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## Research Article

# Evaluation of Phytochemical and Anti-Bacterial Activity of *Bauhinia vahlii*

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## ABSTRACT

*Bauhinia vahlii* is a medicinal plant with therapeutic potential, belonging to the family Fabaceae, and is also known as Thanjedu, recognized for its antibacterial activity. Recently, many biological activities of *Bauhinia vahlii* L. seeds have been reported, including traditionally, said to be medicinal because it can relieve inflammation irritation causes increased passing of urine and gives tonic effects and also cucurbit pepo (pumpkin), *Bauhinia vahlii* Colocynths (bitter cucumber), among from that I have been taken *Bauhinia vahlii* seed (Thanjedu) is one of most effective and potential anti-bacterial activity, particularly in its seed. The result of the antibacterial activity indicates that the effects of Thanjedu seed extract were tested on phytochemicals, and the results showed the presence of alkaloids, flavonoids, corticosteroids, saponins, and tannins in the extracts of Thanjedu. Among those results, alkaloids and flavonoids were the most effective compounds for the antibacterial activity in higher plants. Apart from that, saponins show the most responsible and effective compound for antibacterial activity of Thanjedu. In this study, two solvents, chloroform seed extracts and acetone seed extract from Thanjedu L., have been tested for Anti-bacterial activity. The antibacterial activity of the extract was examined against Gram-positive and Gram-negative bacteria by measuring the zone of inhibition. Results showed acetone extract was increased linearly increasing different concentration of extracts (25 mg/ml, 50 mg/ml, 75 mg/ml, 100 mg/ml) against two pathogenic strains (*Escherichia coli*, *Staphylococcus aureus*), by disc diffusion method, as compared with standard drug such as ciprofloxacin and control as acetone, the antibacterial activity of acetone was good. Both organisms used for screening have similar sensitivity.

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**KEYWORDS:** Anti-antibacterial activity, Thanjedu L. seed. Ciprofloxacin, *Escherichia coli*, and *Bacillus Subtilis*.

## 1. INTRODUCTION

Although they mentioned "*Bauhinia racemosa* Vahl," the name *Bauhinia vahlii* Wright & Arn was first legitimately published in the Isle of Wight without any description or diagnosis, which made a passing reference to the effectively published description by Vahl that came before. Therefore, contrary to

what is usually believed in the majority of published literature, *Bauhinia* was not truly published in Wright & Arn. for the first time. The fact that Wright & Arnott discovered Vahl had incorrectly applied the binomial *Bauhinia racemosa* Lam. to a specimen that needed to be classified as a new species is the cause. So, they gave Vahl a new name to honour him. Only the

Schumacher specimen from "India Orientali" and the plate provided in Vahl (1794) qualified as original materials because Wight only cited the no 628, i.e., Wight's herbarium specimens to which the no 628 is associated, in the protologue of *Bauhinia vahlii*. *Bauhinia sericea*, *Phanera vahlii* (Wight & Arn) Benth, and *Bauhinia vahlii* Wright & Arn, Wight and Arn. Are the three names on the herbarium sheet? The locale had been listed as India Orient; however, a note by A. Fox Maule (AFM) from 1984 explains that the epithet *sericea* was changed to *racemosa* in Vahl's manuscript. Because the calyx was 5-lobed rather than 2 & 3-lobed, the Vahl specimen could not be perfectly matched with the Indian sample. Instead of being narrowly elliptic, the ovary was widely elliptic. While Rexburg's description of the nature of the splitting of the calyx and the shape of the ovary of this species was totally in agreement with Banyopadhyay, Wight & Arnott only note the calyx shape, which is elliptical splitting to the base. (1)

### Classification

The following describes *Bauhinia vahlii*'s taxonomic position:

