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# Organ-dependent variability in mineral composition, phytochemicals and antioxidant potentials in *Polygonum equisetiforme* parts

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## ABSTRACT

*Polygonum equisetiforme* is a perennial herbaceous plant thriving in the arid regions of Tunisia and widely used in health care and self-medication. The objective of the current study was to investigate the distribution of minerals, phenolic compounds, and antioxidant potentials in various plant parts including the fruit, stem, leaf, and root. The mineral composition was determined using flame atomic absorption spectrometry. The phenolic content of the samples was investigated using colorimetric assays and identified and quantified using HPLC-ESI/MS. The study found that the different parts of *P. equisetiforme* contain significant amounts of essential minerals such as sodium, potassium, calcium, magnesium, copper, zinc, and iron. The leaf and root extracts had high amounts of polyphenols, flavonoids, and tannins. Through LC-ESI-MS analysis, eleven flavonoids and eight phenolic acids were characterized. The most abundant compounds were gallic acid, quinic acid, catechin (+), and hyperoside. The findings suggest that different parts of *P. equisetiforme* are valuable sources of essential minerals and phenolic compounds, which can have potential health benefits.

## 1. Introduction

*Polygonum equisetiforme* is a perennial herbaceous plant that grows in the Tunisian dry lands and has long been used in traditional medicine as a remedy for several ailments. *P. equisetiforme*, also known as "Gourdhab," is a common wild plant that grows in drought and salt-affected areas (Boughalleb et al., 2020; Mahmoudi et al., 2018). It exhibits high phenotypic plasticity, adapting to different conditions in terms of seed morphology, germination, size, and reserve. The plant's timing of flower production is also plastic, allowing for faster reproduction and complete life history. It produces a large number of flowers to survive numerous interacting stresses and allocates resources to support vegetative growth over reproductive growth (Bidak et al., 2007). The plant is well adapted to climatic conditions, with a creeping port that helps absorb the maximum amount of moisture and reduces contact with open air to minimize transpiration (Mahmoudi et al., 2020b). Our previous work on this plant has shown

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that seed lipids and proteins are widely affected by the geographical area under different bioclimatic zones, from semi-arid to Saharan regions (Mahmoudi et al., 2018). The plant produces more secondary metabolites under arid conditions (Mahmoudi et al., 2019). Recently, it was found to be a salt-tolerant species able to survive at salinities up to 300 mM, with a root system that allows the plant to thrive in the Saharan and arid environments (Boughalleb et al., 2020). Additionally, the plant establishes a widespread distribution across the flooded soils as well as the active sand dunes of the Tunisian desert. It is considered one of the most palatable plants with high nutritional value for domesticated animals feeding which provides potent economic benefits to rural communities (Mouldi, 2014). It has been reported that the aerial parts of the plants as well as the seeds are very rich in polyphenols (Mahmoudi et al., 2018; Mahmoudi et al., 2019). *P. equisetiforme* is widely used in folk medicine for treating colds, coughs, and sore throats (Khafagi & Dewedar, 2000). Additionally, it is employed as a flavoring for tea (Facciola, 1990). Previous chemical characterizations of *P. equisetiforme* have revealed that this plant is very rich in phenolic acids and flavonoids (Hussein et al., 2017), with significant antioxidant, hepatoprotective, antibacterial, and antifungal effects ( El-Toumy et al., 2017; El-Toumy et al., 2019). The essential oil of the plants showed higher free radical scavenging activities against DPPH and ABTS radicals (Abd-ElGawad et al., 2023). Water-soluble polysaccharides isolated from *P. equisetiforme* extracts showed considerable antioxidant potential and anti-tumoral effects against colon and breast cancer cells (Ibrahim & El-Hela, 2012).

Despite its potential health benefits, little is known about the chemical composition and biological properties of its different organs. Therefore, the present report aims to evaluate the mineral composition, polyphenol contents, as well as antioxidant activities in the fruit, leaf, stem, and root of the Tunisian *P. equisetiforme*. In particular, we focused on the importance of phenolic compounds, which have been shown to play a crucial role in the biological activity of many plants, to provide insight into the potential health benefits of this plant.

