

PHYTOCHEMICAL AND ANTIOXIDANT ACTIVITY OF *HEMIGRAPHIS COLORATA*

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ABSTRACT

Hemigraphis colorata (Blume) H.G. Hallier (syn: *Hemigraphis alternata*, family: Acanthaceae), is an exotic plant adapted to India. It is a versatile low-creeping perennial herb mostly valued as an ornamental plant. Since remote past, the leaves are ground into a paste and applied on fresh cut wounds to promote wound healing. Clinical studies also highlight its significant antimicrobial and anti oxidant activities. The present investigation study was focused to evaluate the pharmacognostic and preliminary phytochemical properties of *Hemigraphis colorata* leaves. The morphological and anatomical characters, quantitative microscopy, powder microscopy and the behavior of powdered sample with different reagents were recorded. Pharmacognostic study of crude drug would be essential for any advanced pharmaceutical research on this plant. The use of herbal plants in the medical system plays an important role in the treatment of illnesses. Researchers all over the globe are exploiting the medicinal plants because of their least side effects, high efficacy and safety. Exploitation of such medicinal plants has helped in treatment of ailments and its management. Among such plants *Hemigraphis colorata* is a plant that has significant part in the medicine system and has made a remarkable impact in traditional medicine treatment. The plant has been used in treatment of bacterial infection, fungal infection, haemorrhoids and as a wound healing agent. Along with these several pharmacological studies also suggest that the plant has potent antioxidant activities these activities can be a results of its vast photochemical composition which includes alkaloids, tannins, glycosides, steroids, flavonoids.

INTRODUCTION

The plants' ecology remains extremely versatile. Approximately 10% of all vascular plants have therapeutic uses. Traditional medicine is used by over 80% of people worldwide (WHO, 2022). All living things throughout the universe benefit from every portion of the plant (Bamola et al., 2018). Because they are excellent sources of numerous strong and effective medications, these medicinal plants are essential to preserving human health. These plants have been used for thousands of years to cure and prevent a wide range of illnesses, including respiratory conditions like asthma (Singh, 2023).

The Indian, Egyptian, Chinese, Greek, and Roman civilizations all used plant products to treat illnesses as well as revitalize body systems, demonstrating the items' therapeutic potential as early as 5,000 years ago (Kanta, 2019). It has been established that using plants as a source of study in the hunt for medicinal active chemicals produces a substantial amount of scientific output (Manzano et al., 2020). Physicians and pharmacists are better equipped to handle the problems that have arisen with the expansion of professional services in the facilitation of human life because they are aware of the evolution of ideas regarding the use of

medicinal plants as well as the evolution of awareness (Nishant, 2016).

Indian food utilizes an enormous number of herbs and spices, including onions, garlic, ginger, turmeric, cloves, cardamom, cinnamon, cumin, coriander, fenugreek, fennel, ajwain, bay leaf, hing, and more. All of these are used as medicine or in diet in Ayurvedic treatment (Petrovska, 2012). Brahmi and ashwagandha are examples of herbal medicines that can promote energy, improve immunity, rejuvenate body cells, and increase nutrients (Patro, 2016). Approximately 50,000 to 80,000 flowering plants are utilized medicinally worldwide. At least 15,000 of these could go extinct as a result of habitat loss and overharvesting (Roberson, 2008). Another excellent substitute for snake venom is found in the realm of plants. Historically, snake bites have been treated with folk medicine made from medicinal herbs (Roshana and Sanmugarajah, 2018).

Therapeutic potential is widely employed in India by all segments of the population, both as processed pharmaceutical products and as traditional medicines in various indigenous medical systems including Siddha, Ayurveda, and Unani. Phytochemicals are naturally occurring substances found in fruits, vegetables, and

plants that combine with fiber and minerals to fight disease. The secondary metabolites found in medicinal plants include diterpenes, alkaloids, phytosterols, glycosides, phenols, and flavonoids. The strong therapeutic power of medicinal plants is attributed to the existence of these secondary metabolites (Singh, 2023).