Kingdom: Plants

Sub kingdom: Tracheobionta

Super division: Spermatophyta

Division: Magnoliophyta

Class: Magnoliopsida

Subclass: Rosidae

Order: Fabales

Family: Fabaceae, Caesalpinaceae

Genus: *Bauhinia*

Species: *Vahlii*, *Variegata* L

### Vernacular names

*B. vahlii* is a giant climber. It goes by several names in many languages

**Sanskrit:** Asmantaka, Malanjhana, Phalgu.

**Assamese Nak:** Kati lewa, Shonapushpaka

**Bengali:** Chehur lata Shimool, Kanchana

**Hindi Malu:** Jallaur, Jallur, Mahul

**Kannada:** Chambolli, Kanchavala, Bilimanda

**Tamil:** Mandarai, Adda, Kattumandarai, Kattaki, Kanjani

**Telugu:** Madapu, Adattige, Adavimandaramu, Devakanchanam

### Origin and geographical description

It is a medium-sized tree called *B. vahlii*. It is deciduous and grows to a height of around 10 to 12 metres. Most of it is grown in tropical areas. There are 600 species in the genus *Bauhinia*, which includes shrubs, trees, and vines. Typically, it is grown as an ornamental plant. It flourishes all over China and India. It is a dependable greenhouse species that thrives in the Himalayas at an elevation of 3000 metres [4]. The Fabaceae (Caesalpinia eae) family member *B.vahlii* is also known as Mountain Ebony (English), Rakta Kanchan (Marathi), and kachnar (Hindi). One of the most prevalent types of Indian *Bauhinia* is the giant climber *Bauhinia vahlii*. The species is found in Assam, Central India, Bihar, the Eastern and Western Ghats, and the Sub-Himalayan region up to 3000 metres above sea level. In Madhya Pradesh, Orissa, and Andhra Pradesh, leaves are collected [2]



Fig 1: *Bauhinia vahlii*

*Escherichia coli*, commonly abbreviated *E. coli*, is a gram-negative, rod-shaped bacterium that is commonly found in the lower intestine of warm-blooded organisms (endotherms). Most *E. coli* strains are harmless and are occasionally responsible for product recalls due to food contamination [4]. The harmless strains are part of the normal flora of the gut, and can benefit their hosts by producing vitamin K and by preventing the establishment of pathogenic bacteria within the intestine. *E. coli*

and related bacteria constitute about 0.1% of gut flora, and fecal-oral transmission is the major route through which pathogenic strains of the bacteria cause disease. Cells can survive outside the body for a limited time, and environmental samples for fecal contamination. There is, however, a growing body of research that has examined environmentally persistent *E. coli*, which can survive for extended periods outside of the host [5].



Fig. 2: *Escherichia coli*, a gram-negative bacterium.

*Bacillus subtilis* is a Gram-positive bacterium, rod-shaped, and catalase-positive. It was originally named *Vibrio subtilis* by Christian Gottfried Ehrenberg, and renamed *Bacillus subtilis* by Ferdinand Cohn in 1872 (*subtilis* being the Latin for "fine") [6]. *B. subtilis* cells are typically rod-shaped, and are about 4-10 micrometers ( $\mu\text{m}$ ) long and 0.25-1.0  $\mu\text{m}$  in diameter, with a cell volume of about 4.6  $\mu\text{L}$  at the stationary phase. As with other members of the genus *Bacillus*, it can form an endospore to survive extreme environmental conditions of temperature and desiccation. *B. subtilis* is a facultative anaerobe under certain conditions. *B. subtilis* is heavily flagellated, which gives it the ability to move quickly in liquids. *B. subtilis* has proven highly amenable to genetic manipulation and has become widely adopted as a model organism for laboratory studies, especially for sporulation, which is a simplified example of cellular differentiation. The number of spores found in the human gut was too high to be attributed solely to consumption through food contamination [7]. *B. subtilis* appears in normal honey bee gut flora in some bee habitats.



Fig. 3: *Bacillus subtilis* is a Gram-positive bacterium

## Historical Aspects

### Vedic period

Vedic and Samhita eras. In the Ayodhyakanda, Sundara Kanda, and Yuddakanda of the Valmiki Ramayana of the Rig Veda, Kodivara flowers are mentioned in literature. Kodivara is described in Varivamsa as a *B. Variegata* tree with lovely blossoms. Vedic literature views it as a stem that should not be used in ceremonies.

