Evaluation of antioxidant potential of *Alangium salvifolium* (L.f.) Wangerin

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Abstract: Medicinal plants have been used as a source of therapeutic agent since time immemorial. *Alangium salvifolium* (L.f.) Wangerin (Family: Cornaceae) is an important plant which have been used as traditional medicine to treat several diseases. The present study deals with the evaluation of antioxidant potential of *A. salvifolium*. The total phenolic content and total flavonoid content was found to be 700 µg/ml ± 1.56 GAE and 256 µg/ml ± 1.26 quercetin equivalent, respectively. The IC50 value of the leaves extract against DPPH was found to be 135± 0.78 µg/ml. The Ferric reduction activity (FRAP assay) was evaluated to be 1350 ± 2.12 µM/L. The results suggest that *A. salvifolium* is a good candidate as a natural antioxidant.
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1. Introduction

Medicinal plants are part of our day to day life. They are useful source of nutrition and therapeutic agent (Vats and Kamal, 2013; Vats and Kamal, 2014a; 2014b). The use of plants dates back to advent of human civilization. Ancient Indian manuscripts like *Ayurveda, Charak Samhita, Rigveda* and others are laden with various uses of plants. Traditional systems of healing of almost all the countries reveal the immense importance of plants as medicinal agent.

Alangium salvifolium (L.f.) Wangerinis a relatively under-explored medicinal plant belonging to the family Cornaceae. This plant is used traditionally against several diseases such as cancer, paralysis, diabetes and microbial infection (Tran et al., 2009). Root is used to treat diarrhoea, vomiting and piles (Pandey et al., 2005). External application is suggested during inflammatory response and rheumatism (Anjaria et al., 2002).

The plant contain 1-Methyl-1H-pyrimidine-2,4-dione and 3-*O*-β-d-glucopyranosyl-(24β)-ethylcholesta-5,22,25-triene having antimicrobial activity (Anjumj et al., 2002). Alkaloids like marckine, markidine, psychotrine, cephaeline, lamarckinine etc. (Daniel, 2005) and flavonoids like kaempferol, kaempferol 3-*O*-β-d-glucopyranoside have also been reported (Tran et al., 2009). Other compounds present in *A. salvifolium* includes betulinic acid, betulinaldehyde, betulin, lupeoland β-sitosterol have been identified (Pakrashi et al., 1968).

Methanol extract of root of *A. salvifolium* showed anti-inflammatory and analgesic activity (Porchezhian et al., 2001). Wound healing property (Inayathulla et al., 2010), antifungal activity and antidiabetic activity (Kumar et al., 2011) have been validated through experimental models.

Free radicals are continuously generated in our body as a result of metabolic processes. These nascent molecules are capable of initiating chain reaction which might lead to pathological conditions if not tackled properly. Antioxidants play a major role against these free radicals. It is always beneficial to take antioxidant rich diet for a healthier life. Antioxidants help in minimizing the adverse effect of several diseases including cough, cold, arthritis, Alzheimer etc. The present work was undertaken to explore the antioxidants and antioxidant potential of *A. salvifolium*.

2 Materials and Methods

2.1 Extraction

The test plants were collected locally. The leaves were air-dried and finely powdered. 1 g sample of the experimental plant was extracted in methanol in orbital shaker at 50°C overnight at 110 rpm. The extract was filtered and kept at 4°C for further use.

2.2 Total phenolic content (TPC)

The total phenolics were determined colorimetrically according to the Folin-Ciocalteaeu method (Vats, 2014). The results were expressed as gallic acid equivalents (GAE).

2.3 Total Flavonoid content (TFC)

Total flavonoid content was estimated using the method of Vats et al. (2012) and expressed as quercetin equivalent (Mean ± S.D). Extracts (0.5 mL) were mixed with ethanol (1.5 mL), aluminum chloride (0.1 mL), potassium acetate (0.1 mL) and distilled water (2.8 mL). After incubation at room temperature for 30 min, the absorbance of the reaction mixture was measured at 415 nm.

2.4 DPPH assay

Plant extract (1ml) was mixed with 1ml of 0.3 mM DPPH reagent and allowed to stand at room

temperature for 30 minutes in dark. The absorbance was taken at 517nm. Radical scavenging activity was expressed as IC 50 (Vats, 2012).

2.5 FRAP (Ferric Reduction Antioxidant Potential)

25ml of acetate buffer, 2.5ml TPTZ solution (10mM in 40 mM HCl) and 2.5ml $FeCl_3.6H_2O$ solution (20mM) was mixed to make working solution. 50µl of sample extract was mixed with 1.5ml of FRAP reagent and monitored up to 5 min at 593nm. Absorbance was compared with calibration curve of aqueous solution of known Fe (II) concentration (µM/L) (Vats and Tiwari, 2014).

2.6 Statistical analysis

The experiments were repeated five times and the results were expressed as mean ±standard deviation.

3. Results and Discussions

Phenolic compounds have always been a source of investigation for researchers. They are universally present in plants and have antioxidant activity. Thus, these phytometabolites are being extensively used as dietary supplements. Presence of hydroxyl functional group in the phenolic compounds serves as hydrogen donators. In the present study a linear calibration curve of Gallic acid with coefficient of determination R^2=0.99 was obtained. The total phenolic content of leaves of *A. salvifolium* was found to be 700 µg/ml ± 1.56 GAE. The total flavonoid content as quercetin equivalent was found to be 256 µg/ml ± 1.26. The calibration curve of quercetin had R^2=0.98. Phenolic compounds prevent decomposition of hydroperoxides into free radicals and neutralize lipid free radicals (Li *et al*., 2009).Flavonoids are known to be effective in several diseases including inflammatory responses, cancer, atherosclerosis etc.

The antioxidant assays revealed that the leaves of the experimental plant had potent free radical scavenging activity. The IC50 value, which is the concentration of the plant extract that scavenges 50% of DPPH radical, was found to be 135 ± 0.78µg/ml (Fig. 1). DPPH is deep violet colored solution which turns lighter in color due to the action of antioxidants. The phenolic compounds present in the plant extract donate the H group to DPPH and neutralize it (Vats and Tiwari, 2014). The IC50 value of standard ascorbic acid was 10 µg/ml. FRAP assay is related to reducing potential of plant extract. In this assay ferric ions are reduced to ferrous ions due to the action of antioxidants. The ferric reducing ability was evaluated to be 1350 ± 2.12µM/L. The higher FRAP may be attributed to the phenolic content including flavonoids. These Low molecular weight antioxidants play a significant role in scavenging the free radicals.

4. Conclusion

The results suggest that *A. salvifolium* is a good candidate as a natural antioxidant. The leaves of the plant can be used as dietary supplements after proper validation.

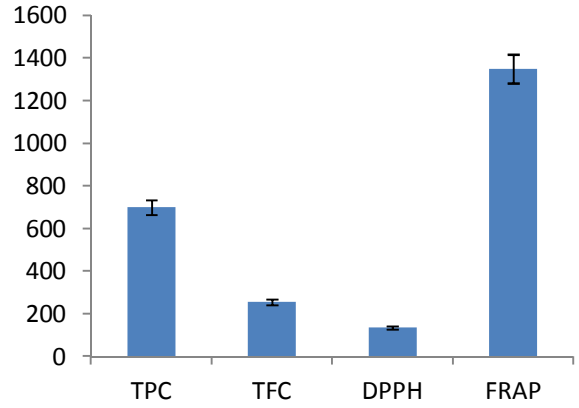

Fig.1: Antioxidant potential of *A. salvifolium* (TPC, TFC, DPPH: µg/ml; FRAP: µM/L)

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