 psi/MS. Column temperature was initially kept at 60°C for 6 min and then gradually increased to 250°C at 2°C/min rate and finally held for 10 min at 250°C. The temperature of the injector was fixed to 250°C and one of the detectors (FID) to 270°C. The compounds were identified by a combined search of retention time and mass spectra (NKS library, 75,000 spectra). The percentages were calculated from the peak areas given by the GC/FID, without the use of correction factor.

RESULTS AND DISCUSSION

The chemical composition of the volatile constituents of the essential oil obtained by gas chromatography is presented in Table 1. Many factors can be responsible for the variability in the chemical composition of thyme extracts. The most important are the climate, the soil, the harvest period and the method of preservation and extraction. Genetic factors [9] and also vegetative cycle [10,11].

The oil was obtained from the aerial parts of *T. fontanesii* with an average yield of $2,256\pm0,094\%$ (w /w average value of three independent hydrodistillations) based on the weight of the dry matter. This oil is dark yellowish with a pleasant scent of thyme. In our work, the yield values were similar to those reported by Sidali [12] with $2,4 \pm 0.8\%$. Our results are lower than 5,20% and 5,46% reported by Bekhechi [13]. Furthermore, authors obtained 0,9%

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yield from stems and leaves [7], while others reported a yield value of 1,9% and 2% respectively [14,15] lower than that obtained in our study.

The average yield of essential oil was calculated according to the dry plant matter of the aerial part of the plant and the result obtained shows that the yield of the essential oils varies according to interaction with the environment (temperature, type of climate, salinity, soil, rainfall), harvest period (season, stage of development), plant state (fresh or dried) [16]. These variations are also observed between the essential oils extracted from different parts of the same plant, leaves, flowers, stems, seeds and roots [17,18].

| Table 1. The Thymus fontanesii essential oil compound analysed by gas |
|---|
| chromatography |

| Pick | Refraction time (min) | Compounds | % |
|------|-----------------------|-----------------------------|-------|
| 1 | 7,1 | 2-Methylbutyrate of Methyle | 0,04 |
| 2 | 7,3 | Methyl Isovalerate | 0,01 |
| 3 | 7,5 | α-PINENE | 0,89 |
| 4 | 7,6 | α-THUYENE | 0,40 |
| 5 | 8,8 | Camphene | 0,05 |
| 6 | 10,2 | β-Pinene | 0,10 |
| 7 | 11,9 | delta3-Carene | 0,06 |
| 8 | 12,1 | 3-Heptanone | 0,04 |
| 9 | 12,5 | β-Myrcene | 1,29 |
| 10 | 12,6 | α-Phellandrene | 0,13 |
| 11 | 13,3 | α-Terpinene | 1,31 |
| 12 | 14,2 | Limonene | 0,54 |
| 13 | 14,6 | 1,8-Cineole | 0,06 |
| 14 | 14,7 | β-Phellandrene | 0,10 |
| 15 | 16,8 | γ-Terpinene | 12,62 |
| 16 | 17,0 | Trans-B-Ocimene | 0,05 |
| 17 | 17,2 | 3-Octanone | 0,22 |
| 18 | 18,2 | P-Cymene | 7,43 |
| 19 | 18,8 | Terpinolene | 0,07 |
| 20 | 23,3 | 1-Hexanol | 0,02 |
| 21 | 23,5 | Cetone Terpenique | 0,04 |
| 22 | 25,5 | 3-Octanol | 0,15 |
| 23 | 28,3 | A-P-Dimethylstyren | 0,02 |
| 24 | 28,6 | Cis-Oxyde De Linalol | 0,02 |
| 25 | 29,1 | 1-Octen-3-OL | 0,44 |
| 26 | 30,1 | Trans-Thuyanol | 0,18 |
| 27 | 30,4 | Trans-Oxyde De Linalo | 0,03 |
| 28 | 31,9 | Ylangene | 0,04 |
| 29 | 33,0 | A-Copaene | 0,03 |
| 30 | 33,2 | Camphre | 0,05 |
| 31 | 33,5 | B-Bourbonene | 0,03 |
| 32 | 34,3 | A-Gurjunene | 0,27 |

