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Research Article

PHYTOCHEMICAL SCREENING AND EVALUATION OF BIOLOGICAL ACTIVITY OF ROOT EXTRACTS OF *SYZYGIUM SAMARANGENSE*

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ABSTRACT

Phytochemical constituents are non-nutritive plant chemicals that have preventive and curative properties of disease. The use of plants and phytochemicals, both with known biological properties, can be of great significance in therapeutic treatment. The present study includes phytochemical screening and quantification of secondary metabolites and their biological activities of root extract of *Syzygium samarangense.* Phytochemical screening of the plant root extracts with ethyl acetate, methanol and water showed the presence of flavonoids, terpenoids and phenolic compounds. In aqueous extracts, Terpenoids with highest quantity (81.923 micrograms per gram extract) were estimated, whereas flavonoids are present only in methanolic extract and with an estimated quantity of 33.687 µg /per gram extract. Anti oxidant, anti inflammatory, and anti diabetic activity of these root extracts reveal that methanoilc extracts with flavonoids showed the high antioxidant, anti inflammatory and antimicrobial activity than the aqueous and ethyl acetate extract. Highest antidiabetic activity was observed in aqueous extract. Antimicrobial activity study reveals that gram positive bacterial strains are more sensitive than the gram negative bacteria to all the three types of root extracts. The present study reveals that the root extract of *Syzygium samarangense* is a potential source for phytochemicals for traditional use as therapeutics.

Keywords: *Syzygium samarangense* (wax jambu), flavonoids, terpenoids.

INTRODUCTION

Since ancient times, about 80 % of individuals use traditional medicine, which has chemical compounds derived from medicinal plants[1, 2]. Several hundred plant species and herbs in the form of whole plant, crude extract or purification, purified constituents are used in indigenous system of medicines and are of great importance to the health of individual and communities, which have ultimately evaluated into the modern therapeutic science. Medicinal plants are important source of life saving drugs for majority of the world population[3, 4].

Fig. 1: *Syzygium samarangense* (wax jambu) plant and fruit

Syzygium samarangense (common name - wax jambu) is a plant species in the family Myrtaceae which is widely cultivated in the tropics[5-13]. There are lot of traditional claims has been reported of leaves, root, bark, fruits of the plant[14-17]. Various Pharmacological Activities like Antidiarrhoeal Activity, Anticholinesterase Activity, Immunopharmacological Activity, Cytotoxic Activity, Anti hyperglycaemic activity, Analgesic and Anti- Inflammatory activity are reported with various parts of the plant[18-25]. And also Investigators have found their principal constituent to be tannins, Quercetin glycosides, monoterpenes secondary metabolites those involved in pharmacological properties[26, 27]. Traditionally the root bark decoction of the *Syzygium samarangense* is used in dysentery and amenorrhea and also used as abortifacient. Root is used as diuretic and is given to alleviate edema. Malayans use powdered dried root preparations for itching. As plant root has significant therapeutic uses, this study is aimed to screen the phytochemicals of the root and study of its biological activities.

The literature review proves that the plant is rich with many medicinal and bioactive compounds like flavanoids, phenolic compounds, glycosides, terpinoids etc**.** various studies are reported these compounds in various areal parts of the plant. **K. M. Moneruzzaman et al**[14], reported the flavonoids (914.1 mg/100g) and phenolic (326.67 mg GAE/100g) content from the Syzygium samarangense plant where in our study root is reported as 3mg/100grams. **Wu YZ et al**[24], investigated the chemical constituents from the branches and leaves with 95% ethanol, then partitioned with petroleum ether, chloroform and ethyl acetate. **M.O. Edema et al**[36], studied on *Syzygium samarangense* juice extracts and evaluated high amounts of saponins content (4.77%). **Mario J. Simirgiotis et al**[37], reported the six quercetin glycosides on the methanolic extracts of the pulp and seeds of the fruits of *Syzygium samarangense* Merr. & Perry (Blume). **Vasanthi et al**[38], reported the total phenolic content (162.58Â±0.51 Î¼g/mg), total flavonoid contents (310Î¼g/mg) of Syzygium samarangense Fruit extract (SSFE) where the study with roots are reported 46.944 µg and 23.056 µg per gram in methanolic and water extract respectively. **G. R Nair et al**[39], reported the two flavonol glycosides. **Ghayur MN et al** [40], reported four flavonoids isolated from the hexane extract. **Evangeline C. Amor et al**[18], isolated four rare C-methylated flavonoids with a chalcone and a flavanone the compounds from the hexane extract of the leaves. **Dennis D. Raga et al**[41], isolated the Cycloartenyl stearate (**1a**), lupenyl stearate (**1b**), sitosteryl stearate (**1c**), and 24-methylenecycloartanyl stearate (**1d**) (sample 1) from the air-dried leaves. **Samy MN et al**[42], isolated three new compounds (one new cyanogenic glucoside) from a MeOH extract of the leaves. **Rachana Srivastava et al**[43], isolated the new triterpene, methyl 3epibetulinate in its native form and 4′,6′dihydroxy2′ methoxy3′, 5′dimethyl chalcone along with ursolic acid, jacoumaric acid and arjunolic acid have been isolated from the aerial parts. However the study with root extracts reported that root reported with high terpenoid content which is not reported earlier. And it is also reported that root is another alternate source for flavonoid and phenoilc groups. Anti oxidant, anti inflammatory, and anti diabetic activity of these three root extracts conducted in order to study the biological activity of the plant. Anti oxidant activity of the extract was conducted by DPPH scavenging activity. Results reveal that methanolic extracts which consist of flavonoids has shown the high antioxidant activity