A crucial component of India's healthcare system is the long-standing practice of using medicinal plants as a source of medication. Due to the rising incidence of adverse drug reactions and the high expense of the modern medical system, interest in traditional remedies is fast increasing among the general population, academic community, and government. There have been surges in interest in the study of natural product chemistry while taking this into account (Singh, 2023). The therapeutic plants linked to traditional knowledge must be documented immediately because they are susceptible to shrinkage (Bisht et al., 2013). Therefore, accurate documentation, plant species identification, and herbal preparation are required to conserve this indigenous knowledge of traditional medicines.

Involvement in the cultivation of the most frequently utilized medicinal plants will be beneficial to the local populations (Jima and Moa, 2018).

PHYTOCHEMISTRY

According to Egbuna et al. (2019), phytochemistry is the study of phytochemicals produced by plants, their structural compositions, biosynthetic pathways, roles, and mechanisms of action in living systems. According to their structure and biosynthesis routes, the compounds found in plants, commonly referred to as phytochemicals, are divided into primary and secondary metabolites (Veluet al., 2018). The structural properties of primary metabolites are very similar. Additionally, they are essential for plant growth, metabolism, development, and survival. Examples of primary metabolites include proteins, carbohydrates, lipids, and nucleic acids (Erb and Kliebenstein, 2020).

Lipids play a crucial role in the integrity of cells and organelles by controlling cell metabolism. Similar to hormones, carbohydrates are signaling molecules and essential building blocks for energy production and storage. In addition to being genetic information carriers and storage components, nucleic acids stimulate the creation of proteins. Although they are not essential to a plant's life, secondary metabolites do influence how the plant interacts with its environment and, ultimately, how it survives in the ecosystem. Secondary metabolites include anthocyanins, cardiac glycosides, alkaloids, polyphenols, flavanoids, and saponins. Different plants and plant parts have different phytochemicals. They protect plants from harmful things like insects, bacteria, and diseases as well as from harsh conditions like salt and high temperatures.

They are also in charge of giving the plant its color. Conventional plant-based drugs are chosen over synthetic ones because they are less harmful to the environment and have fewer negative effects. Plant compounds known as phytochemicals have a wide range of bioactivities and many health benefits. They are also used in traditional medicine to treat a number of illnesses and conditions. According to Thacker and Ram (2024), plants that contain a variety of bioactive phytochemicals have a wide range of bioactivities, including anti-inflammatory, anti-cancer, analgesic, antioxidant, and anti-diarrheal properties.

According to East Malaysia, the mushroom species *Hemigraphis colorata* (H. colorata) has adapted to the Indian subcontinent. Red ivy, purple wavyplant, and murikooti are some of its common names. The herbaceous perennial H. colorata is an annual creeper that reaches a height of 28 to 30 cm. With serrated edges, the leaves are arranged opposite each other. The stems are upright and have a reddish brown color, while the leaf blades are silvery on top and blood crimson underneath. Historically, this plant's leaves have been used to cure bleeding, anemia, kidney stones, red diarrhea, and wound healing. Furthermore, leaf extracts can be used to hasten the healing process of new wounds (Gowthaman et al., 2024).

Taxonomical classification

Kingdom: Plantae
Phylum: Spermatophyta
Subphylum: Angiospermae
Class: Magnoliopsida – Dicotyledons
Subclass: Asteridae
Order: Scrophulariales
Family: Acanthaceae Juss. - Acanthus family
Genus: *Hemigraphis* Nees - *Hemigraphis*
Species: *Hemigraphis colorata*.

Vernacular name

Kannada: Tincture ghida
English: Cemetery plant, Aluminium plant, red flame ivy, Metal leaf, Tincture plant.
Sanskrit: Vranaropani
Malayalam/Tamil: Murikooti

MATERIALS AND METHODS

Plant samples

The leaves of the selected plant *Hemigraphis colorata* was collected from Malappuram district, Kerala. Samples were brought into laboratory and washed with running tap water for the removal of dust and other unwanted particles.

Chemicals

DPPH, linalool, gallic acid, tannic acid, 3, 5 Dinitro salicylic acid, Folin-phenol reagent, Copper sulphate, Trichloro acetic acid, 2, 4-dinitrophenyl hydrazine, Thiourea, 2,2'-dipyridyl, Ferric chloride, Sodium potassium tartarate and sodium hydroxide were

purchased from Himedia, India. And all other chemicals and reagents used are analytical grade purchased from SRL, India.

