QUALITATIVE AND QUANTITATIVE EXAMINATION OF PHYTOCHEMICALS FROM SINARUNDINARIA WIGHTIANA (NEES) CS CHAO & RENVOIZE

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Abstract

Biologically active chemicals derived from natural sources have historically sparked the curiosity of infectious disease researchers. In fact, according to estimates from Wani *et al.*, (2019), around 25% of all prescribed medicines in use today are made from plant-based ingredients. Drug makers are especially fascinated by plant phytochemical analysis because this research is critical for the development of innovative medications that could potentially be utilized for treating a wide range of diseases. The current research examines the phytochemical composition of several portions of *Sinarundinaria wightiana* (Nees) CS Chao & Renvoize, a plant that is indigenous to Tamil Nadu's Nilgiris region. All across the world, bamboo is known for its many medicinal qualities. To identify the plant phytochemical components, aqueous extract samples of the leaves, flowers, young shoots, and rhizomes were employed for analysis. These extracts were put through conventional chemical testing for a preliminary phytochemical examination. According to the data, alkaloids, carbohydrates, proteins, flavonoids, phenols, tannins, and steroids were present in the leaves, flowers, roots, and early shoots. However, mucilage, gum, volatile oil, and saponins were inadequate.

Keywords: Sinarundinaria wightiana, Preliminary phytochemical, Carbohydrates, Aqueous and Flavonoids.

Introduction

Herbal medicines have been used and sold all over the world since the prehistoric era. To provide them with basic healthcare, over 80–90% of people worldwide require traditional medicine, and the vast majority of them use plant extracts. Every plant has phytochemical components, which makes them possible sources of treatment. However, a biological screening is necessary to find out more about how these compounds function. While secondary metabolites work on non-essential functions in plants, primary metabolites participate in vital metabolic processes. By serving as allelopathic and photo-protectant agents, as well as being involved in chemical resistance against illnesses and predators, secondary metabolites assist in pollination, dispersal, and disease prevention in humans. The main bioactive components are alkaloids, phenols, flavonoids, terpenes, steroids, and glycosides (Jerun Nisha *et al.*, 2021).

According to Sungkaew *et al.*, (2009), there are 1,439 species of bamboo, which belong to the family Poaceae and subfamily of Bambusoideae. The tropical and temperate regions of Asia, Africa, Australia, and America are all home to bamboo (Guerreiro and Lizarazu, 2010).

Bamboo species are quite diverse in Brazil; there are 33 genera in addition approximately 250 species were found, of which 160 are thought to remain endemic and some species are not appropriately categorized so far. Additionally, more than 20 species of bamboo, most of which are Asian natives, were brought into Brazil, mostly during the colonial era and later by Japanese immigration (Silva *et al.*, 2011; Tombolato *et al.*, 2012).

The usagesof bamboo remain innumerable. Numerous species are cultivated as decorative plants, while others remain forage, as raw materials for the construction of housing, as sources of fiberin pulp and paper manufacturing, and also as sources of biomass intended for the invention of energy. Edible shoots from some species are consumed as food. Additionally, bamboo is a resource for local economies because it is used as a raw material by entrepreneurs (Filgueiras*et al.*, 2004).

Sinarundinaria wightianais a clumping bamboo with small rhizomes that grows upright. It has 2 to 3 meter long woody culms. The culms are taken from the wild and used for basketry and weaving. They are found mostly as undergrowth in evergreen sholas, are gregarious in damp environments, and are generally found on hills and slopes over 1,800 meters. In the case of certain mature tropical species, the new stem might grow to be as tall as 30 meters, with daily height gains of 30cm or greater over the height of the development period. As a result, they are among the world's fastest-growing species. Several varieties of bamboo are monocarpic, meaning they live for many years before blossoming, then flower and seed lavishly for 1 - 3 years before dying. While most bamboos only blossom every few years, this species has been known to flower every year without the canes withering. Baskets, mats, huts, roofing, and fences are made from the culms (Seethalakshmi and Muktesh Kumar, 2002).

Materials and Methods Sample collection

The experimental material selected for the present study is *Sinarundinaria weightiana* (Nees) CS Chao & Renvoize belongs to the family Poaceae. Various plant parts of *Sinarundinaria wightiana* (Nees), CS Chao & Renvoize, such as the leaves, flowers, young shoot, and root were collected from the sholas of Doddabetta, Mainala, Naduvattam and Pykara areas of the Nilgiri District. The obtained plant parts were left to dry in the shade, pulverized, and stored for further use.