## 2. Materials and methods

### *2.1. Chemicals and Reagents*

All chemicals were of analytical-reagents grade and were purchased from Sigma-Aldrich (St. Louis, MO, USA) and Loba Chimie Ltd (Mumbai, India).

### *2.2. Plant collection*

Seedlings of *P. equisetiforme* were collected in July 2016 at the EL-Fja, Medenine region, Tunisia (33°28′55.20″ N, 10°39′23.78″ E, at 95 m a.s.l.) and were identified by Dr. Raoudha Abdellaoui, (Laboratory of Rangeland Ecosystems and Valorization of Spontaneous Plants and Associated Microorganisms, Arid Regions Institute, Medenine, Tunisia) and voucher specimens of fruit (PEF1601), stem (PES1602), leaf (PEL1603), and root (PER1604) were deposited in the seed bank of the Arid Regions Institute. The fruit, leaf, stem, and root of the plant were cleaned, separated, shade-dried, ground to a fine powder, and stored for subsequent analysis.

### *2.3. Mineral analysis*

Firstly, an oven drying step was done, at 60 °C, for the leaf, stem, fruit, and root of *P. equisetiforme* until a constant weight. After that, the minerals were subjected to an acidic extraction process utilizing hydrochloric acid and nitric acid (Boughalleb et al., 2021). The mineral composition in the different organs of the plant was determined using flame atomic absorption spectroscopy (Shimadzu AA-6800) as detailed in Maher et al. (2023).

### *2.4. Determination of phytochemical contents*

#### *2.4.1. Preparation of methanolic extracts*

The fine powders of the different plant parts of *P. equisetiforme* were extracted by simple maceration in hydromethanolic solution 80% (v/v) in a light-protected flask (at 40 °C for 24 h), the macerate was then subjected to centrifugation at 4500 rpm for 15 min. Finally, the supernatant was filtered through a 0.2 µm syringe filter and stored at –20 °C for subsequent analysis (Mahmoudi et al., 2021b).

#### *2.4.2. Total phenolic content*

To evaluate the polyphenols in *P. equisetiforme* organs, the Folin-Ciocalteu method was adopted (Dewanto et al., 2002). A 125 µl of sample extract was added to 125 µl of Folin-Ciocalteu reagent and 1250 ml of $Na_2CO_3$ (7%). The resultant mixture was subsequently diluted with deionized water until the final volume reached 3 ml. The absorbance was read at 760 nm after an incubation away from the light of 90 min at room temperature. The polyphenol contents in the plant parts were expressed as mg of gallic acid equivalents per gram of dry weight (mg GAE/g DW) (Maher et al., 2023).

#### *2.4.3. Total flavonoid content*

The amount of flavonoids in *P. equisetiforme* parts was evaluated according to (Dewanto et al., 2002; Helrich, 1990). In a 5 ml flask, 100 µl of plant part extract samples were mixed with 75 µl of 7% $NaNO_2$ and 150 µl of 10% aluminum chloride hexahydrate. After that, a volume of 500 µl of 1 M NaOH was added, and deionized water was added to the resulting mixture to obtain a final volume of 3 ml. Absorbance was read at 510 nm versus a blank. The concentration of flavonoids is expressed as quercetin equivalents per gram of dry weight (mg QE/g DW) using the standard curve (Maher et al., 2023).

#### *2.4.4. Condensed tannin content*

The level of tannins in the plant organs was measured based on the vanillin-sulfuric acid colorimetric reaction (Broadhurst & Jones, 1978). A volume of 4% vanillin solution (1.5 ml) and concentrated sulfuric acid (750 µl) were added to a tube containing 25 µL of plant extract. Absorbance was determined at 500 nm versus a blank after incubation at 15 min in the dark. The condensed tannin content is expressed as mg catechin (CE) equivalents (mg CE/g DW) by utilizing the standard curve prepared from authentic catechin.