Glassware and Plasticware

Glassware from Borosil and Schott Duran; plasticware from Tarsons and Laxbro were used after washing with the detergent and rinsing in distilled water. They were then dried in hot air oven at 70°C before use.

Preparation of *Hemigraphis colorata* leaf extracts

The *Hemigraphis colorata* leaf extracts were prepared by using standard procedures as described by Harborne, (1998). The extract from *Hemigraphis colorata* was prepared using maceration method. 50 g of the fresh samples of *Hemigraphis colorata* plant leaves were grinded using mortar and pestle and percolated in 200 ml methanol for 24 hours with occasional shaking. The extracts were then filtered using Whatman no. 41 filter paper. The organic solvent filtrates were concentrated in vacuum using a rotary evaporator and the methanolic extracts were dried using water bath to obtain crude extracts. They were collected and stored for further analysis.

Evaluate the amount of flavonoids

Flavonoid was extracted and estimated by the method of Cameron *et al.*, (1993).

Procedure

10 g of the plant sample was extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through whatman filter paper No 42 (125 mm). The filtrate was later transferred into a crucible and evaporated into dryness over a water bath and weighed to a constant weight.

Estimation of Tannins

The amount of total tannin in the extracts was estimated by Folin-Denis method proposed by Polshettiwar *et al.*, (2007).

Principle

Tannins like compounds reduce phosphotungstomolybdic acid in alkaline solution to produce a blue colour complex and the colour intensity is proportional to the concentration of Tannin and measured at 700nm.

The tannins were determined by slightly modified Folin and Ciocalteu method. Briefly, 0.5 ml of sample extract is added with 0.5 ml of distilled water and added 0.5 ml of Folin Phenol reagent, 4.5 ml of 2 % sodium carbonate solution. The absorbance was measured at 725 nm. Prepare a stock standard of 100 mg /100 ml Tannic acid. Pipet out 10 ml stock and made upto 100 ml with distilled water and used as working standard. Tannic acid dilutions (0 to 1 ml) were used as working standard solutions for standard graph preparation. The results of tannins are expressed in terms of tannic acid in mg/ml of extract.

Estimation of total polyphenol:

The amount of total polyphenols in the plant extracts were estimated by the method proposed by Mallick and Singh, (1980).

Principle

Phenols react with phosphomolybdic acid in Folin-Ciocalteu reagent to produce a blue-coloured complex in alkaline medium, which can be estimated spectrophotometrically at 765nm.

Procedure

The phenols were determined by slightly modified Folin and Ciocalteu method. Briefly, to the 0.5 ml of the sample extract, 800 µl of Folin-Ciocalteu reagent mixture and 2 ml of 2 % sodium carbonate added. The total content is diluted to 7 volumes with distilled water and finally kept the tubes for 2 hrs incubation in dark. The absorbance was measured at 765 nm. Gallic acid dilutions were used as standard solutions. Prepare a stock standard of 100 mg /100 ml Gallic acid. Pipet out 10 ml stock and made upto 100 ml with distilled water and used as working standard. Gallic acid dilutions (0 to 1 ml) were used as working standard solutions for standard graph preparation. The results of phenols are expressed in terms of Gallic acid in mg/ml of extract.

Evaluate the amount of glycosides

The amount of glycoside was determined by using the method of Balbaa *et al.*, (1981).

Principle

Glycosides develop an orange red colour complex with Baljet's reagent (Picric acid in alkaline medium). The intensity (absorbance) of colour produced is proportional to the concentration of glycosides were measured at 495 nm.

Reagents

1. Standard digitoxin: 0.02% digitoxin is prepared in chloroform: methanol (1:1).
2. Baljet's reagent: Freshly prepared 95ml 1% picric acid + 5ml 10% NaOH are mixed immediately before use and filtered through a sintered glass funnel.

Procedure

1 ml of the extract and 1 ml of Baljet's reagent are taken and allowed to stand for one hour. Then dilute the solution with 10ml distilled water and mix. Read the intensity of the colour obtained against blank at 495nm using a spectrophotometer. The difference between test and control is taken for calculation. Standard graph can be prepared using standard digitoxin.

