

Original Article

ANTIOXIDANT AND ANTHELMINTIC POTENTIAL OF THE STEM & LEAVES OF WHITE ABRUS

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ABSTRACT

Introduction: World Health Organization (WHO) appreciates the importance of medicinal plants for public health care in developing nations. White Abrus (*Abrus precatorius* Linn) have important role in traditional medicine, its seeds, bark, leaves and stem are medicinal.

Objective: The potential of the leaves and stem of this plant as an antioxidant and anthelmintic agents were evaluated.

Methods: All the analysis was done according to standard protocols.

Results: The methanolic extract produced significant anthelmintic and antioxidant properties in a dose-dependent manner. DPPH free radical scavenging assay of leaf and stem exhibited IC 50 values of $275 \pm 0.83 \mu\text{g/ml}$ and $170 \pm 0.90 \mu\text{g/ml}$ respectively. At $1000 \mu\text{g/ml}$ concentration both leaf and stem shows maximum radical scavenging activity (93 % & 98 %).

Superoxide anion scavenging assay also resulted promising activity with IC 50 values $85 \pm 0.83 \mu\text{g/ml}$ and $80 \pm 0.90 \mu\text{g/ml}$ respectively for leaf and stem. The percentage scavenging of super oxide radical surged with the enhanced concentration of plant extracts. The maximum scavenging activity of plant extract was at $1000 \mu\text{g/ml}$ concentration, 80 % and 92% respectively in leaf and stem extracts.

Preliminary phytochemical screening revealed the presence of saponins, steroids, flavonoids, terpenoids, cardiac glycosides and phenols in both stem and leaves that may be the reason for its biological properties.

Conclusion: The findings of this study indicate that this plant is medicinal with prominent antioxidant and anthelmintic property.

Keywords: White Abrus, Anthelmintic property, Anti-oxidant property, Phytochemistry, Traditional medicine.

INTRODUCTION

Nature has provided a complete storehouse of remedies to cure all ailments of humankind [1]. Traditional medicine use is a common practice in developed and developing countries at the primary healthcare level [2]. Herbal medicines are prepared from various plant parts like leaves, stem, roots, barks and seeds, which usually contain many bioactive compounds and used primarily for treating mild or chronic ailments. Due to the increasing demand in the field of herbal medicines, it has become necessary and pertinent to probe into the area of systematic knowledge about herbal drugs. There is a need for the application of this knowledge in authentication, detailed study and practical utilization of crude drugs [3].

Abrus precatorius L of Fabaceae family is a vine originally native to India that now commonly found throughout the tropical and subtropical parts of the world [4]. It grows best in dry regions at low elevations. Leaves, roots, stem and seeds used as a medicine in an Indian traditional system for antihelminthic, anti-diarrhoeal, antiemetic and inhibit intestinal motility.

Researchers have reported that seeds used for the treatment of diabetes and chronic nephritis [5]. *A. precatorius* leaves have employed to sweeten foods and certain medicines used for stomach complaints, to treat fevers, cough and cold (used as decoction).

The leaves casually chewed and the vine sometimes sold as a masticatory in Curacao [6, 7]. The plant also traditionally used to treat tetanus, and to prevent rabies. Even though, considerable work did on the seeds for different activities, scientific information on the pharmacological action of leaf and stem of the white variety is still not available or rather scarce. However, the red variety has a broad range of therapeutic effects. Like, antibacterial, antifungal, antitumor, analgesic, anti-inflammatory, antispasmodic, anti-diabetic, antiserotonergic, anti-migraine, including treatment of inflammation, ulcers, wounds, throat scratches and sores [8]. Thus, we screened the potential of the leaf and stem of this plant as an antioxidant and anthelmintic agent.

MATERIALS AND METHODS

Collection and Identification of Plant materials

The plant white Abrus were collected from Mannuthi, Thrissur District of Kerala, India. Taxonomic identification made with Flora of the Presidency of Madras by JS Gamble [9].

