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ASSESSING THE HEALTH BENEFITS AND COSMETIC POTENTIAL OF PARSLEY EXTRACTS FROM ALGERIAN CULTIVAR

Karima BOUZENOURA[1], Aya REDJIMI[1], Yacine TORCHE[1,2*], Houssem Eddine KEHILI[1], Khadidja BEKHOUCHE[3]

[1]Nature and Life Sciences Faculty, Abdelhafid Boussouf University, Mila, Algeria
[2]Laboratory of Natural Sciences and Materials, Abdelhafid Boussouf University, Mila, Algeria
[3]Pharmaceutical Sciences Research Center (CRSP), Ali Mendjli – Constantine, Algeria

Abstract: Parsley (*Petroselinum crispum* Fuss.) is one of the oldest and most widely used aromatic plants, particularly in the Mediterranean region. In this study, we examined multiple extracts from various parts of parsley growing under Algerian environmental conditions to assess their biological activities and potential health benefits, and the possibility of their application in the local cosmetic industry. After carefully selecting and drying samples of parsley roots, leaves, stems, and seeds, we prepared different extracts, including ethanol extract, aqueous extract, essential oil, and hydrosol. The extraction of essential oil revealed its significant presence in the seeds, whereas leaves and stems contained considerably lower amounts, and the roots were completely free of essential oil. All extracts were tested for antioxidant, antibacterial and antifungal activities. The results showed that the seeds exhibited the highest antibacterial, antifungal and antioxidant activities among all extracts, with essential oil and ethanol extracts being particularly effective, while hydrosols and other extracts from the plant's remaining parts demonstrated less effectiveness. Therefore, for Algerian parsley cultivars, seeds appear to be optimal source for health benefits and the cosmetic applications in producing natural parsley-based products.

Keywords: *Petroselinum crispum*, essential oil, antioxidant activity, antibacterial activity, cosmetic industry

INTRODUCTION

Petroselinum crispum (Mill.) Fuss 1886, commonly known as parsley, belongs to the Apiaceae family and is synonymous with *Petroselinum hortense* Hoffm. Parsley is a cool-season crop that thrives at temperatures between 15 and 20°C. This biennial herb has become a global staple, cultivated in diverse climates worldwide, and can be grown all year round as long as the temperature is maintained above 5°C, making it widely used, and systematically available (Farzaei et al., 2013; Park and Ryu, 2022). The plant is typically sown in mid-March, transplanted in June, and harvested from November to March the following year. It is native to Southeastern Europe and North Africa.

Beyond its widespread use as a green vegetable and culinary garnish, parsley serves various medicinal purposes in the traditional and folklore medicine of different countries (Casanova et al., 2024). This versatile herb has been valued for its potential health benefits, and its application in traditional medicine spans a range of uses from digestive aid to anti-inflammatory properties, parsley has found a place in folk remedies for different ailments (Farzaei et al., 2013). Various compounds from different phytochemical categories have been identified in Parsley, making the plant a valuable source of bioactive food and pharmaceutical resources (Yu et al., 2021). Moreover, different biological activities such as antioxidant, antidiabetic, immunomodulating, antimicrobial and cytotoxic activities have been attributed to this plant (Mara De Menezes Epifanio et al., 2020). The leaves also possess anticoagulant and abortifacient activity and are useful in skin diseases widely used in north Africa (Ziyyat et al., 1997).

In addition to its usage in the food industry, *Petroselinum crispum* is widely employed in the cosmetic industry (Park and Ryu, 2022). Products based on parsley, including perfumes, soaps, and creams, are now extensively used in cosmetic manufacturing (Choi and Moon, 2017). Furthermore, consumers show an increasing interest in plant-based, natural ingredients for cosmetics due to their perceived health benefits and lower environmental impact (Amberg and Fogarassy, 2019). Natural and plant-based products contains compounds thought to offer protective effects for skin health, such as polyphenols and flavonoids, which contribute to their antioxidant power (Mert and Timur, 2017). These antioxidants help neutralize free radicals, which are known to contribute to skin aging and inflammation (Kļaviņa et al., 2024). Moreover, it has been shown that plant-based products have antimicrobial effects on skin-related microorganisms, as some are implicated in skin issues like acne and infections (Ahmed et al., 2020). Therefore, it is capital to asses both antioxidant and antimicrobial properties to determine how effective target herb extracts and eventually their use as natural alternatives to synthetic ingredients in cosmetic formulations.

