

PHYTOCHEMICAL SCREENING OF ETHANOLIC EXTRACT OF BUTEA MONOSPERMA
AND SALVADORA PERSICAT.Mohammad Munawar^{1*} and D.Muralidhara rao²^{1*}Department of Biotechnology, JNTUA College of Engineering Pulivendula-516390, Andhra Pradesh, India.²Department of Biotechnology, S.K University, Anantapuramu-515003, Andhra Pradesh, India.

ABSTRACT: Natural drugs play important and vital role in the modern medicine. It is usually used to cure some diseases which may not be treated by conventional medicine. Drugs from the plants are easily available less expensive, safe and efficient and rarely have side effects. The alkaloids, tannins, flavanoids and phenol compounds play a major role in preventing various chronic diseases by a definite physiological action on the human body like anticancer, antimicrobial, antioxidant and anti-diabetic activities. The aim of the present study was to evaluate the phytochemical analysis of ethanolic extract of flowers of *Butea Monosperma* and stem of *Salvodara Persica*. Phytochemical screening was carried out for ethanolic extract. Phytochemical screening was carried out for ethanolic extract revealed the presence of various bioactive compounds include alkaloids, carbohydrates, proteins and saponins.

Key words: Phytochemical screening, Natural drugs, *Butea Monosperma*, *Salvodara Persica*

*Corresponding author T.Mohammad Munawar, Department of Biotechnology, JNTUA College of Engineering Pulivendula-516390, Andhra Pradesh, India munna686@gmail.com

Copyright: ©2017 T.Mohammad Munawar. This is an open-access article distributed under the terms of the Creative Commons Attribution License , which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

INTRODUCTION

Plants have been a valuable source of natural products for maintaining human health, especially in the last decade, with more intensive studies for natural therapies. Since ancient times green leafy vegetables have been used as medicine and the major sources of carbohydrates, proteins, vitamins, minerals, fats, aminoacids and fibres (Sharma and Kumar, 2013). The bioactive compounds of plants have a wide range of biological functions including, antimicrobial, antioxidant, anti-inflammatory activities (Burt, 2004; chanda, et.al, 2010]. Plants have to adapt to the changing environmental conditions for their survival of existence. The oxidative environment presents a range of free radicals including superoxide, hydroxyl radical, nitric oxide and peroxynitrite, for living organisms to deal with. Many evidences are exists to explain the role of free radicals in the development of various diseases including cancer, neurodegeneration and some inflammatory diseases (Halliwell, 2006, 2007; Ferguson, 2010). Antioxidants have therefore gained importance for their capacity to neutralize free radicals. In this context, the antibacterial and antioxidant properties of various medicinal plants are being investigated throughout the world because of the toxicological concerns associated with the synthetic antioxidants and preservatives (Peschel, et.al., 2006).

Butea monosperma (Palas) is a medium-sized deciduous tree belongs to family Leguminosae- Papilionae. This tree is also called 'Flame of the Forest' and Bastard Teak (Farooq, 2005; Khare, 2007). *B. monosperma* is a medium sized deciduous tree, with a somewhat crooked trunk (10-15 feet) in height and 5-6 feet in girth.

The dried flowers and stem bark of the plant contain some important flavanoids (medicarpin, plasonin) and alkaloids (butrin, isobutrin) that have a wide range of pharmaceutical and medicinal utilities. Alcoholic extracts of petals of *B. frondosa* flowers and seeds have shown the antiestrogenic, antiimplantation and antifertility activity (Laumas and Uniyal, 1966; Kamboj and Dhawan, 1982).

The toothbrush tree, *Salvadora persica* is also called miswak, belonging to the Salvadoraceae family, is one of the most important ones among 182 species of plants being used as chewing sticks. It has been widely used in many Asian, African, and Middle Eastern countries. The roots, twigs, and stems of this plant have been used for oral hygiene and small miswak sticks have been used as toothpicks for maintaining oral hygiene (Sher, et.al., 2011; Goyal, et.al., 2011). It has been reported that the aqueous and methanol extracts of miswak possess various biological properties against organisms considered important for the development of dental plaque and periodontitis (Sofrota, et.al., 2008). Therefore, the main objective of the present study was to evaluate the phytochemical analysis of ethanolic extract of flowers of *Butea monosperma* and rhizome or stem of *Salvadora persica*.

MATERIALS AND METHODS

Chemicals

Potassium hydroxide, α -naphthol, Mercuric nitrate, Ferric chloride, Lead acetate, Sulphuric acid, Hydrochloric acid, Picric acid, Chloroform and Nitric acid were purchased from SR Scientifics (Tirupathi, India). All other chemical reagents used were of analytical grade.

