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Research Article

PHYTOCHEMICAL SCREENING AND ANTIBACTERIAL PROPERTIES OF *ADATHODA VASICA* AGAINST FEW HUMAN PATHOGENSSajani Jose^{1*} and Sujatha.K²¹ PG & Research, Department of Zoology, Nirmala College for Women, Coimbatore, Tamil Nadu, India.² PG & Research, Department of Zoology, Government Arts College, Coimbatore, Tamil Nadu, India.

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Abstract:

To investigate the phytochemical screening and antimicrobial activity of *Adathoda vasica*. (Linn) in different solvents viz petroleum ether, chloroform, ethyl acetate, methanol and water. The aim of the present study was to evaluate the quantitative analysis of phytochemicals and antimicrobial activity of various extracts of *Adathoda vasica*. The phytochemical analysis of petroleum ether extract showed the presence of tannin, steroids, flavanoid, terpenoid and cycloglycosides. Ethyl acetate extract showed the presence of steroids, coumarin and cycle glycosides. Chloroform extract showed the presence of steroids, terpenoid and cyclo glycosides. Methanol extract showed the presence of alkaloid, tannin, steroids, flavanoid, terpenoid, phlobotanin and total phenol. Where as in aqueous extract, tannin, flavanoids, coumanin and total phenol were present. Flavanoids were strongly positive in methanol and aqueous extract.

The *in vitro* biological screening effects of different solvents extracts were tested against gram positive microorganism (*Staphylococcus aureus*, *Bacillus subtilis* and *Bacillus cereus*) and gram negative microorganism (*Escherichia coli*, *Pseudomonas aeruginosa* and *Proteus vulgaris*). Petroleum ether, chloroform, ethyl acetate, methanol and water extracts at different concentrations (5, 10, 20, 30, and 50 µg/ml) were used to investigate the antimicrobial activity. The results were compared with standard drug Methicilin (50ug/ml).

It was observed that petroleum ether, ethyl acetate, chloroform, methanol and aqueous extract all showed activity against bacteria. Methanol extract of *Adathoda vasica* showed maximum activity against *Staphylococcus aureus* the zone of diameter 29.47 ± 0.01 mm compared to other solvent extracts and Methicillin (positive control). The study revealed a notable antibacterial inhibitory activity of methanolic extract of the leaf.

Keywords: Antibacterial, Gram positive, Gram negative, Methicillin, Zone of inhibition**Corresponding author:**

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INTRODUCTION:

Nature has a source of medicinal agents since times immemorial. It is clear that the plant kingdom harbours an inexhaustible source of active ingredients invaluable in the management of much intractable disease. There are several reports on the antimicrobial activity of different herbal extracts in different regions of the world[1-4]. There is continuous and urgent need to discover new antimicrobial compounds with diverse chemical structures and novel mechanisms of action due to an alarming increase in the incidence of new and re-emerging infectious diseases and development of resistance to the antibiotics in current clinical use[5]. The medicinal value of plants lies in some chemical substance or group of compounds that produce a definite physiological action in the human body. These chemical substances are called secondary metabolites. The most important of these bioactive groups of plants are alkaloids, terpenoids, steroids, flavonoids, tannins and phenolic compounds [6]. In addition, many biological activities and antibacterial effects have been reported for plant tannins and flavonoids[7-8]. Plants have an almost limitless ability to synthesize aromatic substances, most of which are phenols or their oxygen substituted derivatives[9]. These compounds protect the plant from microbial infection and deterioration [10].

Adathoda vasica (Linn). Nees and *Justica adathoda* Linn. It is commonly known as Basak (Bengali); Aradusi, adusa (Gujrati); Arusa, baansa, adusla (Hindi); Bansa basuti bhekkar (Punjabi), and shwetavasa, vasa, vasaka, (Sanskrit) and Malabar nut (English) in different languages and regions of India[11]. It is an evergreen, gregarious, perennial shrub, 1-2.5m in height, having opposite ascending branches. The plant is distributed throughout India up to an altitude of 1300m and mainly found in sub-Himalayan regions; also found in Nepal, Pakistan, Myanmar and Germany[12]. Leaves are simple, petiolate, ex-stipulate, 10-12 cm long and 3-10 cm broad, lanceolate to ovate lanceolate having crenate margin, tapering base and an acuminate apex with characteristic odour and bitter taste. Transverse section of the leaves showed two layers of palisade cells with diacytic stomata. Glandular and non-glandular trichomes are present on both the surface of the leaf. Elongated cystoliths, acicular and prismatic form of calcium oxalate crystals are present in the mesophyll. The midrib region composed of 4-6 layers of cellonchyma just below the epidermis and 3-5 vascular bundles, central one being largest[13].