Preparation of Extracts

Leaves and stem of the plant were shade dried separately for several days. The dried plant material were ground to a coarse powder and 50g of the powdered plant materials soaked in 95% methanol (1:5) for 72 hours [10]. The solvent removed by rotary evaporation and dried extract was stored in refrigerator for further studies.

Phytochemical screening.

The preliminary Phytochemical analysis of the plant extracts were performed using standard protocol given by Harborne [11].

Anthelmintic property

The standard Albendazole (25mg /ml) and the test solutions of *A. precatorius* (25, 50, 100 mg/ml) were evaluated for anthelmintic activity with Indian adult earthworm *Pheretima posthuma*, the identified worms were collected from Kerala Agricultural University, Mannuthi. Observations made for the time taken for paralysis and death of individual worms up to four hours of test period. Time for paralysis was noted when no movement of any sort could be observed except the worms were shaken vigorously. Time for death of worms was recorded after ascertaining that the worms neither moved when shaken vigorously nor when dipped in warm water at 50°C [12].

Anti-oxidant property screening

DPPH free radical scavenging assay

The free radical scavenging activity of the plant extracts assessed based on the radical scavenging effect of the stable 1, 1-diphenyl-2-picrylhydrazyl (DPPH) - free radical activity by a method given by

Braca et al [13]. The diluted working solutions of the test extracts and 6.34 μ M solutions of DPPH were prepared in methanol, and 100 μ l test, 100 μ l DPPH solution and 800 μ l of methanol were taken in a test tube and mixed well. These solution mixtures were kept in dark for 20 min and optical density was measured at 517 nm using Cecil-Elect spectrophotometer. Methanol (900 μ l) with DPPH solution (6.34 μ M, 100 μ l) taken as control and methanol as blank. The optical density was recorded and % of inhibition was calculated using the formula given below [14].

$$\text{Percentage (\%)} \text{ of inhibition} = A-B/A \times 100$$

Where A = optical density of the control and B = optical density of the sample.

Super oxide anion scavenging activity

In vitro super oxide anion scavenging activity is measured by riboflavin / NBT (Nitro blue tetrazolium) reduction method. Reduction of NBT is the most popular method. This method was based on the generation of super oxide anions by auto oxidation of riboflavin in presence of light. The super oxides reduce NBT to a colored formazon that can be measure at 590 nm. The capacity of extracts to inhibit the color to 50% is measure in terms of IC50 [14].

RESULTS AND DISCUSSION

Preliminary phytochemical screening

The results of preliminary phytochemical screening revealed the presence of number of valuable secondary metabolites; it is given in Table 1.

Anthelmintic property screening

It was seen that the methanolic extract of *A. precatorius* leaf and stem possess dose dependent anthelmintic activity as compared to

a standard drug Albendazole. The mean paralyzing time of *Pheretima posthuma* with the dose of 25mg/ml was found to be 17.6 \pm 2.6 minutes in leaf extract, and 31.16 \pm 1.04 minutes in stem extract. In the meantime, Albendazole at the same concentration causes paralysis in 31.76 minutes.

Table 1: Results of preliminary phytochemical screening

| Phytoconstituent group | Stem | Leaf |
|------------------------|------|------|
| Quinone | + | - |
| Cardiac glycoside | + | + |
| Steroids | + | + |
| Flavonoids | + | + |
| Phenols | + | + |
| Saponins | + | + |
| Terpenoids | + | + |
| Tannin | - | - |
| Alkaloids | - | - |
| Coumarin | - | + |

The mean death time of *Pheretima posthuma* with the same dose was 37.3 \pm 2.08 minutes in leaf extract and 56.43 \pm 1.28 minutes in stem extract. But in case of Albendazole the worms get paralyzed only during the experimental period of 4 hours, no death was observed (Table: 2). Therefore, from these results it is clear that this traditional drug is more effective than the commercially available drug Albendazole.

Phytochemical screening of the leaves and stem of White Abrus revealed the presence of various valuable secondary metabolites, among them phenolic compounds may contribute [15] to the maximum percentage of anthelmintic property.