Extraction and phytochemical analysis

Extraction is the method whereby the preferred constituents of plants are separated using solvents. The powered material was subjected to extract with 95% ethanol, for 24 hours using a soxhlet apparatus. The extract is then concentrated in a rotary evaporator undera constant vacuum. The extract obtained as above was then subjected to qualitative as well as quantitative tests for the identification of severalplant constituents like Alkaloids, Flavonoids, Glycosides, Carbohydrates, Saponins, Proteins, Fixed Oils, Fats, Phenolics, Tannins, Steroids, Gum Mucilages and Volatile oil). Trease and Evans (1978) and Harborne, (1984, 1973) techniques were used to screen the phytochemical components of the plant extract qualitatively.

Quantitative estimation of phytoconstituents

A spectrophotometer is a device that measures the absorption of light as a function of

wavelength in both the UV and visible ranges. It also adheres to the basic rules of light absorption. As an example, consider Beer Lamber's law. Spectrophotometry has evolved into an effective technique for both qualitative and quantitative assessments.

Estimation of Protein (Lowry et al., 1951)

1gm of leaves flowers, young shoots, and rhizome were taken and ground well with mortar and pestle distilled water was added and a sample solution was made into 1ml. With that reagent D (5 ml) and reagent (0.5 ml) was added. Blank was also arranged in the same way by using distilled water. After 30 minutes OD was taken at 750nm along with standards. From the standard graph, the amount of protein present in the given sample was calculated.

Estimation of Carbohydrates (Anthrone Method)

Leaves, flowers, youngshoot, and rhizome of *Sinarundinariawightiana* were taken it was ground with 80% of CHOH. This extract was re-extracted twice using the equivalent volume of CHOH as above the entire collected extract was diluted in a standard flask to 50ml using distilled water. It was used as a sample solution for analysis.

Estimation of Phenol

Total phenol content was performed for extracts and fractions of *Sinarundinariawightiana* (Nees) C. S. Chao & Renvoize at the concentration of microgram/ml.

Total Phenol Content (TP)

Chandler and Dodds' methodology was used to quantify total polyphenol compounds. In a test tube, combine 1 mL of sample solution, 1 mL of 95% ethanol, 5 mL of distilled water, and 0.5 mL of 50% folin-ciocalteu reagent. After allowing the reaction mixture to react for 5 minutes, 1 ml of 5% Naz Cos was added. After carefully mixing the contents and leaving them in the dark for 1 hour, the optical density was measured at 725nm using a UV/VIS Spectrophotometer (Perkin Elmer).

The standard curve was built using 20, 40, 60, 80, and 100 μ g/ml of gallic acid, and the quantifications were represented as gallic acid equivalent per dry weight of the sample (mg GAE/g DW). The averages of duplicate analysis were used to derive the results.

Estimation of Flavonoid

Flavonoid content was estimated in colorimetry using aluminum chloride. 1 ml of sample was mixed with methanol (3 ml), 10% aluminum chloride (0.2 ml), 1M potassium acetate (0.2 ml) and distilled H₂O (5.6 ml). The reaction mixture was kept at room temperature for 30 minutes and the absorbance was measured at 415nm with a UV/VIS spectrophotometer (Perkin Elmer). The standard curve was prepared by preparing Rutin (Sd fine) solution at 20-100 ug/ml concentration in methanol and flavonoid content wasexpressed as mg of Rutin equivalents (RE) per g dry weight if the sample (mg RE/g DW) results were the averages of duplicate analysis.

Alkaloids Determination (Harbone Method)

5g of each extract of the leaves, flowers, young shoots, and roots were weighed into a beaker with a volume of 250 ml, and 200 ml of acetic acid (10%) in ethanol was added. It was covered up and left to stand for 4 hours. Further, the above was filtered, and the extract was reduced toward one-quarter of its original volume througha water bath. Concentrated ammonium hydroxide was added drop by drop to the extract while waiting for precipitation to be complete. The entire solution remained permissible to settle earlier being collected and washed with feeble ammonium hydroxide before being filtered. The residual content isinthe dried and weighed alkaloid.

