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World Journal of Science and Research



Article

Botany

PHYTOCHEMICAL SCREENING AND IN VITRO ANTIPROLIFERATIVE ACTIVITY OF *Bauhinia variegata* (L.) STEM EXTRACT

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Received on 10th Jan. 2025;

Revised on 25th Feb. 2025

Online 16th March. 2025

ABSTRACT

The goal of the current study was to evaluate the antidiabetic properties and diverse phytochemical profiles of *Bauhinia variegata* stem extracts. Tannin, saponins, flavonoids, polyphenol, glycoside, anthraquinone and coumarins in aqueous and ethanol extract while steroids and alkaloids were only present in ethanolic extract of *Bauhinia variegata* stem. Terpenoids was present in aqueous extract only. The seed extract of *Bauhinia variegata* stem contained a notable amount of flavonoids and polyphenols. Using histochemical, UV visible, and fluorescence methods, this study further verified the phytochemicals present in the *Bauhinia variegata* stem extract. Stem of *Bauhinia variegata* reduced yeast cell growth based on concentration. The stem of *Bauhinia variegata* are a significant source of phenolic compounds, and the results indicated that they may have antiproliferative potential.

Keywords: *Bauhinia variegata* stem, Qualitative, Quantitative, Histochemical, UV visible, Fluorescence and antiproliferative.

Citation: M. Dhavadharshini and S. Beatrice Florence Febronia. Phytochemical screening and in vitro Antiproliferative activity of *Bauhinia variegata* (L.) stem extract. World Journal of Science and Research. 10 (1): 08-15 (2025)

INTRODUCTION

Cancers are a class of diseases characterized by aberrant cell proliferation. Without any checkpoints in place, the condition could continue to worsen and eventually cause premature death. People of various ages, socioeconomic backgrounds, and races may be impacted by them, and they can appear anywhere in the body. Globally, cancer is the primary cause of morbidity and mortality. Cancer is one of the leading causes of death worldwide and a significant public health concern. In the world, cancer is one of the main causes of death. Many

of these deaths, however, are preventable. Public health initiatives like vaccination against viruses that cause cancer and healthy lifestyle choices like quitting smoking can prevent between 30 and 50 percent of malignancies. Others are curable, treatable, and early detectable. Good palliative care should alleviate patients' pain, even in cases of advanced cancer (WHO, 2022). Cancer is defined biochemically as any alteration or

anomaly in the cell division process. As a result, the cancer cells create mechanisms to live longer than they should and divide in an irregular manner through mitosis.

Model organisms like yeast are especially attractive due to the shortcomings of existing cancer models based on mammalian cells and methods for changing gene function in these cells (Wassmann and Benezra, 2001). Many human proteins that are not conserved also interfere with the course of the yeast cell cycle and are therefore good targets for cell-based drug screening in yeast. Yeast can be highly helpful for the development of anticancer drugs if target enzymes, pathways, or settings are carefully chosen (Simon and Antonio Bedalov, 2004). Herbal medicine presents an alluring alternative for the treatment of cancer because of the severe side effects of radiation and chemotherapy as well as the emergence of drug resistance (Lynch and Berry, 2007). In medicine, plants have long been used to cure a variety of illnesses. Because of their safety and few side effects, natural plant-based medicines have been more popular in recent years (Kma, 2013). Numerous anticancer bioactive chemicals have been discovered in plants. Natural compounds made from medicinal plants are present in over 75% of anti-cancer medications that are prescribed (Craig, 1999). In this regard, the current work is an attempt that uses *Bauhinia variegata* stem for phytochemical screening, in vitro antiproliferative activity.

MATERIALS AND METHODS

Collection of plant materials and preparation for extract

The *Bauhinia variegata* stem were collected in January 2025 from Thamarankottai, Thanjavur, Tamil Nadu. Take one gram of stem powder in the extract prepared in 50 ml of ethanol and aqueous solvent, the extract shake it well for 30 minutes by free hand and wait for 24 hours. After extracts were filtered using whatman filter paper No.1 and filtrate used for further analysis.