Table 2: Anthelmintic property of *Abrus precatorius*

| Time taken for paralysis (min) | Distilled water | Albendazole (25mg/ml) | Drug (25 mg/ml) | | Drug(50 mg/ml) | | Drug(100 mg/ml) | |
|--------------------------------|-----------------|-----------------------|-----------------|------------------|----------------|----------------|-----------------|------------------|
| | | | Leaf | Stem | Leaf | Stem | Leaf | Stem |
| - | - | 31.76 \pm 0.90 | 17.6 \pm 2.6 | 31.16 \pm 1.04 | 16.2 \pm 1.8 | 20.9 \pm 1 | 14 \pm 1.9 | 17.16 \pm 0.76 |
| Time taken for death (min) | - | - | 37.3 \pm 2.08 | 56.43 \pm 1.28 | 30.6 \pm 1.5 | 38.8 \pm 1.4 | 26.3 \pm 1.87 | 29.6 \pm 1.4 |

Antioxidant property of leaf and stem

Antioxidant compounds may function as free radical scavengers, initiator of the complexes of pro-oxidant metals, reducing agents and quenchers of singlet oxygen formation [16]. Therefore, the importance of search for natural antioxidants has increased in the recent years so many researchers focused the same [17].

DPPH free radical scavenging activity

DPPH is a stable free radical at room temperature often used to evaluate the antioxidant activity of several natural compounds.

The reduction capacity of DPPH radicals was determined by the decrease in its absorbance at 517 nm, which induced by antioxidants.

The percentage of DPPH radical scavenging activity of the methanolic extract of *Abrus precatorius* presented in Table 3. The methanolic extract of *A. precatorius* leaf and stem exhibited a maximum DPPH scavenging activity of 93 % and 98 % respectively at 1000 μ g/ml concentration. The IC50 of the methanolic extract of leaf and stem was found to be 275 \pm 0.83 μ g/ml and 170 \pm 0.90 μ g/ml respectively.

Table 3: DPPH free radical scavenging property of *Abrus precatorius*

| S. No. | Concentration of plant extract(μ g \cdot ⁻¹) | Percentage of inhibition | | IC 50 | |
|--------|---|--------------------------|---------------|----------------|----------------|
| | | Leaf | Stem | Leaf | Stem |
| 1. | 10 | 20 \pm 2.1 | 19 \pm 1 | 275 \pm 0.83 | 170 \pm 0.90 |
| 2. | 15 | 24 \pm 0.7 | 24 \pm 0.57 | | |
| 3. | 25 | 28 \pm 1.4 | 27 \pm 1.3 | | |
| 4. | 50 | 32 \pm 0.95 | 34 \pm 1.07 | | |
| 5. | 75 | 36 \pm 1.8 | 42 \pm 2.70 | | |
| 6. | 100 | 39 \pm 1.4 | 45 \pm 2.08 | | |
| 7. | 250 | 48 \pm 1.15 | 56 \pm 1.10 | | |
| 8. | 500 | 75 \pm 1 | 73 \pm 1.52 | | |
| 9. | 750 | 83 \pm 0.57 | 94 \pm 0.52 | | |
| 10. | 1000 | 93 \pm 0.64 | 98 \pm 0.57 | | |

Superoxide anion scavenging activity

Superoxide is a highly reactive molecule that reacts with various substances produced through metabolic processes. Superoxide dismutase enzymes present in aerobic and anaerobic organisms catalyses the breakdown of superoxide radical [18]. Percentage scavenging of superoxide anion by the leaf and stem methanolic extracts of *A. precatorius* were examined at different concentrations

(10 -1000 µg/ml) and the result is depicted in table 4. The percentage scavenging of superoxide radical surged with the enhanced concentration of plant extracts.

The maximum scavenging activity of the plant extract was at 1000 µg/ml concentration, 80 % and 92% respectively in leaf and stem extracts. The IC50 value of leaf and stem extracts was recorded as 85 ± 0.83µg/ml and 80± 0.90µg/ml respectively.

