Year: 2014; Volume: 1; Issue: 1
Article ID: BT14 06; Pages: 1-6

BioMed Research

The Open Access Publisher

www.bmrjournals.com

BMR Biotechnology

Research Article

Analysis of Phytochemical and High Performance Liquid Chromatography in *Desmostachya bipinnata*

N. Packialakshmi* and Alwin
PG and Research Department of Microbiology, Jamal Mohamed College (Autonomous) Tiruchirappalli-620020, Tamil nadu, India.
Correspondence should be addressed to N. Packialakshmi.

Received 29 April 2014; Accepted 27 May 2014; Published 27 May 2014

Abstract

Desmostachya bipinnata is popularly known as holy grass. It shows antibacterial effect against gram negative and gram positive pathogens. In present investigation preliminary phytochemical screening and HPLC studies of *Desmostachya bipinnata* were analysed. Preliminary Phytochemical analysis showed that carbohydrates, phytosterols, saponins, Tanins , phenolic compounds, flavonoids, lignin were present. The HPLC result shows that a high peak of retention time minutes 2.5 significant 2.641 was found.

Keywords : *Desmostachya bipinnata*, HPLC, Phytochemical analysis.

Introduction

Medicinal plants constitute an important component of flora and are widely distributed in India. The pharmacological evaluation of substances from plants is an established method for the identification of lead compounds which can leads to the development of novel and safe medicinal agents. Traditional systems of medicine continue to be widely practised on my accounts. Population rise, inadequate supply of drugs, prohibitive cost of treatments, side effects of several allopathic drugs and development of resistance to currently used drugs for infectious diseases have led to increased emphasis on the use of plant materials as a source of medicines for a wide variety of human ailments [1].

The herbal plant *Desmostachya bipinnata* belonging to the family poaceae is commonly known as sacrificial grass, as it is being used in yagnas and religious rites [2]. It has several synonyms like *Briza bipinnata* L., *Eragrostis bipinnata* L., *Eragrostis cynosuriodes* (Retz), commonly known in English by names Halfa grass, a old perennial grass. *Desmostachya bipinnata* is native to north east, west tropical, Northern Africa and countries in the Middle east, temperate and tropical Asia including India. According to religious *Desmostachya bipinnata* has long been used in various traditions as a sacred plant. The plant was mentioned in the Rig veda for use in sacred ceremonies and also seat for priest. In arid regions *Desmostachya bipinnata* has been used as a fodder for domesticated live stock. In

agricultural, *Desmostachya bipinnata* is a weed commonly found in wheat crops. The pharmacological studies on plant of *Desmostachya bipinnata* estabilished its antiulcerogenic, analgesic, antipyretic and anti-inflammatory activities [3]. Five main flavanoid glucosides were isolated from ethanolic extract of *Desmostachya bipinnata* and two of the isolated compounds (trycin and trycin-7-glucoside) show very promising antiulcerogenic activity, furthermore, another flavanoid compound 4 methoxy quercetin-7-0-glucoside isolated from the whole plant of *Desmostachya bipinnata* showed good in vitro antihelicobacter activity [4].

Material and Methods

Collection of plant material

The leaves of *Desmostachya bipinnata* were collected from the bank of river Cauvery Tiruchirappalli district at the month of May 2013. The best harvesting or grazing time of *Desmostachya bipinnata* is in May, when it reaches peak vegetative growth, it contains high nutritive value in the middle of May, [5]. Based on the [6]. investigation and the results from literature, it was found that that *Desmostachya bipinnata* is regularly consumed by the African population and this herb contain adequate amounts of protein and do not exhibit any toxic properties. The collected leaf parts were dried it by air without directly shown on the sunlight for 7-10 days.

Extraction Procedure

The leaves were blunder it powder and homogeneously grained by using mortar and pestle.

Preparation of Column

A plug of cotton is placed on the bottom of the column so that silica and soil won't fall out. Slurry of silica was prepared and poured into the column carefully. It is allowed to settle and sand is added.

The sample was added using pasture's pipette carefully above the sand. The eluent is added on top of the sand, the mobile phase slowly flows down through the silica gel column by gravity leaving behind zone of colour and a compound was eluted from the column.

Preparation of Sensitivity Discs

Six millimetre (6mm) diameter whatman No.1 filter paper discs were punched out with a paper puncher and sterilized in an oven at 120˚C for 30 minutes.

Phytochemical Analysis

The compounds that are responsible for therapeutic effect are usually the secondary metabolites. The preliminary phytochemical analysis was carried out by following procedure Sterols, Flavonoids, Alkaloids, Tannins, Phenol, Lignin, Fixed oil and fats, gum and mucilage.