Phytochemical screening

The extract was subjected to chemical tests using standard protocols to determine its constituents, as outlined by Sofowara (1993), Trease and Evans (1989), and Harborne (1973 and 1984). Edeoga et al. (2005) used a spectrophotometric method to determine the total phenols. Boham and Kocipai-Abyazan's (1994)

method is used to determine flavonoids. Histochemical assays (Gersbach et al., 2001; John Peter Paul, 2014). Assessment of plant powder's fluorescence behavior (Rao et al., 2016). UV analysis of the visible spectrum. Functional groups were identified using Ahluwalia and Dhingra's (2004) methodology.

In-vitro Anti-proliferation followed by yeast cell culture method

The assay protocol followed was as previously described by Periyanyagam et al. (2013).

RESULTS

Phytochemicals analysis

The present study was carried out on the *Bauhinia variegata* stem extract revealed the presence of medicinally active constituents. The phytochemical characters of *Bauhinia variegata* stem investigated and summarized in table 1 and figure 1. The presence of tannin, saponins, flavonoids, polyphenol, glycoside, anthraquinone and coumarins in both aqueous and ethanol extract while steroids and alkaloids were only present in ethanolic extract of *Bauhinia variegata* stem. Terpenoids was present in aqueous extract only. Significant amount of flavonoids (50.00 mg/gm) and total phenol (246.00 mg/gm) were found in *Bauhinia variegata* stem extract (Table 2). The importance of histochemistry in solving critical biosystematic problems is as popular as the use of other markers. According to botanical literatures, the use of histochemical characters in taxonomic conclusions is now a common practice. Table 3 and figure 2 represents histochemical studies of *Bauhinia variegata* stem powder. This study further confirmed the presence of phytochemicals in *Bauhinia variegata* stem extract. The fluorescence analysis of powdered *Bauhinia variegata* stem material was subjected to analysis under visible light, Short UV light (254 nm), Long UV light (365 nm) after treatment with various chemical and organic reagents. The fluorescence behavior was noted as in table 4. The results indicates the fluorescence compounds were present in the plant (Table 4 and Figure 3). The *Bauhinia variegata* stem was examined under visible UV-Visible spectrum. The sample was

scanned in the wavelength ranging from 340-800nm. These solutions were scanned in turn at intervals of 10 nm and the characteristic peaks were detected. The peak value of the UV-Visible was recorded. The UV spectrum profile showed the peaks at 340, 400 and 640 nm and identified phytochemicals are Flavonoids, alkaloids, phenolic, terpenoids and chlorophyll respectively

(Table 5). Figure 4 shows the absorption spectrum of *Bauhinia variegata* stem extract and these are almost transparent in the wavelength region of 340-800 nm. The alcohol, phenol, aliphatic amines, aldehyde, ketone, carboxylic acid are presence in *Bauhinia variegata* stem indicating the phytochemicals (Table 6 Figure 5).

Table 1: Qualitative analysis of *Bauhinia variegata* stem extract

S. No	Phytochemicals	Aqueous extract	Ethanolic extract
1	Tannin	+	+
2	Saponin	+	+
3	Flavonoids	+	+
4	Steroids	-	+
5	Terpenoids	+	-
6	Alkaloids	-	+
7	Antroquinone	+	+
8	Polyphenol	+	+
9	Glycoside	+	+
10	Coumarins	+	+

(+) Presence and (-) Absence

Table 2: Quantitative analysis of *Bauhinia variegata* stem

Secondary metabolites	Result (mg/gm)
Flavonoid	50.00 \pm 18.23
Total Phenol	246.00 \pm 47.52

Values were expressed as Mean \pm SD.

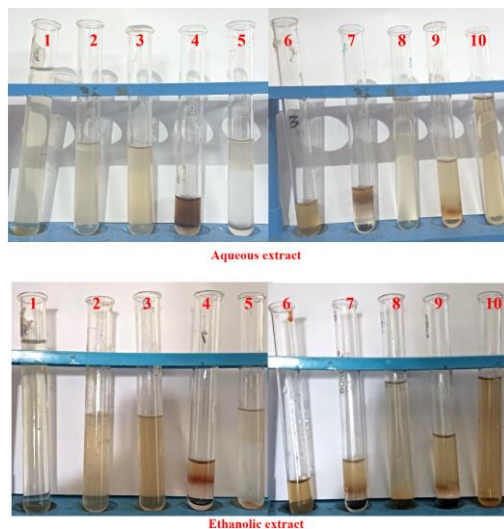


Figure 1: Qualitative analysis of *Bauhinia variegata* stem extract

(1. Tannin, 2. Saponin, 3. Flavonoids, 4. Steroids, 5. Terpenoids, 6. Alkaloids, 7. Anthroquinone, 8. Polyphenol, 9. Glycoside and 10. Coumarins)

