European Journal of Biology
Eur J Biol 2023; 82 (2): 239–242
DOI: 10.26650/EurJBiol.2023.1282708

SHORT COMMUNICATION

Comparison of Fatty Acid Contents of Wild and Cultivated *Arum italicum* Mill. Seed Oils from Kırklareli in Turkey by Gas Chromatography-Mass Spectrometry

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ABSTRACT

Objective: *Arum L.* is the largest genus of Aroids. Tubers, leaves, and fruits of *Arum italicum* Mill. (*Araceae*) are used as a traditional medicine in the treatment of hemorrhoids and in preparing local dishes in Turkey. In this study, the fatty acid contents of the wild and cultivated seed oils of *A. italicum* Mill. species from the Kırklareli region in Turkey were determined and compared by the gas chromatography-mass spectrometry method.

Materials and Methods: Seed oils were obtained via petroleum ether extraction. Fatty acid methyl esters were prepared with boron trifluoride/methanol derivatization and analyzed with TC WAX capillary gas chromatographic column.

Results: The amount of 13-phenyl tridecanoic acid, which is the specific fatty acid of *Arum italicum* Mill. species, was found to be similar in both samples. In addition, 2-hydroxy palmitic acid was identified for the first time in the same species.

Conclusion: This is the first report on the presence of the 2-hydroxy palmitic acid content of *Arum italicum* Mill. seeds from Turkey.

Keywords: *Arum italicum* Mill., Seed, Fatty acids, 13-Phenyl tridecanoic acid, 2-Hydroxy palmitic acid

INTRODUCTION

The *Araceae* family is represented in Turkey by 32 taxa, consisting of five genera, 22 species, five subspecies, and 12 varieties. *Arum* L. is the largest genus among other aroids of Turkey. Representatives of the genus are distributed in Central Asia, Europe, Macaronesia, the Mediterranean, and the Middle East regions. The plant can be found mainly in shady places, near walls, abandoned gardens, cemeteries, and at the base of olive and oak trees. *Arum italicum* Mill. is a well-known species, grown not only for the attractive foliage, but also for rich colored berries that are produced in the autumn in Turkey. The plants sprout in early autumn or early winter from a rhizomatous tuber. It produces arrow or broad hastate or sagittate-hastate deep green leaves, usually with silver or grey blotches, 9–35 (-40) cm long, 2–29 cm wide, and with 15–40 cm long petiole. They produce oblong cylindric fruiting spikes consisting of deep orange or red berries, which are also attractive to birds. The individual fruits contain between two and five seeds. Each seed consists of a leathery, reticulate testa. *Arum* L. taxon has been familiar to Eastern Mediterranean people for centuries and has many vernacular names, such as Serpent knife (*Yılan bıçağı*), Serpent dagger (*Yılan burçağı*), Serpent vetch (*Yılan kaması*), and Serpent pillow (*Yılan yastığı*). Plants have been used as a traditional medicine and food, mostly in rural areas, regardless of the species in the country. The tubers, leaves, and fruits of *A. italicum* Mill. are mainly used as a traditional medicine and in preparing local dishes. In some areas of Turkey, ripe fruits are considered as an effective treatment for hemorrhoids: two or three are swallowed with a glass of water each morning for a week.[1–4]

The aim of this study was to compare the fatty acid contents of the seed oils of wild and cultivated *A. italicum* Mill. from the Kırklareli region of Turkey by gas chromatography-mass spectrometry (GC-MS).

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Submitted: 13.04.2023 • **Revision Requested:** 22.05.2023 • **Last Revision Received:** 16.06.2023 • **Accepted:** 12.09.2023 • **Published Online:** 01.12.2023

MATERIALS AND METHODS

Samples

Ripe fruit of the wild *A. italicum* Mill. were collected from a field around the Kırklareli region (Northwestern Turkey) when the plants were in the fruiting state and could be identified (July 2018). Ripe fruits of the cultivated specimens were also collected from the same population above, and the seeds were sown for germination, blooming, and growth at the Alfred Heilbronn Botanical Garden, Istanbul University (2015).

Oil Extraction

The dried and crushed seeds were extracted with petroleum ether (40-60 °C) for four hours in a shaker. The solvent was removed and evaporated at 40 °C under nitrogen flow. The obtained oil was kept in a vacuum desiccator with P_2O_5 and then weighed.

Derivatization of Fatty Acids

The dried and crushed seeds were derivatized with 20% BF3/MeOH according to the method of Soukup and Holman.[5] The obtained methyl esters were extracted with petroleum ether (40-60 °C), and the solvent part was evaporated under nitrogen flow at 40 °C.

