ORIGINAL ARTICLE

Quality assessment of *Achyranthas aspera* Linn. seed

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Achyranthas aspera Linn. (Amaranthaceae) seeds were studied to determine the various parameters for pharmacognostical standards. The whole plant of *A. aspera* is reported to have good medicinal values in traditional systems of medicine. The present paper deals with pharmacognostical examination of morphological and microscopical characters and physic-chemical investigations of seeds including determination of loss on drying, ash values, extractives values, foreign organic matter, crude fiber content, and hemolytic activity. The preliminary phytochemical screening, elemental analysis, microbial contamination of powdered drug, were also carried out. High performance thin layer chromatography profile developed for the seeds was to aid in identification of the drug and also in isolating and identifying the biomarker compound responsible for the bioactivity.

Key words: *Achyranthas aspera*, amaranthaceae, apamarg, elemental analysis

INTRODUCTION

Achyranthas aspera Linn. (Amaranthaceae) commonly known as 'Apamarg' is a stiff, erect, weed found throughout India up to 900 m.[1] *Achyranthas aspera* is an important medicinal plant, used in Ayurveda, Unani, Siddha, and in folk medicine for treating several ailments including renal dropsy, bronchial affections, urinary tract infections, and leprosy.[2] Literature survey has demonstrated the presence of two triterpene oleanolic acid saponins as major phyto-constituents in seeds.[3] The plant possesses various medicinal properties; hence, the present study was undertaken to standardize the seeds of *A. aspera* by evaluating various quality control parameters according to WHO guidelines.[4]

MATERIALS AND METHODS

Plant Material

The seeds were collected from the campus of Guru Jambheshwar University of Science and Technology, Hisar. The plants were taxonomically identified and authenticated by Dr. H.B. Singh, Head, Raw Materials Herbarium and Museum Division of National Institute of Science Communication and Information Resources. The voucher specimen has been deposited in the herbarium section of the Pharmacognosy Division, Department of Pharmaceutical Sciences, Guru Jambheshwar University of Science and Technology, Hisar under ref number Pg-10-01. The fresh seeds were taken for macroscopic and microscopic studies. The dried seeds were ground to a coarse powder and subjected to physico-chemical parameters.

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Pharmacognostical Evaluation

Various morphological features viz. shape, size, colour, odour, taste, and fracture were studied.

Microscopic studies were done using the method described by O Brien *et al*.[5] Photomicrographs were obtained by observing free-hand sections of drug under compound trinocular microscope (Zeiss Primostar). Powder study was done according to Evans.[6]

Standardization Parameters

Various standardization parameters viz. loss on drying, ash values, extractives values, foreign organic matter, crude fiber content, swelling index, hemolytic activity, foaming index, microbial contamination were determined according to procedure mentioned in WHO guidelines.[4] Preliminary phytochemical analysis was carried out using standard conventional protocol.[7] The seeds were analyzed for the presence of nine elements by using Atomic Absorption Spectroscopy.[6] The HPTLC studies were performed for ethanolic extract on pre-coated silica gel GF_{254} plates using the suitable solvent system. R_f values, peak area, peak height, and spectrum of each peak were determined

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for the extract.[8] Glycyrrizinic acid (Sigma Aldrich) was used as a biomarker.

RESULTS

Macroscopic Characters

The seeds are brown in color; size: 2.5 mm long; sub-cylindrical in shape, truncate at the apex, round at the base; taste: Mucilaginous; odour: Characteristic.

Microscopic Characters

The transverse and longitudinal sections of seed show a single layered testa, endosperm having polygonal endospermic cells without intercellular space and embryo [Figure 1].

Powder Microscopy

Microscopic examination of powder showed the presence of endospermic cells and pollen grains and stomata.

Physicochemical Parameters

The physicochemical parameters total ash, acid-insoluble ash, water-soluble ash, and sulfated ash values were found to be 7.5, 3.5, 1.25, and 8.6% w/w, respectively. The alcoholic and water soluble extractive values by hot and cold methods were found to be 2.5 and 7.25% w/w; 1.5 and 2.5% w/w, respectively. The foreign organic matter was less than 2%. The percentage moisture content was found to be 3.0% w/w. The crude fiber content was 5.0% w/w, swelling index was 1.5, and foaming index was found to be less than 100. The hemolytic activity was found to be 2.0 units/g.