Table 3: Histochemical studies of *Bauhinia variegata* stem powder

S. No.	Secondary metabolites	Result
1	Tannin	+
2	Flavonoids	+
3	Saponnin	+
4	Polyphenol	+

(+) Indicates Presence

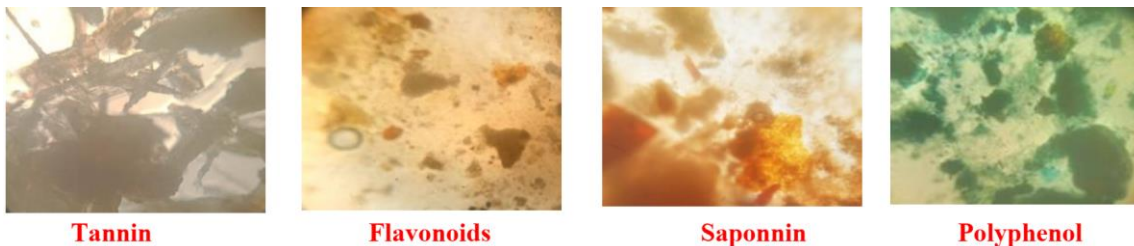


Figure 2: Histochemical studies of *Bauhinia variegata* stem powder

Table 4: Fluorescence analysis of *Bauhinia variegata* stem

S. No	Test	Visible Light	Short UV Light (254 nm)	Long UV Light (365 nm)
1	Plant powder	Ash	Ash	Black
2	Plant powder treated with distilled water	Brown	Brown	Black
3	Plant powder treated with Hexane	Ash	Ash	Black
4	Plant powder treated with Chloroform	Ash	Ash	Black
5	Plant powder treated with Methanol	Ash	Ash	Black
6	Plant powder treated with Acetone	Ash	Ash	Black
7	Plant powder treated with 1N Sodium Hydroxide	Brown	Black	Black
8	Plant powder treated with 1N HCL	Ash	Black	Black
9	Plant powder treated with sulphuric acid with equal volume of water	Black	Brown	Black
10	Plant powder treated with HNO ₃ diluted with an equal volume of water	Yellow	Light green	Black