It has been used in Ayurvedic system of medicine for the treatment of various ailments of respiratory tract

in both children and adults. All the parts of the plant have been used for their therapeutic beneficiary effect from ancient times. The plant is used as an ingredient of numerous popular formulations including cough syrups used in combination with ginger and tulsi where it exerts its action as an expectorant and antispasmodic[14]. Its leaves are extensively used for treating cold, cough, whooping cough and chronic bronchitis and asthma as sedative expectorant, antispasmodic and anti-inflammatory drug. There is considerable demand for this plant within the country[15].

MATERIALS AND METHODS:**Plant Material - *Adathoda vasica***

The leaves of *Adathoda vasica* was collected from in and around Kerala during September to October-2015. The leaves were authenticated by the Department of Botany. The voucher specimens were kept in the Department of Botany in Nirmala College for Women, Coimbatore, Tamilnadu, India.

Preparation of the extract

The leaves were washed with fresh water and thin finally with double distilled water. The leaves were dried in shade at room temperature for 10 days and homogenized to fine powder using an electronic blender. The fine powder was used for phytochemical screening. The leaf powder was subjected to extraction in soxhlet extractor. Extraction of the plant parts were done with different solvents based on the polarity of the solvents. The solvents used were petroleum ether, ethyl acetate, chloroform, methanol and water.

Phytochemical Screening

The concentrated extracts were subjected to qualitative test for the identification of various phytochemical constituents as per standard procedures[16-18]. Based on the results obtained in the qualitative phytochemical analysis, these extract were taken for antimicrobial study.

Test Organism

The test microorganism like gram positive microorganism (*Staphylococcus aureus*, *Bacillus subtilis* and *Bacillus cereus*) and gram negative microorganism (*Escherichia coli*, *Pseudomonas aeruginosa* and *Proteus Vulgaris*). The microorganisms were collected from PSG Medical college and Research Centre Laboratory, Coimbatore, Tamilnadu, India.

Antibacterial Assay

The antibacterial assay was performed by agar well diffusion method. The nutrient agar was inoculated with 100ul of the inoculum and poured into the Petri plate. A well was prepared in the plates with the help of a cork-borer (6mm). About 50µl of the extract (5, 10, 20, 30 and 50µg/ml) was dispensed into the well.

Mecithylin 50µg/ml was used as the standard. The plates were incubated overnight at 37°C. For each bacterial strain, Mecithylin 50µg/ml were used as positive control. The diameters of the inhibition zones were measured in mm.

RESULT AND DISCUSSION:

Preliminary Phytochemical analysis revealed the presence of secondary metabolites like alkaloid, saponin, tannin, steroid, flavanoids, terpenoids, phlobotanin and total phenol, whereas absence of coumanin, cycloglycosides and quinine (Table-1). Phytochemical constituents such as tannins, flavanoids, alkaloids, phenols and several other aromatic compounds are secondary metabolites of plants that serve as defence mechanism against predation by microorganism, insects and herbivore[19-20]. The medicinal value of plant lies in some chemical substances that produce a definite physiological action on the human body. Many leaves have antimicrobial principles such as essential oils and aromatic compounds[21]. Plants are

important source of potentially useful structures for the development of new chemotherapeutic agents.[22].

Plants contain many biological active compounds which have potential for development as medicinal agents. Herbal medicines already form the basis of therapeutic use in the developing countries, but recently there has been an increased in the use of herbal medicines in developed world too [23]. The presence of their secondary metabolites in the selected leaf extract serves to protect the plant against infection by microorganisms, predation by insects and herbivores, while some give plants their odours or flavours[24].

The results of antibacterial activity are given in the (Table-2) (Fig-1), which clearly show that all the extracts have shown antibacterial activity against the active tested organisms. Methanol extract have shown better activity among all solvents. Methanolic extract was more effective against *Staphylococcus aureus* than other organism followed by ethyl acetate, aqueous, petroleum ether and chloroform.

Table 1: Preliminary phytochemical analysis of *Adathoda vasica* (Linn.)

