ABSTRACT
The antioxidant potential of methanolic extract of seeds of *Abutilon muticum* was evaluated by two model systems like 1, 1-diphenyl-2-picryl hydrazyl (DPPH) scavenging assay and total reducing power assay. The results clearly indicate that methanolic extract of seeds possesses significant antioxidant potential relative to standard ascorbic acid.

Key words: *Abutilon muticum*, Antioxidant potential, DPPH assay, Total reducing power.

INTRODUCTION
*Abutilon muticum* (Family: Malvaceae), nutritionally important plant used in number of Indian herbal medicines. It is locally known as Ashwagandha and is traditional used for analgesic, anthelmintic, hepatoprotective, and hypoglycemic properties. Seeds of the plant are major source of active chemical substances and are traditionally used to cure ulcers, fever, cough, dyspnoea, consumption, dropsy, impotence, rheumatism, toxicosis and leucoderma. Seeds show various pharmacological effects like antiaging, cardio protective, anxiolytic and antidepressant, and antioxidant effect. The seeds of the plant have been used at large extent because of its nutritional contents. It contains calcium, potassium, phosphorus, magnesium, iron and aluminum in the amount 320, 312.32, 235.02, 183.23, 4.97 and 4.13 mg/100g, respectively. In addition, seeds are rich in neutral lipids (94.7%) and polar lipids (3.6%).

In living cells, reactive oxygen species (ROS) are continuously produced in numerous processes such as mitochondrial respiration, metabolism of xenobiotics by cytochrome P450, inflammation, phagocytosis. ROS damage cellular macromolecules (lipids, proteins, nucleic acids) leading to oxidative stress. It has been demonstrated that oxidative stress is involved in many diseases such as cardiovascular diseases, rheumatoid arthritis, neurodegenerative diseases, alcoholic and non-alcoholic steatohepatitis, diabetes mellitus and cancer.

Literature survey revealed that very little work on its seeds and no antioxidant, no phytochemical work had so far been carried out on other parts of this plant species. The screening of herbal extracts and their components by the 1, 1-diphenyl-2-picryl hydrazyl (DPPH) scavenging assay and reducing power assay has become a routine parameter for testing their antioxidant efficacy. In the present study, the antioxidant activities of methanolic extract of *A. muticum* was evaluated in terms of activity of scavenging DPPH radicals and ferrous reducing power.

MATERIALS AND METHODS
Collection of materials
Seeds of *Abutilon muticum* were purchased from local market and was authenticated from official agencies. Solvent methanol was procured from Merck’s Chemicals Ltd. The seeds were dried in
shade, crushed to coarse powder and used for further studies.

**Extraction of materials**

Powdered material of dried seeds of *A. muticum* (500 gm) was subjected to continuous hot extraction with methanol for 48 h in a soxhlet apparatus. The *A. muticum* methanol extract (AME) was filtered and the solvent was evaporated under reduced pressure and dried in a vacuum desiccator. The dried *A. muticum* extract (AME) thus obtained was used for the evaluation of antioxidant activity.

**Evaluation of antioxidant potential**

(1) **DPPH scavenging assay**

DPPH scavenging ability of extract was determined by the reported method. About 1 mL of different concentration (20, 40, 60, 80 and 100 μg/μL of methanol) of extract was taken in different test tubes. To the tubes, 5 mL of DPPH solution was added, shaken and immediately kept in dark at 27°C for 20 min. After incubation, the absorbance of solution was measured at 517 nm. Control solution was prepared and zero was set using methanol. The percent DPPH scavenging effects of sample solution was calculated from following formula:

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\text{%DPPH scavenging effect} = \frac{Ac - As}{Ac} \times 100
\]

Where, \(Ac\) is the absorbance of control solution, and \(As\) is the absorbance of sample solution.

(2) **Reducing power assay**

The reducing capability of extract was measured by transformation of Fe3+ to Fe2+ as per the method reported. The different concentration (20, 40, 60, 80 and 100 μg/μL of methanol) of extract was mixed with phosphate buffer (2.5 mL, 0.2 M, pH 6.6) and potassium ferricyanide (2.5 mL, 1%). The mixture was incubated at 50°C for 20 min by adding 10% trichloroacetic acid (2.5 mL, 1%). The mixture was then centrifuged at 6000 rpm for 10 min. The upper supernatant layer of solution (2.5 mL) was mixed with distilled water (2.5 mL) and FeCl₃ (0.5 mL, 0.1%), and absorbance was measured at 700 nm. The increase absorbance of the reaction mixture indicates increased reducing power.

**RESULTS AND DISCUSSION**

DPPH is a stable nitrogen-centered free radical. When DPPH is reduced by either the process of hydrogen or electron donation its color changes to yellow from violet. Substances to perform this above reaction can be considered as antioxidants and therefore radical scavengers. The DPPH radical scavenging activity was known to correlate well with the inhibitory capacity of lipid peroxidation of a test compound.

The DPPH scavenging activity of methanolic extract of seeds of *A. muticum* was compared with standard ascorbic acid. The methanolic extract of *A. muticum* showed significant activity on scavenging DPPH radical, which implicates an essential defence against the free radicals. The methanolic extract of seeds of *A. muticum* and ascorbic acid was found to decreases the concentration of DPPH significantly from the concentration range of 20 μg/μL to 100 μg/μL. The IC\(_{50}\) value of the seeds extract and ascorbic acid was 48.18 and 38.73, respectively. The free radical scavenging activity was decreased with decrease in concentration (Fig 1).

![Fig 1: DPPH scavenging potential of *A. muticum* extract (AME)](image-url)

In conclusion, the methanolic extract of *A. muticum* shows the significant antioxidant activity and thus, it can protect cells against oxidative stress.
Fig 2: Total reducing power of A. muticum extract (AME)

REFERENCES