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SHORT REPORT

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Phenolic compounds of natural knotweed (*Polygonum cognatum* Meissn.) populations from Turkey

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ABSTRACT

The phenolic compositions of two different *Polygonum cognatum* samples collected from the Cumra and Manisa regions of Turkey were investigated for the first time. Both tested samples were rich in different phenolic compounds, mostly rutin, isorhamnetin, and catechin. The rutin content of Cumra's sample was relatively higher than that of Manisa's sample. *P. cognatum* has potential regarding rutin content as a functional dietary food or may be used as an ingredient to enrich functional foods. Our study will contribute to the previous works performed by different researchers on *P. cognatum*, commonly consumed in Turkey, to reveal its beneficial properties.

1. Introduction

Polygonum genus contains over 300 species spread out in Europe, North and South America, Asia and North Africa (Jansone, 2015). Also, the *Polygonum* genus has 27 species that are spread in Turkey. Knotweed (*Polygonum cognatum* Meissn.) is one of these 27 species, a perennial plant with a slender woody stock. Its stems are prostrate and green like the leaves. The ochreae on the stems are hyaline, 3-nerved, conspicuous, longer than or as long as the internodes (rarely shorter in very lax). Its leaves are oblong-elliptic in shape and petiolate, often slightly mucronate. There are many flowers in fascicles in the leaf axils. The perianth is pinkish, 4-5 mm in length, hardening, and accrescent in fruit. It grows widespread in the roadsides, slopes, cliffs, cultivated lands, and 720-3000 m altitudes (Davis, 1967).

P. cognatum belongs to the Polygonaceae family (Yıldırım et al., 2003), and it is locally named "madimak" in Turkey (Macar and Kalefetoglu, 2018; Dereli et al., 2019). The knotweed young shoots and leaves are used to treat several diseases such as diabetes, stomachache, abdominal pain, and anemia in central parts of Turkey (Önen et al., 2014; Sargin et al., 2015, Polat, 2019). The plant is commonly consumed as salad and dishes by local people in many provinces. It was also reported to possess antioxidant, antimicrobial, diuretic, antifungal, insecticidal, and antidiabetic activities (Yıldırım et al., 2003; Baytop, 1999; Dereli et al., 2019). These mentioned properties are attributed to tannins, flavonoid glycosides sterols, triterpenes, polyuronides, and saponins of *P. cognatum* (Dereli et al., 2019). The knotweed was previously reported to contain phenolic compounds, vitamin C, and carotenoids (Yıldırım et al., 2003; Önen et al., 2009). *P. cognatum* samples collected from the Sivas region in Turkey were recorded to be rich in vitamin E and Zn,

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Fe, and Mn elements (Ulusoy et al., 2017). Similarly, folic acid levels of *P. cognatum* samples of the Sivas district were found to be 176, 25, 41, 28, and 10 times higher than those of fruit juices, spinach, broccoli, green beans, brussels sprouts, and soya bean tissue, respectively (Ulusoy et al., 2018). Bioactive components of "madimak" consumed in high amounts and is very famous locally still need to be clarified because of attracting the attention of consumers as potential functional food. Although there have been some studies on bioactive properties and components of natural knotweed (*P. cognatum*) populations from Turkey, to the best of our knowledge, the phenolic composition of *P. cognatum* has not been studied so far. This research was designed to determine the phenolic components of *P. cognatum* collected from two different regions, Cumra and Manisa, Turkey.