Sl.No	Phytochemical	Tests Performed	Ethyl Acetate	Petroleum Ether	Chloroform	Methanol	Water
1	Alkaloid	Mayers	-	-	-	+	-
2	Saponin	Foam	-	-	+	+	-
3	Tannin	Ferric chloride	-	+	-	+	+
4	Steroid	Liebermann Burchard	+	+	+	+	-
5	Flavanoid	Lead acetate	-	+	-	++	++
6	Terpenoid,	Copper acetate	-	+	+	+	-
7	Phlobotanin	Hcl	-	-	-	+	-
8	Coumanin	NH4OH	+	-	-	-	+
9	Cycloglycosides	Fehlings	+	+	+	-	-
10	Total phenol	Ferric chloride	-	-	-	+	+
11	Quinone	Sodium hydroxide	-	-	-	-	-

(-) Negative (+) Positive (++) Strongly positive

Table 2: Zone of inhibition of extracts against bacteria

Antibacterial activity of different extracts of *Adathoda* against different Human pathogenic bacteria (Mean± SEM) (mm)

Organism	Ethyl Acetate	Petroleum Ether	Chloroform	Methanol	Water	Methicillin
<i>Escherichia coli</i>	10.07±0.14	7.17±0.10	11.10±0.29	13.20±0.23	10.10±0.10	22.17±0.29
<i>Pseudomonas aeruginosa</i>	15.12±0.23	8.0±0.00	6.06±0.00	22.10±0.10	10.03±0.12	20.10±0.10
<i>Proteus vulgaris</i>	12.27±0.15	9.0±0.16	8.03±0.06	25.77±0.29	12.17±0.15	16.17±0.15
<i>Staphylococcus aureus</i>	23.20±0.20	16.17±0.15	13.20±0.15	29.47±0.10	13.20±0.17	21.17±0.29
<i>Bacillus subtilis</i>	16.83±0.25	13.20±0.20	11.10±0.10	20.17±0.15	14.07±0.12	25.90±0.01
<i>Bacillus cereus</i>	16.17±0.15	13.20±0.19	10.10±0.12	21.17±0.29	12.20±0.20	23.07±0.12