Evaluate the amount of alkaloids

Total Alkaloid Content Determination using Harborne, (1973) method

Procedure

40 ml of 10% acetic acid in ethanol was added to 10 g of powdered sample, covered and allowed to stand for 4 hours. The filtrate was then concentrated on a water bath to get 1/4th of its original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and collected precipitate was washed with dilute ammonium hydroxide and then filtered. The residue was dried and weighed.

Estimation of Ascorbic acid (Vitamin C)

Ascorbic acid was analysed by the spectrophotometric method described by Roe and Keuther, (1943).

Principle

Ascorbate is converted into dehydroascorbate on treatment with activated charcoal, which reacts with 2,4-dinitrophenyl hydrazine to form osazones. These osazones produce an orange coloured solution when dissolved in sulphuric acid, whose absorbance can be measured spectrophotometrically at 540nm.

Reagents

1. TCA (4%)
2. 2, 4-dinitrophenyl hydrazine reagent (2%) in conc H₂SO₄
3. Thiourea (10%)
4. Sulphuric acid (85%)
5. Standard ascorbic acid solution: 100µg / ml in 4% TCA

Protocol

For the estimation of ascorbic acid, 1 g of the different parts of sample were homogenized by 4% of TCA after centrifugation a pinch of activated charcoal was added, mixed vigorously using cyclo mixer and stand for 5 minutes. The tubes were centrifuged again to pellet the charcoal particles. Aliquots of supernatant were taken for the estimation and as adopted by Roe and Keuther, (1943). To 0.5 ml of charcoal treated supernatant 2.0 ml of 4% TCA, 0.5 ml of Di Nitro Phenyl Hydrazine was added followed by 2 drops of thiourea solution and mixed well. The tubes were incubated for 3 hours. Removed, placed in ice cold water and added 2.5 ml of 85% H₂SO₄ drop by drop and the absorbance were recorded at 540nm. Concentration of ascorbic acid in the samples were calculated and expressed as mg/g tissue.

Estimation of α-Tocopherol (Vitamin E)

The levels of α-tocopherol were estimated by the method of Baker *et al.*, (1980). This method involves the reduction of ferric ion to ferrous ion by α-tocopherol and the formation of a red coloured complex with 2,2'-dipyridyl. Absorbance of the chromophore was measured at 520nm.

Reagents

1. Petroleum ether 60-80°C
- Double distilled ethanol

3. 0.2% - 2,2'-dipyridyl in ethanol

4. 0.5% ferric chloride in ethanol

5. Stock standard: 100 mg of α-tocopherol in 100 ml of distilled ethanol.

6. Working standard: Stock solution was diluted to a concentration of 10µg/ml in distilled ethanol.

Procedure

To 0.1 ml of sample, 1.5 ml of ethanol and 2.0 ml of petroleum ether were added, mixed and centrifuged. The supernatant was evaporated to dryness at 80°C. To this was added 0.2 ml of 2, 2'-dipyridyl solutions and 0.2 ml of ferric chloride solution. Mixed well and kept in dark for 5 minutes and added 2 ml of butanol. The intense red color developed was read at 520nm. Standard tocopherol in the range of 10-100µg were taken and treated similarly along with blank containing only the reagent. The amount of α-tocopherol was expressed as mg/ml plasma or mg/mg extract.

Evaluate the amount of terpenes

The amount of total terpenes in the extracts was estimated by Ghorai *et al.*, (2017). Terpenes and terpenoids are primary constituents of essential oils of different type of plants and flowers. Some qualitative estimation methods of terpenoids in plant tissue have been previously described but there is no protocol of estimating the same quantitatively till date. In the present study a protocol has been attempted to estimate the total terpenoids concentration of different resin producing plants using a monoterpene, Linalool as standard reagent.