Table 4: Super oxide anion scavenging property of *Abrus precatorius*

| S. No. | Concentration of plant extract(µg ^L ⁻¹) | Percentage of inhibition | | IC 50 | |
|--------|--|--------------------------|-----------|-----------|----------|
| | | Leaf | Stem | Leaf | Stem |
| 1. | 10 | 5 ± 2.1 | 16 ± 1 | 85 ± 0.83 | 80± 0.90 |
| 2. | 15 | 19 ± 0.7 | 24 ± 0.57 | | |
| 3. | 25 | 23 ± 1.4 | 28 ± 1.15 | | |
| 4. | 50 | 26 ± 1.32 | 36 ± 0.57 | | |
| 5. | 75 | 36 ± 1.8 | 49 ± 1.7 | | |
| 6. | 100 | 64 ± 1.4 | 59 ± 0.96 | | |
| 7. | 250 | 68 ± 1.4 | 65 ± 1.5 | | |
| 8. | 500 | 71 ± 0.57 | 76 ± 0.70 | | |
| 9. | 750 | 76 ± 1.30 | 83 ± 1.15 | | |
| 10. | 1000 | 80 ± 0.90 | 92 ± 0.57 | | |

The antioxidant assays reveals that both Leave and stem of *A. precatorius* posses prominent anti oxidant property, when comparing their activity (fig 1 & 2) it is clear that stem of *A. precatorius* is more antioxidant than leaf with least IC 50 values in both DPPH and NBT reduction methods (170 & 80 µg/ml).

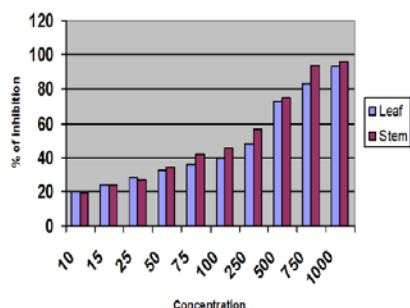


Fig. 1: Comparison of dpph free radical scavenging property of leaf and stem

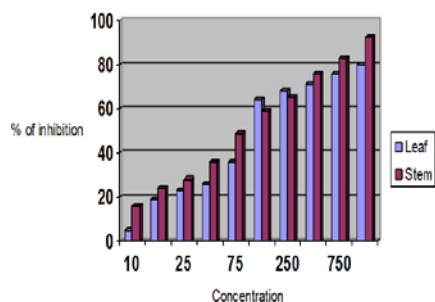


Fig. 2: Comparison of super oxide anion scavenging property of leaf and stem

A perusal of the literature reveals that, the majority of the antioxidant activity is due to the flavones, isoflavones, flavonoids, anthocyanin, coumarins, lignans, catechins and isocatechins [19]. The antioxidant activity of plants might be due to their phenolic compounds [20]. It recognized that, flavonoid shows antioxidant activity and their effects on human nutrition and health was considerable. The mechanism of action of flavonoids is through scavenging or chelating process [21]. Phenolic compounds are a

class of antioxidant agents, which act as free radical terminators [22]. Flavonoids are a group of polyphenolic compounds with known properties, which includes free radical scavenging, inhibition of hydrolytic and oxidative enzymes and anti-inflammatory action [23, 24]. The preliminary phytochemical analysis of the methanol extract of both stem and leaves showed the presence of flavonoids, phenols, cardiac glycosides, steroids, saponins, terpenoids and tannins this may account for the antioxidant potential of the extracts.

CONCLUSION

The present study shows that stem and leaves of white Abrus posses prominent anthelmintic and anti-oxidant properties. Phytochemical studies portray the presence of several biologically active secondary metabolites. Therefore, there is no doubt that this plant is a reservoir of potentially useful chemical compounds, which serve as drugs, provide newer leads and clues for modern drug design.

CONFLICT OF INTERESTS

Declared None

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