#### *2.4.5. Analysis by high-performance liquid chromatography coupled with electrospray ionization–mass spectrometry*

The LC-MS system consists of two LC-20ADXR pumps for mobile phase delivery, SIL-20AXR autosampler for automated sample injection, SCL-10A system controller serves as the central control unit, CTO-20 AC column oven, electrospray ionization source for sample ionization, and DGU-20AS degasser enhance separation efficiency. The MS was operated in negative mode electrospray ionization (ESI). The experiment involves the use of two mobile phases, A and B, in linear gradient chromatography. Mobile phase A comprises of water with formic acid (0.1%), while mobile phase B

consists of water with formic acid (0.1%) and methanol with formic acid. The linear gradient involves a stepwise increase in the proportion of mobile phase B in the following sequence: from 10 to 20% (0-14 minutes), from 20 to 55% (14-27 minutes), from 55 to 100% (27-37 minutes), 100% (37-45 minutes), and a decrease to 10% (45-50 minutes). The characterization of the polyphenol compounds was conducted using a comparative approach, whereby the MS spectra as well as the retention times of the detected compounds were matched against those of reference standards and the concentrations of the identified compounds were expressed as µg/g DW (Mahmoudi et al., 2020a; Mahmoudi et al., 2021a).

### 2.5. Determination of antioxidant potential

#### 2.5.1 Total antioxidant activity

The total antioxidant potential of *P. equisetiforme* parts was investigated according to the phosphomolybdenum assay (Prieto et al., 1999). In brief, 200 µl of samples were added to 2 ml of reagent solution composed of sulfuric acid (0.6 M), sodium phosphate (28 mM), and ammonium molybdate (4 mM). The mixture was incubated for 90 minutes at 95 °C and the absorbance was determined at 700 nm versus a blank. Results are expressed as equivalents of gallic acid (mg GAE/g DW) (Maher et al., 2023).

#### 2.5.2. DPPH anti-radical activity

The antiradical potential effect of the different plant parts was evaluated against the DPPH free radical according to Sánchez-Moreno et al. (1998) as detailed in Maher et al. (2023).

#### 2.5.3. Reducing power potential

The reducing power potential of the plant part extracts was evaluated using the procedure outlined by Oyaizu (1986). A volume (2.5 ml) of phosphate buffer (0.2 M, pH = 6.6) and potassium ferricyanide (1%) were mixed with 1 ml of plant extracts and incubated for 20 minutes at 50 °C, After the incubation step (20 min at 50 °C), 2.5 ml of TCA (10%) was added to the mixture. After that, the solution was centrifuged at 13.000 rpm for 10 min, and 2.5 ml of the supernatant was mixed with an equal volume of distilled water and 0.5 ml of $FeCl_3$ solution (0.1%). The DO was read at 700 nm. The reducing potential in the extracts was expressed as $EC_{50}$ values (mg/ml) which correspond to the concentration at which ferrous ions were chelated by 50% (Maher et al., 2023).

### 2.6. Statistical analysis

The statistical analysis was carried out using IBM SPSS statistical software (version 20.0, IBM Corp., Armonk, NY, USA). The data were presented as means ± standard deviation (SD) from three replicates and subjected to analysis of variance (ANOVA). To compare the means, Duncan's post-hoc tests were performed. Linear regression analysis was employed to determine the $EC_{50}$ values for the reducing power assays. The significance level was set at 5% to determine the differences between means.

## 3. Results and discussion

### 3.1. Variations in mineral composition

In the current study, the macro and microelement contents in the leaf, stem, fruit, and root of *P. equisetiforme* were quantified using flame atomic absorption spectrometry (FAAS). As shown in Table 1, the stem and fruit parts contained higher amounts of Na, Ca, and Mg compared to the leaf and root. While leaf and stem contained higher K levels (6.59 and 6.08 mg/kg), followed by fruit (4.87 mg/kg) and root (2.81 mg/kg). It was shown that Cu and Fe were higher in the root part with respective values of 0.22 and 0.65 mg/kg. The stem has the highest Zn content (0.44 mg/kg) compared to the leaf and root parts (0.36 and 0.22 mg/kg). However, the studied microelements were not detected in the fruit. The present findings corroborate our previous study on Tunisian *Polygonum* species, which highlighted significant differences in mineral composition among the leaves, stems, and roots of *P. maritimum* and *P. aviculare* across various plant organs. Among *P. maritimum* parts, the leaf had higher Na, K, Ca, and Mg levels with respective values of 14.2, 9.5, 12.5, and 5.8 mg/Kg. However, the root part possessed a higher concentration of Fe. Additionally, in *P. aviculare* parts, the stem showed higher mineral amounts compared to the other organs (Mahmoudi et al., 2021b). These contents were similar to the value that had been previously recorded in several Polygonaceae i.e. *Fagopyrum tataricum* (Huang et al., 2014) and *Persicaria tinctoria* (Park et al., 2016). Moreover, Corlett et al. (2002) recorded higher amounts of the mentioned minerals in the aerial parts of other *Polygonum* species including *P. odoratum* and *P. runcinatum*.