HPLC Analysis

High performance liquid chromatography (HPLC) is now one of the most powerful tools in analytical chemistry, with the ability to separate, identify and quantitate the compounds that are present in any sample that can be dissolved in a liquid. In Isocratic HPLC the analyse is forced through the column of the stationary phase (usually a tube packed with small round particles with a certain surface chemistry) by pumping a liquid (mobile phase) at high pressure through the column. The sample to be analysed is introduced in a small volume to the stream of mobile phase and is restarted by specific chemical or physical interactions with stationary phase as it traverses the length of the column. The time at which a specific analyse elutes (comes out of the end of the column) is called the retention time and is considered a reasonably unique identifying characteristics of a given analyse. Often a series of tests are performed on the analyse and a number of generic runs may be processed in order to find the optimum HPLC method for the analyse the method which gives the best separation of peaks.

Procedure

An isocratic HPLC with 240 wavelengths and a reverse phase Luna 5 mc 18 column was used. The HPLC system equipped with software class VP series version 6.1(shimadzu). The mobile phase components acetonitrile : water (1:3) were filtered through 0.2m membrane filter before use and pumped from the solvent reservoir to the column at a flow rate 1ml/min which yielded a column pack pressure of 16-165 Kgf/cm². The column was maintained at 27˚C. 20ml of sample was injected using Rheodyne syringe. The graph obtained was used for further studies.

Result and Discussion

The present study shows, at 64 µg concentration the zone of inhibition of the crude extract of *Desmostachya bipinnata* against the *Escherichia coli* is 17mm and the Zone of inhibition of compound obtained through column chromatography against the

Escherichia coli is 20mm. The Zone of inhibition of the crude extract of *Desmostachya bipinnata* against the *Klebsiella sp* is 15mm and the zone of inhibition of compound obtained through column chromatography against the *Klebsiella* sp is 17mm. The zone of inhibition of the crude extract of *Desmostachya bipinnata* against the *Staphylococcus aureus* is 16mm and the zone of inhibition of compound obtained through column chromatography against the *Staphylococcus aureus* is 18mm. (Table 1 and Table 2) [7].

The HPLC result were analysed from St. Joseph's College, Tiruchirappalli District. The qualitative HPLC *Desmostachya bipinnata* profile was detected at a wave length of 230nm due to sharpness of the peaks and proper baseline and recorded its retention time (Rt min). The HPLC study of the isolated compound using methanol as a solvent showed the value of retention time of 2.641 and the area of 95722 and of height 3046 (Figure 1).

Preliminary phytochemical analysis of *Desmostachya bipinnata* compounds shows various types of chemical compounds which provide the base for the occurrence medicinally active constituents like Alkaloids, Carbohydrates, Phytosterols, Saponins, Tannins and Phenolic compounds, Flavonoids, lignin, Protein and Free amino acids as shown in the (Table 3). Therefore compounds generated from these experiments have provided the chemical basis for the wide use of this plant as therapeutic agent for treating various diseases.

Table 1. Zone of inhibition formed by crude extract of *Desmostachya bipinnata* plant powder against bacteria

S.No.	Anti bacterial agent	Name of the bacteria	µg	Zone of inhibition		$X^2= (0-E)^2/E$
				Standard value	Observed crude	
1.	*Desmostachya bipinnata*	*E-Coli*	64	21	17	0.7619
2.		*Klebsiella sp*	64	21	15	1.7142
3.		*Staphylococcus aureus sp*	64	21	16	1.1904

Table value X^2 (0.05) = 3.841

Chi Square value significance at 5% level.

Table 2. Zone of inhibition formed by compound extract of *Desmostachya bipinnata* plant powder against bacteria

S.No.	Anti bacterial agent	Name of the bacteria	µg	Zone of inhibition		$X^2=(0-E)^2/E$
				Standard value	Observed compound	
1.	*Desmostachya bipinnata*	*E-coli*	64	21	20	0.0476
2.		*Staphylococcus aureus sp.*	64	21	18	0.428
3.		*Klebsiella sp.*	64	21	17	0.7619

Table value X^2 (0.05) = 3.841

Chi Square value significance at 5% level.