Protocol

The previously prepared sample for quantitative analysis was transferred from assay tube to Colorimetric cuvette [95% (v/v) Methanol will be used as blank] to read the absorbance at 538 nm. For the standard curve 200 µl of previously prepared Linalool solution in methanol will be added to 1.5 ml Chloroform & serial dilution must be done [dilution level- 100mg/200µl to 1mg/200 µl Linalool Conc.] In case of serial dilution total volume of 200 µl will be made up by addition of 95% (v/v) Methanol.

Evaluate the amount of saponin

Saponin was analysed by the gravimetric method described by Obadoni and Ochuko., (2001).

Protocol

The samples were ground and 20 g of sample put into a conical flask followed by the addition of 200 ml of 20% aqueous ethanol. They were then heated over a hot water bath for 4 hours with continuous stirring at about 55°C. The mixture was filtered and the residue re extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90°C. The concentrate was transferred into a 250 ml separatory funnel and 20 ml of diethyl ether added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification

process was repeated. 60 ml of n-butanol was added. The combined n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath. After evaporation the samples were dried in the oven to a constant weight. Saponin content was calculated as percentage.

Estimation of antioxidant activity

DPPH (2, 2-diphenyl-1-picryl hydrazyl) Radical Scavenging Assay by (Blois, 1958). The antioxidant activity of the extracts was estimated using the DPPH radical scavenging protocol. DPPH solution (0.004% w/v) was prepared in 95% ethanol. 500 µl of freshly prepared DPPH solution (0.004% w/v) was added in each of these test tubes which contain 25 µl of extract and 475 µl buffered saline. The reaction mixture was incubated in the dark for 30 min and thereafter the optical density was recorded at 520 nm against the blank. For the control, 500 µl of DPPH solution in ethanol was mixed with 500 µl buffered saline and the optical density of the solution was recorded after 30 min. The assay was carried out in triplicate. The decrease in optical density of DPPH on addition of test samples in relation to the control was used to calculate the antioxidant activity, as percentage inhibition (% IP) of DPPH radical. The capability of scavenging DPPH radical was calculated using the following equation

$$\text{DPPH Scavenged (\%)} = \frac{(\text{A control} - \text{A test}) \times 100}{(\text{A control})}$$

Where —A control is the absorbance of the control reaction and —A test is the absorbance of the sample of the extracts.

RESULTS AND DISCUSSION

Plants are source of natural products for various therapeutic processes and they play a major role in the treatment of human disease. The plant *Hemigraphis colorata* is a versatile tropical low creeping perennial herb that reaches a height of 15 to 30 cm, which is the native of tropical Malaysia. It is a prostrate growing plant with spreading, rooting stems. Its stained leaves are

slender and lance shaped with toothed, scalloped or lobed margins. They are grayish green stained with red purple above and darker purple beneath. Literally Hemigraphis means 'half writing' because the filament of the outer stamen bear brushes. *H. colorata* blooms irregularly throughout the year in the tropics. Flowers are small (1 to 1.5 cm diameter), five lobed, bell shaped with imbricate bracts. These are white in colour with faint purple marks within and appear in terminal 2 to 10 cm long spikes. Seeds are small, flat and white in colour. The leaves are opposite, ovate or cordate, serrate crenate, about 2 to 8 cm long and 4 to 6 cm wide, bearing well defined veins. They are grayish green stained with red purple above and darker purple beneath (Shana *et al.*, 2022).



Fig. 1: Plant picture of *Hemigraphis colorata*.

Estimation of phytochemical constituents

Phytochemistry is associated with the study of phytochemicals produced by the plants. Phytochemicals are often categorized as primary and secondary metabolites. Carbohydrates, lipids, proteins, nucleic acids are primary metabolites that means they are essential for the survival of the plants. Secondary metabolites are not pivotal for the survival of plants but determine their existence in the ecosystem. Phytochemicals present in plants possess varied bioactivities with abundant health advantages and have also been employed in conventional medicine system to treat a variety of ailments and diseases (Thacker and Ram, 2024).

Table 1: Estimation of Phytochemical constituents of *Hemigraphis colorata*.