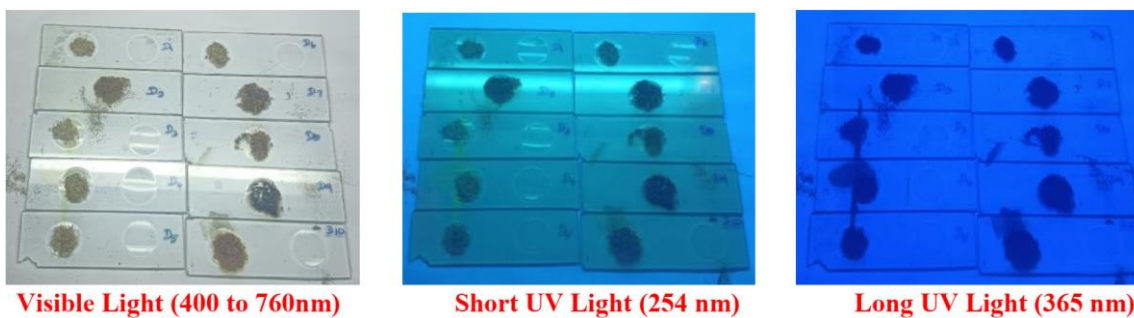


Figure 3: Fluorescence analysis of *Bauhinia variegata* stem

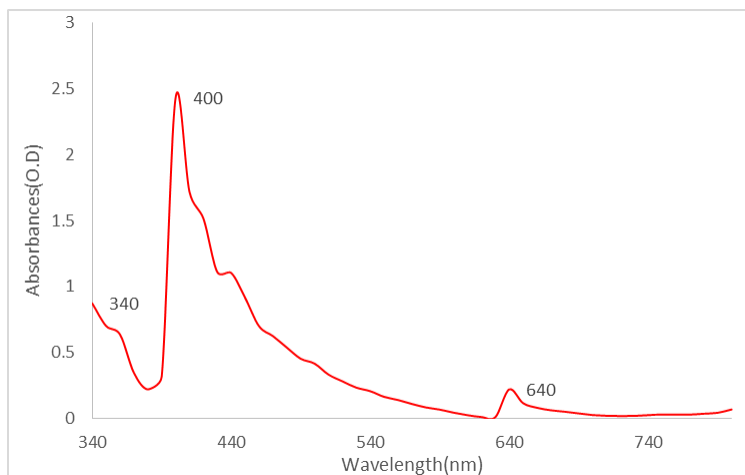


Figure 4: UV visible spectrum analysis *Bauhinia variegata* stem extract

Table 5: UV visible spectrum analysis *Bauhinia variegata* stem extract

S. No.	Absorption maxima (Wavelength ranges) nm	Phytochemical compounds (metabolites)
1.	340	Flavonoids, alkaloids and phenolic
2.	400	Terpenoids
3.	640	Chlorophyll

Table 6: Functional group analysis of *Bauhinia variegata* stem

S. No	Functional group	Results
1	Alcohol	++
2	Phenol	++
3	Aliphatic amines	++
4	Aldehyde	+
5	Ketone	+
6	Carboxylic acid	++

(+) Presence; (++) present with high intensity of the colour

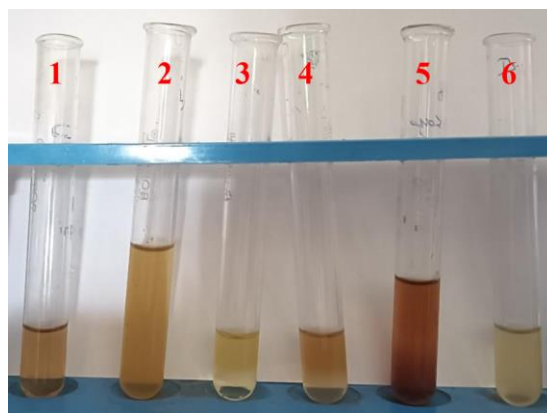


Figure 5: Functional group analysis of *Bauhinia variegata* stem

Plants have a wide range of phytochemical compounds that are rich in antioxidant activity, including vitamins, terpenoids, phenolic acids, lignins, stilbenes, tannins, flavonoids, quinones, coumarins, alkaloids, amines, betalains, and other metabolites (Zheng and Wang, 2001). Numerous studies have demonstrated the anti-inflammatory, anti-atherosclerotic, anticancer, antimutagenic, anticarcinogenic, antibacterial, and antiviral properties of these antioxidant substances (Sala *et al.*, 2002). Research on the pharmacological properties of secondary plant metabolites, or phytochemicals, as a source of therapeutic medicines has been extensive (Krishnaraju *et al.*, 2005). According to Panche *et al.* (2016), the amount of free hydroxyl groups in flavonoids and their location on the flavonoid skeleton both affect how strong the antioxidant activity is. We found that *Bauhinia variegata*

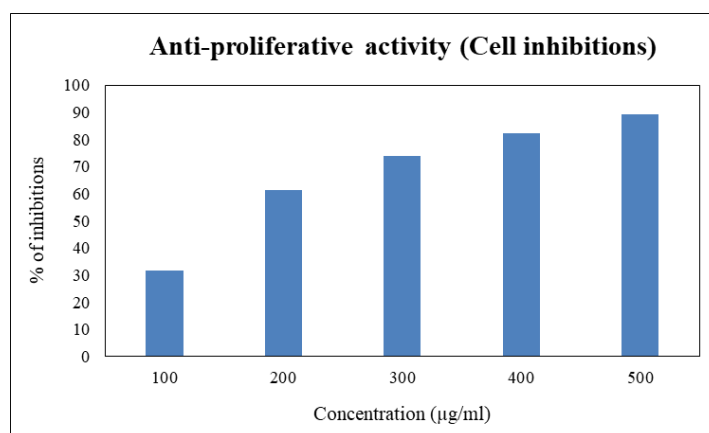
stem were a high source of flavonoids and total phenol, and that they had excellent *in-vitro* biological active characteristics overall.