GC-MS Analysis

The GC-MS model with mass selective detection (ionization energy, 70 eV; source temperature, 300 °C) was used (Agilent 7890B Series GC System). The fatty acid methyl esters were analyzed with TC WAX capillary gas chromatographic column (Capillary column: 30 m x 0.25 mm id, 0.25 mm film thickness; temperature program: 170-210 °C, 1 °C/min) (GL Sciences Inc.). Helium was used as the carrier gas (split ratio 1/20). The injector and detector temperatures were 230 °C and 250 °C, respectively. The fatty acid methyl ester standard mixtures were used for the identification of key fatty acids (Sigma). Calculations were made according to the peak area normalization technique.

RESULTS AND DISCUSSION

In our previous study, we had only determined the fatty acid compositions of *A. italicum* Mill. which grows naturally in Istanbul.6 However, in this study, the fatty acid compositions of the wild and cultivated species were compared, and the naturally grown species from Kırklareli, were investigated for the first time. This is the first report on the 2-hydroxy palmitic acid content of *A. italicum* Mill. seeds from Turkey.

The total oil amounts of wild and cultivated A. italicum Mill. were found to be 4.08 g and 3.57 g/100 g seed, respectively. The fatty acid contents of the wild and cultivated *A. italicum* Mill. seeds from Kırklareli are in Table 1, and the comparison of the fatty acid percentages is illustrated in Figure 1.

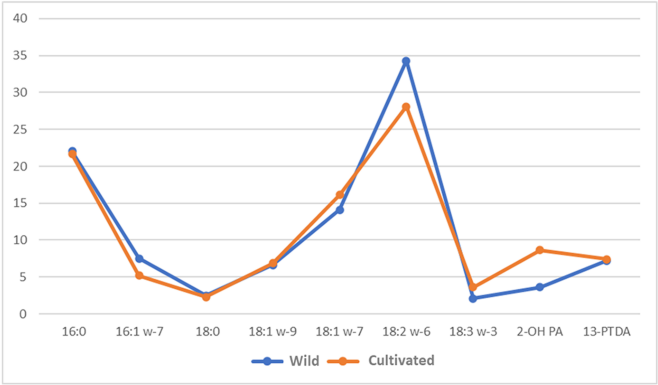

Figure 1. The comparison of fatty acid percentages of the wild and cultivated *Arum italicum* Mill. seed oils.

The main fatty acids were found as palmitic acid, cis-vaccenic acid, and linoleic acid. While the amounts of palmitic acid and cis-vaccenic acid were similar in both samples, the amount of linoleic acid was found to be higher in the wild sample with lower contents in the samples from Istanbul.[6]

It is noteworthy that cis-vaccenic acid, which is an eighteen-carbon monounsaturated fatty acid, is one of the main fatty acids. It is the main fatty acid in bacteria, and it has bacterial biomarker properties. It is found in very small amounts in plant and animal tissues.[7,8]

The amounts of 13-phenyl tridecanoic acid, which is a specific fatty acid to *Arum* species, were found to be similar in both samples, which contrasts with the literature findings.[6,8] The mass spectrum and formula of 13-phenyl tridecanoic acid are provided in Figures 2a and 2b, respectively. ω-phenyl fatty acids are cyclic acids with a terminal phenyl group. They are found in seed oils of the *Araceae* especially.[9,10]

The presence of 2-hydroxy palmitic acid (which is effective on inflammation and diabetes)[11] was determined for the first time in *A. italicum* Mill. species. It was found to be approximately three times more in the culture. The mass spectrum and formula of 2-hydroxy palmitic acid are provided in Figures 3a and 3b, respectively. The 2-hydroxy palmitic acid consists of a C16 chain with a hydroxyl group at position 2.

Table 1. Fatty acid contents of the wild and cultivated *Arum italicum* Mill. seed oils (% of total major FAME).

Fatty acid		Retention time (min)	Wild *Arum italicum* (%)	Cultivated *Arum italicum* (%)
Palmitic acid	(C16:0)	7.73	22.05±0.05	21.65±0.05
Palmitoleic acid	(C16:1w-7)	8.33	7.50±0.02	5.21±0.01
Stearic acid	(C18:0)	13.21	2.50±0.01	2.31±0.02
Oleic acid	(C18:1w-9)	13.89	6.63±0.03	6.88±0.04
cis-Vaccenic acid	(C18:1w-7)	14.14	14.10±0.04	16.15±0.07
Linoleic acid	(C18:2w-6)	15.58	34.30±0.05	28.12±0.06
Linolenic acid	(C18:3w-3)	18.17	2.09±0.01	3.62±0.02
2-Hydroxy palmitic acid (2-OH PA)		21.47	3.63±0.02	8.66±0.02
13-Phenyl tridecanoic acid (13-PTDA)		37.57	7.20±0.03	7.40±0.03

The mean values of the results are given as mean ± standard deviation.

