# *Helenium L.* plant flower and leaf growing in Uzbekistan

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**Abstract.** This study investigates the antiradical activity of Inula helenium L. (elecampane) leaves and flowers using the 2.2-diphenyl-1-picrylhydrazyl (DPPH) assay. The HPLC-UV method was employed to monitor the discoloration of the purple DPPH solution, which indicates hydrogen atom donation or electron transfer by antioxidant compounds. Samples were prepared at concentrations of 50, 100, 150, and 200 μL by mixing plant material with 3 mL of DPPH solution. Absorbance was measured at 517 nm wavelength to determine the concentration required for 50% inhibition (IC50) of radical activity. Measurements were recorded after 10 minutes of reaction time across all tested concentrations. Results demonstrated significant antiradical activity in Inula helenium L., with an IC50 value of 645.2 μL. These findings confirm the potent radical-scavenging capacity of elecampane leaves and flowers, suggesting their potential value in preventive healthcare and therapeutic applications for oxidative stress-related disorders.

## 1 Introduction

The World Health Organization estimates that approximately 80% of the global population depends on traditional medicine for primary healthcare, with particular reliance on plant extracts and their bioactive components [1]. Recent research has increasingly focused on the therapeutic potential of medicinal plants, many of which serve as rich sources of natural antioxidants containing phenolic compounds, tocopherols, carotenoids, and ascorbic acid [2].

Inula helenium L. (elecampane) has been widely recognized for its significant medicinal and nutritional value. The plant's flowers and leaves have demonstrated therapeutic potential for various conditions, including peptic ulcers, gastric disorders, hyperlipidemia, cardiovascular diseases, and cancer [3]. Previous studies have primarily examined the antioxidant activity of elecampane sprouts, particularly focusing on sinapic acid esters and flavonoids [4]. However, no research has documented the antiradical properties of Inula helenium L. cultivated in Uzbekistan, nor has its chemical composition been fully characterized - a key objective of this study [5].

Various plant components (flowers, leaves, stems, and roots) hold substantial ecological and economic importance. These organs contain abundant fibers, vitamins, and minerals

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suitable for human consumption, while also serving as sources of natural colorants, pectins (for food gels), and polyphenol antioxidants [6]. Additionally, many medicinal plants provide valuable fodder for livestock [7]. This multifunctionality underscores their critical role in sustainable agriculture, biodiversity conservation, and economic development, while offering natural alternatives for healthcare and nutrition [8-10].

## 2 Methods

The discoloration of the purple 2.2-diphenyl-1-picrylhydrazyl (DPPH) solution serves as an indicator of hydrogen atom donation or electron transfer by antioxidant compounds. DPPH, a stable free radical, functions as a key reagent in spectrophotometric antioxidant assays. This study employed a modified Blois method [8] for evaluating free radical scavenging activity using DPPH spectrophotometric analysis.

A 0.812 mM DPPH solution was prepared in ethanol using a 100 mL volumetric flask. The solution was protected from light by wrapping the flask in aluminum foil and stored at room temperature for 30 minutes prior to analysis. For spectrophotometric measurements:

A blank sample was prepared by adding 3 mL of DPPH solution and 200 µL ethanol to a 4 mL quartz cuvette;

Baseline absorbance ($D_1$) at 517 nm was recorded every 10 seconds for 10 minutes using a YOKE K7000 spectrophotometer (China);

Test samples (50, 100, 150, and 200 µL) were mixed with 3 mL DPPH solution;

Ethanol was added to maintain a constant total volume of 3.2 mL in all cuvettes;

Sample absorbance ($D_2$) at 517 nm was measured following the same protocol.

Radical scavenging activity was calculated using the following equation (1):

$$AA\% = \frac{D_1 - D_2}{D_1} \cdot 100\% \qquad (1)$$

## 3 Results and discussion

The results obtained are presented in Table 1.