**Charaka Samhita:** In Sutrastana's Vamanapoga Desaimani, Kodivara is addressed. Kodivara is also mentioned in the Samhitas and Chakrapani. According to a report, Kodivara blossoming happens in Sarat ritu.

**Susrutha Samhit:** Both the Kashaya Varga and the Urdwa Bhagaharangana mention Kodivara. Rakapitta chikitsa uses kodivara leaves. Devakanchanara was mentioned in Kalpastana for sarpa visha chikitsa. In addition, he recommended Kodivara flowers for internal bleeding. Karbudhara was viewed by Dalhana as a specific type of kanchanara or slesmataka.

**Astanga Hrudaya:** For arsha chikitsa, kovidara root powder was utilised. Kovidara Picchabasti administered treatment for rectal prolapse. For the treatment of fever, anorexia, goitre, malignant tumours, and abdominal enlargement, kovidara flower decoction was used.

## Botanical description:

### Leaves

This enormous climber has thickly hairy branches and circinate tendrils that are typically opposite the foliage. Leaves are petiolate and alternating, orbicular in shape, 7.5 to 9 cm long, cordate at the base, lobed at the apex, 10 to 46 cm long and nearly as wide, sparsely and densely hairy on the top and below surfaces. A terminal inflorescence, subcorymbose, thickly hairy raceme with persisting bracteoles. The leaves vary in size, typically up to 18 inches in diameter, as broad as long, profoundly cordate, 11 to 15 nerved, cleft through approximately 1/3 of the length, sub-coriaceous, dark green and glabrescent above, more or less downy beneath, lobes obtuse, rounded petiole 3 to 6 inches long, strong [3, 6].

### Flowers

The plant produces several axillary raceme inflorescences, each of which has 20–35 blooms, the majority of which are in the bud state and have three or more fully grown flowers. The flowers are huge, zygomorphic, white when young, turning buff as they age, and on long cylindrical pedicels. When fully blown, they measure 10-12 cm long and 4-5 cm wide. The calyx has thick hairs and is made up of two lobes that are created by the remaining three green sepals and two combined green sepals that comprise the calyx. A standard petal is encased within the lateral ones (wings) of the corolla's five white, densely hairy petals, which is a frequent morphological feature for the Caesalpinae subfamily [9, 10]. Two little infertile staminodes and three free, lengthy, fertile stamens

make up the androecium. Recently, several taxonomists separated the genus *Phanera* from *B. vahlii* based on the number of fertile and sterile stamens, among others.

### Stem

The main stem (trunk) is robust, solid, and cylindrical with thick brown cork showing longitudinal fissures and transverse cracks. It grows obliquely or vertically for around 50–70 cm before continuing primarily horizontally, reaching lengths of up to 1.5–3 m with bases that are between 22–28 cm in diameter. The stem subsequently splits into smaller branches that are between 17 and 20 metres long and 10 to 15 centimetres wide, which continue to develop horizontally and/or climb vertically on adjacent supports. The monopodial branching has pairs of revolute tendrils, petioles, leaves, and/or inflorescences on each branch. The surfaces of old branches are rough, fissured, and cracked, and they are cylindrical and greyish brown in colour. Younger branches climb using many pairs of tendrils, are flexible, green, densely hairy, 0.5-1 cm in diameter, cylindrical (deeply grooved when very young), and fracture with fibrous tissue. The morphological segregation of *B. vahlii* to the genus *Phanera* was targeted by the presence of tendrils in addition to the quantity and fertility of stamens [11].

### Fruit

Fruit is a 20-30 cm long, flat, woody pod with fine rusty hairs. Pod woody, dehiscent, reddish velvety, and 6–12 seeded, measuring 22.5-30 cm in length, 5-7.5 cm in width. Seeds are flat, dark brown, polished, and 2.5 cm in diameter.