Results

The phytochemical analysis was carried out for Qualitative and Quantitative estimation of *Sinarundinaria wightiana* (Nees) C. S. Chao &Renvoize. The powdered material of leaf 97gm, young shoot 60gm, root 62gm, and flower 54gm, was subjected to extract with 95% Ethanol for 24 hours using a soxhlet unit. The extract then evaporated to dryness in the water bath, and a 2.5gm extract was obtained from the root. 5.4gm extract from the flower, 2.2gm extract from the shoot, and 3.9gm extract obtained from the leaf, and then subjected to Qualitative as well as Quantitative tests for the identification of various plant constituents.

It was observed that the leaves, flowers, roots, and young shoots exhibited the presence of Alkaloids, Carbohydrates, proteins, flavonoids, phenols, tannins, and steroids. But saponins, volatile oil, gum, and mucilage were completely absent [Table. 1].

Quantitative estimation of major phytoconstituents Estimation of Protein

The maximumprotein concentration was observed in young shootsas 52.0 mcg/ml andthe lowermostprotein concentration was observed in leaf as 24.8 mcg/ml [Fig.1] [Table. 2].

Estimation of Carbohydrate

The highest concentration of Carbohydrates in rootsat 709 mcg/ml and the lowest concentration of carbohydrates in flowers (183.75 mcg/ml) were observed [Fig. 2] [Table. 2].

Estimation of phenol

The highest concentration of phenol in the flower (13.92 mcg/ml) and the lowest concentration of phenols of 10 mcg/ml was observed in the root [Fig. 3; Table. 2]

Estimation of Flavonol

The highest flavonol concentration 146.916 mcg/mlwas observed in the leaf and the lowest concentration was obtained in the root (53.3 mcg/ml) [Fig. 4; Table. 2]

Estimation of Alkaloids

The highest concentration of alkaloids in the flower and the lowest concentration of Alkaloids in the leaf [Table. 3].

Discussion

Zang et al., (2002) observed that the presence of phytochemicals in bamboo may have biological implications due to its antioxidant capacity. Abundantinvestigators have described the antioxidant capabilities of extract of bamboo leaf namely Zang, et al., (2002), Kim, et al (2001), Kweon, et al., (2001), and Kimet al., (1996), identified phytochemicals intended for antioxidative activity. Fu- Zhong Wu, et al., (2009) studied the influence of the density of the

stem on leaf dynamics in terms of nutritional and usage of nutrient efficiency in dwarf bamboo.

According to D. J. C. (1883), Indian *Dendrocalamus Strictus* with a yard clump of 40 square may generate seeds of around 320 pounds and the numbers are 800-1000. Seeds to a single Deogun, (1936), cause a population explosion of rats (with short life cycles), resulting in starvation. Because the seed - shed attracts seed predators, mostly rats (Species of Mus and Rattus). Ghosh, (2008) has reported Bamboos are also considered a medicinal plant. Many ancient and modern literaturerecognizes this fact; it can cure several diseases and disorders. Bamboo has historically been utilized as a folk remedy in the treatment of high blood pressure and coronary artery disease in the countries of the Orient.

Yu Zhang, et al., (2008) isolated and purified four flavone C-glycosides from antioxidants of bamboo leaves using macroporous resin column chromatography and preparative HPLC.

Sinarundinariawightiana (Nees) C. S. Chao &Renvoize, exhibited the presence of Alkaloids, Flavonols, Phenols, Proteins, Carbohydrates, Tanins, and Steroids but Saponins, Volatile oil, GM and Mucilage were completely absent. The Quantitative estimation showed that the young shoots have high protein and the leaves have low concentration of protein. It is also observed that the concentration of Carbohydrates in rhizomes is very high and the concentration of carbohydrates is very low. The highest phenol concentration has been observed in the flowers it is lowest in the rhizomes. In the leaves, the flavonol is in higher concentration and it is lower in rhizomes. Similarly, there is the highest concentration of Alkaloids in the flowers and the lowest in the leaves.

