



PHYTOCHEMICAL SCREENING AND *IN-VITRO* ANTIMICROBIAL ACTIVITY OF  
HYDROALCOHOLIC EXTRACT OF STEM OF *BAUHINIA VARIEGATE*

Dr. Peenu Mahendra Joshi\*, Dr. Jaswinder Mehta, Naresh Kumar, Aaradhana Vishwakarma,  
Dr. Bhawna Sharma, Dr. Ruchi Acharya

Department of Botany, Career College, Bhopal (M.P.)

**\*Correspondence Info:**

**Peenu Mahendra Joshi**

Department of Botany, Career  
College, Bhopal (M.P.)

*Email:*

Joshi-drpeenu1@gmail.com

**ABSTRACT**

Infectious diseases are the second leading cause for worldwide death. Treatment of infections continues to be difficult in modern time because of the severe side effects of some drugs and the growing resistance to antibacterial agents. Over the past few decades the use of antibiotics is under threat as many commonly used antibiotics have become less effective against certain illnesses due to emergence of multi drug-resistant bacteria. *Bauhinia variegata* showed the presence of flavonoid, diterpenes, proteins, carbohydrate, saponins while, Alkaloids, Glycosides and Phenol were not detected. The total flavonoids estimation of stem hydroalcoholic extract of *Bauhinia variegata* showed the content values of 5.062 mg/100 mg respectively. Results of antimicrobial activity of the plant extract were found more effective against *Klebsiella pneumoniae* followed by *Staphylococcus aureus*. Phytochemicals present in extracts showed antimicrobial activities. MIC value shows that plant extracts have antimicrobial activity against *Klebsiella pneumoniae* and *Staphylococcus aureus*.

**Key words:** *Bauhinia variegata*, *Antibacterial*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Total flavonoid content*.

**\*Article History:**

Received: 22/03/2021

Revised: 18/04/2021

Accepted: 26/04/2021

**INTRODUCTION:**

From the classical times the nature stands a golden mark and provided the armoury of remedies to cure all ailments of mankind. Herbs have always been the predominant form of medicine in India and currently they are becoming popular globally. India has an age old system of medicine known as Ayurveda, Siddha and Unani system. There is extensive change in the international interest of herbal medicines (Kamboj, 2000). The medicinal value of plants lies in some chemical

substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannin, flavonoids and Phenolic compounds (Okwu, 1999; Hill, 1952). Today, plants material remains an important source for combating illness, including infectious diseases and many of these plants have been investigated for novel drugs or used as templates for the development of new therapeutic agents, food additives, agrochemicals and industrial chemicals (Arora

et al., 2012; Habila et al., 2011). Keeping in the view the importance of phytochemical. It was compulsory to provide their scientific data base line which may play a significant role in drug preparation. Alkaloids play some metabolic role and control development in living system. It is used as antioxidant, anti-inflammatory & reduces aerodigestive tract cancer risk in smokers (Hadi and Bremner, 2001). Tannin have shown antiviral, antibacterial and anti parasitic effects, anti-inflammatory and antiulcer activity (Lata and Dubey, 2010; Souza et al., 2006). The Flavonoids and phenolic compounds in plants have reported to exert multiple biological effects including antioxidant, free radical scavenging abilities, anti-inflammatory, anticarcinogenic (Asha et al., 2011). Medicinal plants represent a rich source of antimicrobial agents. The development and spread of resistance to the existing antibiotics by microorganisms are due to haphazard use of commercial antimicrobial drugs commonly used in the treatment of various diseases. Although number of plants with antimicrobial activities has been identified, great number still remains unidentified (Yadav et al., 2011).

There are about one hundred species under the genus *Bauhinia* (Caesalpiniaceae) and eight are native to India. *B. variegata* Linn., commonly known as 'Kachnar', is a small sized deciduous tree with dark brown and more or less smooth bark, up to 8m tall; propagated by seed. The roots and bark are astringent, acrid, cooling, constipating, depurative, anthelmintic, vulnerary, anti-inflammatory and styptic (Chang, 1995). They are useful in vitiated conditions of pitta and kapha, diarrhoea, dysentery, skin diseases, leprosy, intestinal worms, tumours, inflammations,

scrofula, proctoplosis, haemorrhoids, haemoptysis, cough, menorrhagia and diabetes. Root decoction is used in dyspepsia and flatulence and act as an antidote to snake poison. Plant extract act as an effective measure for controlling insect pest like *Plutella xylostella*. *B. variegata* var. *candida* is a promising source of edible wild vegetable flowers with plenty of nutrients. This plant may serve as a potential source for low cost proteins. The tree is susceptible to 'Brown Root Rot' caused by *Phellinus noxius*. The abundance of phytophagous mites is higher, being *Lorryia Formosa* Cooreman the dominant species (Rogrigo et al., 2007). Therefore, the objectives of the present investigation were to evaluate the antibacterial activity of *B. variegata* in addition to provide data on total flavonoids contents and preliminary phytochemical profile of the extracts.