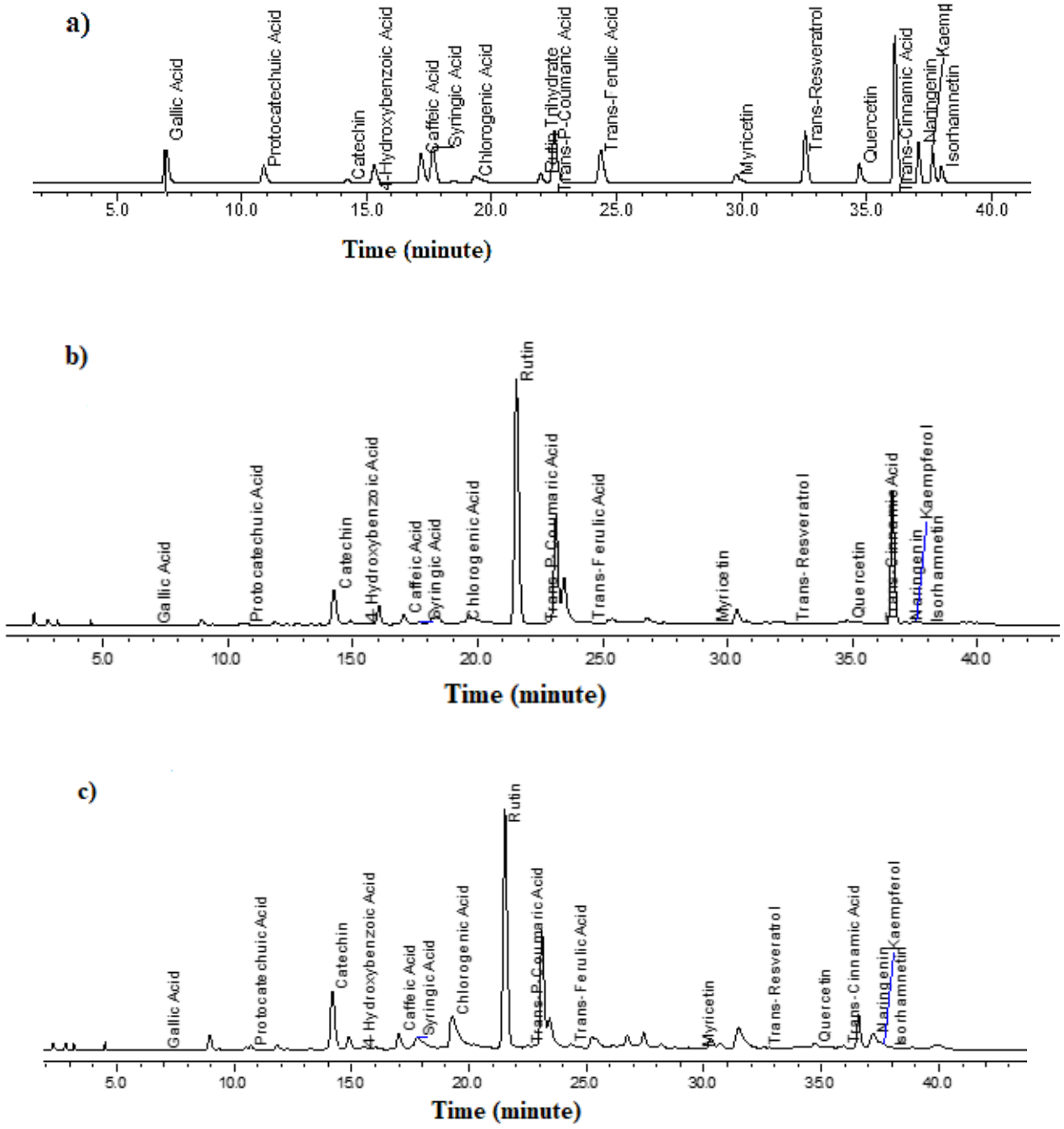

Figure 1. HPLC chromatogram of phenolic compounds of a) standard solutions, b) *P. cognatum* from Manisa, c) *P. cognatum* from Cumra

2. Materials and methods

2.1. Plant materials

This study was performed in Advanced Technology, Research and Application Center laboratories, Selçuk University, in 2016. Knotweed samples were collected from wild flora in Manisa and Çumra (Konya) localities. The sample from Manisa was collected from Salihli-Ödemiş road, near Allahdiyen village, roadside, and altitude was 781 m, on 4th June. The sample from Cumra was collected from the near cemetery, roadside, and altitude was 1016

m, on 26th May. Plant samples were comprised of aerial parts at the flowering stage, and each of them was 100 grams.