***In-vitro* Anti-proliferative followed by yeast cell culture method**

Anti-proliferative activity of there was reduction in the rate of proliferation of cultured *S. cerevisiae* cells exposed to *Bauhinia variegata* stem extracts for 24 hrs as concentration of the extract increased compared to the untreated control. Proliferation of yeast cells was significantly inhibited at all the concentrations of *Bauhinia variegata* stem extracts (Table 7) examined. However, activity was observed at the concentrations depended. In this study, *Bauhinia variegata* stem extract depended on concentration of inhibited proliferation in yeast cells at all the concentrations examined compared with control (Table 7 and Figure 6, 7).

Table 7: *In vitro* Anti-proliferative activity of *Bauhinia variegata* stem extracts using yeast cell

Concentration (µg/ml)	<i>Bauhinia variegata</i> stem extracts (%)	
	Cell survival	Cell inhibitions
100	68.18	31.82
200	38.63	61.37
300	26.08	73.92
400	17.77	82.23
500	10.81	89.19
Control	100	-

**Table 6: *In vitro* Anti-proliferative activity (Cell inhibitions) of *Bauhinia variegata* stem extracts using yeast cell**

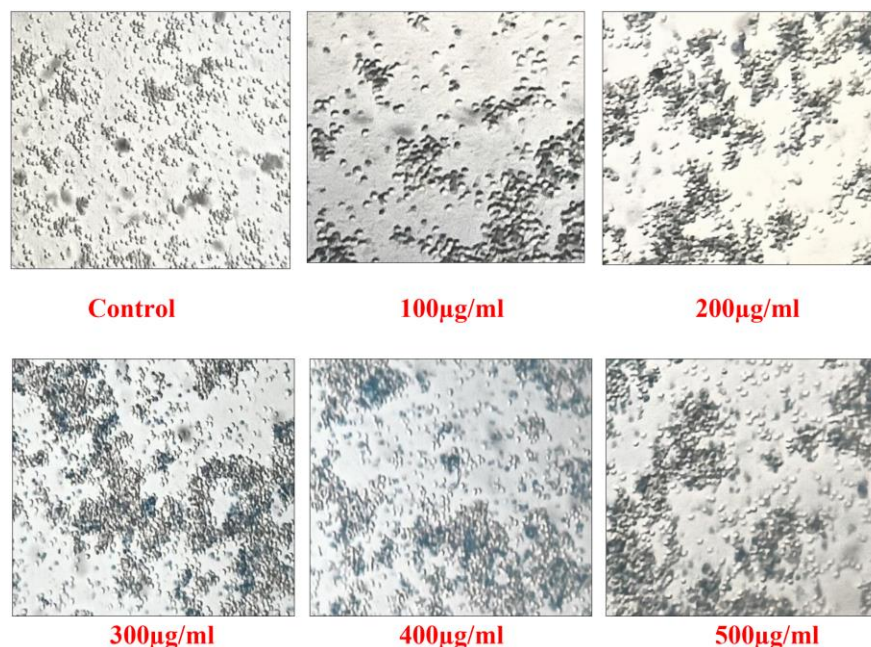


Figure 7: In-vitro Anti-proliferative activity of *Bauhinia variegata* stem extracts using yeast cell

According to Choudhari *et al.* (2020), phytochemicals naturally occurring substances derived from plants are essential resources for new medications and cancer treatment. Therefore, the cancer cells create mechanisms to live longer than they should and multiply in an irregular manner through a process called mitosis (Feitelson *et al.*, 2015). Cell lines are inhibited by its anti-proliferative activity. Numerous studies have documented the use of traditional plants from northeastern India to cure cancer (Dolui *et al.*, 2004; Sharma *et al.*, 2001; Jamir *et al.*, 1999). Historically, practically every plant part has been utilized to cure different kinds of cancer.

CONCLUSION

The phytochemicals found in several stem extracts of *Bauhinia variegata* have strong anti-proliferative properties, according to the current study. The study concludes that anti-proliferative secondary metabolites may be present in extracts of the stem of *Bauhinia variegata*.

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