

Study of Antioxidant activity of *Amoora rohitaka* and *Prunus cerasoides* plants

Dr. Prem Singh Meena, Associate Professor
Department of Botany, Government College, Tonk

Abstract

Several in vitro assays were used to determine the antioxidant potential of some plant extracts. The *Amoora rohitaka* and *Prunus cerasoides* extract demonstrated greater antioxidant activity as measured by the scavenging of DPPH, ABTS radical scavenging assay and Ferric ion reducing antioxidant power (FRAP) assay. The highest ABTS scavenging activity (82%) was observed in the ethanol extract of *Amoora rohitaka*, whereas the lowest scavenging activity (65%) was observed in the aqueous extract of *Prunus cerasoides*. In FRAP assay highest activity (95%) was observed in the water extract of *Amoora rohitaka*, whereas the lowest scavenging activity (62%) was observed in the aqueous extract of *Prunus cerasoides*. The highest DPPH activity (89%) was observed in the ethanol extract of *Amoora rohitaka*, whereas the lowest scavenging activity (72%) was observed in the aqueous extract of *Prunus cerasoides*.

Keywords: *Amoora rohitaka*, *Prunus cerasoides*, DPPH, FRAP, ABTS, Extract, Free radical, Antioxidant activity

1. INTRODUCTION

It is well known that oxidative stress is one of the primary initiating agents of a number of chronic and degenerative diseases, such as cancer, atherosclerosis, ischemic heart disease, ageing, and diabetes mellitus, disorders such as immunosuppression and neurological conditions. Numerous plants that are used for flavouring, medicine, spices, and other purposes also contain chemical components that have antioxidant effects. (1) One of the most significant mechanisms for creating free radicals in foods, medications, and even living systems is the oxidative process (2). Antioxidative defence mechanisms are the most efficient way to stop and lessen the effects of free radicals that cause oxidative stress. Antioxidants are chemicals with the ability to prevent free radical chain reactions. Ascorbic acid, carotenoids, and phenolic compounds are among the many naturally occurring antioxidants that are more efficient than others (3). They have been shown to scavenge free radicals and active oxygen species by promoting a reaction cycle, prevent lipid peroxidation (by inactivating lipoxygenase), and chelate heavy metal ions. According to research on medicinal plants and vegetables, plant elements with antioxidant activity are capable of protecting biological systems against oxidative stress (4). As part of our ongoing experimental research on the

antioxidant properties of medicinal plants, we looked at two different plant extracts.

Due to the complexity of the phytochemicals, it is impossible to assess the antioxidant activity of plant extracts using a single technique. Consequently, a method involving numerous assays for assessing the antioxidant capacity of extracts would be more useful and even required. Various in vitro tests can be used to assess the antioxidant capabilities of various plant extracts. Utilizing the DPPH radical scavenging test, ABTS radical scavenging assay, reducing power assay, and ferric reducing antioxidant potential (FRAP) method, antioxidant activity of medicinal plants under research was assessed in the current work.

Only the FRAP assay is capable of measuring antioxidants or reductants in a sample directly. It is based on phenolics' ability to convert blue ferrous complex (Fe (II)-TPTZ) from yellow ferric tripyridyltriazine complex (Fe (III)-TPTZ) in the presence of antioxidants that donate electrons. The results of the FRAP assay were presented as the sum of the concentrations of all electron-donating reductants present in the samples.

The stable free radical DPPH (a, a-diphenyl-β-picrylhydrazyl) can remove the labile hydrogen atom from chemical compounds. It is a deep purple organic nitrogen radical with a long life. This test, which gauges the scavenging impact on DPPH, is a quick and efficient way to find out whether or not raw plant extracts have any free radical-scavenging capacity.

An important predictor of a compound's potential antioxidant activity is its reducing power. According to the test specimen's reducing power, the test solution's yellow hue in this assay turns green. The Fe³⁺/ferricyanide complex is reduced to the ferrous form in the presence of reductants in the solution

2. MATERIALS AND METHODS

2.1. Plant sample collection

In Rajasthan, India, the leaves of *Amoora rohitaka* and *Prunus cerasoides* were harvested in good health and transported to the lab. The leaves were rinsed under running water and allowed to air dry at room temperature. In preparation for future research, the dried plant samples were ground into a powder in a blender and stored in airtight plastic bags.

2.2. Preparation of plant extract

The dry powder was successively extracted in Ethanol, Petroleum ether and Water. 10 g of the dried and powdered plant material was extracted with 150 ml of each ethanol Petroleum ether and Water using a Soxhlet apparatus for 6 to 8 hours at a temperature below the boiling point of the solvents. The obtained crude extracts were concentrated using a rotary evaporator at 40° C while under vacuum, filtered using Whatman No. 1 filter paper, and then kept at 4° C for further use.

2.3 Antioxidant analysis

2.3.1 ABTS radical scavenging assay

The method created by Re et al. (5) was used to test the plant extracts' ability to scavenge ABTS radicals. Initially, 4.9 mmol/L of potassium persulfate and 14 mmol/L of ABTS solution in phosphate-buffered saline (PBS, pH 7.4) were combined to create ABTS radical cations. The mixture was then left undisturbed at room temperature for 12 to 16 hours. Then, 1ml of the plant extract was combined with 4 mL of the ABTS+ working solution. After 6 min, the absorbance of the samples was measured at 734 nm using a spectrophotometer

2.3.2 Ferric ion reducing antioxidant power (FRAP) assay

With a few modest modifications, Benzie and Strain's method of determining the ferric reducing antioxidant power test (6) was used. The plant extract (3 mg diluted in 2 mL 70% ethanol) was mixed with three millilitres of the FRAP reagent, which was made by combining 10 mmol/L TPTZ solution in 40 mmol/L HCl, 300 mmol/L acetate buffer (pH 3.6), and 20 mmol/L FeCl₃ solution in proportions of 1:10:1 (v/v/v). spectrophotometer was used to measure the absorbance at 593 nm after the reaction had been running for 10 minutes at room temperature

2.3.3 DPPH radical scavenging assay

With a small adjustment, DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity was carried out as Gow-Chin and Hui-Yin (7) stated. At various concentrations, one millilitre of freshly made DPPH-methanol solution (0.3 mmol/L) was combined with 3.8 millilitres of methanol and 0.2 millilitres of the plant extract solution (70% ethanol). The samples were then kept at room temperature and kept in the dark. A spectrophotometer was used to measure the absorbance at 517 nm after 10 minutes.