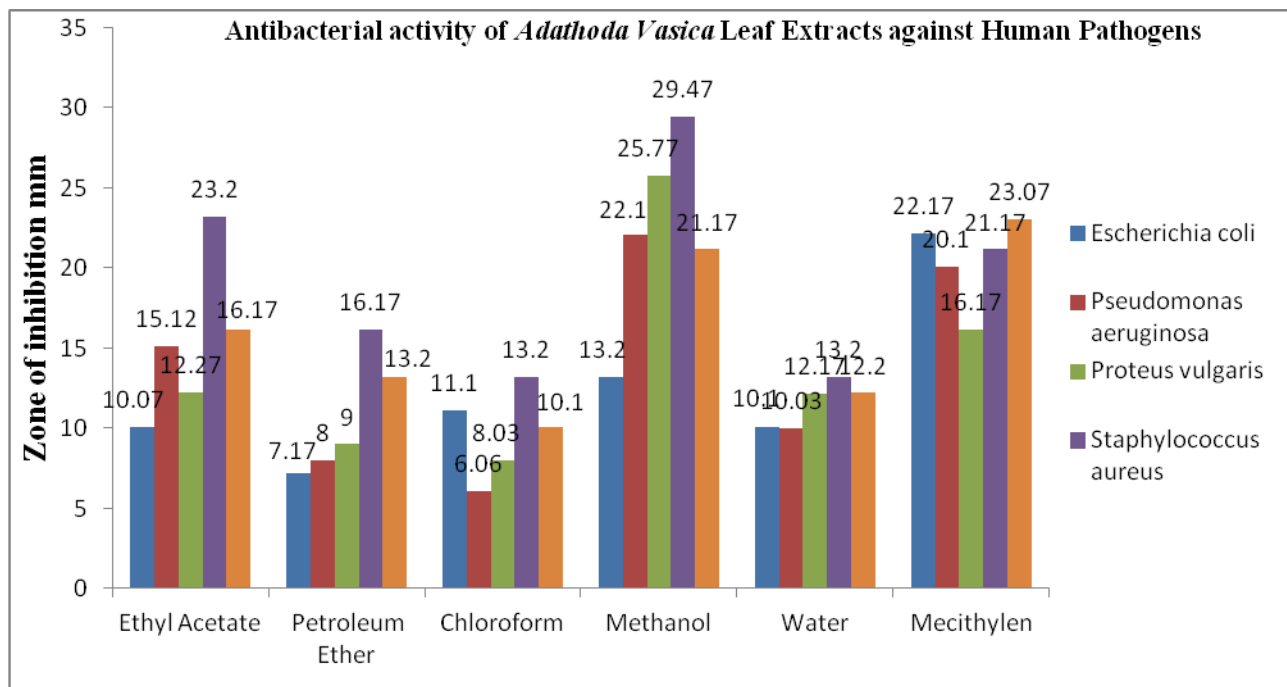


Fig 1: Antibacterial Activity

Plants rich in tannins have antibacterial potential due to their character that allows them to react with proteins to form stable water soluble compounds there by killing the bacteria by directly damaging its cell membrane^[22]. Alcoholic extracts of leaves and roots showed antibacterial activity against *Staphylococcus aureus* and *E.coli*, where water extract showed activity against *Staphylococcus aureus* only^[25]. So from this study it is clearly understood that each plant has its own inhibition activity against micro organisms.

Several authors have also reported that plant extracts are most effective against gram positive than gram negative bacteria and attributed this to the differences in the cell wall structure, the quantity of the active ingredients required to effect^[26-27] complete kill may not matter since medicinal plants have been reported to have little or no side effects^[28-29].

Flavonoids are a major group of phenolic compounds reported for their antiviral^[30] antimicrobial^[31] and spasmolytic^[32] properties. Alkaloids isolated from plant are commonly found to have antimicrobial properties^[33]. Extract of the seeds of *Vitex agnus-castus* was reported to possess antimicrobial activity which was associated with its alkaloids, saponins, tannins, flavonoids and glycosides content^[34]. This may be true in this study, since the extract of *Adathoda vasica* in all its solvents have shown antibacterial activity. So this plant extract with its suitable solvents, for selected microbes can be

exploited as bio pesticide agent in future and also can be recommended for pharmaceutical use.

CONCLUSION:

It is concluded that the plant extract possess microbial activity against tested organisms. The zone of inhibition varied suggesting the varying degree of efficacy and different phytoconstituents of herb on the target organism. The antimicrobial activity of the plants may be due to the presence of various active principles in their leaf. Further studies are needed to isolate and characterize the bioactive principles to develop new antimicrobial drugs.

REFERENCES:

- 1.Chung PY, Chung LY, Ngeow YF *et al.*, Anti-microbial activities of Malaysian plant species. *Pharm Bio.* 2004; 42: 292-300.
- 2.Nair R and Chanda SV. Antibacterial activity of some medicinal plants of Saurashtra region. *J. Tissue Res.* 2004; 4:117-120.
- 3.De Boedr HJ ,Kool, A *et al.*, . Antifungal and Antibacterial activity of some herbal remedies from Tanzania. *J. Ethnopharmacol.* 2005; 96: 461-469.
- 4.Nair R, Kalariya T and Chanda S. Antibacterial activity of some selected Indian medicinal flora. *Turk J Biol.* 2005; 29: 41-47.
- 5.Bauer J, Roias R and Bustamante B. Antimicrobial activity of selected Peruvian medicinal plants. *J Ethnopharmacol*, 2003; 88: 199-204.
- 6.Edeoga HO, Okwu DE and Mbaebie BO. Phytochemical constituents of some Nigerian