**Table 1.** Determination of Antiradical Activity of Inula Helenium L. Plant Flower and Leaf Growing in Uzbekistan.

| Time, sec | $D_1$ (HPLC) | $D_2$ (50 mcl) | AA% | $D_2$ (100 mcl) | AA% | $D_2$ (150 mcl) | AA% | $D_2$ (200 mcl) | AA% |
|---|---|---|---|---|---|---|---|---|---|
| 0 | 0.876 | 0.841 | 4.00% | 0.853 | 2.63% | 0.822 | 6.16% | 0.829 | 5.37% |
| 10 | 0.876 | 0.835 | 4.68% | 0.842 | 3.88% | 0.808 | 7.76% | 0.811 | 7.42% |
| 20 | 0.876 | 0.831 | 5.14% | 0.835 | 4.68% | 0.797 | 9.02% | 0.797 | 9.02% |
| 30 | 0.876 | 0.828 | 5.48% | 0.829 | 5.37% | 0.79 | 9.82% | 0.786 | 10.27% |
| 40 | 0.875 | 0.826 | 5.60% | 0.824 | 5.83% | 0.784 | 10.40% | 0.777 | 11.20% |
| 50 | 0.875 | 0.824 | 5.83% | 0.82 | 6.29% | 0.778 | 11.09% | 0.77 | 12.00% |
| 60 | 0.875 | 0.822 | 6.06% | 0.816 | 6.74% | 0.772 | 11.77% | 0.764 | 12.69% |
| 70 | 0.875 | 0.82 | 6.29% | 0.813 | 7.09% | 0.767 | 12.34% | 0.758 | 13.37% |
| 80 | 0.875 | 0.818 | 6.51% | 0.81 | 7.43% | 0.763 | 12.80% | 0.752 | 14.06% |
| 90 | 0.875 | 0.817 | 6.63% | 0.807 | 7.77% | 0.759 | 13.26% | 0.747 | 14.63% |
| 100 | 0.875 | 0.816 | 6.74% | 0.805 | 8.00% | 0.756 | 13.60% | 0.743 | 15.09% |
| 110 | 0.875 | 0.814 | 6.97% | 0.802 | 8.34% | 0.753 | 13.94% | 0.738 | 15.66% |
| 120 | 0.875 | 0.813 | 7.09% | 0.8 | 8.57% | 0.751 | 14.17% | 0.734 | 16.11% |
| 130 | 0.875 | 0.812 | 7.20% | 0.797 | 8.91% | 0.748 | 14.51% | 0.73 | 16.57% |
| 140 | 0.875 | 0.811 | 7.31% | 0.795 | 9.14% | 0.745 | 14.86% | 0.726 | 17.03% |
| 150 | 0.874 | 0.81 | 7.32% | 0.793 | 9.27% | 0.743 | 14.99% | 0.722 | 17.39% |