**Table 1.** Mineral element composition *P. equisetiforme* parts (mg/kg dry matter)

| | Na | K | Ca | Mg | Cu | Zn | Fe |
|---|---|---|---|---|---|---|---|
| **Fruit** | $8.62 \pm 0.7^{a}$ | $4.87 \pm 1.47^{b}$ | $7.69 \pm 3.44^{b}$ | $2.6 \pm 0.58^{a}$ | - | - | - |
| **Leaf** | $4.626 \pm 0.3^{c}$ | $6.599 \pm 0.04^{a}$ | $4.552 \pm 0.04^{d}$ | $1.22 \pm 0.09^{d}$ | $0.104 \pm 0.06^{c}$ | $0.364 \pm 0.006^{b}$ | $0.43 \pm 0.02^{b}$ |
| **Stems** | $8.72 \pm 0.91^{a}$ | $6.08 \pm 0.06^{a}$ | $8.37 \pm 0.68^{a}$ | $2.12 \pm 0.1^{b}$ | $0.18 \pm 0.06^{b}$ | $0.44 \pm 0.11^{a}$ | $0.28 \pm 0.02^{c}$ |
| **Root** | $7.76 \pm 0.74^{b}$ | $2.81 \pm 0.05^{c}$ | $7.0 \pm 0.08^{c}$ | $1.93 \pm 0.12^{c}$ | $0.22 \pm 0.04^{a}$ | $0.26 \pm 0^{c}$ | $10.65 \pm 0.29^{a}$ |

Data expressed as means ± standard deviation ($n = 3$). The different lower-case letters in the same row indicate significantly different values ($p < 0.05$)

Many research studies have established a significant correlation between the pharmacological properties of plants and their mineral composition. Additionally, several minerals are crucial components for various human physiological processes in the human body. Mineral elements play a vital role in various biological processes, including the formation of macromolecules (i.e. proteins, nucleic acids, and carbohydrates). The chemical characteristics of these elements significantly impact the structure as well as the function of macromolecules, which are essential for the proper functioning of living organisms (Kolasani et al., 2011).

For instance, the mineral element Mg is an essential cofactor for many enzymes that are involved in carbohydrate metabolism. Mg ions help stabilize the structure of enzymes, which enables them to function efficiently in the catalysis of biochemical reactions involved in the breakdown and synthesis of carbohydrates. Similarly, Ca ions are essential for the proper functioning of enzymes involved in blood clotting and muscle contraction. Mineral elements are also involved in the regulation of enzyme activity through the modulation of protein structure and function. Additionally, iron is indispensable as enzymes that participate in cellular respiration, which is the process by which cells convert glucose into energy (Kanjaksha, 2006). Furthermore, mineral elements are also involved in the formation and maintenance of the structure of carbohydrates, which are macromolecules that play crucial roles in energy storage, structural support, and cell recognition (Kolasani et

al., 2011). The findings of this study highlight the importance of *P. equisetiforme* parts as a potential source of essential minerals for human consumption and medicinal purposes.