Collection of plant material

The fresh flowers of *Butea monosperma* and rhizome or stem of *Salvadora persica* were collected from the Western Ghats of Karnataka, India, during winter season. Plant parts were packed immediately after picking and kept in cold (-20°C) dark storage until processed. The plant specimen was identified with the help of an expert, Prof. C.Sudhakar (Department of Botany, S.K University, India). Collection was performed by pulling plants out of the soil and transferring them into sealable plastic bags.

Preparation of extracts

Flowers and stems of the plant were collected and dried under shade at room temperature (Fig-1). The plant material was then chopped and ground to fine powder using a mechanical blender. 20gm of powder of flowers of *Butea monosperma* and rhizome or stem of *Salvadora persica* was taken into conical flask. The phytoconstituents were extracted by adding 100ml of ethanol to the powder. The flask was incubated in orbital shaker for 48 hrs. The extract was filtered through five layers of muslin cloth. The process was repeated twice. The collected extract was pooled and concentrated by evaporation (Siddiqui, et.al., 2009). The extract was preserved and stored at 4°C in airtight bottles for further study (Fig-2).



Fig-1: Flowers and Stem of *Butea monosperma* and *Salvadora persica*

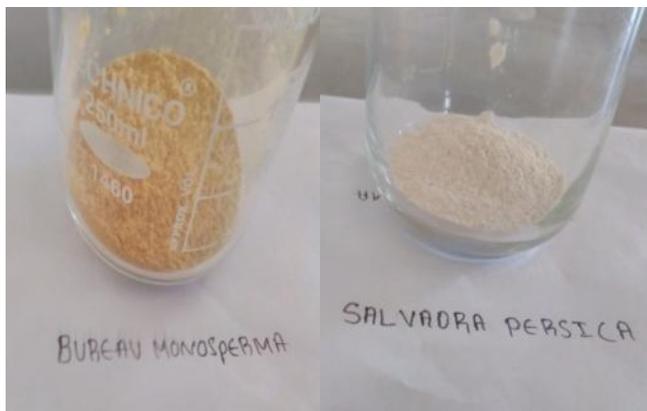


Fig-2: Extract powder form of *Butea monosperma* and *Salvadora persica*

Phytochemical screening

Phytochemical analysis of flowers of *Butea monosperma* and rhizome or stem of *Salvadora persica* extracts were carried out to identify various phytoconstituents. The methods for screening were carried out according to the method of (Siddiqui, 2009; Edeoga,2005) with some modifications.

The test for phytochemical screening includes:

a) Test for carbohydrates

Molisch's Test

Extracts were dissolved individually in 5ml distilled water and filtered. Filtrates were treated with two drops of alcoholic α -naphthol solution in a test tube. Add 0.2 ml of concentrated sulfuric acid slowly through the sides of the test tube, a purple to violet colour ring appears at the junction.

b) Test for Proteins and Amino acids

Millons test

Test solution with 2ml of Millons reagent (Mercuric Nitrate in Nitric acid containing traces of nitrous acid), white precipitate appears, which turns red upon gentle heating.

c) Test for fats and fixed oils

Stain test

Press the small quantity of extract between two filter papers. The stain on one filter paper indicates the presence of fixed oils.

Saponification test

Add a few drops of 0.5N of alcoholic potassium hydroxide to small quantities of various extracts along with a drop of phenolphthalein separately and heat on a water bath for 1-2 hours. The formation of soap or partial neutralization of alkali indicates the presence of fixed oils and fats.

d) Test for Alkaloids

Hager's test

Extracts were dissolved individually in dilute hydrochloric acid and filtered. Filtrates were then treated with Hager's reagents (saturated picric acid solution). The presences of alkaloids were confirmed by the formation of yellow colored precipitate.

Wagner's test

Filtrates were treated with wagner's reagent. Formation of brown/reddish brown precipitate indicates the presence of alkaloids.

e) Test for Flavonoids

Lead acetate test:

Extract were treated with few drops of lead acetate solution. Formation of yellow colour precipitate indicates the presence of flavonoids.

Sulphuric acid solution:

Extracts were treated with few drops of sulphuric acid. Formation of orange colour indicates the presence of flavonoids.

f) Test for steroids

2ml of acetic anhydride was added to 0.5g of the extracts of each with 2ml of sulphuric acid. The change of colour from violet to blue or green in samples indicates the presence of steroids.

g) Test for Terpenoids

Salkowski's test

0.2 g of the extract of the whole plant sample was mixed with 2 ml of chloroform and concentrated sulphuric acid (3 ml) was carefully added to form a layer. A reddish brown coloration of the interface indicates the presence of terpenoids.

h) Test for phenols

Ferric chloride test

Extracts were treated with few drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenol.

