



THE ANTIOXIDANT STUDIES OF TWO MEDICINAL PLANTS, SPHAERANTHUS INDICUS AND HELIANTHUS ANNUS

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ABSTRACT

Objective: The present study deals with the antioxidant assays of the different plant extracts of two medicinal plants, *Sphaeranthus indicus* and *Helianthus annus*.

Methods: Dried leaves of *S. indicus* and *H. annus* were packed in separate round bottom flasks for sample extraction using ethanol, methanol, hexane, and distilled water as solvents for 72 h, and the extracts were collected after evaporating the solvents. Antioxidant studies of the various extracts were performed by 1-diphenyl-2-picrylhydrazyl and Ferric Reducing Ability of Plasma assays.

Results: The IC₅₀ value of *S. indicus*, *H. annus* and ascorbic acid was 87.25, 188.67, and 18.69 respectively in methanol extracts. In chloroform extracts IC₅₀ value was 940.35 and 460.45 respectively. Among the two plants studied, *S. indicus* and *Helianthus annus* showed better 2-diphenyl-1-picrylhydrazyl (DPPH), scavenging activity than *H.annus* with IC₅₀ values. The FRAP assay results for both the plants indicated that the methanol fractions showed good results when compared with standards, ascorbic acid. These results clearly indicate that *S. indicus* methanol fraction had better antioxidant activity when compared to both standards.

Conclusion: It is concluded that *S. indicus* and *H. annus* have excellent antioxidant activities which could be the major contributing factors for their medicinal roles. Further studies in this direction are being carried on.

Keywords: *Sphaeranthus indicus*, IC₅₀, *Helianthus annus*, 1-diphenyl-2-pi, ascorbic acid.

Introduction: The complementary and alternative medicine, which is also known as traditional medicinal practice, depends mostly of plants and other natural products such as minerals as sources of medicines. Ayurveda and Sidhha forms of medical practices are age-old and time-tested practices. The use of herbs, shrubs, trees, and roots as sources of medicine is a common practice for the folklore. However, the fact remains that these forms of medicines require rigorous standardization to eliminate the ambiguity about their veracity. Tremendous advancements have taken place toward analytical procedures, and these technologies must be used for proving the efficacy of the Ayurvedic and other forms of alternative medicine forms. Some work in this regard is forthcoming, which is a welcome sign [1-7]. This exercise will help in delivering cheap, affordable medicines with the additional advantage of their being less toxic in contrast to the modern-day molecular medicines. The present work is a step in this direction. Two medicinal plants, namely *Sphaeranthus indicus* and *Helianthus annus*, were taken for the present study. These plants are used as folklore medicine for various ailments in India and other countries. There are numerous scientific reports on the medicinal roles of these two plant.

S. indicus is known as Maha Mundi or Mundi in Ayurveda. The medicinal properties such as antiviral, antibacterial, antifungal, neuroprotective, central nervous system depressant, anticonvulsive, fertility enhancing, analgesic, antipyretic, hepatoprotective, antidiabetic, antioxidant, and anticancer are



reported [8-16]. The phytochemical and gas chromatography–mass spectrometry (GC-MS) analysis of various extracts of the leaves of *S. indicus* was reported by Rao and Vijayalakshmi, 2018 [17]

In vitro study, ethanolic extract of *Sphaeranthus indicus* L. (1000gm/ml) appeared maximum scavenging of the radical 1,1 diphenyl,2-picryl hydrazyl (DPPH) (2,2-azinobis-3-ethylebenzothiazoline -6-sulphonate) (ABST), superoxide and nitric oxide radical. The extract also revealed moderate scavenging activity of iron chelation (Shirwaikar *et al.*, 2006). In vitro study, methanolic extracts of *Sphaeranthus indicus* L. exhibited a significant antioxidant effect appearing increasing levels of glutathione peroxidase, catalase and superoxide dismutase and by reducing malondialdehyde levels in rats (Tiwari and Khosa, 2009). The alcoholic extract of flowers of *sphaeranthus indicus* L. is evaluated to have hypotensive, cathartic activities and peripheral vasodilatory (Shrivastava *et al.*, 1971). This plant is also reported to have antiprotozoal activity and anticancer activity against *Entamoeba histolytica* (Dhar *et al.*, 1968). In connection to above findings present investigation deals with the phytochemical screening of *Helianthus annuus* L. and *Sphaeranthus indicus* L. and the pharmacological potential of both the plants of family Asteraceae. At the other hand this work also included the comparative potency of *Helianthus annuus* L. and *Sphaeranthus indicus* L.