### 3.2. Variations in phytochemical contents

Polyphenols are a large and diverse group of naturally existing organic compounds and abundantly present in numerous plant-derived food sources including fruits, vegetables, whole grains, seeds, and tea. They are characterized by several phenol groups and possess a wide range of chemical structures. The beneficial health effects of these components are well documented. Polyphenols are known to possess antioxidant, anti-inflammatory, and anticancer properties, and may help to protect against a range of chronic diseases. Common polyphenols include flavonoids, phenolic acids, stilbenes, and lignans, and their levels can vary widely depending on factors such as the type of plant, the growing conditions, and the processing and preparation methods used. The different plant extracts of *P. equisetiforme* are rich in polyphenols, flavonoids, and condensed tannins. The levels of these compounds vary among different parts of the plant. The highest concentration of polyphenols was found in the leaves (51.75 mg GAE/g DW), followed by the root with 48.6 mg GAE/g DW, stem with 40.15 mg/g GAE/g DW, and fruit with 14.55 mg GAE/g DW (Figure 1). Similarly, the highest concentration of flavonoids was found in the leaves with a content of 62.75 mg quercetin equivalents (QE) per gram of DW, followed by the root with 59.55 mg QE/g DW, stem with 55.95 mg mg QE/g DW, and fruit with 21.05 mg mg QE/g DW. In terms of condensed tannins, the root contains the highest levels at 15.1 mg/g, followed by the stem and leaf, with the fruit having the lowest levels. The amounts of polyphenols, flavonoids, and tannins in different populations of Tunisian *P. equisetiforme* from distinct bioclimatic zones, namely the Saharan, arid, and semi-arid areas, were found to be in the range of 31-113 mg GAE/ g DW, 29-130 mg QRE/g DW and 8-33 mg CTE/g DW, respectively (Mahmoudi et al., 2018). Compared to other Tunisian *Polygonum* species, the halophyte sea knotgrass (*P. maritimum*) exhibited higher TPC levels in its stem, leaf, and root parts. In contrast, knotgrass (*P. aviculare*) exhibited lower levels which ranged between 14.1 mg GAE/g DW (in leaves) and 31.90 mg GAE/g DW (in stems). Additionally, the stems of both species possessed higher TFC amounts compared to the other plant parts while the condensed tannins were higher in the roots (Mahmoudi et al., 2021b). Furthermore, these contents are consistent with those measured in other Mediterranean *Polygonum* sp. (El-Haci et al., 2013; Maria Joao Rodrigues et al., 2019a; Maria Joao Rodrigues et al., 2019b). These findings indicate that the leaves, roots, and stems of *P. equisetiforme* are valuable sources of polyphenols, flavonoids, and condensed tannins that may have potential health benefits for humans.

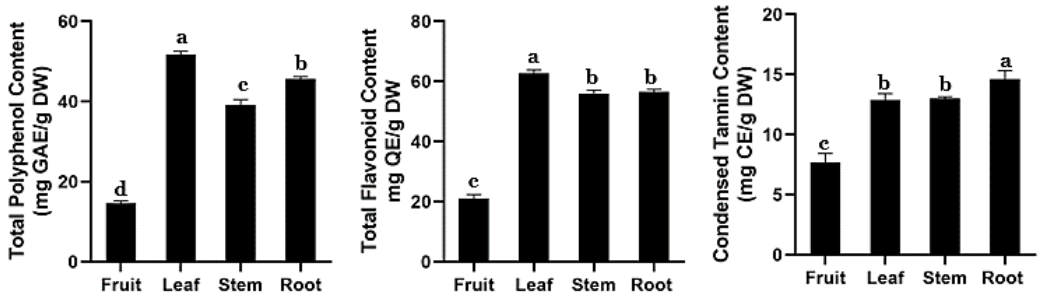


Figure 1. Total polyphenol, total flavonoid, and condensed tannin contents in different parts of *P. equisetiforme*
The different lower-case letters indicate significant ($p < 0.05$) differences. Data expressed as means ± SD ($n = 3$).