The present study aims to assess different biological activities of Parsley growing under Mediterranean North African environmental conditions for both health benefits and cosmetic uses.

METHODS AND MATERIALS

To make an exhaustive phytochemical screening, all *P. crispum* plant parts was used and different extractions methods were tested for biological activities.

***Correspondence**: Yacine Torche, Nature and Life Sciences Faculty, Laboratory of Natural Sciences and Materials, Abdelhafid Boussouf University, Mila, Algeria, email: torche.yacine@centre-univ-mila.dz*

Plant material, geographical and climate conditions

Samples of the Parsley including leaves, stems, roots and seeds were collected during 2023 winter from North East Algeria in North Africa from farms of two regions (Fig 1). The climate is typically Mediterranean, characterized by hot, dry summers and mild, wet winters. Summers typically span from June to September, with temperatures often exceeding 30°C. Winters, running from December to February, are milder, with daytime temperatures ranging from 10°C to 15°C. The region receives most of its precipitation during the winter months, with occasional rainfall in summer.

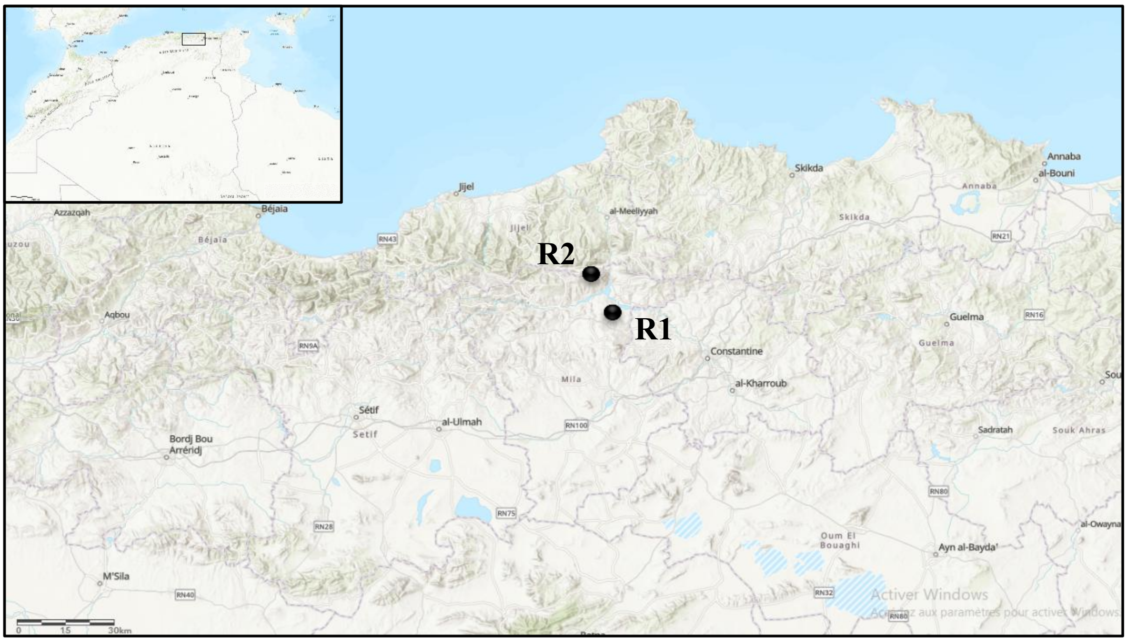

Fig 1. Geographical location of parsley samples from two regions (black dots) in North East Algeria. Map created in ArcGIS Online (Esri, California, USA).