Lead acetate test

Extracts were treated with few drops of lead acetate solution. Formation of yellow precipitate indicates the presence of phenol.

i) Test for Saponins

Foam test:

About 0.2 g of the extract was shaken with 5 ml of distilled water. Formation of frothing shows the presence of saponins.

j) Test for Anthroquinone glycosides

Extracts were treated with 5ml chloroform and shaken for minutes. The extracts were filtered and filtrate was added with equal volume of 10% ammonia solution. A pink violet or red colour was observed for the presence of anthraquinone.

RESULTS**Preliminary phytochemical screening**

The results of preliminary phytochemical screening of ethanolic extract of flowers of *Butea monosperma* showed the presence of carbohydrates, proteins, alkaloids, Flavanoids, saponins, phenols and tannins and ethanolic extract of stem of *Salvadora persica* showed the presence of carbohydrates, alkaloids, flavanoids and saponins. The pharmacological activities of medicinal plants are due to presence of secondary metabolites such as alkaloids, carbohydrates, proteins and saponins, there is possibility of antioxidant and antimicrobial activity. The phytochemicals play a vital role in preventing renal diseases, cholesterol and carcinomas (Titanji, 2008). The results of tests are shown in Table 1.

DISCUSSION

In present study, the phytochemical screening of ethanolic extract of flowers of *Butea monosperma* showed the presence of carbohydrates, proteins, alkaloids, flavanoids, saponins, phenols and tannins and ethanolic extract of stem of *Salvadora persica* showed the presence of carbohydrates, alkaloids, flavanoids and saponins. Phenolic compounds are important plant constituents because of their free radical scavenging ability facilitated by their hydroxyl groups and the total phenolic concentration could be used as a basis for rapid screening of antioxidant activity (Yi, et.al., 2007). Phenolic compounds are also involved in conferring plants with oxidative stress tolerance. Flavonoids are highly effective scavengers of most oxidizing molecules, including singlet oxygen, and various other free radicals implicated in several diseases (Bravo, 1998). Flavonoids, on the other hand, suppress reactive oxygen formation, chelate trace elements involved in free-radical production, scavenge reactive species, and up-regulate and protect antioxidant defenses (Agati, et.al., 2012). Crude extracts of fruits, herbs, vegetables, cereals, and other plant materials rich in phenolics and flavonoids, are increasingly being used in the food industry for their antioxidative properties and health benefits.

Table 1. Phytochemical screening of extract flowers of *Butea monosperma* and stem of *Salvadora persica*

S.No	Phytochemical test	Reagent used (test performed)	Observation	Result of <i>Butea monosperma</i>	Result of <i>Salvadora persica</i>
1.	Test for Carbohydrates	Molisch’s test	Formation of violet ring	++	++
2.	Test for Proteins and amino acids	Millon’s test	Formation of red colour	++	-
3.	Test for fats and oil	Saponification test	No formation of soap	-	-
4.	Test for Alkaloids	Wagner’s test	Formation of cream precipitate	+++	+++
		Hager’s test	Formation of yellow colour	+++	+++
5.	Test for Flavanoids	Lead acetate test	No formation of Yellow precipitate	++	++
		Sulphuric acid test	No formation of orange colour	++	++
6.	Test for Terpenoids	Salkawoski test	No formation of reddish brown	-	-
7.	Test for glycosides	Borntrager’s test	No formation of Pink colour	-	-
8.	Test for steroids	Acetic ahydride test	Formation of blue or green colour	-	-
8.	Test for Saponins	Foam test	Formation of foam	+++	+++
9.	Test for Phenols and Tannins	Ferric chloride test	No formation of bluish black colour	++	-

+ sign indicates fairly presence, ++sign indicates moderately presence, +++sign indicates more quantity and – sign indicates absence

CONCLUSION

The results suggest that flowers of *Butea monosperma* and stem of *Salvadora persica* is a potential source of useful drugs due to the presence phytochemicals and can be utilized in the treatment of various diseases and also be oppressed for use in the pharmaceutical and cosmetic industries. However further studies mandatory to isolate the active principle from the crude extract for proper drug development.

ACKNOWLEDGMENTS

We acknowledge the Department of Biotechnology, JNTUACE, Pulivendula for providing necessary facilities for carrying out the work. We also acknowledge Prof.C.Sudhakar, Department of botany, S.K University for the identification of plant material.