### 3.3 Variations of polyphenolic compounds

Polyphenols in the fruit, leaf, stem, and root of *P. equisetiforme* were identified through an LC-ESI/MS analysis as summarized in Table 2. Eight compounds were characterized and the most abundant component in all plant parts is quinic acid, with the highest concentration in the fruit (259.81 µg/g) and the lowest in the leaf (26.61 µg/g). Gallic acid is also present in all plant parts, with the highest concentration in the fruit (48.34 µg/g) and the lowest in the leaf (8.34 µg/g). Among the other phenolic acids, the concentrations vary depending on the plant part. For example, 4-*O*-caffeoylquinic acid is found only in the fruit, stems, and root, with the highest concentration in the stems (44.89 µg/g). Caffeic acid is not identified in the stems, but is present in the fruit, leaf, and root, with the highest concentration in the fruit (0.82 µg/g). Syringic acid is found in all plant parts, with the highest concentration in the fruit (2.28 µg/g) and the lowest in the root (1.13 µg/g). *trans*-Ferulic and *p*-coumaric acids are also present in all plant parts, with the highest concentrations in the fruit (6.14 µg/g and 1.85 µg/g, respectively) and the lowest in the root (2.8 µg/g and 2.48 µg/g, respectively). Overall, these results suggest that *P. equisetiforme* contains a diverse array of phenolic acids in its different plant parts and that the identified compounds exhibit high variation based on the plant parts. Furthermore, in this study, the presence and quantity of eleven flavonoid compounds were identified in the fruit, leaf, stems, and root of *P. equisetiforme* using LC-MS. Catechin (+) was found to be the most abundant flavonoid compound in all four parts of the plant, with the highest concentration in the stems (1706.35 µg/g) and lowest in the leaf (4.38 µg/g). Epicatechin and hyperoside were also present in all parts of the plant, with the highest concentrations found in the root and stems, respectively. Rutin was identified in all parts of the plant, but its concentration was highest in the fruit (0.82 µg/g). Quercetin was identified in the leaf with the highest amounts among all the plant parts, measuring 22.37 µg/g. However, detectable levels of quercetin were also seen in the root and stem, although at relatively lower concentrations. Both naringin and quercetin were present in all parts of the plant but in relatively lower amounts. Naringenin was identified in the fruit, stems, and roots but was not found in the leaf. Cirsiliol was present in all parts of the plant, with the highest concentration found in the root (24.63 µg/g). The compounds that were identified have been previously documented in both the seeds and the shoots of the plant. They are significantly affected by environmental fluctuations in different bioclimatic conditions across Tunisia (Mahmoudi et al., 2018; Mahmoudi et al., 2019). Furthermore, various phenolic acids and flavonoid compounds have been identified in other *Polygonum* species, with varying amounts found in different parts of the plant, including *P. maritimum*, *P. aviculare* (Mahmoudi et al., 2021a; Mahmoudi et al., 2021b) and *P. cognatum* (Gümüşçü et al., 2022).

The identified compounds have been documented to possess diverse biological activities. For instance, quinic acid and gallic acid are widely distributed in nature and are found in many fruits, vegetables, and plants. These compounds have been shown to have anti-inflammatory, antioxidant, antimicrobial, and antifungal effects (Cao & Prior, 1998). Furthermore, these compounds have been shown to exhibit high gastrointestinal and neuroprotective effects which may have potential benefits for the management of gastrointestinal disorders and improving cognitive function in animal models of neurodegenerative diseases (Jang et al., 2017; Song et al., 2019). Also, catechin has been shown to possess several biological activities, such as antioxidant, cardiovascular, and anti-cancer effects (Bernatoniene & Kopustinskiene, 2018; Vickers, 2017).

It is well known that phenolic content, as secondary metabolites of plants, is widely influenced by environmental factors such as salinity and aridity. Our previous study demonstrated that antioxidant activities and phytochemical contents, including phenolic acids and flavonoid compounds, in *P. equisetiforme* extracts increased with salinity. Specifically, an increase in phenolic acids, particularly quinic, gallic, and protocatechuic acid, was observed, followed by quercetin-3-*O*-galactoside, catechin, and epicatechin (Boughalleb et al., 2020). Additionally, these compounds were found to increase with aridity in the Tunisian Saharan climatic range (Mahmoudi et al., 2019).

These results provide valuable information on the distribution patterns of polyphenolic components in different *P. equisetiforme* parts, which could have important implications for the use of this plant in folk medicine and the development of new therapeutics.