Figure 1. Hplc Analysis Report

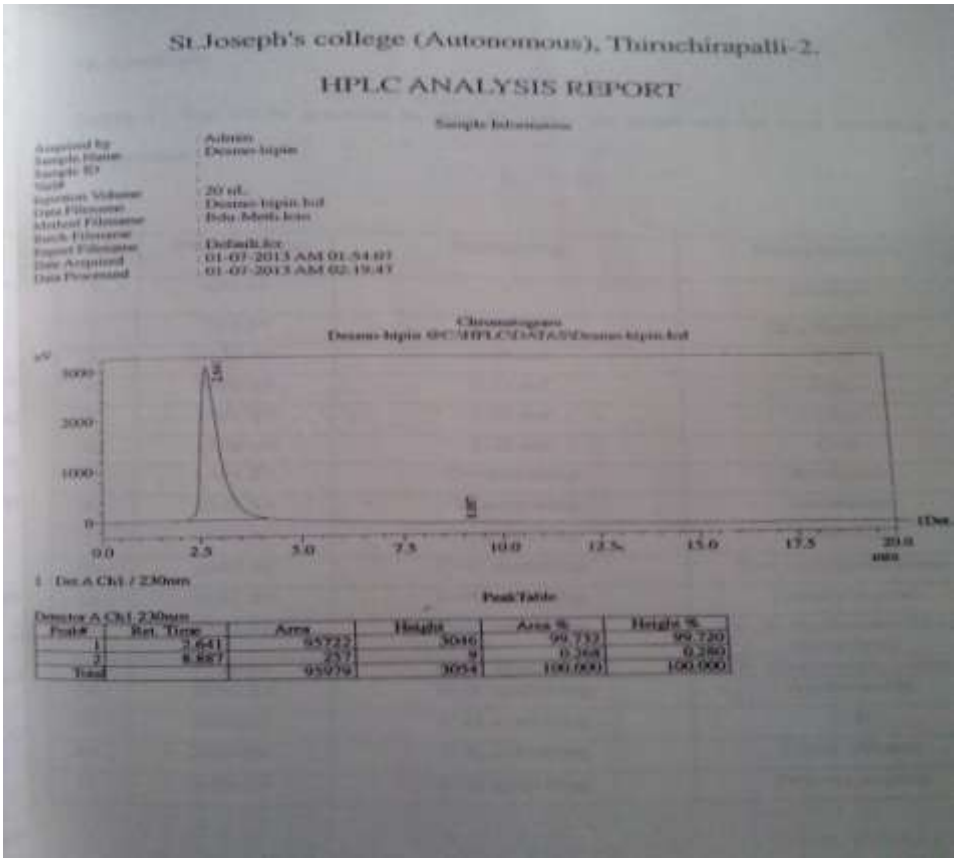

Table 3. Phytochemical screening of *Desmostachya bipinnata*.

S.No.	Phytoconstituents	Results
1.	Alkaloids	positive
2.	Carbohydrates	positive
3.	Glycosides	Negative
4.	Phytosterols	Positive
5.	Saponins	Positive
6.	Fixed oils & Fats	Negative
7.	Tannins & Phenolic compounds	Positive
8.	Protein & Free amino acids	Positive
9.	Gums & mucilage	Negative
10.	Flavonoids	Positive
11.	Lignin	Positive
12.	Volatile oil	negative

Conclusion

The present investigation concluded that the isolated compounds from the plant *Desmostachya bipinnata* are pure and the plant *Desmostachya bipinnata* shows the various antibacterial effects against different bacteria and found that different phytochemical compunds. Further study is needed for the isolation of the constituents present in the plant and its individual pharmacological activity should need to consider and ultimately it should be implemented for the benefit to human beings.

References

1 Joshi, S.G. 2003 *Medicinal plants*. New Delhi, India: Oxford and IBH publishing Co. Pvt. Ltd., 318.

2 Prajapati, N.D., Purohit, S.S., Sharma A.K. and Kumar, T. 2003**.** Handbooks of medicinal plants Agarobios, India, pp. 127**.**

3. Amani, S.A., Nawal, H.M., Derek, J.M. and Gamal, A.S. 2008. *Anti-ulcerogenic Activity of Extract and Some Isolated Flavonoids from Desmostachya bipinnata* (L.) Stapf. Res. Nat. Prod.3: 76 - 82.

4. Mohamed A. Ramadan. and Safwat, N.A. 2009 *Antihelicobacter activity of a flavonoid compound isolated from Desmostachya bipinnata*. Austr J Basic and Applied science. 3(3) : 2270-7.

5. Gulzar S., Khan, M.A. and Liu, X. 2007. Seed germination strategies of *Desmostachya bipinnata* a fodder crop for saline soils. *Rangeland Ecology and Management* 60**:** 401- 407.

6. Golla, U., Kumar, A.K. and Solomon Suder Raj, B. 2011**.** Assessment of antioxidant and hypnotic activity of Unani formulation Arq gulab. *Pharmacologyonline***.** 1: 930-941.

7. Panda, S., Choudhury, N.S.K., Patro, V.J., Pradhan, D.K. and Jana, G.K. 2009**.** analgesic,antipyretic, and anti-inflammatory effect of the whole plant extract of *Desmostachya bipinnata* Stapf (Poaceae) in albino rats. *Drug Invention Today* **1**: 150-153.