PHYTOCHEMICALS	Concentration (%)
Flavonoid	6.14
Alkaloid	1.21
Saponin	3.97
Tannin	0.76
Polyphenol	0.33
Glycoside	1.24
Terpene	0.88
Vitamin C	0.54
Vitamin E	0.023

Table 1 showed the result of phytochemical constituents in *Hemigraphis colorata*. The results showed that *Hemigraphis colorata* plant leaf possess flavonoid

concentration is 6.14 %, alkaloid content is 1.21 %, saponin content is 3.97 %, tannin concentration is 0.76 %, polyphenol concentration is 0.33 %, glycoside

concentration is 1.24 %, terpene concentration is 0.88 %, vitamin C concentration is 0.54 % and vitamin E concentration is 0.023 %.

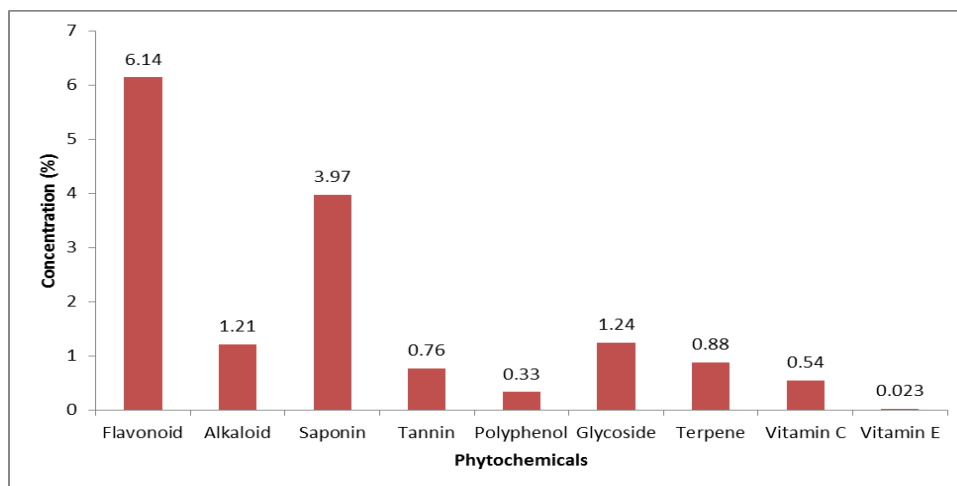


Figure 2: Estimation of phytochemical constituents.

Free radical scavenging activity of plant extract by DPPH method

Free radicals play a key role in oxygen dependent (aerobic) living systems. They are part of cell respiration and other important cellular processes but are also involved in aging and disease development. Free radicals are unstable molecules with free unpaired electrons. They are highly reactive because the free electrons always attempt to bond with other electrons and form covalent pairs. The antioxidant activity of leaf extract has been studied by its ability to reduce DPPH. Interaction of

antioxidant compounds with DPPH is based on the transfer of hydrogen atom or electron to DPPH radical and converts it to 1, 1-diphenyl-2-picryl hydrazine. The result of reduction DPPH radicals causes discoloration from purple colour to yellow pale colour which demonstrates the scavenging activity (Hussen and Endalew, 2023). The antioxidant activity of the methanolic leaf extract against DPPH assay was tested with concentrations ranging from 10 to 100 µg/ml as the results shown in Table 2.

Table 2: Free radical scavenging activity of plant extract by DPPH method.

Sl. No	Concentration (µg/ml)	Absorbance (517 nm)	% inhibition	Linear regression equation	IC ₅₀ (µg/ml)
1	Control	0.739		Y=0.456X+17.64	70.96
2	10	0.658	10.96		
3	15	0.611	17.32		
4	20	0.501	32.21		
5	25	0.447	39.51		
6	50	0.380	48.58		
7	100	0.312	57.78		

Table 2 showed six varying concentrations (10, 15, 20, 25, 50 and 100 µg/ml) of plant extract demonstrated different percentage of inhibition. Interestingly, scavenging activity of extract was increased in a concentration dependent manner. The 100 µg/ml extract showed the best antioxidant activity in DPPH method.