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| 160 | 0.874 | 0.809 | 7.44% | 0.792 | 9.38% | 0.741 | 15.22% | 0.718 | 17.85% |
| 170 | 0.874 | 0.808 | 7.55% | 0.79 | 9.61% | 0.737 | 15.68% | 0.715 | 18.19% |
| 180 | 0.873 | 0.807 | 7.56% | 0.788 | 9.74% | 0.735 | 15.81% | 0.712 | 18.44% |
| 190 | 0.874 | 0.806 | 7.78% | 0.786 | 10.07% | 0.732 | 16.25% | 0.709 | 18.88% |
| 200 | 0.874 | 0.805 | 7.89% | 0.784 | 10.30% | 0.73 | 16.48% | 0.705 | 19.34% |
| 210 | 0.874 | 0.804 | 8.01% | 0.783 | 10.41% | 0.728 | 16.70% | 0.701 | 19.79% |
| 220 | 0.874 | 0.803 | 8.12% | 0.781 | 10.64% | 0.727 | 16.82% | 0.698 | 20.14% |
| 230 | 0.874 | 0.802 | 8.24% | 0.779 | 10.87% | 0.725 | 17.05% | 0.694 | 20.59% |
| 240 | 0.874 | 0.801 | 8.35% | 0.778 | 10.98% | 0.723 | 17.28% | 0.688 | 21.28% |
| 250 | 0.874 | 0.801 | 8.35% | 0.777 | 11.10% | 0.721 | 17.51% | 0.684 | 21.74% |
| 260 | 0.874 | 0.8 | 8.47% | 0.775 | 11.33% | 0.719 | 17.73% | 0.682 | 21.97% |
| 270 | 0.874 | 0.799 | 8.58% | 0.773 | 11.56% | 0.717 | 17.96% | 0.68 | 22.20% |
| 280 | 0.873 | 0.798 | 8.59% | 0.771 | 11.68% | 0.715 | 18.10% | 0.677 | 22.45% |
| 290 | 0.874 | 0.798 | 8.70% | 0.77 | 11.90% | 0.713 | 18.42% | 0.675 | 22.77% |
| 300 | 0.873 | 0.797 | 8.71% | 0.768 | 12.03% | 0.711 | 18.56% | 0.673 | 22.91% |
| 310 | 0.873 | 0.796 | 8.82% | 0.766 | 12.26% | 0.71 | 18.67% | 0.672 | 23.02% |
| 320 | 0.873 | 0.796 | 8.82% | 0.764 | 12.49% | 0.708 | 18.90% | 0.671 | 23.14% |
| 330 | 0.873 | 0.795 | 8.93% | 0.762 | 12.71% | 0.706 | 19.13% | 0.67 | 23.25% |
| 340 | 0.873 | 0.794 | 9.05% | 0.761 | 12.83% | 0.705 | 19.24% | 0.669 | 23.37% |
| 350 | 0.873 | 0.794 | 9.05% | 0.76 | 12.94% | 0.703 | 19.47% | 0.667 | 23.60% |
| 360 | 0.873 | 0.793 | 9.16% | 0.759 | 13.06% | 0.701 | 19.70% | 0.665 | 23.83% |
| 370 | 0.873 | 0.792 | 9.28% | 0.758 | 13.17% | 0.7 | 19.82% | 0.663 | 24.05% |
| 380 | 0.873 | 0.792 | 9.28% | 0.757 | 13.29% | 0.698 | 20.05% | 0.662 | 24.17% |
| 390 | 0.874 | 0.791 | 9.50% | 0.756 | 13.50% | 0.697 | 20.25% | 0.66 | 24.49% |
| 400 | 0.874 | 0.791 | 9.50% | 0.756 | 13.50% | 0.696 | 20.37% | 0.658 | 24.71% |
| 410 | 0.873 | 0.79 | 9.51% | 0.755 | 13.52% | 0.695 | 20.39% | 0.656 | 24.86% |
| 420 | 0.873 | 0.79 | 9.51% | 0.754 | 13.63% | 0.693 | 20.62% | 0.654 | 25.09% |
| 430 | 0.873 | 0.79 | 9.51% | 0.754 | 13.63% | 0.692 | 20.73% | 0.651 | 25.43% |
| 440 | 0.873 | 0.789 | 9.62% | 0.753 | 13.75% | 0.69 | 20.96% | 0.649 | 25.66% |
| 450 | 0.874 | 0.788 | 9.84% | 0.752 | 13.96% | 0.689 | 21.17% | 0.648 | 25.86% |
| 460 | 0.874 | 0.788 | 9.84% | 0.752 | 13.96% | 0.688 | 21.28% | 0.646 | 26.09% |
| 470 | 0.873 | 0.788 | 9.74% | 0.751 | 13.97% | 0.687 | 21.31% | 0.645 | 26.12% |
| 480 | 0.873 | 0.787 | 9.85% | 0.75 | 14.09% | 0.686 | 21.42% | 0.643 | 26.35% |
| 490 | 0.873 | 0.787 | 9.85% | 0.749 | 14.20% | 0.684 | 21.65% | 0.642 | 26.46% |
| 500 | 0.873 | 0.786 | 9.97% | 0.748 | 14.32% | 0.683 | 21.76% | 0.64 | 26.69% |
| 510 | 0.873 | 0.786 | 9.97% | 0.747 | 14.43% | 0.682 | 21.88% | 0.638 | 26.92% |
| 520 | 0.873 | 0.786 | 9.97% | 0.746 | 14.55% | 0.681 | 21.99% | 0.637 | 27.03% |
| 530 | 0.873 | 0.785 | 10.08% | 0.745 | 14.66% | 0.68 | 22.11% | 0.635 | 27.26% |
| 540 | 0.873 | 0.785 | 10.08% | 0.745 | 14.66% | 0.679 | 22.22% | 0.634 | 27.38% |
| 550 | 0.873 | 0.784 | 10.19% | 0.744 | 14.78% | 0.678 | 22.34% | 0.632 | 27.61% |
| 560 | 0.873 | 0.784 | 10.19% | 0.743 | 14.89% | 0.677 | 22.45% | 0.631 | 27.72% |
| 570 | 0.873 | 0.784 | 10.19% | 0.742 | 15.01% | 0.676 | 22.57% | 0.629 | 27.95% |
| 580 | 0.873 | 0.783 | 10.31% | 0.742 | 15.01% | 0.674 | 22.79% | 0.628 | 28.06% |
| 590 | 0.873 | 0.783 | 10.31% | 0.741 | 15.12% | 0.674 | 22.79% | 0.626 | 28.29% |
| 600 | 0.873 | 0.783 | 10.31% | 0.741 | 15.12% | 0.673 | 22.91% | 0.625 | 28.41% |