Conclusion

The results revealed the presence of medicinally important constituents in the plants studied. Much evidencewas gathered in earlier studies which confirmed the identified phytochemicals to be bioactive. Several studiesconfirmedthepresenceofthese phytochemicals contributes medicinal as well as physiological properties to the plants studied in the treatment of different ailments. Therefore, extracts from these plants couldbe seen as a good source of useful drugs. Traditional medicine practice is recommended strongly for this plant as well and it is suggested that further work should be carried out to isolate, purify, and characterize the active constituents responsible for the activity of these plants. Also, additional work is encouraged to elucidate the possible mechanism of action of these extract

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Table 1. Qualitative phytochemical screening of S. wightiana

S.No.	Phytochemical test	Leaves	Flowers	Young shoot	Rhizome
1	Detection of Alkaloids				
	a) Mayers reagent	+	+	+	+
	b) Wagner's reagent	+	+	+	+
	c) Hagner's reagent	+	+	+	+
	d) Dragendroff's reagent	+	+	+	+
2	Detection of Carbohydrates and				
	glycosides				
	a) Molish's test	+	+	+	+
	b) Fehling's test	+	+	+	+
	c) Barfoed's test	+	+	+	+
	d) Benedict's test	+	+	+	+
	e) Legal's test	+	+	+	+
	f) Bontrager's test	+	+	+	+
3	Detection of Saponin's				
	a)Foam test	-	-	-	-
4	Detection of Proteins and Amino				
	acids				
	a) Millions test	+	+	+	+
	b) Biuret test	+	+	+	+
	c) Ninhydrin test	+	+	+	+
5	Dectection of Fixed Oils and Fats				
	a) Saponification test	-	-	-	-
	b) Spot test	-	-	-	-
6	Detection of Flavonoids				
	a)Alkaline test	+	+	+	+
	b) Zinc test	+	+	+	+
	c) Shinoda test	+	+	+	+
7	Detection of Compounds of				
	Phenolic and Tannins				
	a) Gelatin test	+	+	+	+
	b) Ferric Chloride	+	+	+	+
	c) Lead acetate test	+	+	+	+
	d) Mg and HCl reduction test	+	+	+	+
8	Detection of steroids				
	a) Salkowskis test	+	+	+	+

	b) Libermanns test	+	+	+	+
9	Detection of gum and mucilages	-	-	-	-
10	Detection of volatile oil	-	-	-	-

Table 2. Quantitative Estimation

Quantitative test	Wavelength	Sample ID	Ordinate	Concentration
		Young shoot	0.0264	52.0 mcg/ml
Protein	750.0	Leaf	0.0124	24.8 mcg/ml
Protein		Rhizome	0.0238	47.6 mcg/ml
		Flower	0.0230	46 mcg/ml
	630.0	Young shoot	0.0828	207 mcg/ml
Carla alazzelmata		Leaf	0.1200	300 mcg/ml
Carbohydrate		Rhizome	0.2836	709 mcg/ml
		Flower	0.0735	183.75 mcg/ml
	725.0	Young shoot	0.0120	10 mcg/ml
Phenol		Leaf	0.0029	2.42 mcg/ml
Phenoi		Rhizome	0.0028	2.33 mcg/ml
		Flower	0.0167	13.92 mcg/ml
	451.0	Young shoot	0.1614	67.25 mcg/ml
Flavonoids		Leaf	0.3526	146.916 mcg/ml
riavolioids		Rhizome	0.1280	53.3 mcg/ml
		Flower	0.1020	42.5 mcg/ml

Table 3: Estimation of Alkaloids

S.No.	Plant parts used	% of Alkaloids
1	Young shoot	11.2 %
2	Leaf	3.8%
3	Rhizome	13.46%
4	Flower	14.68%

Figure 1, Estimation of Protein

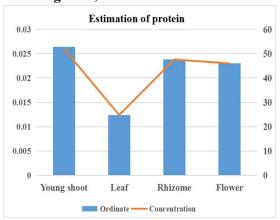


Figure 2, Estimation of Protein

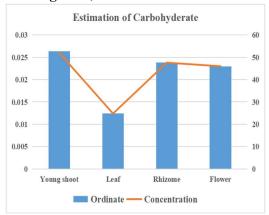


Figure 3, Estimation of Phenol

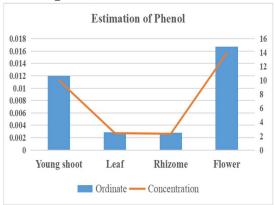


Figure 3, Estimation of Flavonids