### Cultivation and Collection

When given the right conditions, *B. Variegata* can be naturally propagated through its seeds; however, artificial multiplication is accomplished by stump planting, or the direct sowing of seeds. Branch cuttings typically have trouble establishing roots, but when auxins are applied in August, November, and February, these cuttings thrive. Direct sowing can be carried out in lines with a 3 m separation. Within a week of the beginning of the monsoon rains, germination begins, guaranteeing enough soil saturation. The soil ball and the plants as a whole must be transplanted. Seeds from the previous year are sown in March or April in preparation for planting out in July or August [12]. Seeds, stem planting, and branch cutting are all used to propagate the ornamental plant. In March and April, seeds are sown. In July and August, the seedlings are then replanted. When the monsoon season begins, they begin to sprout. *In vitro* regeneration was seen in explants of mature *B. variegata* nodal trees. On media enhanced with 13.3 micrometre IBA, optimal shooting was accomplished in 15-20 days. When transplanted to MS media with 4.9 micrometre IBA within 45 days, single shoots with 3-4 nodes begin to root [13].

**Flowers:** V asantha r utu.

**Flowering:** February-April.

**Fruiting:** May-June [13].

**Chemical constituents** [14]

The roots of *B. Variegata* were found to contain flavonoids such as flavanone, 5, 7-dimethoxy-30, 40 methylenedioxyflavanone, and a novel dihydrodibenzoxepin 5, 6-dihydro-1, 7-dihydroxy-5, 6-dihydro-1, 7-dihydroxy-methylidibenzoxepin [32]. The roots of *B. Variegata* were utilized to produce the new flavonol glycoside 5, 7, 3', 4'-tetrahydroxy-3-methoxy-7-O- $\alpha$ -lpharhamnopyranosyl (1-->3) - O-beta-galactopyranoside. A triterpene saponin that was isolated from the leaves of *B. Variegata* Linn. It was credited with the plant's anti-inflammatory and antinociceptive qualities. From *B. Variegata*, a phenanthraquinone called bauginione has been discovered [15].

## 2. MATERIALS AND METHODS

**Materials:** *Bauhinia vahlii* (Mandarai) seeds were procured from the local market, acetone and chloroform were procured from S.D. Fine chemicals, Mumbai. Ciproflaxin drug was obtained as a gift sample from Aurobindo Pharma Ltd., Hyderabad. Clinical isolates of *Staphylococcus* sp and *Escherichia coli* were obtained from Acharya Nagarjuna University, Guntur. All the other chemicals were procured from St Xavier's Institute of Pharmacy, Guntur.

**Methods:** *Bauhinia vahlii* (Mandarai) seeds of good quality were purchased from the local market, and eight watermelon fruits were purchased. They were cleaned, and then cut and opened to obtain the seeds, washed and air dried for 7 days, then pulverized using a mortar and pestle under aseptic conditions and ground to powder using a blender. Powdered seed materials were then weighed (175 g) and kept in air-tight containers until further usage.

### Extraction of seeds

#### Chloroform extract

10 g of seed powder were weighed and transferred into a Soxhlet apparatus, and the seed powder was extracted with chloroform at 35°C for 3-4 cycles. The extract was collected, and the chloroform was evaporated after extraction by using a rotary evaporator connected to a vacuum pump. The final extract in semi-solid form was dried by placing it in desiccators. A rotary evaporator, yielding the extracted compound and its percentage yield, is calculated respectively and used for further extracted crude drug phytochemical evaluation studies.

#### Acetone extract

10 g of seed powder were weighed and transferred to the Soxhlet apparatus, and the seed powder was extracted with acetone at 35°C for 3-4 cycles. The extract was collected, and the acetone was evaporated after extraction by using a rotary evaporator connected to a vacuum pump. The final extract in semi-solid form was dried by placing it in desiccators. A rotary evaporator, yielding the extracted compound and its percentage yield, is calculated respectively and used for further extracted crude drug phytochemical evaluation studies.



### Phytochemical Screening

Phytochemical analysis of various solvent extracts of *Bauhinia vahlii*

S. No	TEST	ACETONE	CHLOROFORM
1.	alkaloids	+++	++
2.	corticoids	+	+
3.	tannins		+
4.	flavonoids	+++	++
5.	saponins	++++	

The result of antibacterial activity indicates that the effects of *Bauhinia vahlii* seed extract were tested on phytochemicals. Results showed the presence of alkaloids, flavonoids, corticoids, saponins, and tannins in the extracts of *Bauhinia vahlii*. Among the results, alkaloids and flavonoids were the most effective compounds for the antibacterial activity in higher plants. Apart from that, saponins are the most effective compound for antibacterial activity of *Bauhinia vahlii*.