| Pick | Refraction time (min) | Compounds | % |
|----------------------|-----------------------|-------------------------------|------|
| 33 | 35,3 | Linalol | 4,26 |
| 34 | 35,5 | 1-Nonen-3-OL | 0,16 |
| 35 | 35,9 | 1-Octanol | 0,02 |
| 36 | 36,2 | Trans-P-Menth-2-En-1-OL | 0,02 |
| 37 | 37,0 | Epsilon-Cadinene | 0,02 |
| 38 | 38.1 | A-Trans-Bergamotene | 0.02 |
| 39 | 38.2 | Methyl Thymol ETHER | 0.05 |
| 40 | 38.4 | B-Carvophyllene | 0.57 |
| 41 | 38.7 | Terpinene-4-OL | 0.42 |
| 42 | 39.0 | Cis-Dihvdrocarvone | 0.07 |
| 43 | 39.0 | Aromadendrene | 0.50 |
| 44 | 39.6 | Sesquiterpene | 0.03 |
| 45 | 40.1 | Trans-Dihydrocaryone | 0.14 |
| 46 | 40.2 | Cis-P-Menth-2-En-1-Ol | 0.03 |
| 47 | 41 2 | Trans-Pinocarveol | 0.01 |
| 48 | 41.3 | Allo-Aromadendrene | 0.09 |
| 49 | 41.8 | Sesquiternene | 0.03 |
| 50 | 42.3 | Zonarene | 0,00 |
| 51 | 42,0 | A-Humulene | 0.03 |
| 52 | 43.2 | Trans-Verbenol | 0,00 |
| 53 | 13 Q | Gamma-Muurolene | 0,00 |
| 54 | 40,0 AA 1 | Borneol | 0,00 |
| 55 | | Ledene | 0,00 |
| 55 | 44,4 | | 0,52 |
| 57 | 44,0 | A Muurolene | 0,11 |
| 58 | 40,1 | R Bisabolene + Carvone | 0,03 |
| 50 | 40,5 | a Cadinana | 0,07 |
| 60 | 40,0 | v Cadinene | 0,10 |
| 61 | 40,1 | y-Cadillelle Cuminal | 0,07 |
| 62 | 40,9 | Myrtapol | 0,02 |
| 63 | 49,2 | Cadina 1.4 DIENE | 0,02 |
| 64 | 49,5 50 7 | | 0,02 |
| 0 4 65 | 50,7 | Colomonono | 0,03 |
| 66 | 52,5 | Caldinenene R Cymono & Ol | 0,02 |
| 67 | 55,1 | | 0,07 |
| 60 | 54,0 55.0 | Ascalluur | 0,02 |
| 00 | 55,0 56 5 | Myrcenor Fri Cubabal | 0,02 |
| 09 70 | | Company Aramatia | 0,02 |
| 70 | 50,8 | Compose Aromatic Delustral | 0,08 |
| 71 | 57,1 | | 0,03 |
| 12 | 0,1C | I Talls-Jasilione | 0,03 |
| 13 | 0,0 | | 0,13 |
| 74 75 | 01,U | | 0,03 |
| /5 70 | 01,8 00,0 | Epoxyde Sesquiterpenique | 0,02 |
| /6 | 62,9 | | 0,05 |
| | 65,3 | Viridifiorol | 0,04 |

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| Pick | Refraction time (min) | Compounds | % |
|------|-----------------------|-----------------------|-------|
| 78 | 66,4 | Cuminol | 0,04 |
| 79 | 67,0 | 10-Epi-Gamma-Eudesmol | 0,02 |
| 80 | 67,7 | Spathulenol | 0,36 |
| 81 | 69,8 | Isothymol | 0,10 |
| 82 | 70,0 | Heneicosane | 0,06 |
| 83 | 70,6 | Thymol | 1,76 |
| 84 | 71,5 | Isocarvacrol | 0,08 |
| 85 | 72,1 | Carvacrol | 62,25 |
| 86 | 72,7 | Aromatic compound | 0,05 |
| 87 | 72,9 | Isospathulenol | 0,05 |
| 88 | 73,2 | Phenolique compound | 0,07 |
| 89 | 75,3 | Aromatic compound | 0,07 |
| 90 | 94,2 | Aromatic compound | 0,20 |
| 91 | 101,5 | Acide Palmitique | 0,09 |
| | | Total | 99,92 |





Fig. 1. The chromatographic profile of the Thymus fontanesii essential oil

Chromatographic analyses of the essential oils in our sample identified ninetyone (91) compounds which represents approximately 99,92% of the total detected oil components mainly represented by carvacrol with 62,25% being by far the main component. The composition of the four samples of *T. fontanesii* collected from the Tlemcen's region at the extreme west of Algeria was quite similar to ours with 66,7% -69,5% [13]. Those results are higher than the carvacrol amount obtained from the same species such as in Ain Defla at the center region of Algeria with $54,7\pm1,2\%$ reported by Sidali [12] and higher than 42,9% and 44% respectively reported [8,19].

CONCLUSION

In our study, the other appreciable constituents identified were γ -terpinene with 12,62%, β -cymene with 7,43%, Linalool with

4,26%, β -myrcene with 1,29% and α terpinene with 1,31%. However, thymol represented only 1,76% of the composition. Dob et al. [7] have identified thymol about 29,2% and monoterpenes in the essential oils of Thymus fontanesii from Djelfa region of Algeria, the same results were reported for the same plant harvested in Setif region of Algeria, which revealed the existence of a single major compound, thymol with 67.8% followed by y-terpinene with 15,9% and pcymene with 13,0% [14]., this results show that the chemical composition of thyme extract can vary according to the geographical localization.

The chemical characteristics makes possible to highlight the quality of the essential oil of *Thymus fontanesii* harvested in Tiaret at the western region of Algeria that is chemotyped with Carvacrol.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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