Material and Method

Material

All Chemicals were used of analytical grade. These chemicals were procured from Hi-media, Merk, Fisher scientific Mumbai, Sigma-Aldrich USA, SD, fine chemicals Mumbai etc and all glass wares were used of Borosil. All glassware was baked in oven overnight, culture media and buffer solutions were autoclaved at 121⁰ for 20 min

Equipment and instruments

The equipment and instrument for the study included refrigerator, orbital shaking incubator (REMI, CIS-24 BL), UV- Visible Spectrophotometer (Systronic type-2203), IR Spectrometer (Systronic type-7600), Autoclav, Analytical balance, Hot air oven, Thermostate, water bath etc.

Method

DPPH scavenging activity was measured by the spectrophotometer (Olufunmiso *et al.*, 2011). Stock solution (6 mg in 100ml methanol) was prepared such that 1.5ml of it in 1.5ml of methanol gave an initial absorbance. Decrease in the absorbance in presence of sample extract at different concentration (10-100ug/ml) was noted after 15 minutes. 1.5ml of DPPH solution was taken and volume made till 3 ml with methanol, absorbance was taken immediately at 517nm for control reading 1.5ml of DPPH and 1.5ml of the test sample of different concentration were put in a series of volumetric flasks and final volume was adjusted to 3 ml with methanol. Three test samples were taken and each processed similarly. Finally the mean was taken. Final decrease in absorbance was noted of DPPH with the sample at different concentration after 15 minutes at 517nm.

Calculation of percentration = $\frac{\text{Control absorbance} - \text{Test absorbance}}{\text{Control Absorbance}} \times 100$



Results

Results of antioxidant activity using DPPH method

Table No. 1: Percentage Inhibition of ascorbic acid and *Sphaeranthus indicus* extract using DPPH method

| S. No. | Concentration (µg/ml) | Percentage Inhibition | | | |
|------------------------|-----------------------|-----------------------|--------------------|------------------|-----------------|
| | | Ascorbic acid | Chloroform extract | Methanol extract | Aqueous extract |
| 1 | 10 | 30.42 | 1.8 | 1.6 | 2.77 |
| 2 | 20 | 59.11 | 2.58 | 8.83 | 3.96 |
| 3 | 40 | 67.48 | 5.16 | 24.09 | 8.33 |
| 4 | 60 | 75.25 | 7.38 | 28.91 | 16.26 |
| 5 | 80 | 77.58 | 9.59 | 47.38 | 22.61 |
| 6 | 100 | 79.63 | 11.07 | 57.83 | 32.93 |
| IC₅₀ | | 18.69 | 460.45 | 87.25 | 158.44 |

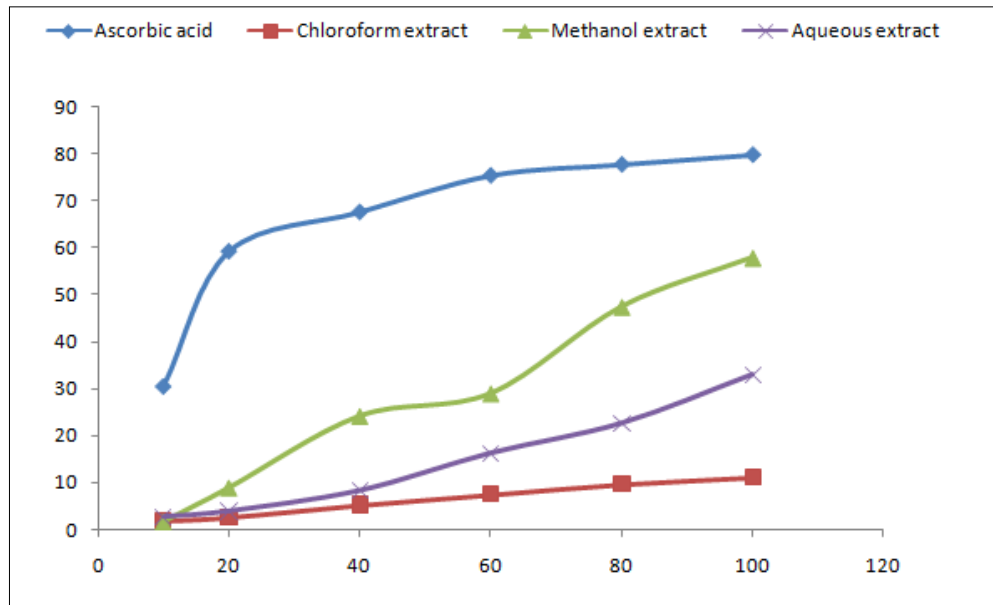


Figure 1: % Inhibition of ascorbic acid and *Sphaeranthus indicus* extracts using DPPH method