(88.021%) (table-3 and graph A) than the aqueous and ethyl acetate extracts at 40 µg/ml extract concentration. Similar studies are reported by the **Fonseca A et al**[44], investigated the antioxidant activity (0.9 µMol TROLOX®/mg) of the fruit. **Vasanthi et al**[38], reported that IC50 value of antioxidant activity of the fruit extract found to be 140 Î¼g/ml in standard (Lascorbic acid), 280 Î¼g/ml in SSFE by DPPH and ABTS.+ scavenging activity found to be the IC50 value 175 Î¼g/ml and 250 Î¼g/ml respectively. No one has reported with root extracts and found that similar antioxidant activity as fruit. Antidiabetic activity (table-4 and graph B) of the root extracts was studied by *In vitro* α- amylase inhibition activity by Spectrophotometric method. Results showed that dose dependent inhibition was observed with all the samples where as aqueous extract (92.626%) exhibited the highest activity than remaining two extracts. No antidiabetic activity studies are reported with *S. Samarangense***.** Anti inflammatory activity (table-5) of the root also studied with all three extracts and among them methanolic extracts (84.552%) showed the highest anti inflammatory activity. In literature **Shabnam Mollika et al**[45], reported evaluated the moderate effect anti-inflammatory activity of the methanolic extract of leaves in mice. **Dennis D. Raga et al**[41], reported that the leaves exhibited potent analgesic and anti-inflammatory activities at effective doses of 6.25 mg/kg body weight and 12.5 mg/kg body weight, respectively. This reveals theat no one has reported the anti inflammatory activity with root and potential activity was reported in our study. Antimicrobial activity result (table-6 and figure-3) reveals that Gram-positive bacteria were more sensitive than Gram-negative ones towards the plant extracts studied. *Bacillus subtilis* and *Staphylococcus aureus* strains are shown more inhibition zone than gram negative *Escherichia coli, Pseudomonas aeruginosa, Salmonella typhi.* Previous studies are also reported the antimicrobial activity of the different areal plant extracts. **S. Adeola Adesegun et al**[46], evaluated the antimicrobial effect of the volatile oil from the leaf of *Syzygium samarangense* on *Escherichia coli.* **Abd Aziz et al**[47], evaluated antimicrobial properties of ethanolic extracts of the leaves of minimum inhibitory concentration (MIC) value was determined to be 20 mg/mL. **K. Venkata Ratnam et al**[48], evaluated the antimicrobial properties of fruits, against certain bacterial and fungal strains with petroleum ether and methanol extracts found to be effective on both Gram positive and Gram negative bacteria. **M.O. Edema et al**[36], reported that juice extracts have significant (P<0.05) antimicrobial activities against *Escherichia coli, Salmonella typhi* and *Candida albicans.* **Consolacion Y. Ragasa et al**[49], elucidated the dichloromethane extract of the leaves exhibited moderate antifungal activity against C. albicans, low activity against T. mentagrophytes and low antibacterial activity against E. coli, S. aureus and P. aeruginosa. It was inactive against B. subtilis and A. niger. In comparison with the literature studies root extracts of *S. samarangense* also have significant antimicrobial active compounds with low minimum inhibition concentration. Studies proved that compounds in plant extracts have potential activity against gram positive bacteria than gram negative.

