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Evaluation of Antioxidant Activity and Total Phenolic Content of Salvia rosmarinus, Heracleum persicum, Lavandula, Citrus aurantium and Crocus sativus Extracts

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ABSTRACT —

The current study compared the antioxidant activity and total phenolic content of extracts of Rosemary (Salvia rosmarinus), Angelica (Heracleum persicum) or Persian Hogweed or Golpar, Lavender (Lavandula), Orange-spring (Citrus aurantium) and Saffron (Crocus sativus). The antioxidant activity of extracts has been evaluated by ABTS method. Total phenolic content was also determined by Folin–Ciocalteu (F–C) reagent using UV/Vis spectrophotometric method in the mentioned plant samples. The highest phenolic content was related to Angelica(Heracleum persicum) with 233.42±2.16 gallic acid equivalent (GAE) mg/mL and the lowest was related to orange-spring (Citrus aurantium). In the same way, the analysis of ABTS assays showed that Angelica and Saffron extracts are more active than other extracts and show higher antioxidant activity ranging from 65.23±0.16 to 75±0.08%. The highest antioxidant activity was related to Angelica with 65.23±0.16 and the lowest was related to Lavender (Lavandula) with 43.75±0.08. In general, Angelica (Heracleum persicum) extract had higher phenolic content and antioxidant activity than other extracts. Therefore, it can be used as a food additive to prevent the rancidity of food products.

Keywords: Medicinal plants, antioxidant activity, Total phenolic content, Angelica, plant extracts

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1. Introduction

Medicinal plants are the best natural sources of antioxidants and polyphenolic compounds. Natural antioxidants have the ability to scavenge free radicals before they start oxidative chain reactions in the cell membrane or lipid-containing parts of the cell. In fact, the timely action of antioxidants in inhibiting the oxidation reactions of radicals is a guarantee of existing health. The use of herbal plants and natural supplements has increased over the past. Medicinal

plants have been popular because of their fewer toxic and side effects, widespread availability, and relatively low costs (Karimlar et al., 2019). Nowadays, synthetic antioxidants such as BHT, BHA, or TBHQ are used in the food industry to delay the oxidation of lipids, but due to the bad nutritional effects and carcinogenicity of these compounds, natural antioxidants, especially polyphenols and carotenoids, that exhibit a wide range of biological effects, including anti-inflammatory, anti-aging, and anticancer has become important (Jiménez-Escrig et al., 2000).

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Rosemary (*Salvia rosmarinus*) is a herb that has been traditionally used as an anti-inflammatory agent, and currently is being studied for anti-cancer properties (Allegra et al., 2020). Rosemary due to its high anti-oxidant and anti-microbial activities has been reported to be an effective food preservative. These properties allow rosemary to prevent microbial growth while decreasing food spoilage through oxidation (Veenstra et al., 2021).

Angelica (*Heracleum persicum*) or Persian Hogweed or Golpar, a flowering plant of the family Apiaceae, naturally grows under humid conditions and has traditionally been used as a medicinal herb and flavoring agent. H. persicum has been extensively used as food additives, food preservatives, and for the treatment purposes of gastrointestinal, neurological, respiratory, urinary, and rheumatologically dysfunctions (Nanakali et al., 2023).

Lavender (Lavandula) is a medicinal plant that has a long history of consumption in Persian medicine. The most prominent medicinal use has been for neurological disorders in a way that it has been called "the brain scavenger (Amin et al., 2019). Bitter orange or Orangspring or (Citrus aurantium) blossoms normally appear on the trees usually in early spring. The blossoms are famous for their "magical sweet fragrance".