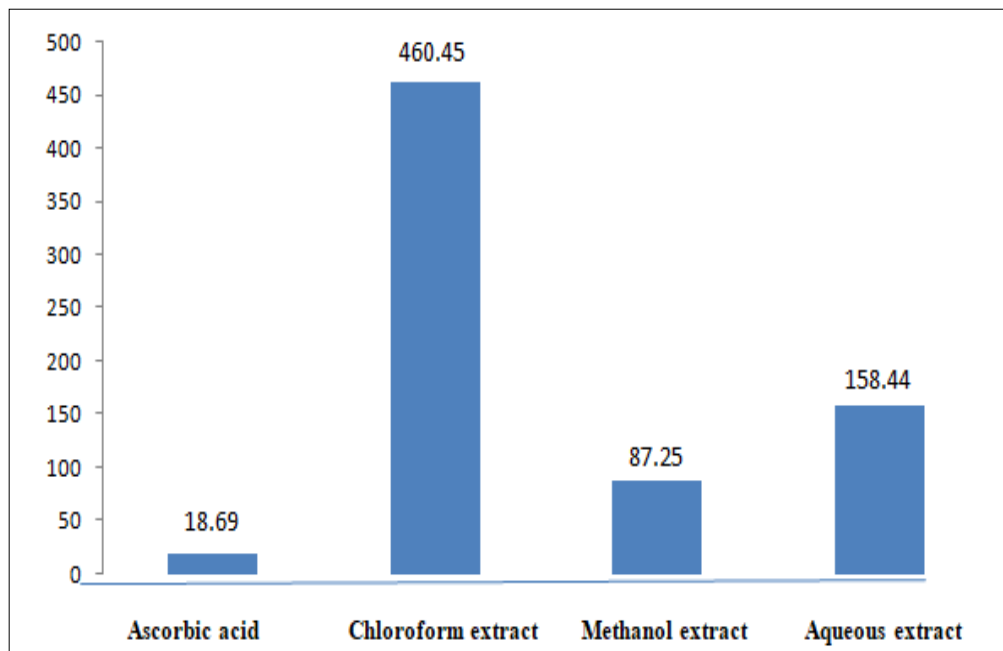


Figure 2: IC₅₀ value of ascorbic acid and *Sphaeranthus indicus* extracts



Table No. 2: % Inhibition of ascorbic acid and *Helianthus annus* extract using DPPH method

| S. No. | Concentration (µg/ml) | Percentage Inhibition | | | |
|------------------------|-----------------------|-----------------------|--------------------|------------------|-----------------|
| | | Ascorbic acid | Chloroform extract | Methanol extract | Aqueous extract |
| 1 | 10 | 30.42 | 0.91 | 8.03 | 1.86 |
| 2 | 20 | 59.11 | 1.21 | 10.24 | 3.11 |
| 3 | 40 | 67.48 | 2.13 | 17.72 | 4.36 |
| 4 | 60 | 75.25 | 3.35 | 18.83 | 7.16 |
| 5 | 80 | 77.58 | 4.26 | 25.76 | 8.72 |
| 6 | 100 | 79.63 | 5.79 | 28.8 | 15.88 |
| IC₅₀ | | 18.69 | 940.35 | 188.67 | 362.21 |