Samples preparation

All parsley parts samples were initially shade-dried at room temperature for 15 days, then dried in an oven at 37°C until a stable dry weight was recorded. Subsequently, the samples were finely grinded and stored at 4°C for further analysis.

Extraction methods

To facilitate further use in the local cosmetic industry, polar solvents (water and ethanol) were chosen for their safety and low cost (Joshi and Adhikari, 2019). In addition, hydrosol and essential oil were also extracted.

Aqueous extracts

For aqueous extracts, exact (10 g) powder from each parsley part was macerated in 100 mL of distilled water with mechanical agitation for 24 h at room temperature, the resulting solution was then filtered and stored at 4°C.

Ethanolic extracts

For ethanolic extracts, 5 g of *P. crispum* shoot part (leaves, stem), roots and seeds were soaked in 20 mL of ethanol at room temperature for 24h, after which the mixture was filtered. Ethanol was removed by evaporation under reduced pressure using a rotary evaporator (LAB1ST Scientific, IG-RE-100Pro, China), resulting in the formation of a dry extract with a brownish-green gummy consistency, the operation was repeated until a transparent filtrate was obtained.

Essential oil and hydrosol

Finally, essential oil (EO) and hydrosol were obtained by steam distillation from 50 g of different plant parts, using a Clevenger-type apparatus (Molyneux Philip, 2004) (isolation ratio 1:15). In this process, two immiscible phases are formed during condensation, where the hydrosol is the aqueous phase byproduct. Essential oil yield as well as pH were determined.

Qualitative phytochemical analysis

To identify the presence or absence of certain compounds belonging to secondary metabolite chemical families, we performed specific phytochemical tests based on coloration, turbidity, or precipitation reactions, using methods described in the literature (Shaikh and Patil, 2020).

Biological activities

The different extracts from each part of *P. crispum* (aerial parts, roots, and seeds) were used to assess antibacterial and antioxidant activities known for their high potential in health care (Ahmed et al., 2020; Kļaviņa et al., 2024).

Antibacterial assessment

To determine the presence of antibacterial and antifungal activity in plant extracts, the disc diffusion method for antimicrobial susceptibility testing was used, as described by Zaidan *et al.*, 2005. A sterile swab was used to evenly spread a four bacterial culture and a yeast (adjusted to the 0.5 McFarland standard) onto Mueller–

Hinton agar plates as presented in table 1. The plates were allowed to dry for 15 minutes before conducting the sensitivity test. The plant extract was subjected to a two-fold serial dilution in DMSO to obtain concentration ratios of 1:2 (v/v) (D1), 1:4 (v/v) (D2), and 1:8 (v/v) (D3). A positive control (gentamicin), in the form of a commercial antibiotic disc, was included. Discs impregnated with the plant extract were then placed on the surface of the Mueller–Hinton agar, and each test plate contained five discs (SS, D1, D2, D3, and the gentamicin disc). The plate was incubated at 37°C for 24 hours. After incubation, the plates were inspected for the inhibitory zone (IZ). The inhibitory zone was measured with calipers and recorded. Tests were conducted three times to verify dependability (Inhibition zone determination < 8 mm: not sensitive, < 9 - 14 mm: sensitive, > 15 - 19 mm: very sensitive, > 20 mm: extremely sensitive) (Szczepanski and Lipski, 2013). SS of the essential oil was used on two volume 20 and 10 µL.

Table 1.

Bacterial strains used in the study

Family	Bacteria strains	Abbreviations	Gram
Micrococcaceae	*Staphylococcus aureus* ATCC 25923	Sa	+
Enterobacteriaceae	*Escherichia coli* ATCC 25922	Ec	-
Bacillaceae	*Bacillus spizizenii* ATCC 6633	Bs	+
Pseudomonadaceae	*Pseudomonas aeruginosa* ATCC 27853	Pa	-
	Fungal strain		
Saccharomycetaceae	*Candida albicans* ATCC 10231	Ca	/