Preliminary Phytochemical Screening

The preliminary phytochemical screening of the alcoholic and water extract indicated the presence of alkaloids, steroids, saponins, glycosides, phenols, and flavonoids. Carbohydrates, proteins, and tannins were completely absent in both extracts while fixed oils were absent in water extract.

Elemental Analysis

The Atomic Absorption Spectroscopy study showed the presence of cadmium, lead, zinc, copper, nickel, sodium, magnesium, iron, cobalt, manganese, and mercury in aqueous and alcoholic extracts of seed but below the WHO permissible limits and therefore are safe to use.

Microbial Contamination

Determination of microbial contamination of alcoholic and aqueous extracts of seeds showed complete absence of *Escherichia coli*, *Salmonella typhi*, *Psuedomonas aerginosa*, *Staphylococcus aureus*, *Clostridia*, *Shigella*.

HPTLC Studies

HPTLC profile was developed for alcoholic extract of seeds as a preliminary fingerprinting of the extract. Toluene:ethylacetate (5:15) was found to be a suitable solvent system for the separation of constituents of seed extract. At 254 nm, alcoholic extract showed 8 bands at R_f values of 0.07, 0.16, 0.19, 0.61, 0.81, 0.82, 0.92, and 1.00 indicating the presence of eight phytoconstituents [Figure 2]. The band at R_f 0.80 is corresponding to standard, i.e., Glycyrrhizinic acid (R_f 0.81) indicative of presence of triterpene oleanolic acid saponins.

CONCLUSION

An important information on microscopic, physicochemical parameters, preliminary phytochemical analysis, and elemental analysis of the seeds is provided in the present paper. Pharmacognostical parameters and phytochemical screening can serve as a basis for proper identification, collection, and investigation of the plant. HPTLC profile helps in standardization and identification the bioactive compounds. Pharmacognostical anatomical work on the seeds of *Achyranthas aspera* Linn. is scanty. The outcome of the above study on the seeds of *Achyranthas aspera* Linn. might be useful in determining the authenticity of the drug.

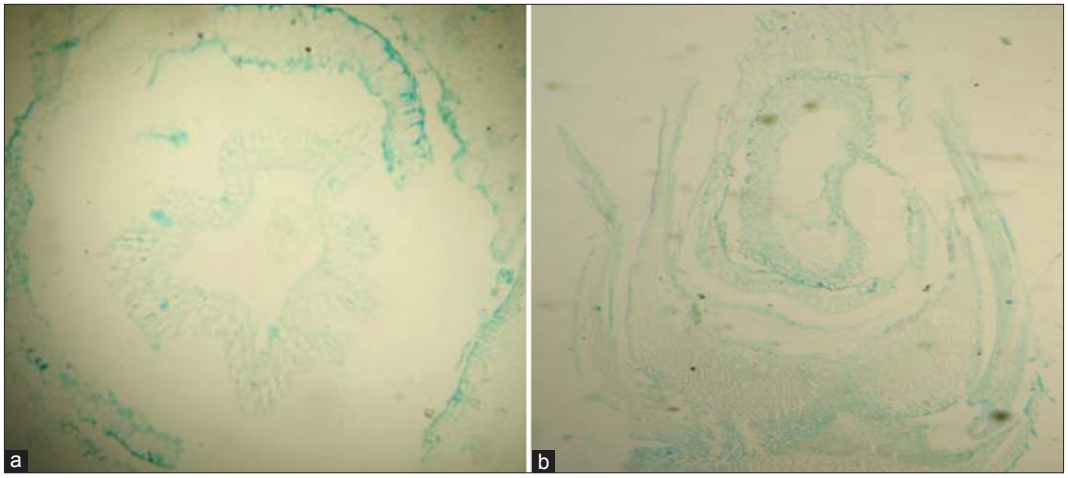

Figure 1: (a) T.S of *Achyranthas aspera* seed (b) L.S of *Achyranthas aspera* seed

