WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 5.990
Volume 4, Issue 11, 836-842. Research Article ISSN 2277– 7105

PHYTOCHEMICAL SCREENING AND ANTIMICROBIAL ACTIVITY OF *SENNA OCCIDENTALIIS*

Rachamallu Ranganath Reddy[1*], P.Laxmi[2], G. Karthik Reddy[2], G. Venkateshwarlu[2], H. Ramana[2], Dr. Ch. Shashikala[3]

[1]Rayalaseema University, Kurnool, AP

[2]Venkateshwara Institute of Pharmaceutical Sciences, Cherlapally, Nalgonda-508001, Telangana, India.

[3]IST, JNT University, Kukatpally Hyderaba.

Article Received on
24 Aug 2015,

Revised on 17 Sep 2015,
Accepted on 11 Oct 2015

***Correspondence for Author**
Rachamallu Ranganath Reddy
Rayalaseema University, Kurnool, AP.

ABSTRACT

Phytochemical screening and antimicrobial activity of methanol and ethanol crude extract of *Senna occidentalis (L).*leaves were studied in this work. The preliminary studies of various extracts revealed the presence of carbohydrates, tannins alkaloids, proteins, flavonoids, steroid and triterpenoids. The antimicrobial screening was carried out using the *E.Coli*. *Senna occidentalis* (L) leaf extract have interesting pharmacological activity compounds with great antimicrobial effect and as such could be used in ethno medicine for treatment of some infections and ailments.

KEYWORDS: Phytochemical, antimicrobial, *senna occidentalis*, organism, ethno medicine.

1. INTRODUCTION

Plants are important source of drugs, especially in tradition medicine. It is a common practice in Nigeria and other parts of the world to use plant in the form of crude extracts, decoction or tincture to treat common infectious and chronic conditions.[1]

According to WHO, over 75% of the world population rely on medicinal plants for primary health care and there are reports from various researches on natural substances of plant origin which are biologically active, with desirable antimicrobial property.[2] Despite tremendous progress in the human medicine, infectious diseases caused by bacteria, fungus, virus and parasites are still a major threat to public health. There impact is particularly large in

developing countries due to relative unavailability of medicines & the emergence of wide spread drug resistance.

During the last two decades the development of drug resistance as well as appearance of undesirable side effects of antibiotics has lead to search for new antimicrobial agents mainly among plant extracts with the goal to discover new chemical structures.[3]

Senna occidentalis (L.) a small shrub about 3 ft. high belong to Leguminaesiea family. It is native to the tropical regions of America and naturalized in Australia, eastern Africa, southern and eastern USA. Plants belonging to the family have been extensively investigated because of their rich medicinal anti-inflammatory,[4] anti-carcinogenic,[5] anti-mutagenic,[6] anti-plasmodia,[7] anti-rheumatic,[8] and hepato protective,[9,10] and economic uses. It is part of our continuing effort and mandate to investigate Nigeria medicinal floras, since studies on this plant showed that the nature and amount of the phytochemicals varies according to the season, geographical location and because much work has not been done on this particular genus. This paper reports on the phytoconstituents, antimicrobial activities of *Senna occidentalis*.

2. MATERIALS

The medicinal plant part used for the experiment was leaves of *senna occidentalis*. Ethanol, Methanol, distilled water, chemical reagents and some glassware used from our laboratory.

3. METHODOLOGY

3.1 Collection of plant

Fresh leaves of *Senna occidentalis* (L.) were collected from botanical garden. The leaves were chopped into pieces using knife and then air dried under shade for 10 days and grounded into mesh size and kept in a non-absorptive nylon for subsequent use.

3.2 Solvent Extracts

Dried and milled leaf materials were extracted successively with Soxhlet extractor at temperature of 55 ^{0}C. Each of the solvent; methanol and ethanol were allowed to remain in contact with the plant material for 12 hours; the extracts were evaporated to dryness.

