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Extraction and characterization of tannins from *Cassia tora* Linn leaves

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ABSTRACT

Cassia tora Linn is a medicinal plant used as laxative for the treatment of leprosy and various skin disorders, belongs to family *Caesalpin : aceae* used in Ayurved and Siddha system. It is common weed of rainy season and even blooms in hot arid environment of drought prone area of Baramati. It is rich in tannins and other biomolecules which may help in its medicinal potential. The objective of this study is to extract tannins from normal and oven dried method and to characterize the extracted tannins using UV and FT-IR methods. *Cassia tora* Linn grow as weed in waste places and thus tannin extraction and its characterization may further be utilized for its bioprospeting potential.

Introduction

Cassia tora Linn. is annual foetid herb which grow in low lying coastal areas, river banks, abundant in waste places and other uncultivated fields. It is most commonly known as 'Sickle pod' due to Sickle shape of pods. Kirtikar and Basu (1975) reported that the leaves of *C. tora* are reported to have antirheumatic activity in folklore practice and decoction of the leaves is used as laxative. The extract of *C. tora* leaves has been found to possess significant hepatoprotective activity and anti-inflammatory activity (Maitya,1998)

Das et al., (2011) proposed that various phytochemicals present in this plant such as anthraquinone glycosides, naphthopyrone glycosides, phenolic compounds, flavonoids, sennosides, rubrofusarinetriglucoside, etc. present in the plant significantly contribute to the diverse biological functions such as antioxidant, antibacterial, antifertility, antitumor, antiinflamatory, antifungal activities. *Cassia tora* leaves are used as antifungal agent due to presence of chrysophanic acid –9-anthrone (Acharya et al.,1975). Leaves and seeds are used in the treatment of ringworm and itch (Hooker,1979). Sennosides, which are well known for their medicinal importance, have been detected in the leaves of the plants(Lohar,1975).

Besides of its biological evaluation tannin extraction and its characterization from leaves of *Cassia tora Linn* is worth to study for its industrial exploitation. In the present investigation, two

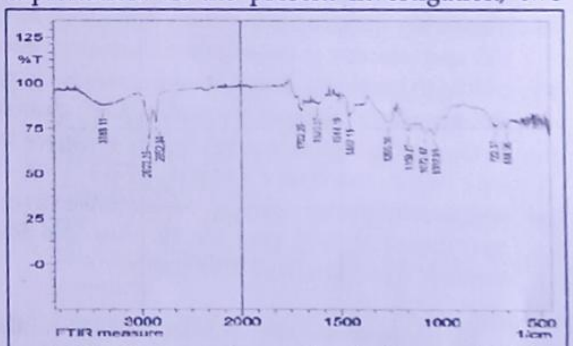

Fig. 1: FTIR spectrum of extracted tannins from normal dried leaf powder of *Cassia tora Linn.*

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methods were used to dry the leaves and comparative study on characterization of extracted tannins was carried out.

Materials and Methods

The fresh plant of *Cassia tora* Linn. was collected from Baramati area during the month of July-August 2019 and identified by the taxonomist of Post Graduate Research Centre, Department of Botany, Tuljaram Chaturchand College, Baramati using floras. The leaves were cleaned and dried by natural sun drying and oven dried method. It was grinded to make fine powder filtered through musclin cloth and stored in a vacuum desicator for further studies.

Extraction of Tannins

For extraction of tannins *Cassia tora* L. normal and oven dried leaf powder of 10gms was macerated in 100 ml of acetone for 24 hours; The supernatant was separated from the residue by filtration using Whatman no.1 filter paper, the fraction concentrated, dried to a constant weight in a vacuum oven at 45°C and the residues obtained stored in refrigerator. The extract obtained was used for further analysis of UV and FT-IR spectroscopy.

UV analysis

Normal and oven dried powder of *Cassia tora* L. leaves were scanned by UV-visible spectrophotometer at the wavelength of 200 - 800 nm on Perkin – Elmer Lambda 25 spectrophotometer. It is basically done for monitoring the extract as UV – VIS spectroscopy is used for the characterization of colloidal particles. Nobel metal particles possess strong surface plasmon resonance (SPR) absorption in the visible region and are highly sensitive to the surface modification. For UV-VIS spectrophotometer analysis, the acetone extract was centrifuged at 3000 rpm for 10 mins and filtered through Whatman No. 1 filter paper.

FT-IR Spectroscopy: Fourier transform infrared measurement of normal and oven dried powder of *Cassia tora*L. leaves was performed on the instrument (Shimatzu). A few mg of sample wasplaced in the sample holder and scanned the sample between range 4000-500 cm-1 by ATR technique was used to identify the characteristic functional groups in the extract. It provides information about the structure of molecule and obtained from its absorption spectrum.