Antioxidant assessment

The ability of all extracts, in terms of hydrogen donating or radical scavenging capacity, was measured using the stable radical DPPH according to the published method (Brand-Williams et al., 1995; Molyneux Philip, 2004). For this purpose, an ethanolic solution of DPPH reagent (0.1 mM) was freshly prepared. The varying concentrations of the diffrent plant extracts were added to 2 mL of the DPPH solution, allowing the mixture to react for 30 minutes at room temperature. Subsequently, the measure the absorbance was recorded at 517 nm with a spectrophotometer. The percentage reduction was calculated using the formula:

$$\text{Percentage reduction} = \frac{(Ao - Ac)}{Ao} x\ 100$$

where Ao is the initial DPPH absorbance and Ac is the absorbance after reaction with the sample.

The EC_{50} (concentration required to obtain a 50% antioxidant effect), or scavenging activity, indicates the quantity of parsley extract needed to reduce the absorbance of DPPH by 50%. This value can be determined either graphically by plotting absorbance against extract concentration or calculated using the slope of the linear regression.

RESULTS

Essential oil yield and pH

The EO yield and pH of *P. crispum* from each part are presented in Table 2. Seeds exhibited the highest EO yield with 1.8%, whereas parsley roots were completely free of EO. The arial part showed low essential oil content (0.6%) compared to the seeds. For pH, both seeds and areal parts showed a value of 5.

Table 2.

EO yield and pH of parsley different parts under study

Plant parts	pH	Yield (%)
Seeds	5	1.8
Stems and Leaves	5	0.6
Roots	-	0

Phytochemical screening

The results of the qualitative phytochemical analysis are presented in Table 3. Phenols were present in all parsley parts, while tannins, terpenes, and sterols were present in the shoot part and seeds only. None of the anthocyanins or saponins were found in any of the extracts. Moreover, seeds showed deeper coloration for phenols, tannins, terpenes, and sterols compared to the shoot part of the parsley.

Antibacterial activity

All extracts were screened for their antimicrobial properties “in vitro” towards two infective agents isolated from biological material including Gram-(+)ve and Gram-(-)ve bacteria (Table 1). These results were compared with the antimicrobial activity of gentamicin. From all the extracts used in the study only ethanolic extracts of stem + leaves and essential oil of the seeds had an antibacterial effect and hence are presented in Table 4.

Comparing extracts Seeds essential oil exhibited the widest and highest antimicrobial and antifungal activity. The EO stock solution (SS) showed strong inhibition against *S. aureus* (16 mm) and *B. spizizenii* (20 mm), while its diluted forms (D1–D3) maintained moderate activity across several microorganisms, including *S. aureus* (12–13 mm), *B. spizizenii* (12 mm), *E. coli* (12–14 mm), *P. aeruginosa* (10–11 mm), and *C. albicans* (25–29 mm). This indicates a broad-spectrum antimicrobial effect that persists even after dilution. In contrast, the ethanolic extract (EE) from stems and leaves showed limited activity, with inhibition observed only against *S. aureus* (13–14 mm) in the diluted forms, while the stock solution showed no inhibition and no activity was detected against the other tested strains. It is worth noting that the essential oil (EO) extracts exhibited a greater inhibitory effect than gentamicin (GM) against *C. albicans*.

When using 20 µL of the EO stock solution, no antibacterial or antifungal activity was observed against *Ec*, *Pa*, and *Ca*. However, when the volume was reduced to 10 µL, only *Ec* did not exhibit any biological activity. A similar observation was noted for EE against *Sa*.

Table 3.

Result of phytochemical screening of ethanolic extracts of the different parts of Parsley

Phytochemicals	Ethanolic Extracts		
	Roots	Stem + Leaves	Seeds
Phenols Ferric chloride test	**+**	++	+++
Anthocyanins HCl test	-	-	-
Tannins Braymer's test	-	++	+++
Saponins Foam test	-	-	-
Terpenes and sterols Salkowski's test	-	++	+++

+ indicates presence and - indicates absence of phytochemicals.

Table 4.