2.2. Chemicals

Standards of caffeic acid, catechin, gallic acid, kaempferol, myricetin, naringenin, *p*-hydroxybenzoic acid, rutin trihydrate, *trans*-*p*-coumaric acid, *trans*-ferulic acid, *trans*-resveratrol, *trans*-cinnamic acid were bought from Ehrenstorfer GmbH, protocatechuic acid was from HWI Analytik, rutin trihydrate and syringic acid were from Alfa Aesar. Methanol, glacial acetic acid, and acetonitrile were obtained from Merck (Germany).

2.3. Extraction of phenolic compounds

Extraction of phenolic compounds in dried *P. cognatum* was carried out using methanol as solvent at room temperature for 24 h in a reflux condenser. After the evaporation of methanol in a rotary evaporator (IKA RV 05 Staufen, Germany), liquid-liquid extraction was performed with diethyl ether and ethyl acetate, respectively. After the evaporation of the solvent, the dried extract was dissolved in methanol for further HPLC analysis (Kara et al., 2015).

Table 1. Phenolic compounds of *P. cognatum* from Cumra and Manisa (mg/100 g sample)

Parameter	Retention time (min)	Cumra	Manisa
Gallic acid	6.973	0.016 ± 0.001	-
Protocatechuic acid	10.880	0.152 ± 0.002	0.037 ± 0.006
Catechin	14.242	8.492 ± 0.113	2.635 ± 0.032
4-Hydroxybenzoic acid	15.304	0.097 ± 0.002	0.021 ± 0.003
Caffeic acid	17.178	0.289 ± 0.004	0.110 ± 0.006
Syringic acid	17.651	0.064 ± 0.008	0.043 ± 0.003
Chlorogenic acid	19.326	0.432 ± 0.078	0.043 ± 0.005
Rutin trihydrate	21.961	20.353 ± 0.151	11.404 ± 0.401
trans-*p*-Coumaric acid	22.513	0.019 ± 0.001	0.391 ± 0.005
trans-Ferulic acid	24.364	0.231 ± 0.012	0.088 ± 0.001
Myricetin	29.776	0.074 ± 0.002	0.133 ± 0.018
trans-Resveratrol	32.543	0.43 ± 0.004	0.046 ± 0.004
Quercetin	34.712	0.036 ± 0.001	0.167 ± 0.021
trans-Cinnamic acid	36.129	0.006 ± 0.001	0.038 ± 0.001
Naringenin	37.081	0.034 ± 0.002	0.024 ± 0.001
Kaempferol	37.643	0.234 ± 0.021	0.212 ± 0.046
Isorhamnetin	37.991	0.453 ± 0.022	7.086 ± 0.433
Total		31.407 ± 0.149	22.480 ± 0.806

2.4. Phenolic compounds of P. cognatum

Separation of phenolic compounds of *P. cognatum* was conducted on HPLC system (Shimadzu, Japan) equipped with UV-Vis detector along with ODS 3 column (250 mm × 4.6 mm id, 5 µm particle; Inertsil) using the gradient HPLC method (Kara et al., 2015; Dinc et al., 2018). 0.05% glacial acetic acid in water (A) and acetonitrile (B) mixtures were used as mobile phase at 1.0 ml/min with the following gradient: 0-2 min 8-10% B; 2-27 min 10-30% B; 27-37 min 30-56 %B; 37 min 8% B, before it returned to the initial conditions. The temperature of the column was kept constant at 30 °C. Each 20 µl of the sample was injected, and detection was performed at 280 nm. Seven different concentrations of calibration solutions were used for the calibration curve. The calibration curve was prepared using concentration ratio versus peak area. The calibration equation and R^2 value were calculated using linear regression analysis. R^2 values were greater than 0.99. The solutions of standards and samples were injected three times. Limit of detection (LOD; S/N=3) and limit of quantification (LOQ; S/N=10) were calculated using a certain ratio of signal to noise (S/N). LOQ of standard solutions were between 0,000000156375 ppm (*trans*-resveratrol) and 0,000166248 ppm (catechin).