Saffron (Crocus sativus) is an expensive spice that is cultivated mainly in Iran and on a smaller scale in countries such as Greece, India and Italy. It is mainly used for flavoring and food coloring, as well as in traditional medicine including appetite suppression, asthma, cancer treatment, pain, menstrual disorder and mental depression (Mohaqiq et al., 2020). This study aimed to compare antioxidant and phenolic compounds of extracts of Rosemary (Salvia rosmarinus), Angelica (Heracleum persicum), Lavender (Lavandula), Orange-spring (Citrus aurantium) and Saffron (Crocus sativus).

2.Material and Methods

2.1. Preparation of extract

All dried samples (Salvia rosmarinus, Heracleum persicum, Lavandula and Citrus aurantium) were extracted by maceration method.

2.2 Determination of the antioxidant activity of the extracts

In this study, the ABTS (2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulfonic) acid) method was used to determine the antioxidant properties of the extracts. ABTS is a widely used compound for determining the radical scavenging activity of plant extracts, food, etc. ABTS solution was prepared in the presence of 2 mg potassium persulfate and 3 mL distilled water and stored at dry and dark place for 24 hours and the absorption was adjusted to 0.7 ± 0.2 at wavelength 734 nm. Samples were added to this solution and it is calculated on the basis of the percentage reduction in absorption of antioxidant properties (Cano et al., 2023).

Radical Scavenging Inhibition $\% = (Abs_{control} - Abs_{sample} / Abs_{control}) \times 100$

2.3 Determination of the total phenolic content

The total phenolic content of the samples was measured using the Folin-Ciocalteu (F-C) reagent. 1.97 mL of water with 1.25 mL of Folin's reagent and 0.025 mL of sample (each extract) mixed and wait for 10 minutes until the reaction is complete, then add 20%

calcium carbonate and keep it in the dark-room for 2 hours. After that, the sample is placed in the UV visible spectrophotometer and the absorbance was read at 760 nm. Gallic acid was used to calculate the standard calibration curve. Fig. 1

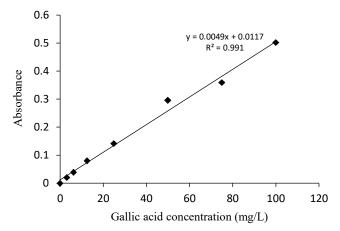


Fig 1. Standard curve between concentration and absorption of gallic acid

2.3. Statistical analysis

In this study, the normal distribution of the data was examined and according to the normality of the data distribution, analysis of variance was used to compare the treatments. Then Duncan's mean comparison test was used to compare the averages at a significance level of $\alpha = 0.05$.

4. Results and Discussion

The standard curve shows the relationship between gallic acid concentration and absorption and is used to determine the phenolic content of all samples *which* are shown in Figure 1, and the results of the evaluation of total phenolic content in extracts are shown in Table 1 and the results of the antioxidant evaluation of the extracts are shown in Table 2. The results showed that Angelica (*Heracleum persicum*) extract contains higher phenolic content than other samples. After that, saffron (*Crocus sativus*) has more phenolic content, and the other samples, lavender (*Lavandula*) and rosemary (*Salvia rosmarinus*), respectively contain less phenolic content. Finally, Orange-spring (*Citrus aurantium*) does not contain any phenolic content.

Table 1. Comparison of the Total Phenolic Content (mg/g)

| Total Phenolic Content |
|-------------------------------|
| 66.08±0.14 ^b |
| 37.09±0.14 ^C |
| 233.42±2.16 ^a |
| 00.00±0.00e |
| 4.95 ± 0.86^{d} |
| |

Dissimilar English letters indicate a significant difference at the level of 0.05

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Table 2: Comparison of the Antioxidant Properties(%)

| Extracts Names | Extract evaluation |
|--------------------|-------------------------|
| Crocus sativus | 60.53±0.25 ^b |
| Lavandula | 43.75±0.08e |
| Heracleum persicum | 65.23±0.16 ^a |
| Citrus aurantium | 50.47±0.16 ^d |
| Salvia rosmarinus | 53.33±0.16° |

Dissimilar English letters indicate a significant difference at the level of 0.05.