### Anti-bacterial activity:

The antibacterial activity of *Bauhinia vahlii* was carried out against the *Staphylococcus aureus* and *Escherichia coli*. The antibacterial activity of the compounds can be assessed by the disc diffusion method.

### Preparation of Paper Discs:

Discs of 5-6 mm in diameter were punched from No.1 Whatman filter paper with a sterile corn borer of the same size. These discs were sterilized by heating at 140°C for 60 min.



Fig 3: Whatman filter paper

### Preparation of Nutrient Agar Media:

Nutrient agar, which served as the basal medium, was prepared by dissolving.

1. Peptone (Bacteriological) - 20 gm
2. Meat extract (Bacteriological) - 8.5 gm
3. Agar - 25 gm
4. Distilled Water up to - 500 ml

Nutrient agar medium was prepared by dissolving all these ingredients in water, and then subjected to sterilization using an autoclave at 15 lbs pressure for 20 mins, and the pH was adjusted to 6.



Fig 5: Nutrient agar medium preparation

### Disc Diffusion Method:

The antibacterial activity of the compounds can be assessed by the disc diffusion method. The sterilized media (about 15-20 ml) were transferred to sterilized Petri dishes, which were previously labelled, and then allowed to solidify after the inoculation of microorganisms was done by using a cotton swab. Then the discs that were previously prepared were carefully kept on the solidified medium, and these Petri dishes were kept as such for 1 hour at room temperature, and then kept for incubation at 37°C for 24 hrs in an incubator. The zone of inhibition was measured in millimeters (mm) after 24 hrs.

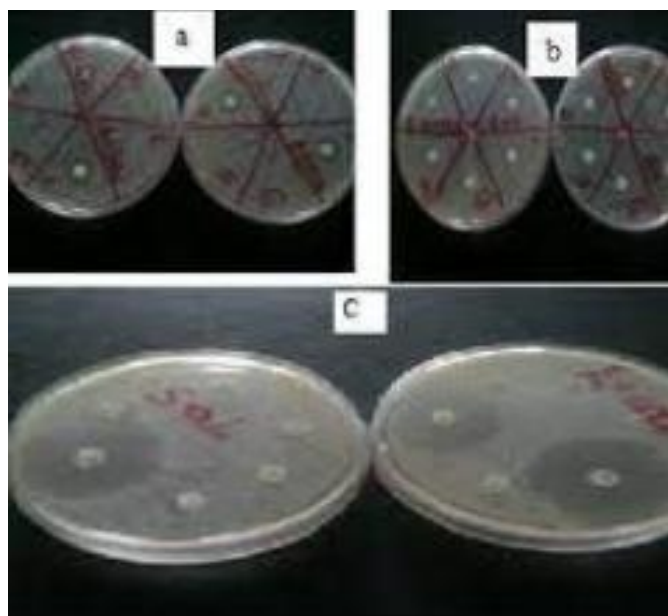
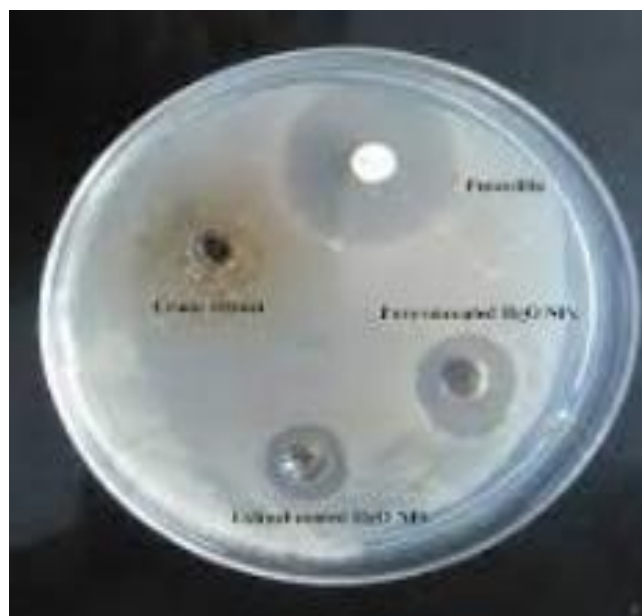
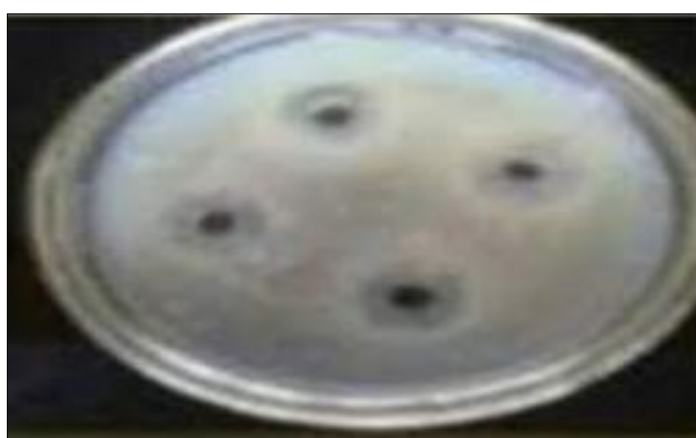