MATERIAL AND METHODS

Chemicals: The solvents used for root extraction are Methanol and Ethyl Acetate. The reagents used for phytochemical screening and estimation were of laboratory reagent grade and were purchased for Merck chemicals private limited, Mumbai, Fisher scientific, Mumbai and SD fine chemicals Mumbai. Distilled water has been used for aqueous extraction. Alpha amylase enzyme was purchased from Sigma Aldrich chemicals, Ofloxacin drug.

Apparatus: Denver electronic balance, TECHCOMP – UV 2301 Double Bean UV Visible Spectrophotometer with HITACHI 2.2 software, Tech-comp UV visible spectrophotometer, soxhlet extraction apparatus, heating mantle, incubator, autoclave.

Sample collection: Root material of *Syzygium samarangense* plants were collected from farms in various places of East Godavari district, Andhra Pradesh, India. The roots are separated and allowed to shade dry. The root sample was ground and powdered for solvent extraction.

Solvent extraction: The phytochemicals present in the roots of the collected plants were isolated using different solvents like ethyl acetate, methanol and water in a series of extraction method from low polarity to high polarity using soxhlet extraction method.

Microbial test strains for antibiotic activity

The bacterial strains used for screening of antimicrobial activity are *Salmonella typhi, Escherichia coli, Pseudomonas aeruginosa, Bacillus subtilis.*

Phytochemical analysis of the root extracts was performed by following standard methods

Preliminary phytochemical screening

1. Test for steroids

Salkowski Test

Few drops of concentrated sulphuric acid are added to the plant extract, shaken and on standing; lower layer turns red in colour.

Liebermann Burchard's Test

To the extract, few drops of acetic anhydride was added and mixed well. 1 ml of concentrated sulphuric acid is added from the sides of test tube, a reddish brown ring is formed at the junction of two layers.

2. Tests for triterpenoids

Salkowski Test

Few drops of concentrated sulphuric acid are added to the extract, shaken and on standing, lower part turns golden yellow colour.

Lieberman Burchard's Test

To the extract, few drops of acetic anhydride was added and mixed well. 1 ml of concentrated sulphuric acid is added from the sides of test tube, a red ring indicates triterpenes.

Ischugajiu Test

Excess of acetyl chloride and pinch of zinc chloride are added to the extract solution, kept aside for reaction to subside and warmed on water bath, cosin red colour is produced.

Brickorn and Brinar Test

To the extract, few drops of chlorosulfonic acid in glacial acetic acid (7:3) are added, red colour is produced.

3. Test for Saponins

Foam Test

Small amount of extract is shaken with little quantity of water; the foam produced persists for 10 minutes. It confirms the presence of saponins.

Haemolysis Test

To 2ml of 1.8% Sodium chloride solution in two test tubes, 2ml distilled water is added to one and 2ml of 1% extract to the other, 5 drops of blood is added to each tube and gently mixed with the contents. Haemolysis observed under the microscope in the tube containing the extract indicates the presence of saponins.

4. Test for Steroidal Saponin

The extract is hydrolysed with sulphuric acid and extracted with chloroform. The chloroform layer is tested for steroids.

5. Tests for Triterpenoidal Saponin

The extract is hydrolysed with sulphuric acid and extracted with chloroform. The chloroform layer is tested for triterpenoids.

6. Tests for Alkaloids

Mayer's Test

The acid layer when mixed with Mayer's reagent (Potassium mercuric iodide solution) gives creamy white precipitate.

Dragendroff's Test

The acid layer with a few drops of Dragendroff's reagent (Potassium bismuth iodide) gives reddish brown precipitate.

Wagner's Test

The acid layer when mixed with few drops of Wagner's reagent (solution of iodide in potassium iodide) gives brown to red precipitate.

Hager's Test

The acid layer when mixed with few drops of Hager's reagent (Saturated solution of pricric acid) gives yellow coloured precipitate.

7. Tests for Carbohydrates

Fehling's Test

The extract when heated with Fehling's A and B solutions gives an orange red precipitate showing the presence of reducing sugar.

Molisch's Test

The extract is treated with Molisch's reagent and concentrated sulphuric acid along the sides of the test tube, a reddish violet ring shows the presence of carbohydrate.

Benedict's test

The extract on heating with Benedict's reagent, brown precipitate indicates the presence of sugar.

Barfoed's Test

Barfoed's reagent is added and boiled on water bath for few minutes; reddish precipitate is observed for the presence of carbohydrate.