### *3.4. Variations in the antioxidant potential*

The results indicate that *P. equisetiforme* contains antioxidants in all its parts – fruit, leaf, stem, and root. The antioxidant activities were measured through three in vitro assays: total antioxidant capacity (TAC), DPPH, and reducing power assays. The TAC values were found to be in the range of 11.85-24.55 mg GAE/g DW, with the highest potential found in the stem **(Figure 2)**. The DPPH radical scavenging activities ranged between 11.58 to 20.6 mg TRE/g DW, with the highest value again found in the stem. The reducing power values, measured by $EC_{50}$, ranged from 40.6 to 58.5 µg/ml, with the highest reducing power found in the stem extract.

**Table 2.** Phenolic compounds identified by LC-ESI/MS in the different parts of *P. equisetiforme*

| | RT | m/z | Fruit | Leaf | Stem | Root |
|---|---|---|---|---|---|---|
| Quinic acid | 2.065 | 191 | 259.81 ± 2.14$^{a}$ | 26.61 ± 0.46$^{d}$ | 91.31 ± 0.16$^{c}$ | 191.63 ± 10.61$^{b}$ |
| Gallic acid | 4.056 | 169 | 48.34 ± 0.8$^{a}$ | 8.34 ± 0.16$^{d}$ | 17.06 ± 0.04$^{c}$ | 45.43 ± 0$^{b}$ |
| Protocatechuic acid | 7.072 | 153 | 7.36 ± 0.2$^{a}$ | 0.84 ± 0.02$^{d}$ | 4.57 ± 0.1$^{c}$ | 6.33 ± 0.22$^{b}$ |
| Catechin (+) | 11.439 | 289 | 28.95 ± 0.59$^{c}$ | 4.38 ± 0.16$^{d}$ | 44.3 ± 0.2$^{b}$ | 1706.35 ± 39.23$^{a}$ |
| 4-*O*-Caffeoylquinic acid | 12.198 | 353 | 0.49 ± 0.01$^{c}$ | nd | 0.1 ± 0$^{b}$ | 44.89 ± 0.1$^{a}$ |
| Caffeic acid | 14.841 | 179 | 0.82 ± 0.01$^{b}$ | 0.65 ± 0.14$^{c}$ | nd | 2.56 ± 0.08$^{a}$ |
| Syringic acid | 16.506 | 197 | 2.28 ± 0.03$^{a}$ | 1.37 ± 0.03$^{c}$ | 1.87 ± 0.02$^{b}$ | 1.13 ± 0.03$^{c}$ |
| Epicatechin | 16.856 | 289 | 2.69 ± 0.03$^{d}$ | 3.92 ± 0.02$^{c}$ | 11.77 ± 0.04$^{b}$ | 42.69 ± 1.06$^{a}$ |
| *p*-Coumaric acid | 21.185 | 163 | 6.14 ± 0.2$^{a}$ | 5.59 ± 0.06$^{b}$ | 3.38 ± 0.02$^{c}$ | 2.8 ± 0.05$^{d}$ |
| *trans*-Ferulic acid | 23.451 | 193 | 1.85 ± 0.04$^{b}$ | 2.36 ± 0.05$^{a}$ | 1.29 ± 0.07$^{c}$ | 2.48 ± 0.04$^{a}$ |
| Hyperoside | 24.634 | 463 | 15.83 ± 0.65$^{c}$ | 41.63 ± 0.21$^{a}$ | 20.1 ± 0.13$^{b}$ | 4.44 ± 0.96$^{d}$ |
| Rutin | 24.52 | 609 | 0.82 ± 0.03$^{b}$ | 0.36 ± 0.02$^{c}$ | 0.21 ± 0.01$^{c}$ | 1.39 ± 0.15$^{a}$ |
| Quercetrin | 27.216 | 447 | 0.93 ± 0.02$^{d}$ | 22.37 ± 0.59$^{a}$ | 1.47 ± 0.02$^{c}$ | 11.09 ± 0.12$^{b}$ |
| Naringin | 26.516 | 579 | 0.05 ± 0$^{d}$ | 1.62 ± 0.01$^{b}$ | 0.31 ± 0$^{c}$ | 1.91 ± 0.02$^{a}$ |
| Quercetin | 32.67 | 301 | 3.68 ± 0.12$^{b}$ | 6.53 ± 0.4$^{a}$ | 3.27 ± 0.05$^{b}$ | 1.06 ± 0.02$^{c}$ |
| Naringenin | 34.37 | 271 | 0.33 ± 0.01$^{b}$ | nd | 0.23 ± 0.01$^{b}$ | 0.79 ± 0.05$^{a}$ |
| Luteolin | 36.95 | 285 | nd | 0.38 ± 0.13$^{a}$ | nd | nd |
| Cirsiliol | 35.811 | 329 | 2.53 ± 0.05$^{c}$ | 2.06 ± 0.11$^{c}$ | 4.27 ± 0.07$^{b}$ | 24.63 ± 0.84$^{a}$ |
| Cirsilineol | 38.952 | 343 | nd | 0.12 ± 0.02$^{a}$ | 0.09 ± 0.01$^{b}$ | nd |