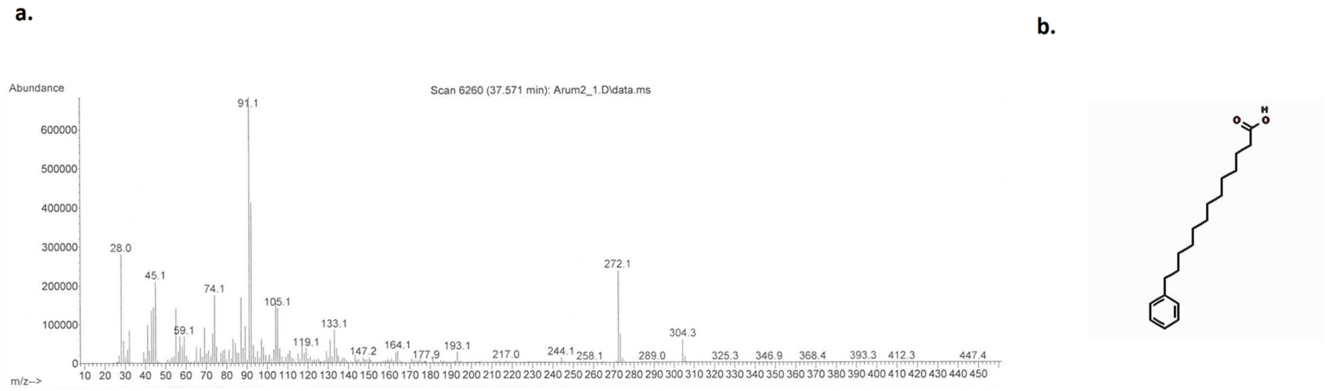

Figure 2. Mass spectrum (a) and formula (b) of 13-phenyl tridecanoic acid.

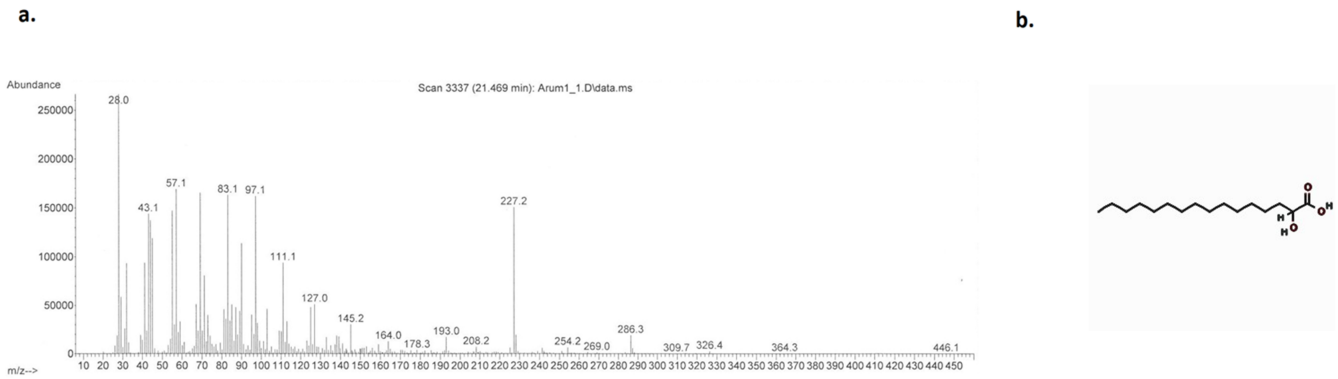

Figure 3. Mass spectrum (a) and formula (b) of 2-hydroxy palmitic acid.

CONCLUSION

In this study, the amount of 13-phenyl tridecanoic acid, is the specific fatty acid of *A. italicum* Mill. species, was found to be similar in the wild and cultivated samples from the Kırklareli region in Turkey. For the first time, 2-hydroxy palmitic acid was identified in this species. It is planned to investigate the fatty acid compositions of other wild and cultivated Aroid species in Turkey for chemotaxonomic and also for therapeutic purposes. Since suitable herbarium specimens could not be prepared from the fruiting plants in this investigation, it is planned to create a herbarium sample by collecting from the same locality in Kırklareli at the appropriate time (when the plant is in bloom). A systematic evaluation can be performed when the oil compositions of the seeds of all *Arum* L. species are revealed.

Peer Review: Externally peer-reviewed.

Author Contributions: Conception/Design of Study- K.A., S.S.A.; Data Acquisition- S.S.A.; Data Analysis/Interpretation- S.S.A.; Drafting Manuscript- K.A.; Critical Revision of Manuscript- S.S.A.; Final Approval and Accountability- K.A., S.S.A., B.K.P, N.N., I.D.,D.O.U.

Conflict of Interest: Authors declared no conflict of interest.

Financial Disclosure: Authors declared no financial support.

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How to cite this article

Alpinar K, Saglik Aslan S, Kulaksiz Piskin B, Nesetoglu N, Danis I, Ozer Unal D. Comparison of Fatty Acid Contents of Wild and Cultivated *Arum italicum* Mill. Seed Oils from Kırklareli in Turkey by Gas Chromatography-Mass Spectrometry. Eur J Biol 2023; 82(2): 239–242. DOI:10.26650/EurJBiol.2023.1282708