## Materials and Methods

### Plant Material

Stem of *Bauhinia variegata* were collected from local area of Bhopal (M.P.) month of February, 2021.

### Chemical reagents

All the chemicals used in this study were obtained from HiMedia Laboratories Pvt. Ltd. (Mumbai, India), Sigma-Aldrich Chemical Co. (Milwaukee, WI, USA), SD Fine-Chem Chem. Ltd. (Mumbai, India) and SRL Pvt. Ltd. (Mumbai, India). All the chemicals used in this study were of analytical grade.

### **Extraction by maceration method**

105 gram of powdered stem of *Bauhinia variegata* were extracted with hydroalcoholic solvent (Methanol: Aqueous: 65:35) by maceration method. The extract was evaporated above their boiling points. Finally, the percentage yields were calculated of the dried extract (Sharma et al., 2020).

### **Phytochemical screening**

Phytochemical examinations were carried out extracts as per the standard methods by standard methods.

### **Total flavonoids content estimation**

Determination of total flavonoids content was based on aluminium chloride method (Parkhe and Bharti, 2019). 10 mg quercetin was dissolved in 10 ml methanol, and various aliquots of 5- 25µg/ml were prepared in methanol. 10 mg of dried extract was dissolved in 10 ml methanol and filter. Three ml (1mg/ml) of this extract was for the estimation of flavonoids. 1 ml of 2% AlCl<sub>3</sub> solution was added to 3 ml of extract or each standard and allowed to stand for 15min at room temperature; absorbance was measured at 420 nm.

### **Antimicrobial activity of prepared extract**

The well diffusion method was used to determine the antimicrobial activity of the

extract prepared from the stem of *Bauhinia variegata* using standard procedure<sup>56</sup>. It's essential feature is the placing of wells with the antibiotics on the surfaces of agar immediately after inoculation with the organism tested. Undiluted overnight broth cultures should never be used as an inoculum. The plates were incubated at 37°C for 24 hr. and then examined for clear zones of inhibition around the wells impregnated with particular concentration of drug (Mehta et al., 2019).

## **RESULTS AND DISCUSSION**

The crude extracts so obtained after the maceration extraction process, extracts was further concentrated on water bath for evaporate the solvents completely to obtain the actual yield of extraction. Phytochemical analysis of hydroalcoholic extracts of *Bauhinia variegata* showed the presence of flavonoid, diterpenes, proteins, carbohydrate, saponins while, Alkaloids, Glycosides and Phenol were not detected. In the plant polyphenols are also strongly support the contribution of to the prevention of cardiovascular diseases, cancer, osteoporosis, neurodegenerative diseases, and diabetes mellitus and suggest a role in the prevention of peptic ulcer etc. The total flavonoid content of the extracts was expressed as percentage of Quercetin equivalent per 100 mg dry weight of sample. Total flavonoid

content was calculated as quercetin equivalent (mg/100mg) using the equation based on the calibration curve:  $Y=0.032X + 0.018$ ,  $R^2=0.998$ , where X is the quercetin equivalent (QE) and Y is the absorbance. The total flavonoids estimation of stem hydroalcoholic extract of *Bauhinia variegata* showed the content values of 5.062 mg/100 mg

respectively. Results of antimicrobial activity of the plant extract were found more effective against *Klebsiella pneumoniae* followed by *Staphylococcus aureus*. Phytochemicals present in extracts showed antimicrobial activities. MIC value shows that plant extracts have antimicrobial activity against *Klebsiella pneumonia* and *Staphylococcus aureus*.

**Table No. 1: % Yield of extract of *Bauhinia variegata* (Stem)**

S. No.	Extract	% Yield (W/W)
1.	Hydroalcoholic	0.952

**Table No. 2: Result of phytochemical screening of *Bauhinia variegata***

S. No.	Constituents	Hydroalcoholic extract
1.	<b>Alkaloids</b> Hager's Test:	-ve
2.	<b>Glycosides</b> Legal's Test:	-ve
3.	<b>Flavonoids</b> Lead acetate Test:	+ve
4.	<b>Diterpenes</b> Copper acetate Test:	+ve
5.	<b>Phenol</b> Ferric Chloride Test:	-ve
6.	<b>Proteins</b> Xanthoproteic Test:	+ve
7.	<b>Carbohydrate</b> Fehling's Test:	+ve
8.	<b>Saponins</b> Froth Test:	+ve