8. Test for Flavonoids

Shinoda Test

The extract solution with a few fragments of magnesium ribbon and concentrated hydrochloric acid produced magenta colour after few minutes.

Ferric chloride test

Alcoholic solution of extract reacts with freshly prepared ferric chloride solution and given blackfish green color.

Lead Acetate Test

Alcoholic solution of extract reacts with 10% lead acetate solution and given yellow precipitate.

9. Test for Glycosides

Anthraquinone test

Drug is powdered and extracted with either ammonia or caustic soda. The aqueous layer shows pink color

Keller-killiani test

This is for cardiac glycosides. The extract and 0.4 glacial acetic acid are mixed with ferrous chloride and 0.5 mi of concentrated sulphuric acid. The acetic acid layer shows blue color.

10. Test for Phenolic Compounds

Ferric chloride test

Treat the extract with ferric chloride solution then blue color appears if hydrolysable tannins are present and green color appears if condensed tannins are present.

Gelatin test

To the test solution add 1% gelatin solution containing 10% NaCl, and then precipitate is formed.

Test for chlorogenic acid

Treat the test solution with aqueous ammonia and expose to air gradually, green color is developed.

Quantitative analysis of Phenoilc Compounds

The total phenolic content in different solvent extracts was determined with the Folin-Ciocalteu's reagent (FCR). In the procedure, 1ml of extract was mixed with 0.4 ml FCR (diluted 1:10 v/v). After 5 min 4 ml of sodium carbonate solution was added. The final volume of the tubes were made up to 10 ml with distilled water and allowed to stand for 90 min at room temperature. Absorbance of sample was measured against the blank at 765nm using a spectrophotometer. A calibration curve was constructed using gallic acid solution as standard and total phenolic content of the extract was expressed in terms of milligrams of gallic acid per gram of dry weight.

Determination of total flavonoid content

Total flavonoid content was determined using aluminium chloride ($AlCl_3$) according to a known method using quercetin as a standard. The plant extract (1 ml) was added to 3 ml distilled water followed by 5% $NaNO_2$ (0. 3ml). After 5 min at 25°C, $AlCl_3$ (0.3 ml, 10%) was added. After further 5 min, the reaction mixture was treated with 2.0 ml of 1 M NaOH. Finally, the reaction mixture was diluted to 10ml with water and the absorbance was measured at 510 nm. A calibration curve was constructed using quercetin solutions as standard and total phenolic content of the extract was expressed in terms of milligrams of quercetin per gram of dry weight.

Quantitative estimation of terpenoids

To 1ml of plant extract 2 ml of chloroform was mixed in extract of plant sample and 3 ml of sulphuric acid were added in sample extract. Reddish brown color was obtained in the test tube. Final volume in the test tube was made up to 10ml with water. The absorbance of the formed color was measured at 538nm against reagent blank prepared similarly without plant extract. Linalool was used as standard terpenoid.

Measurement of Antioxidant Activity using DPPH method

The antioxidant activity of the different root extracts was determined on the basis of their scavenging activity of the stable 1, 1- dipheny1-2-picryl hydrazyl (DPPH) free radical. DPPH is a stable free radical containing an odd electron in its structure and usually utilized for detection of the radical scavenging activity in chemical analysis. lml of each solution of different concentrations (1-500g/m1) of the extracts was added to 3 ml of 0.004% ethanolic DPPH free radical solution. After 30 minutes the absorbance of the preparations were taken at 517 nm by a UV spectrophotometer which was compared with the corresponding absorbance of standard ascorbic acid concentrations (1-500µg/m1).The method described by **Hatano et al** was used to measure the absorbance with some modifications. Then the % inhibition was calculated by the following equation:

$$\%\ \textbf{Radical Scavenging Activity} = \frac{\textbf{Absorbance of blank} - \textbf{Absorbance of sample}}{\textbf{Absorbance of blank}} \times 100$$

Antidiabetic activity

In vitro study α- amylase inhibition activity by Spectrophotometric method

1ml of alpha amylase and 1 ml of plant extract in a test tube and incubated at 37 ˚C for 10 min. After pre-incubation, 1ml of 1% (v/v) starch solution was added to each tube and incubated at 37◦C for 15min. The reaction was terminated with 2 mL DNSA reagent, placed in boiling water bath for 5min, cooled to room temperature, diluted, and the absorbance measured at 546 nm. The control reaction representing 100% enzyme activity did not contain any plant extract. To eliminate the absorbance produced by plant extract, appropriate extract controls were also included.