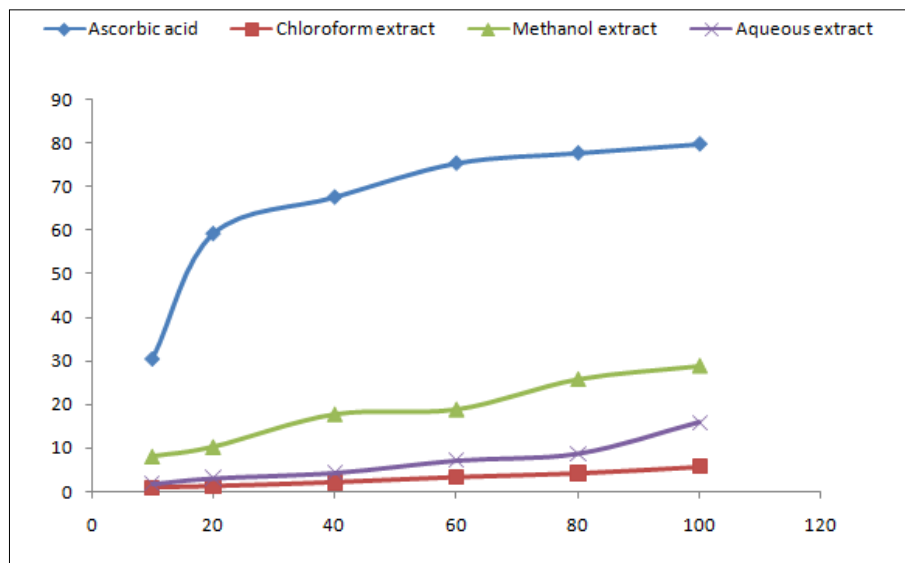


Figure 3: % Inhibition of ascorbic acid and *Helianthus annuus* extract using DPPH method

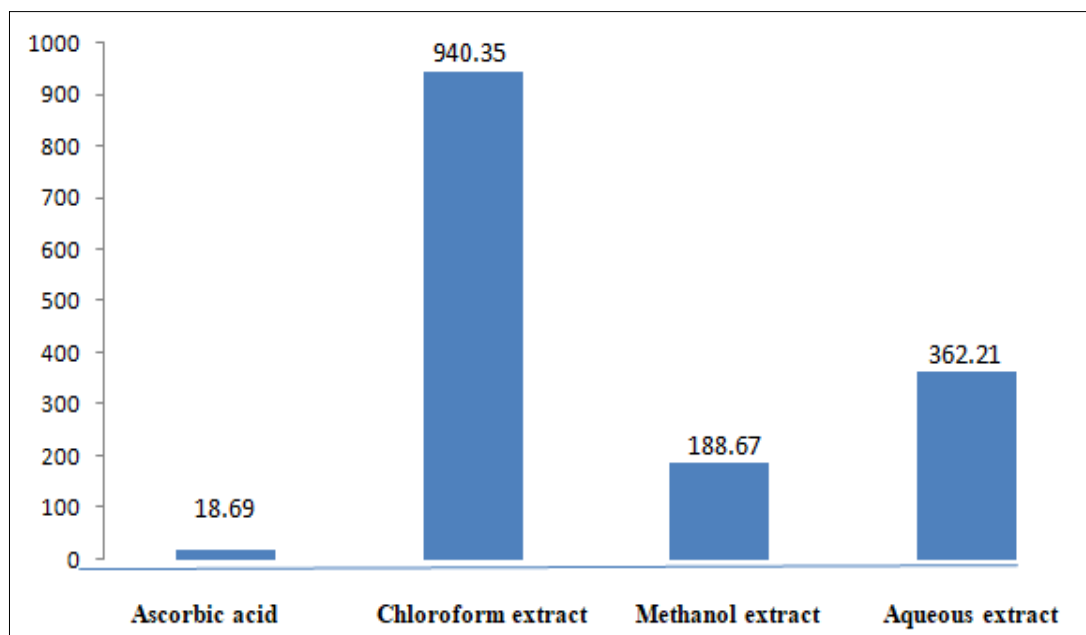


Figure 4: IC₅₀ value of ascorbic acid and *Helianthus annuus* extracts



Discussion:

The study was investigated by DPPH scavenging radical effect method was done with diphenyl picrylhydrazyl solution, it reacts with antioxidant compound then it is converted into purple to light yellow colour. Our study shows when the ascorbic acid and plant extract was taken with varying concentration of different extracts chloroform, Methanol and Aqueous then percentage inhibition is increases with respect to IC₅₀ value of *Sphaeranthus indicus* L. as well as *Helianthus annus* L. In case of *Sphaeranthus indicus* L. it showed maximum zone of inhibition with chloroform extract. Aqueous extract of *Sphaeranthus indicus* L. showed moderate inhibition but very low inhibition showed by methanol extract. The percentage inhibition of *Helianthus annus* L. is more effective comparism to *sphaeranthus indicus* L. So the result indicated that *Helianthus annus* L. is more potential and noticeable effect on scavenging free radicals as well as *Sphaeranthus indicus* L.

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