**Table No. 3: Estimation of total flavonoids content of hydroalcoholic extract of *Bauhinia variegata***

S. No.	Extract	Total flavonoids content (mg/ 100 mg of dried extract)
1.	Hydroalcoholic	5.062

**Table No. 4: Antimicrobial activity of *standard drug* against selected microbes**

S. No.	Name of drug	Microbes	Zone of inhibition		
			30 µg/ml	20 µg/ml	10 µg/ml
1.	Ciprofloxacin	<i>Staphylococcus aureus</i>	22±2.16	18±2.62	17±1.69
		<i>Klebsiella pneumoniae</i>	36±1.699	28±1.24	19±4.71

**Table No. 4: Antimicrobial activity of hydroalcoholic extract of *Bauhinia variegata* against selected microbes**

S. No.	Name of microbes	Zone of inhibition		
		100mg/ml	50 mg/ml	25mg/ml
1.	<i>Staphylococcus aureus</i>	12±0.5	8±0.57	7±0.86
2.	<i>Klebsiella pneumoniae</i>	16±0.74	10±0.47	8±0.5

## Conclusion

In this study, antimicrobial activities of traditional medicinal plant *Bauhinia variegata* were assessed by maceration method. The result showed potential antibacterial effects of *Bauhinia variegata* extract against bacterial strains tested, MIC value shows that plant extracts have antimicrobial activity against *Klebsiella pneumonia* and *Staphylococcus aureus*.

## References

1. Kamboj, V.P. (2000). Herbal medicine. Current science. 78(1): 35-39.
2. Okwu, D.E. (1999). Flavoring properties of spices on cassava fufu, Afr. J. Root Tuber crops, 3(2): 19-21.
3. Hill AF. (1952). Economic Botany: A textbook of useful plants and plants product, 2nd Edition, Mcgraw Hill, Book company, Inc, New York.
4. Arora, C. C., Mehta S and Dua, H. (2012). Phytochemical screening and Evaluation of biological activities of some medicinal plants of Pharwara, Punjab, Asian Journal of Chemistry. 24(12): 5903-5905.
5. Habila, J.D., Bello, I.A., Dzikwe, A.A., Ladan Z. and Sabiu, M. (2011). Aust. J. Basic Appl. Sci. 5:537.
6. Hadi, S. and John, B. (2001). Bremner. Initial Studies on Alkaloids from Lombok Medicinal Plants. Molecules. 6: 117-129,
7. Lata, N. and Dubey, V. (2010). J. Pharm. Res. 3, 6, 1240-1242.
8. Souza, S.M.C., Aquino, L.C. Milach Jr, A.C., Bandeira, M.A., Nobre, M.E. and Viana, G.S. (2006). *Phytotherapy Research*, 21 (3): 220–225,
9. Asha, K., Rasika, C.T., Nirmala, R.D. and Jyoti, P.S. (2011). Ann. Biol. Res. 2(1): 176-180.
10. Yadav, S., Kumar, S., Jain, P., Pundir, R.K., Jadon, S., Sharma, A., Khetwal, K.S. and Gupta, K.C. (2011). Antimicrobial activity of different extracts of roots of *Rumex nepalensis* spreng. Indian Journal of Natural Products and Resources. 12 (1); 65-69.
11. Chang, T.T. (1995). Decline of Nine tree species with brown root rot caused by *Phellinus noxius* in Taiwan, Plant Diseases. 79(9): 962- 965.
12. Rogrigo, D.D., Reinaldo, J.F.F. and Renato B. (2007). Mites (Acari: Arachnida) As sociated with *Bauhinia variegata* L.

- (Leguminosae) in Northeast of State of São Paulo, Neotropical Entomology. 36(2): 322- 325
13. Sharma, S., Jain, P.K. and Parkhe, G. (2020). Extraction, Phytochemical Screening and Anti-Inflammatory Activity of Hydro-Ethanollic Extract of Roots of *Dactylorhiza hatagirea*, Journal of Drug Delivery and Therapeutics. 10(3-s):86-90
14. Parkhe, G. and Bharti, D. (2019). Phytochemical Investigation and Determination of Total Phenols and Flavonoid Concentration in Leaves Extract of *Vitex trifolia* Linn, Journal of Drug Delivery and Therapeutics. 9(4-A):705-707
15. Mehta, J., Joshi, P.M., Kushwaha, P., Parkhe, G. (2019). In-vitro Antioxidant Activity and Antimicrobial Activity of Hydroalcoholic Extracts of *Vernonia cinerea*, Journal of Drug Delivery and Therapeutics. 9(1):225-228