- medicinal plants. African Journal of Biotechnology, 2005; 4: 685-688.
7. Scalbert A. Antimicrobial properties of tannin. Phytochemistry. 1991; 30: 3875-3883.
8. Chung KT, Wong TY, Wei Y, Huang YW and Lin Y. Tannins and human health. A review. Crit Rev Food Sci Nutr, 1998; 8: 421-464.
9. Geissman TA. 1963. Flavonoid compounds, tannins, lignins and related compounds. In: Florkin Mand Stotz EH (eds). Pyrrole pigments, Isoprenoid compounds and Phenolic plant constituents, New York, USA: Elsevier Press, pp 265.
10. Cowan M.M. Plant products as antimicrobial agents. Clin Microbiol Rev. 1999; 12: 564-582.
11. Anjarai J and Bhatt G, A Glossary of selected indigenous medicinal plants of India, Nature Heals, SRISTI, Ahmedabad, 1995, p. 11.
12. Ali M. 1998. Text book of Pharmacognosy. CBS Publishers, 2nd Edn, New Delhi, p. 355.
13. Satakopan S and Thomas PJ, Note on adulterant of Vasaka, Indian J Pharm. 1970; 32 (3): 66-67.
14. Atal CK. 1980. Chemistry and Pharmacology of Vasicine – A new oxytocic and abortifacient, Indian Drugs, 15 (2): 15-18.
15. Shabir AL, Yadav AS, Ajit KS, Malik T, Yogesh B and Raghuvanshi DK. A review on *Adhatoda vasica* Nees- An important and high demanded medicinal plant. Indo American Journal of Pharm Research. 2013; 3(3): 2600-2615.
16. Harborne JB. 1973. Phytochemical Methods. Chapman and Hall, Ltd., London, pp. 49-188.
17. Trease GE and Evans WC. 1989. Pharmacognosy. 13th edition, Balliere Tindall, London, pp. 176-80.
18. Sofowora A. 1993. Screening plants for bioactive agents. Medicinal plants and traditional medicinal in Africa. 2nd Ed. Spectrum Books Ltd, Sunshine House, Ibadan. Nigeria, pp. 134-156.
19. Lutterodt, GD, Imai A, Basheer RH, Baharudian HM. Antimicrobial effects of *Psidium guajava* extracts as one mechanism of its anti-diarrhoeal action. Malaysian J. Med Sci. 1999; 6(2): 17-20.
20. Marjorie MC. Plant Products as Antimicrobial Agent. Clin. Microbiol Rev. 1999; 12(4): 564-582.
21. Kumar R, Singh NP. Effect of Tannins in pala leaves (*Ziaphus nummularia*) on ruminal proteolysis. Indian J Anim Sci, 1984; 54: 881-884.
22. Tona L, Kambu K, Ngimbi N, Cimanga K and Vlietinck AJ. Antimicrobial and phytochemical screening of some Congolese medicinal plants. J. Ethnopharma. 1998; 61: 57-65.
23. De N and Ifeoma E. Antimicrobial effects of components of the bark extracts of Neem. J. Technol. Dev. 2002; 23-28.
24. Ketkar AY and Ketkar CM. 1995. Various Uses of Neem product: Medicinal Uses Inducing Pharmacology in Asia. In: Chmutterer HS (Ed.). pp. 518-525.
25. George M, Venkatraman PR and Pandalai KM, Investigations on plant antibiotics, part II, A search for antibiotic substances in some Indian medicinal plants. J Sci Ind Res. 1947; 6 B: 42-46.
26. Rabe T and Van Staden J. Antibacterial activity of South African Plants used for medicinal purposes. J. Ethnopharmacol. 1997; 56: 81-87.
27. Parekh L and Chanda S. *In vitro* antimicrobial activities of extracts of *Launaea procumbens* Roxb. (Labiataceae) *Vitis vinifera*, L. (Vitaceae) and *Cyperus rotundus* L. (Cyperaceae). Afr. J. Biomed. Res. 2006; 9(2): 89-93.
28. Hassian- Eshrat, HMA. Hypoglycaemic, hypopolydemic and antioxidant properties of curcumin from *Abroma auguta* streptozotocin induced diabetes. Indian J. Clin. Biochem. 2002; 17(2): 33-43.
29. Obbonnia SO, Enwuru N, Onyemenen EU, Oyedele, GA and Enwuru CA. Phytochemical evaluation and antibacterial profile of *Treculia africana* decna bark extracts on gastrointestinal bacterial pathogens. Afr. J. Biotechnol. 2008; 7(10): 1385-1389.
30. Mehrangiz KK, Seyed AE, Masoud SG, Esmaeel AS, Amirhossein S Asi Biomed. 2011; 5: 63-68.
31. Maria Lysete AB, Maria Raquel FL. Ann Clin Microb Antimic. 2009; 8: 16.
32. Juliane TDL, Jackson RGS, Kelly S, Ana Silvia SC *et al*. Selective spasmolytic effect of a new furano flavoquinone derivative from diplotropin on guinea-pig trachea. J Chem Pharm Res. 2011; 3(1): 249-258.
33. Ahmed el-HM, Nour BY, Mohammed YG, Khalid HS. Antiplasmodial activity of some medicinal plants used in Sudanese folk-medicine. Env Health Insts. 2010; 4 (4): 1-6.
34. Arokiyaraj S, Perinbam K, Agastian P, Kumar RM. Phytochemical analysis and antibacterial activity of *Vitex agnus-castus*. Int J Green Phar. 2009; 3 (2): 162-164.