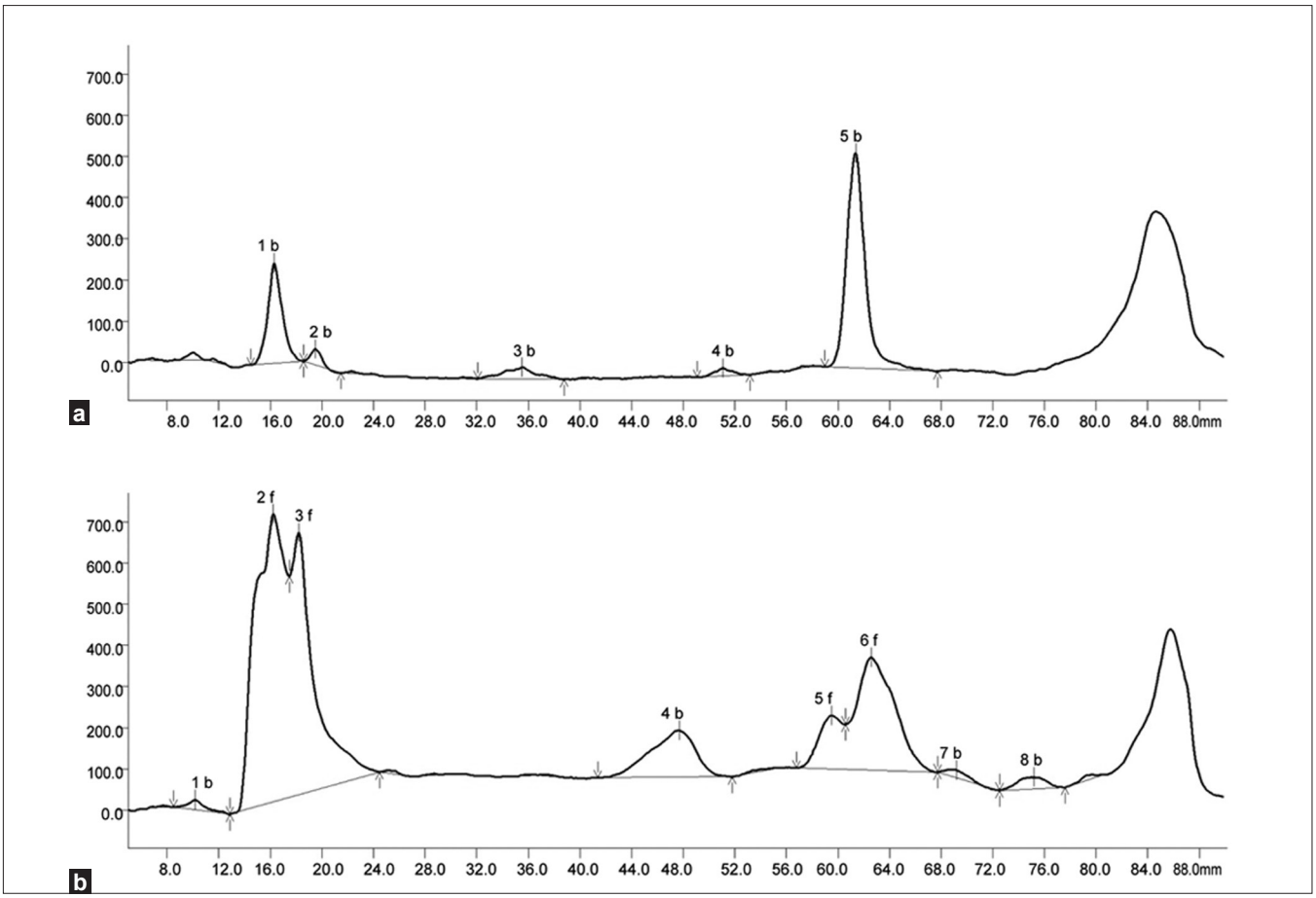

Figure 2: (a) HPTLC of glycyrrizinic acid and (b) ethanol extract of *Achyranthas aspera* seed

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