3.3. Preparation of standard culture inoculums of test organism

Three or four isolated colonies were inoculated in the 2ml nutrient broth.

3.4. Separation of active compounds from *senna occidentalis* extracts suspension preparative thin layer chromatography (TLC).

Preparation of chromaplate

The glass plates were cleaned and dried in hot air oven. Slurry was prepared by mixing silica gel (silica gel for thin layer chromatography incorporating 13% calcium sulphate as binder) with double distilled water in clean beaker with continuous stirring. One larger drop of slurry was placed on the slide and by using another clean slide edge the drop of slurry was scattered all over the slide to make thin film and left as such for some time. This procedure is applied for the preparation of all chromo plates with microscope slide, the plates were activated by heating them in the hot air oven at 1200c for 30 minutes.

Loading of sample

The plate was allowed to cool at room temperature and marked about 2cm from the bottom as the origin. The working suspension was loaded at the centre of the slide about 2cm above from the edge.

Development of chromatogram

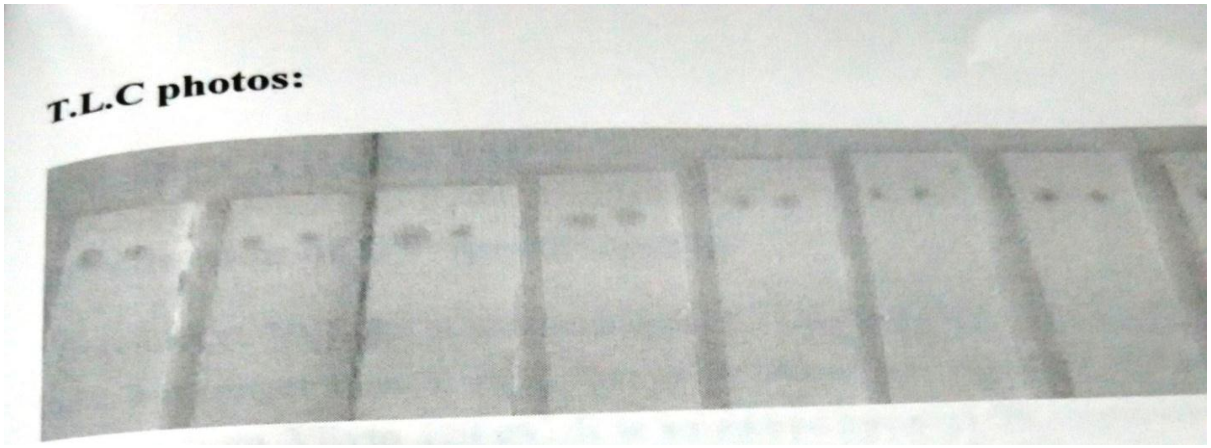
The development tank was saturated with suitable solvent system chloroform, methanol and water (10:10:3) for the analysis of lipid present in plant extract. The plate was kept in the tank without touching baseline by solvent and left for development. The final solvent front was marked and the plate was dried.

Spot visualization

Few pieces of iodine crystals were kept in the tank and covered with glass plate to saturate the tank with iodine vapour. The plate was then kept in iodine vapour saturated tank and left for few hours.

Collection of the active compounds

Spots on the preparative silica gel plate were scratched with the help of clean and dry spatula and collected in beaker containing 70% ethanol and left over night. The content in the beaker was stirred filtrated through what man no.1 filter paper. The filtrate was collected in clean and dry beaker. The filtrate containing active compound was used for the determination of antimicrobial effect.



Rf Value $= \frac{Distance\ travelled\ by\ solute\ front}{Distance\ travelled\ by\ solvent\ front}$

Rf .Value $= \frac{0.5}{0.6} = 0.8$

PHYTOCHEMICAL ANALYSIS

The extracts were analyzed for the presence of alkaloids, tannins, flavonoids, steroids, carbohydrates, proteins and triterpenoids.