3. RESULTS

The highest ABTS scavenging activity (82%) was observed in the ethanol extract of *Amoora rohitaka*, whereas the lowest scavenging activity (65%) was observed in the aqueous extract of *Prunus cerasoides*. In FRAP assay highest activity (95%) was observed in the water extract of *Amoora rohitaka*, whereas the lowest scavenging activity (62%) was observed in the aqueous extract of *Prunus cerasoides*. The highest DPPH activity (89%) was observed in the ethanol extract of *Amoora rohitaka*, whereas the lowest scavenging activity (72%) was observed in the aqueous extract of *Prunus cerasoides*.

Table :1 Antioxidant activity of medicinal Plants in different extracts

Medicinal plant	ABTS radical scavenging activity %	FRAP assay / mg equi. Fe (III) red. / g dw	DPPH radical scavenging activity / %
<i>Amoora rohitaka</i>			
Ethanol	82	76	89
Petroleum ether	79	85	87
Water	75	95	79
<i>Prunus cerasoides</i>			
Ethanol	78	72	81
Petroleum ether	69	68	78
Water	65	62	72

Graph : 1 Antioxidant activity of medicinal Plants in different extracts

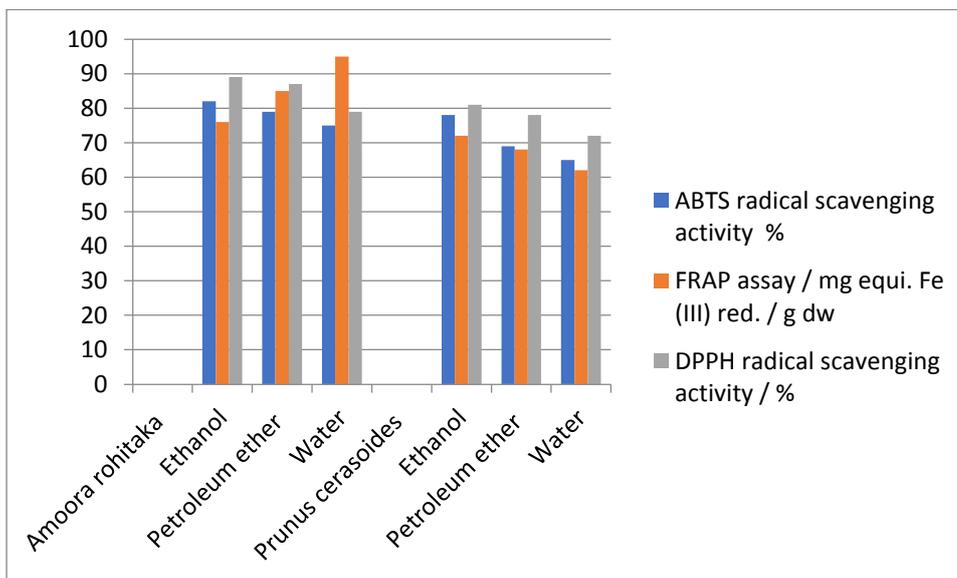


Table : 2 IC₅₀ values for all antioxidant assays (IC₅₀ value / mgml⁻¹)

Medicinal plant	ABTS assay	FRAP assay	DPPH assay
<i>Amoora rohitaka</i>			
Ethanol	0.248	0.621	0.287
Petroleum ether	0.560	0.657	0.512
Water	0.619	0.513	0.545
<i>Prunus cerasoides</i>			
Ethanol	0.342	0.642	0.311
Petroleum ether	0.340	0.667	0.342
Water	0.434	0.689	0.382

4. DISCUSSION

Free radical scavenging abilities were assessed in the current study. Free radicals play a role in a variety of illnesses, including cancer, AIDS, and neurological diseases. Due to their scavenging function, antioxidants are helpful in the treatment of certain disorders. A sensitive approach to assess the antioxidant activity of plant extracts is the DPPH stable free radical assay (8,9). The amount of each extract needed to suppress DPPH activity by 50% is shown in table 2 (IC₅₀). Many plants exhibit antioxidant activities that may be used therapeutically. This is supported by the therapeutic potential of natural medicinal herbs as an antioxidant in lowering such free radical-induced tissue harm(10).

5. Conclusion

According to this study, examined plant components exhibit moderate to considerable antioxidant and free radical scavenging activity. The current study's findings indicate that specific plants can serve as a source of antioxidants for pharmaceutical preparations, and this is amply supported by the current work. It is preferable to put a plant extract to tests that assess a variety of activities such as scavenging reactive oxygen species, inhibiting membrane LPO, and metal ion chelation in order to describe the antioxidant activity of the extract. Nutraceuticals that reduce oxidative stress and hence prevent or delay degenerative illnesses are derived from antioxidant-rich plant extracts. The antioxidant capacities of commercially available herbal extracts have been investigated.

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