% inhibition of alpha amylase by each plant extract can be calculated using the formula:

$$\% \text{ Indibition} = \frac{(\text{Enzyme activity of control} - \text{Enzyme activity of extract}}{\text{Enzyme activity of control}} \times 100$$

Anti inflammatory Activity by Albumin denaturation Assay

A solution of 0.2% W/V of BSA was prepared in Tris buffer saline and PH was adjusted to 6.8 using glacial acetic acid. Stock solutions of plant extract were prepared by using methanol as a solvent. From these stock solutions 6 different concentrations of 10-500µg/ml were prepared by using methanol as a solvent. 50µl of each extract was transferred to Eppendorf tubes using 1ml micro pipette. 5ml of 0.2% W/V BSA was added to all the above Eppendorf tubes. The control consists of 5ml 0.2% W/V BSA solution with 50 µl methanol. The test tubes were heated at 72° C for 5 minutes and then cooled for 10 minutes. The absorbance of these solutions was determined by using UV/Vis Double beam spectrophotometer (Elico SL-196) at a wavelength of 660nm. The % inhibition of precipitation (denaturation of the protein) was determined on a % basis relative to the control using the following formula.

$$\% \text{ inhibition of denaturation} = \frac{(\text{Abs of control} - \text{Abs of extract})}{\text{Abs of control}} \times 100$$

Antimicrobial Activity

Root extracts of *Syzygium samarangense* were tested by agar well-diffusion method to determine the antimicrobial activity. Nutrient agar (NA) plates were seed inoculated. Wells (10mm diameter and about 2 cm a part) were made in each of these plates using sterile cork borer. Stock solution of each plant extract was prepared at a different concentrations 1000, 500, 200, 100, 50,10µg/ ml in different plant extracts viz. Methanol, ethyl acetate, water. About 100 µl of different concentrations of plant solvent extracts were added with sterile syringe into the wells and allowed to diffuse at room temperature for 2hrs. Control experiments comprising inoculum without plant extract were set up. The plates were incubated at 37°C for 18-24 h. The diameter of the inhibition zone (mm) was measured and the activity index was also calculated. Triplicates were maintained and the experiment was repeated thrice, for each replicates the readings were taken in three different fixed directions and the average values were recorded. Oflaxacin drug was used as standard antibacterial agent and compared with the standard results.

RESULT AND DISCUSSION

Secondary metabolites present in the plants are responsible for the biological activities such as hypoglycaemic, antidiabetic, antioxidant, antimicrobial, antiinflammatory, anticarcinogenic, antimalarial, anticholinergic, antileprosy etc. The preliminary screening of phytochemicals and evaluation of bioactive may lead to medicinal plant drug discovery and development of phytomedicine. In the present study the root of *Syzygium samarangense* (wax Jambu) was screened for determination of it's phytochemicals in three different solvent systems. Among the three, aqueous extracts were proved to contain more number of compounds than other two solvents extracts. In aqueous extract, alkaloids, carbohydrates, saponins, tannins, roteins and aminoacids, terpenoids, phenolic compounds were identified. Tannins, Flavonoids, terpenoids, Phenolic Compounds were indentified in the methanolic

extract. In Ethyl acetate extract only Terpenoids are identified. In continuation quantitative analysis of the extracts has conducted to estimate terpenoids, phenolic compounds and flavonoids (table 1 and fig 2).

Terpenoids were detected with highest quantity (81.923 micrograms per gram extract) in aqueous extracts than other phytochemicals. Terpenoids were detected with 48.461µg quantity in methanolic extract and 35.385 µg in ethyl acetate extract. Whereas flavonoids are only present in methanolic extract and amount of flavanoid quantified was 33.687 µg /per gram extract. Similarly phenolic compounds were reported in methanolic and water extracts with estimates as 30.156µg/g and 23.056 µg per gram extract respectively. Our results were in agreement with findings of the medicinal value of plants lies in some chemical substances that have definite physiological functions in the human body. Different phytochemicals have been found to possess a wide range of medicinal properties, which may help in protection against various diseases.

CONCLUSION

In the present investigation, primary and secondary metabolites of the root were qualitatively and quantitatively analyzed then the biological activity (anti oxidant, anti inflammatory, and anti diabetic) was studied. Further evaluation of phytochemicals and their activity is needed for knowing the nutritional potential as well as helpful in manufacturing new drugs.