1. Test for Alkaloids

Take few ml of test solution and add 2-4 drops of dragendrffs reagent. It forms the reddish brown colour precipitate in the test solution .It indicates the presence of alkaloids.

2. Test for carbohydrates

To the test solution add few drops of alcoholic α-naphtha and few drops of concentrated sulphuric acid through the walls of the test tube. It forms purple to violet colour ring appears at the junction of the test tube. It indicates the presence of carbohydrates in the solution.

3. Test for proteins

Heat the test solution in boiling water bath proteins gets coagulated. It indicates the presence of proteins in the test solution.

4. Test for tannins

To the test solution 2ml of ferric chloride solution. It turns to blue colour it indicates the presence of hydrolysable tannins. If it turns to green colour it indicates the presence of condensed tannins.

5. Test for steroids and triterpinoids

To the test solution add few drops of sulphuric acid drop by drop through the walls of the test tube. If red colour forms at lower layer in the solution. It indicates the presence of steroids. If yellow colour forms in the test solution it indicates the presence of triterpinoids.

6. Tests for Flavonoids

To 1 ml of aqueous extract, 1 ml of 10% lead acetate solution was added. The formation of a yellow precipitate was taken as a positive test for flavonoids.

Phytochemical screening of Solvent extract:

1	Alkaloids	+
2	Proteins	+
3	Tannins	+
4	Carbohydrates	+
5	Flavonoids	+
6	Triterpenoids	+

Bacterial culture

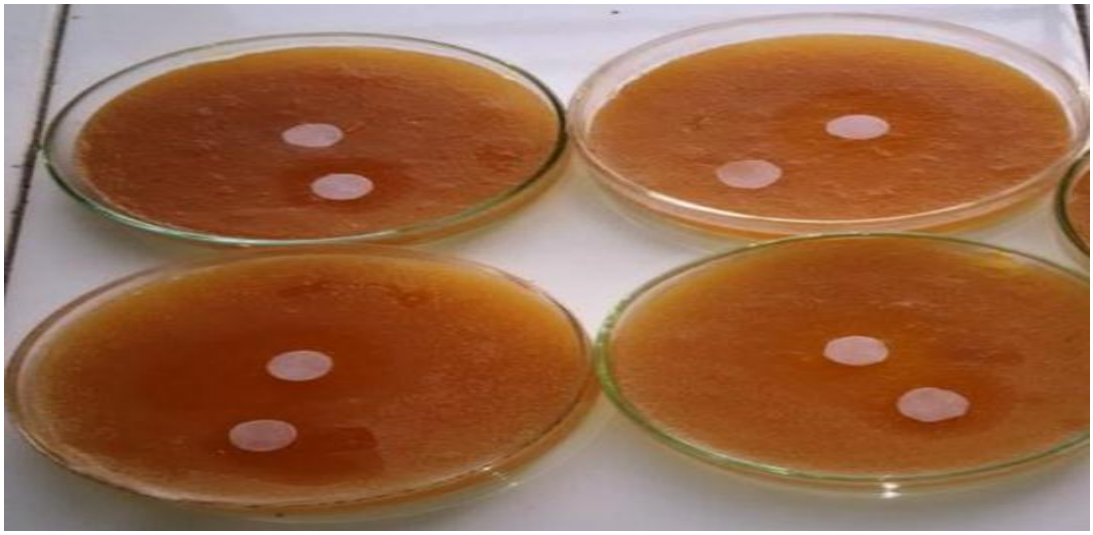
Prior to sensitivity testing each of the bacteria strains were cultured onto nutrient agar plate and incubated for 18 to 370C. A single colony was then cultured in 25ml nutrient broth for 4 hours at 37^0C. The density of bacteria culture required for the test.

Disc diffusion method

Disc diffusion method for antimicrobial susceptibility testing was carried out according to the standard method by Bauer *et al.* to assess the presence of antimicrobial activities of the plant extract. A bacteria culture + was used to lawn Muller Hinton agar (18) plates evenly using a sterile swab. The plates were dried for 15min and then used for the sensitivity test. The discs which had been impregnated with a series of plant extract were placed on the nutrient agar surface. Each test plate comprises of 6 discs.