REFERENCES

[1] Sharma HP, Kumar RA. (2013). Health security in ethnic communities through nutraceutical leafy vegetables. J Environ Res Develop, 7(4), 1423–1429.

[2] Burt S. (2004). Essential oils: their antibacterial properties and potential applications in foods. A review, Int J Food Microbiol, 94(3), 223–253.

[3] Chanda S, Baravalia Y, Kaneria M, Rakholiya K. (2010). Fruit and vegetable peels-strong natural source of antimicrobics. In: Mendez-Vilas A, editor. Current research, technology and education topics in applied microbiology and microbial biotechnology. Spain: Formatex Research Center.

- [4] Halliwell, B. (2007). Oxidative stress and cancer: have we moved forward? *Biochem. J.* 401, 1–11.
- [5] Halliwell, B. (2006). Oxidative stress and neurodegeneration; where are we now? *J. Neurochem.* 97, 1634–1658.
- [6] Ferguson, L.R. (2010). Chronic inflammation and mutagenesis. *Mutat. Res. Fund. Mol. M.* 690, 3–11.
- [7] Peschel, W., Sanchez-Rabaneda, F., Dieckmann, W., Plescher, A., Gartzia, I., et al. (2006). An industrial approach in the search of natural antioxidants from vegetable and fruits wastes. *Food Chem.* 97, 137–150.
- [8] Farooq S.(2005). *Medicinal Plants: Field and Laboratory Manual, (Identification with its Phytochemical and In vitro studies data.)*, International Book Distributors, Uttaranchal; p. 202.
- [9] Khare CP. (2007). *Indian Medicinal Plants, 1st edition.* Springer (India), Pvt. Ltd, p. 105-106.
- [10] Laumas KR, Uniyal JP. (1996). A preliminary report on the antiestrogenic activity of the petals of *Butea frondosa* flowers. *Indian J Exp Biol* , 4, 246.
- [11] Kamboj VP, Dhawan BN.(1982). Research on plants for fertility regulation in India. *J Ethnopharmacol*, 6, 191-226.
- [12] Sher,H., AlYamani, M. N., L. Wijaya.(2011). Ethnobotanical and antibacterial potential of *Salvadora persica*: a well-known medicinal plant in Arab and union system of medicine,” *Journal of Medicinal Plants Research*, vol. 5, no. 7, pp. 1224–1229.
- [13] Goyal,M., Sasmal, D., B. P. Nagori. (2011). *Salvadora persica* (meswak): chewing stick for complete oral care,” *International Journal of Pharmacology*, vol. 7, no. 4, pp. 440–445.
- [14] Sofrata,A., Brito,F., Al-Otaibi,M., Gustafsson,A. (2011). Short term clinical effect of active and inactive *Salvadora persica* miswak on dental plaque and gingivitis,” *Journal of Ethnopharmacology*, vol. 137, no. 3, pp. 1130–1134.
- [15] Siddiqui S, Verma A,Rather A.A, Jabben F, Meghvansi MK.(2009). Preliminary phytochemicals analysis of some important medicinal and aeromatic plants, *Advances in Biological Research*, 3, 188-195.
- [16] Edeoga H.O, Okwu D.E, Mbaebie B.O. (2005). Phyto constituents of some Nigerian medicinal plants, *African Journal of Biotechnology*, 4, 685-688.
- [17] Titanji, V.P.; Zofou, D.; Ngemenya, M.N. (2008). "The Antimalarial Potential of Medicinal Plants Used for the Treatment of Malaria in Cameroonian Folk Medicine". *African Journal of Traditional, Complementary and Alternative Medicines.* 5 (3), 302–321.
- [18] Yi, O., Jovel, E.M., Towers, N.G.H., Wahbe, T.R., Cho, D. (2007). Antioxidant and antimicrobial activities of native *Rosa* sp. From British Columbia, Canada. *Int. J. Food Sci. Nutr.* 58, 178–189.
- [19] Bravo, L.(1998). Polyphenols: chemistry, dietary sources, metabolism and nutritional significance. *Nutr. Rev.* 56, 317–333.
- [20] Agati, G., Azzarello, E., Pollastri, S., Tattini, M. (2012). Flavonoids as antioxidants in plants: location and functional significance. *Plant Sci.* 196, 67–76.

ISSN : 0976-4550

INTERNATIONAL JOURNAL OF APPLIED BIOLOGY AND PHARMACEUTICAL TECHNOLOGY



Email : ijabpt@gmail.com

Website: www.ijabpt.com