Antimicrobial activity (inhibition diameter, mm) of the different extracts of *P. crispum* compared to Ampicillin

Extracts	**Dilutions**	**Bacteria and fungal strains**				
		Sa (mm)	**Bs (mm)**	**Ec (mm)**	**Pa (mm)**	**Ca (mm)**
EE (Stem and leaves)	SS	/	/	/	/	/
	D1	13				
	D2	14				
	D3	14				
EO (Seeds)	SS 10 µL	16	13	/	10	27
	SS 20 µL	16	20	/	/	/
	D1	16	12	12	10	29
	D2	12	12	13	11	25
	D3	13	12	14	10	25
GM (Antibiotic)	/	25	22	25	22	25

Area of inhibition including the diameter of the filter paper disk (6 mm); EE: ethanolic extract, EO: essential oil; SS: Stock solution; D1–D3: Dilutions; Sa: *S. aureus*, Bs: *B. spizizenii*, Ec: *E. coli*, Pa: *P. aeruginosa*, Ca: *C. albicans*, GM = gentamicin; / = no inhibition.

Table 5.

Antioxidant activity of parsley different extracts by DPPH

Parsley parts	**Extracts**	**DPPH assay**
		EC_{50} (mg/ml)
Roots	Aqueous	78
	Ethanolic	95
	Hydrosol	99.49
	EO	-
Stem + Leaves	Aqueous	83
	Ethanolic	62
	Hydrosol	99.54
	EO	66
Seeds	Aqueous	70
	Ethanolic	52
	Hydrosol	98.48
	EO	56

Antioxidant activity

Antioxidant activity of the different *P. crispum* extracts and expressed as CE_{50} is showed in Table 5. The results shows that ethanolic extracts generally exhibit the strongest activity, with the lowest EC_{50} values. Specifically, the ethanolic extracts from seeds (52 mg/mL) and stem + leaves (62 mg/mL) demonstrate the most potent antioxidant properties. Aqueous extracts also showed good activity, particularly from seeds (70 mg/mL) and roots (78 mg/mL), while hydrosol extracts tend to be weaker, especially from roots and shoot (99.49 and 99.54 mg/mL, respectively). EO also display moderate antioxidant activity, with the seed EO showing a low EC_{50} value of 56 mg/mL and the shoot EO at 66 mg/mL.

DISCUSSION

Throughout history, people worldwide have relied on plant-based substances, or natural products, to enhance their appearance. Evidence from ancient Egypt, dating back to around 3000 BC, shows the use of cosmetics, and these practices have been integral to daily life in various cultures (Pal and Nayak, 2021).

Essential oils, are a key component of plants, largely used in the production of perfumes, hair care products, and skin moisturizers. Indeed, traditional herbal remedies such as *Emblica officinalis* (Amla), *Acacia concinna* (Shikakai), and *Callicarpa macrophylla* (Priyangu) have long been used for skin and hair care (Pal and Nayak, 2021). The analysis of *P. crispum* essential oil seeds revealed a yield of 1.8% and 0.6% for the areal parts, signifying a noteworthy essence content, however, our results are slightly lower than other research with parsley seeds (Mert and Timur, 2017) (2.25%) and shoot (Mara De Menezes Epifanio et al., 2020) (0.03~3.2%). When compared to several other plants, the *P. crispum* essence content surpassed that of *O. basilicum* (0.37%) largely used in healthcare (Egata, 2021), in contrast, *Thymus vulgaris*, exhibited comparable yields with 1.8% (Gladikosti and Zekovi, 2023). Our results showed an acidic pH for seeds and arial parts, the acidic pH plays a pivotal role in chemical and biochemical reactions and may influence the stabilizing properties of essential oils, including antioxidant and antimicrobial effects (Gladikosti and Zekovi, 2023).