One positive control, which is a standard commercial antibiotic disc and 5 treated discs. The standard antibiotic disc was Amikacin 20µg. The negative control was DMSO (100%). Besides the control each plate had five treated discs placed about equidistance to each other. The plates were then incubated at 370 C for 24 hours depending on the species of bacteria used in the test. After the incubation, the plates were examined for inhibition zone. The inhibition zones were then measured using callipers and recorded. The tests were repeated 3 times to ensure reliability.

Sample	S1	S2	S3	S4
Control	100µg/ml	150µg/ml	200µg/ml	250µg/ml
No Growth	No Growth	0.5mm	0.8mm	0.9mm
Standard/Amikacin	S1	S2	S3	S4
Control	50µg/ml	75µg/ml	100µg/ml	150µg/ml
No Growth	0.9mm	1mm	1mm	1.3mm



RESULTS AND DISCUSSION

The phytotochemical constituents of *Senna occidentalis* of extract showed various chemical constituents present in ethanol and methanol extracts are alkaloids, carbohydrates, tannins, steroids, triterpenoids, proteins and flavonoids are present. The values obtained at different stages were determined. The chromatography Rf value is 0.8. The sample absorbed in the IR studies of flavonoids of amino, carboxylic group wave length is 1168.86 per cm-1. When test compared with standard drug, plant extract showed maximum dose 250 µg/ml of dose produced the 0.9 mm zone of inhibition.

CONCLUSION

In industry Pharmacognosy plays a major role in bringing out of a safe and effective dosage form. The project work has given the importance for studying the phytochemical analysis and IR studies of Senna occidentalis. The Rf value of flavonoids is 0.89 and IR studies used to determine the wave length1168.86 cm-1. When test compared with standard drug, plant extract showed maximum dose 250 µg/ml of dose produced the 0.9 mm zone of inhibition.

REFERENCES

1. Bako SP, Bakfur MJ, John I, Bala EI. Ethno medicinal and phytochemical profile of some savanna plant species in Nigeria. Int J Bot., 2005; 1(2): 147-150.

2. World Health Organization. Traditional Medicine. Media Centre Fact Sheet No 134. Accessed from http://www.who.int/mediacentre/factsheets/fs134/en/ 2008.
3. Mahesh B, Satish S. Antimicrobial activity of some important medicinal plant against plant and human pathogens. World J Agri Sci., 2008; 4: 839-843.
4. Hamid AA, Aiyelaagbe OO, Usman LA, Ameen OM, Lawal A. Antioxidants: Its medicinal and pharmacological applications. African J of Pure and Applied Chemistry., 2010; 4(1): 7-10.
5. Zampini IC, Cuello S, Alberto MR, Ordonez RM, Almeida RD, Solorzano E *et al*. Antimicrobial activity of selected plant species from the Argentine puna against sensitive and multiresistant bacteria. Journal of Ethno pharmacology., 2009; 124: 499-505.
6. El-olemy MM, Al-Muhtadi FJ, Afifi AA. Experimental phytochemistry; a laboratory Manual king Saud University press., 1994; 350-359.
7. Harbone JB. Phytochemical methods. A guide to modern technique of plant Analysis. Chapinan and Hall, London., 1973; 33-185.
8. Okemo PO, Bais HP, Vivanco JM. *In Vitro* activities of *Maesa lanceolata* extracts against fungal plant pathogens. Fitoterapia., 2003; 74: 312-316.
9. Bouamama H, Noel T, Villard J, Benharref A, Jana M. Antimicrobial activities of the leaf extract of two Moroccan *Cistus* L species. Journal of Ethno pharmacology., 2006; 104: 104-107.
10. Arora DS, Kaur GJ. Antibacterial activity of some Indian medicinal plants. Journal of Natural Medicine., 2007; 61: 313-317.