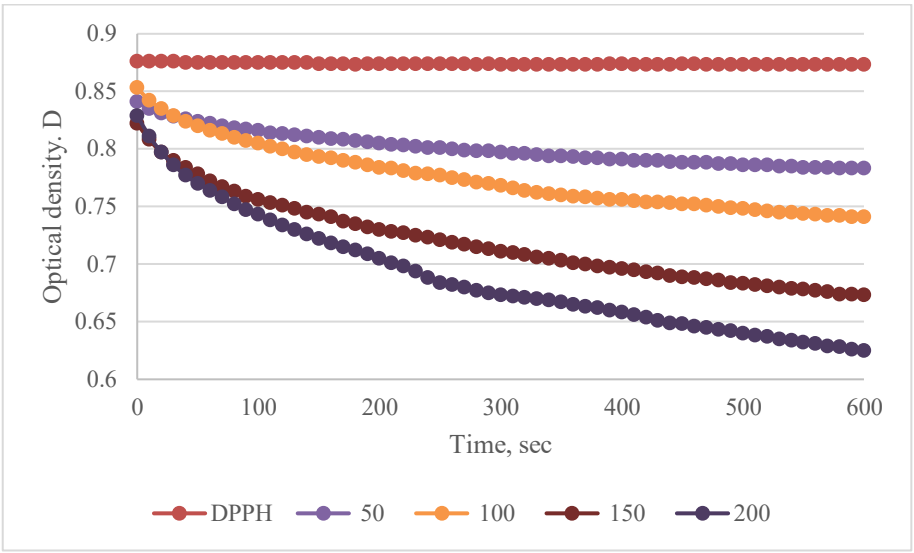


**Fig. 1.** Determination of Antiradical Activity of Inula Helenium L. Plant Flower and Leaf Growing in Uzbekistan, representation of measured light absorption of loose and examined sample solutions added to the HPLC solution.

The half-maximal inhibitory concentration (IC50) was determined by analyzing the relationship between sample concentration and radical scavenging activity. For each experimental trial, a dose-response curve was generated by plotting the 10-minute absorbance measurements ($D_2$) against corresponding antiradical activity values (AA%). The IC50 value, representing the sample concentration required to achieve 50% inhibition of the HPLC solution, was calculated using the linear regression function derived from the trend line of this curve.

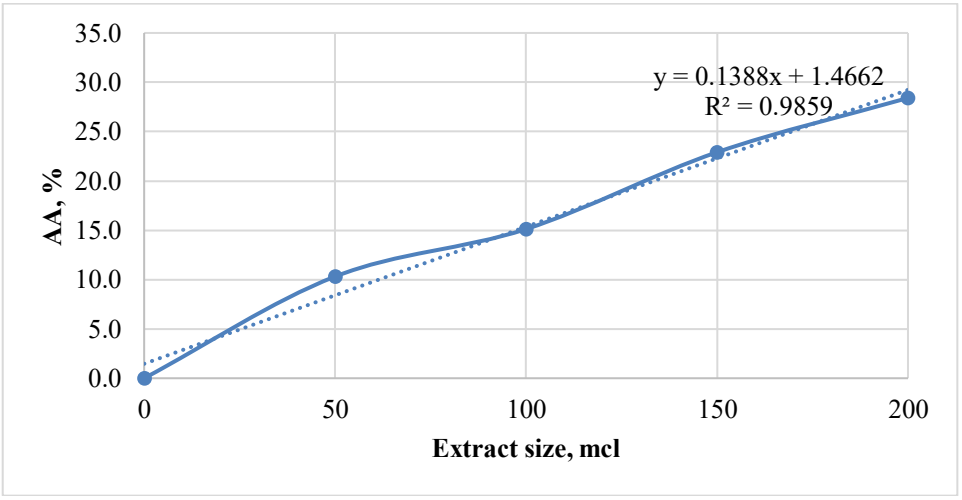


**Fig. 2.** Determination of Antiradical Activity of Inula Helenium L. Plant Flower and Leaf Growing in Uzbekistan on the mesh of AA% s and volumes determined in the 10th minute of the sample.

The trend line transferred to the graph was calculated from the function (2) formula y=mx+b to 50% AA% manifesting volume (IC50) X=(y-b)/M:

$$IC_{50} = \frac{(50-0.6415)}{0.0765} = 645.2\ mcl \qquad (2)$$

The experimental results demonstrate significant antiradical activity in Sample 1, as evidenced by its IC50 value of 645.2 µL. This quantitative measurement indicates the sample's capacity to scavenge free radicals at moderate concentrations, suggesting potential antioxidant properties worthy of further investigation.

## 4 Conclusion

Inula helenium L. (elecampane) has been traditionally consumed for its medicinal properties. Scientific studies indicate that the flowers and leaves of Inula helenium L. contain higher concentrations of bioactive compounds compared to the roots. Given the well-documented radical-scavenging capacity of vitamins and flavonoids, this study evaluated the antiradical activity of OECD-cultivated Inula helenium L. using the 2.2-diphenyl-1-picrylhydrazyl (DPPH) assay.

The analysis determined the concentration required for 50% inhibition (IC50) of DPPH radicals by Inula helenium L. flowers and leaves. Measurements of absorbance at 517 nm were recorded after 10 minutes of reaction time across concentrations of 50–200 µL. The results demonstrate significant antiradical activity, with an IC50 value of 645.2 µL. These findings confirm the potent antioxidant properties of Inula helenium L. flowers and leaves, highlighting their potential for preventive and therapeutic applications in human health.

Further research is warranted to investigate the full spectrum of bioactive compounds in Inula helenium L. and to develop optimized formulations for nutraceutical applications. The preparation of food additives derived from this plant represents a promising area for future study, particularly given its traditional use and demonstrated bioactivity.

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