Plants have long been a source of food and medicine, and a wide range of plant-based natural products are now incorporated into cosmetics, including skin care treatments for dryness, eczema, and acne, as well as products with antioxidant, anti-inflammatory, and anti-aging properties. Antibacterial activities were observed to vary across different parsley extracts, namely aqueous, ethanolic, hydrosol, and essential oil from seeds (Pal and Nayak, 2021). This variation can be attributed to the distinct active substances present in each extract, as well as the different plant parts from which they were derived (roots, arial parts, seeds) (Bitwell et al., 2023). Additionally, the pH level of each extract contributes to the differences in antibacterial effects. Despite limited literature on parsley, existing studies, such as those conducted by Sidra et al. (Sarwar et al., 2016) and Linde et al. (Linde et al., 2016) confirm the antibacterial potency of parsley against various bacteria particularly against *E. coli*, *P. aeruginosa*, and *S. aureus*. The experiment's specific observation regarding parsley essential oil's effectiveness against *S. aureus*, *E. coli*, *P. aeruginosa* and *B. spizizenii* suggests potential factors influencing its efficacy, including bacterial evolution, cultivation conditions, and extraction methods (Barra, 2009). *C. albicans* acts on the skin by adhering to epidermal keratinocytes and secreting secreted aspartic proteinases (SAPs) and phospholipases, which compromise the integrity of the stratum corneum and facilitate deep tissue penetration (Naglik *et al.*, 2003), therefore, parsley seeds essential oil seems to be a power agent against *C. albicans* infection, our results match previous results in Abdu and Hauwa, 2019 and Arismunandar *et al.*, 2021. the Stock solution which is more concentrated shows no antibacterial and antifungal activity while less amount and dilutions showed a strong antibacterial and antifungal activity, this pattern is rather explained by the test method than a true loss of antimicrobial power. In agar disk/well diffusion, the result depends on how well the compound diffuses through aqueous agar, and might be non-polar/hydrophobic and diffuse poorly, so a more concentrated stock can give a smaller or even absent inhibition zone while a dilution diffuses better and looks more active (Bonev *et al.*, 2008).

In the context of antioxidant activities, the ethanolic extract of seeds demonstrated the highest potency, followed by the essential oil, ethanolic extract of stems and leaves, and aqueous extract of seeds. These variations in antioxidant power can be attributed to the diverse components present in each part of the plant, as well as the different methods and solutions used for extraction. Essential oil and hydrosol exhibited distinct characteristics, with the latter showing no antioxidant activity, possibly due to greater dilution and lower concentration of active substances compared to other extracts (Mara De Menezes Epifanio et al., 2020).

Hence, when considering Algerian parsley cultivars, it becomes evident that seeds emerge as the optimal choice for realizing health benefits and catering to the cosmetic industry in the formulation of natural products derived from parsley. This recommendation is grounded in the observed high yield of essential components in the seeds, making them particularly advantageous for applications in both health and cosmetics.

CONCLUSION

This research was centered on the investigation of diverse extracts of parsley, encompassing ethanolic extract, aqueous extract, hydrosol, and essential oil, derived from various plant parts, including roots, shoots, and seeds. Notably, the study underscores that the essential oil obtained from parsley seeds outperforms other plant parts in terms of yield. Furthermore, the research elucidates the notable efficacy of extracts from parsley in both antibacterial antifungal and antioxidant activities. The overarching objective is to discern the most promising extracts suitable for the development of a cosmetic product, prioritizing substances with proven safety for skin and cellular health. Among the extracts, the essential oil, ethanolic extract, and hydrosol derived from parsley seeds exhibited active effects against specific bacteria, aligning with the desired antibacterial properties. Additionally, in the realm of antioxidant activity, both the ethanolic extract and essential oil from seeds demonstrated clear and significant effects. Consequently, based on the observed efficacy, the essential oil, ethanolic extract, and hydrosol derived from parsley seeds were singled out for the formulation of a cosmetic product. This strategic selection aims to create a cosmetic product rich in active and safe substances, emphasizing effectiveness for skin health and overall well-being.

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AUTHOR'S CONTRIBUTION

These authors contributed equally to this work.

CONFLICT OF INTERESTS

The authors declare that they have no conflict of interests.

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