Data expressed as means ± standard deviation ($n$ = 3). nd: not detected. The different lower-case letters in the same row indicate significantly different values ($p <$ 0.05), nd: Not determined

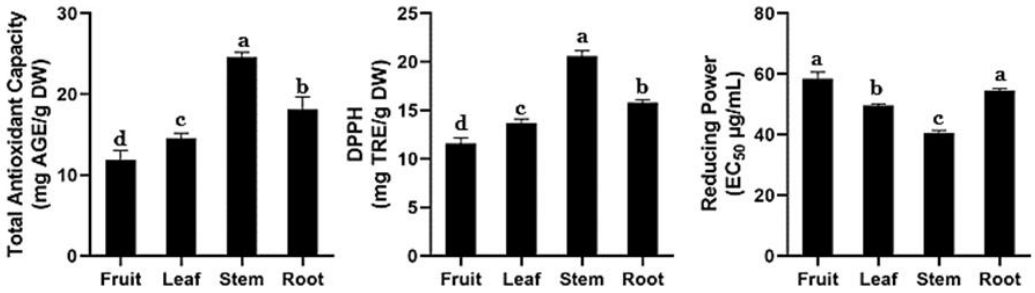


**Figure 2.** Total antioxidant capacity, DPPH radical scavenging activity, and reducing power in *P. equisetiforme* parts
The different lower-case letters indicate significant ($p <$ 0.05) differences. Data expressed as means ± SD ($n$ = 3).

Our findings corroborate the results of many studies that have reported high antioxidant potential in several other species of *Polygonum* (El-Haci et al., 2013; Mahmoudi et al., 2018; Mahmoudi et al., 2019; Mahmoudi et al., 2021b; Maria João Rodrigues et al., 2016). In many studies investigating the antioxidant potential of plant extracts, polyphenols have been identified as the major contributors to the observed effects. This is likely a result of the remarkable ability of polyphenols to neutralize free radicals and chelate transition metals, both of which are important mechanisms of antioxidant activity (Yang et al., 2004). Overall, the study suggests that this plant has significant antioxidant activity in all of its parts, with the stem extract exhibiting the highest potential for antioxidant activity. These results may have implications for the use of *P. equisetiforme* as a source of natural bioactive compounds for

therapeutic or functional food applications, although further research is required to provide a more comprehensive understanding of the specific bioactive components responsible for these observed effects.

## 4. Conclusions

In conclusion, the analysis of mineral and polyphenolic contents of different parts of *P. equisetiforme* demonstrated that this plant has the potential to be a sustainable source of minerals and polyphenols. Specifically, stem and fruit parts were found to contain higher amounts of certain minerals such as Na, Ca, and Mg, while leaf and stem contained higher levels of K. Cu and Fe were found to be higher in the root part, while Zn was highest in the stem. Moreover, the phytochemicals varied among different parts of the plant with the highest concentration found in the leaves. These findings suggest that incorporating *P. equisetiforme* into sustainable agricultural practices in Tunisian arid regions may provide a promising way to promote both human health and environmental sustainability. Additionally, the antioxidant, anti-inflammatory, and anticancer properties of the plant's polyphenols may provide an important means to combat a range of chronic diseases.

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## Conflict of interest

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

## Statement of ethics

In this study, no method requiring the permission of the “Ethics Committee” was used.

## Availability of data and materials

All data generated or analyzed during this study are included in this published article.

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## CRediT authorship contribution statement

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## Supplementary File

None.

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