Figure 2 showed the inhibition percentage of plant extract extracted from *Hemigraphis colorata* leaf against DPPH. The IC₅₀ of the plant extract was obtained as 70.96 µg/ml. This means that the plant extract could exhibit a greater antioxidant activity. This can be explained by the phytochemical content of *Hemigraphis colorata*, as it was reported to contain a high percentage of flavonoid, vitamin C, vitamin E and polyphenolic

compounds (Fathallah, 2023). The presence of these components in the plant extract solution would make up a network of cooperated material that are work together to detoxify DPPH, which make them more powerful as equal as ascorbic acid by its own. Low IC₅₀ values correspond to high antioxidant activity (Rahman *et al.*, 2015). There is strong evidence that many dangerous patho-physiological processes, such as cancer, diabetes and cardio-vascular and neurodegenerative diseases, are associated with the accumulation of free radicals. A free radical is an atom or molecule that has an unpaired electron and is therefore unstable. This unstable radical has the tendency to become stable through electron pairing with biological macromolecules such as proteins,

lipids, and DNA in healthy human cells, thus causing protein and DNA damage (Rahman *et al.*, 2015).

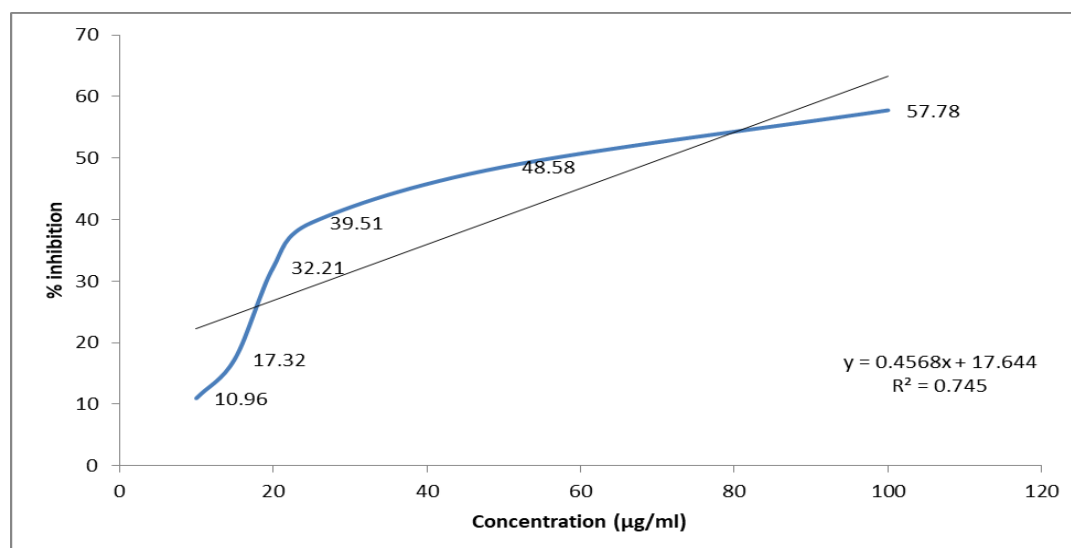


Figure 3: Free radical scavenging activity of plant extract by DPPH method.

SUMMARY AND CONCLUSION

Hemigraphis colorata, a perennial plant adapted to India, is a versatile tropical low creeping exotic herb that reaches a height of 15 to 30 cm. In Kerala, the plant is popular in the name 'Murikooti' or 'Muriapacha' because of its incredible potency to heal wounds. *Hemigraphis* means 'halfwriting' because the filament of the outer stamen bear brushes. With leaf of metallic purple lustre on upper surface and a solid dark purple on ventral side, the leaves are opposite, ovate to cordate, serrate, crenate, about 2 to 8 long and 4 to 6 cm wide, bearing well-defined veins. Flowers are small (1 to 1.5 cm diameter), five lobed, bell shaped with imbricate bracts. These are white in color with faint purple marks within and appear in terminal 2 to 10 cm long spikes. It can be used to treat anaemia, gallstones, haemorrhoids etc. The traditional and medicinal system uses the plant products for the treatment of various infectious diseases. The main goal of this study is to evaluate the phytochemicals constituents analysis of antioxidant activity by DPPH method and extracted from leaves of *Hemigraphis colorata*. The main objectives of the present study are preparation of leaf extract of *Hemigraphis colorata* using maceration method, phytochemicals study by plant extract, evaluation of antioxidant activity by DPPH method.

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