Results and Discussion

Cassia tora Linn is rich in tannins.Mainly there are three types of tannin present in plants hydrolysable, condensed tannins and phlorotannins plays an important role in defense mechanism against microbial infection. Their molecular weight ranges between 500-3000 Da. Three solvents are commonly used to extract tannin from plant samples: boiling aqueous methanol, aqueous acetone, or acidic methanol. Boiling aqueous methanol is thought to be the most effective solvent for condensed tannin (Bate- Smith, 1975). Aqueous acetone is routinely used to extract hydrolyzable tannins (Foo and Porter, 1980) thus in present investigation 80 % acetone is used for extraction of tannins from leaves of *Cassia tora* Linn. A great interest has been emerged in the protective role of tannins against free radicals and ROS generation helps in anticancer studies (Singh and Kumar, 2019)

Hagerman (1988) concluded that aqueous acetone appears to be the best solvent for extracting tannin from leaf tissue. She also observed that the amount of tannin extracted from fresh leaves of Burr oak,

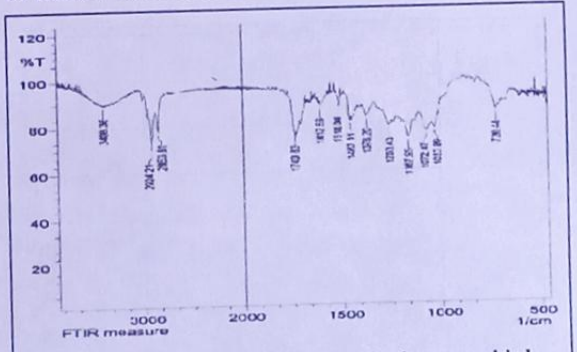

Fig. 2: FTIR spectrum of extracted tannins from oven dried leaf powder of *Cassia tora Linn.*

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Quercus macrocarpa; sugar maple, *Acer saccharum; shagbark hickory, Carya ovata* collected late in the season was greater than from fresh leaves collected early in the season. The extraction of tannin from the dried leaves was quite efficient for some samples and less efficient for other samples.

Acetone is an effective solvent because it inhibits interaction between tannin and proteins thus prevents tannin from binding to leaf proteins during homogenization (Hagerman and Robbins, 1987). Acetone does not interfere with most chemical assays for tannin (Price et al.,1978).

UV analysis: The qualitative UV-Vis profile of extracted tannins from normal and oven driedleaf powder of *Cassia tora* Linn. was taken at 200 to 800 nm. The peaks are at 210.5nm with max absorption of 4.00 in oven dried sample and in normal dried sample max absorption at 3.68and 3.42 with max absorption at 214 and 210.5nm. It is very near to standard tannic acid as similar peaks were shown at 214, 271, 235, 323 nm in dried leaf powder of *C. tora* Linn.

Comparative UV analysis indicate that normal drying method for extraction of tannins is quite suitable for extraction of tannins as having same chromophores or functional groups as that of standard tannic acid but oven dried sample is with only one peak.

FT-IR analysis: FTIR measurement of normal and oven dried powder of *Cassia tora*L. leaves is depicted in figure 1 and 2. It is clear from figures that strong absorption around 3388.00and 3408 cm-1 assigned to NH stretching, 2923.25 and 2924.21 cm-1.due to C-H stretching and 2852.84 and 2853.81 cm-1assigned for O-H stretching in both samples whereas 1702.25 cm-1 and

1740.83 cm-1 from Normal and 1740.83 cm-1 from oven dried leaf sample is due to C = O stretching. C= C stretching was observed at 1620.27and 1612.56 cm-1 while 1514.19 and 1518.04 cm-1 peaks assigned for N-H stretch in normal and oven dried leaf sample.

Interestingly in oven dried leaf powder additional peak at 1462.00 cm-1 for C-H bending,1318.20 cm-1for OH bending,1263.43 for C-O stretch whereas the peaks at 1167.95,1072.47,1033.89 cm-1assigned to C-N stretch which may be amines and C=C bond stretching at 720.44 cm-1. It showed that both the samples have same vibrational frequencies.

Conclusion

Global market for tannins is increasing day by day. As compared to synthetic chemicals natural source of tannins can be an alternative green material in new emerging industries for sustainable environment. *Cassia tora* Linn in grows as weed in many parts of India and abroad unexploited for its utilization as source of tannin. Present studies will be helpful for higher yield of tannins from *Cassia tora* Linn leaves using normal drying method with accuracy with which a chemical component in a biological matrix remain altered in its biological activity and structure. Further microbial studies will throw more light on understanding medicinal potential of tannins.

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