Quantification of phenolic compounds was performed with different concentrations of phenolic standards (1-55 mg/l-8 point) based on the peak areas of samples (Figure 1a).

3. Results and discussion

In this study, we first investigated the phenolic composition of the whole part of *P. cognatum* collected from two different districts of Turkey in 2016. Plant phenolics, known for their health benefits, exist omnipresent in plant foods and are composed of phenolic acids and polyphenols. Flavonoids constitute the largest subclass (Bondonno et al., 2020). Phenolic acids possess free radical scavenging capacity and hinder oxidative deterioration in emulsion model systems. In recent years, the food industry has taken advantage of their utilization as natural antioxidants to retard the oxidative deterioration of foods (Kiokias et al., 2020). 17 phenolic compounds were investigated in this study, and their chromatograms are presented in Figures 1b and 1c, also and the quantitative results are also given in Table 1. Both samples collected from Cumra and Manisa regions are found to contain almost all phenolic compounds tested. The total quantity of phenolic compounds of the Cumra sample was higher than that of Manisa. Rutin, a flavonol glycoside, was found at the highest level in both samples. The ratio of rutin in the Cumra sample was considerably higher than that of Manisa. Due to the lack of published data on phenolic components of *P. cognatum,* we compared our data with those of buckwheat which also belongs to Polygonaceae (knotweed family). Rutin quantity of sample from Cumra was considerably higher than those of different buckwheat cultivars previously reported (Kreft et al., 2006). On the other hand, the rutin content of buckwheat species *F. tataricum* was higher than Cumra's sample. Rutin, present in substantial quantity in plants, has a broad range of physiological activities. It can diminish the cytotoxicity of oxidized LDL cholesterol and the risk of heart diseases (Atanassova and Bagdassarian, 2009). Medicinally utilization of rutin has the advantage of reducing capillary fragility associated with some hemorrhagic diseases or hypertension in humans (Jiang et al., 2007). The other phenolic compound at the highest ratio in Cumra's sample was catechin. Although Manisa's sample was also high in catechin, its level was lower than Cumra's sample. Catechins have many health benefits in cardiovascular diseases and cancer (Lorenzo and Munekata, 2016). Isorhamnetin content of Manisa's sample was relatively higher than Cumra's sample. Isorhamnetin has many pharmacological characteristics such as antimicrobial, antioxidant, anticancer, neurological, cardiovascular, etc. (Kandakumar and

Manju, 2017). Resveratrol is another phenolic compound previously investigated in *Polygonum* species. In our study, the resveratrol quantity of Cumra's sample was higher than that of Manisa's sample. Resveratrol has prevalent utilization in medicine, health products, and cosmetic industries considering pharmaceutical characteristics such as anti-inflammatory, anticancer, and cardioprotective activities (Wang et al., 2013). Resveratrol levels in our samples were relatively lower than those of *P. cuspidatum* species studied by other researchers (Wang et al., 2013; Chu et al., 2005; Kuo et al., 2014).

4. Conclusions

In this study, the phenolic composition of *P. cognatum*, commonly named "madimak" in Turkey, was analyzed for the first time using the samples from Cumra and Manisa. *P. cognatum* is found to be one of the plants which are rich in the phenolic compound in Turkey. The total quantity of phenolic compounds of *P. cognatum* collected from Cumra was much higher than that collected from Manisa. The highest rutin content of *P. cognatum* may be considered an important dietary source of rutin.

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None.

Conflict of interest

The authors confirm that there are no known conflicts of interest.

CRediT authorship contribution statement

Ahmet Gümüşçü: Resources, Investigation, Data curation, Supervision.
Saliha Dinç: Conceptualization, Visualization, Investigation, Methodology.
Meryem Kara: Visualization, Formal analysis, Writing- original draft
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Supplementary File

None.

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