The sample containing angelica (*Heracleum persicum*) and saffron (*Crocus sativus*) had the highest antioxidant properties, respectively. The aim of this study was evaluated the antioxidant activity and total phenolic content of aqueous and oil extracts. The results of the current study showed that Angelica (*Heracleum persicum*) extract contains higher phenolic content than other samples and the highest antioxidant properties was found in this herbal.

The phenolic content of 5 extract samples that were evaluated by spectrophotometer is sorted in descending order:

Angelica (*Heracleum persicum*)> saffron(*Crocus sativus*)> lavender> rosemary(*Salvia rosmarinus*)> orange-spring(*Citrus aurantium*). The antioxidant activity of 5 extract samples is sorted in descending order:

Angelica (*Heracleum persicum*) > saffron(*Crocus sativus*)>, rosemary (*Salvia rosmarinus*)> *orange-spring* (*Citrus aurantium*)> lavender.

Heracleum persicum has been known as "Golpar" in Iran and distributed in Alborz regiaon, the northern parts of Iran, commonly known as Persian Hogweed is a flowering plant. The fruits are widely used as spices, preparation of pickle (Asgarpanah et al., 2012). It is a perennial herb that generally grows up to 50–120 cm. The red-brown stems are thick, straight and hollow, and have joints with septa. Its leaves are alternate. The blades are longer than wide, densely haired on lower side, glabrous on top, and pinnate with blunt-toothed margins. The flowers are small, pale white and limegreen, with five petals and five stamens. The fruits are broadly obovate, with slightly ridged schizocarp and anise-like smell (Sayyah et al., 2005). For a long time, H. persicum has been used as a folklore medicine for treatment of various conditions such as stomachs and infections. It is used as flavoring agent, spices digestive, antiseptic, anti-helminthes diuretic and appetizer and pain killer (Hajhashemi et al., 2009). The commonly known phytochemical compounds from *H. persicum* are volatile substances, terpenoids, triterpenes, furanocoumarins, flavonoids and alkaloids, Hexyl butyrate (56.5%), octyl acetate (16.5%), hexyl-2methylbutanoate (5.2%) and hexyl isobutyrate (3.4%) were identified as the major constituents of the H. persicum fruit essential oil (Hemati et al., 2015). Angelica (Heracleum persicum) properties illustrate its potential as a therapeutic agent. Studies show that H. has pharmacological functions including inflammatory, analgesic, anti-convulsant and antibacterial, antifungal and antioxidant activities and Recently, a study has been conducted on the use of *Heracleum persicum* essential oil nanoemulsion as nanocarrier System for the delivery of promising anticancer and antioxidant bioactive agents (Bashlouei et al., 2022). After reviewing the published literature, it was apparent that *Heracleum persicum*(golpar) has a positive effect on the levels of glutathione (GSH), peroxidation lipids (MDA), the total antioxidant capacity of plasma or ferric reducing ability of plasma (FRAP) and glutathione s-transferase (GST), in addition to modulatory effects on liver enzymes including alanine transferase (ALT) and aspartate transferase (AST) (Changxing et al., 2019).

Free radicals play a major role in the pathogenesis of many diseases including cancer, ischemic heart disease, atherosclerosis, Alzheimer's disease, and Parkinson's disease (Aruoma et al., 2003). This study shows that Angelica (*Heracleum persicum*) fruits have the highest antioxidant properties. As previous studies confirm, this property can be attributed to the presence of compounds such as furanocoumarins that isolated from this valuable plant.

5. Conclusion

This current study emphasizes the potential of Angelica (Heracleum persicum) or Persian Hogweed or Golpar to be employed in new therapeutic drugs and provide the basis for future research on the application of transitional medicinal plants and encourages veterinarians to undertake further work to demonstrate the promising beneficial effects of golpar at effective levels to potentially replace the synthetic antibiotic growth promoters in commercial poultry diets.

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