Fig. 6: Antibacterial activity

Fig. 7: Antibacterial activity of *Bauhinia vahlii* seed against *E. coli*, Ciprofloxacin against *E. coli*Fig. 8: Antibacterial activity of *Bauhinia vahlii*Fig. 9: Antibacterial activity of Seeds against *Bacillus subtilis*. Ciprofloxacin against *Bacillus subtilis*

### 3. RESULTS AND DISCUSSION

Preliminary phytochemical results showed the presence of Alkaloids, Flavonoids, Corticoids, Saponins, and Tannins in the extracts of *Bauhinia vahlii*. Available reports tend to show that alkaloids and flavonoids are the responsible compounds for the antibacterial activity in higher plants. Saponins are responsible for the antibacterial activity of *Bauhinia vahlii*.

The antibacterial activity of the extract was examined against Gram-positive and Gram-negative bacteria by measuring the zone of inhibition. The antibacterial activity of chloroform extract of the seeds of *Bauhinia vahlii* was studied in different concentrations (25 mg/ml, 50 mg/ml, 75 mg/ml, 100 mg/ml) against two pathogenic strains (*Escherichia coli*, *Staphylococcus aureus*), by the disc diffusion method. The result of antibacterial activity is presented in the above figures.

The antibacterial activity of the extract increased linearly with an increase in the concentration of the extract (mg/ml). As compared with standard drugs such as Ciprofloxacin and control, such as acetone, the antibacterial activity of the acetone extract was good. Both organisms used for screening have similar sensitivity. The inhibitory effect of *Bauhinia vahlii* seed acetone extract showed at 25, 50, 75, 100 mg/ml (15, 17, 18, 20 mm) for *Escherichia coli* and (14, 16, 17, 21 mm) for *Staphylococcus aureus*, for bacterial strain, respectively.

### 4. CONCLUSION

The result of antibacterial activity indicates that the effects of *Bauhinia vahlii* seed extract were tested on phytochemicals. The results showed the presence of alkaloids, flavonoids, corticoids, saponins, and tannins in the extracts of *Bauhinia vahlii*. Among those results, alkaloids and flavonoids were the

most effective compounds for the antibacterial activity in higher plants. Apart from that, saponins are the most effective compound for antibacterial activity of *Bauhinia vahlii*. The antibacterial activity of the extract was examined against Gram-positive and Gram-negative bacteria by measuring the zone of inhibition. Results showed acetone extract was increased linearly increasing different concentration of extracts (25 mg/ml, 50 mg/ml, 75 mg/ml, 100 mg/ml) against two pathogenic strains (*Escherichia coli*, *Staphylococcus aureus*), by disc diffusion method, as compared with standard drug such as ciprofloxacin and control as acetone, the antibacterial activity of acetone was good. Both organisms used for screening have similar sensitivity.

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