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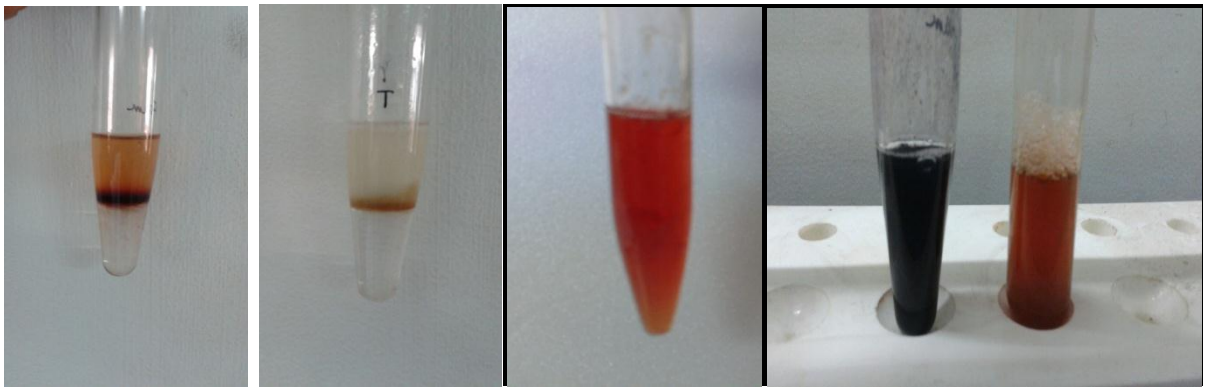
Table 1: Phytochemical screening of root extracts of *Syzygium samarangense*

S. No.	Name of the tests	Ethyl acetate extract	Methanolic extract	Water extract
1	Alkaloids	-ve	-ve	+ve
2	Carbohydrates	-ve	-ve	+ve
3	Glycosides	-ve	-ve	-ve
4	Saponins	-ve	-ve	+ve
5	Tannins	-ve	+ve	+ve
6	Proteins & Aminoacids	-ve	+ve	+ve
7	Flavanoids	-ve	+ve	-ve
8	Terpenoids	+ve	+ve	+ve
9	Phenolic Compounds	-ve	+ve	+ve



Flavanoid test **Terpenoid test**

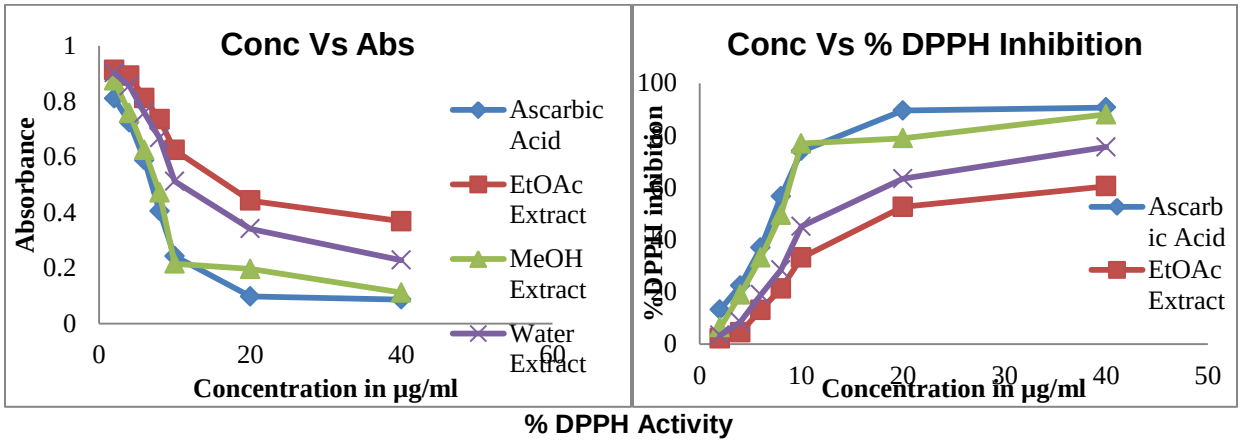
Fig. 2: images of phytochemical screening of root extract of *Syzygium samarangense* (flavonoids, terpenoids tests)

Table 2: Quantitative analysis of root extracts of *S. samarangense*

Phytochemical	Extract	Amount found µg / g of extract
Terpenoids	Ethyl Acetate	35.385
	Methanolic	48.461
	Aqueous	81.923
Flavonoids	Methanolic	33.687
Phenolic Compounds	Methanolic	30.156
	Water	23.056

Table 3: Antioxidant activity (% DPPH scavenging activity) of *S. samarangense* root extracts

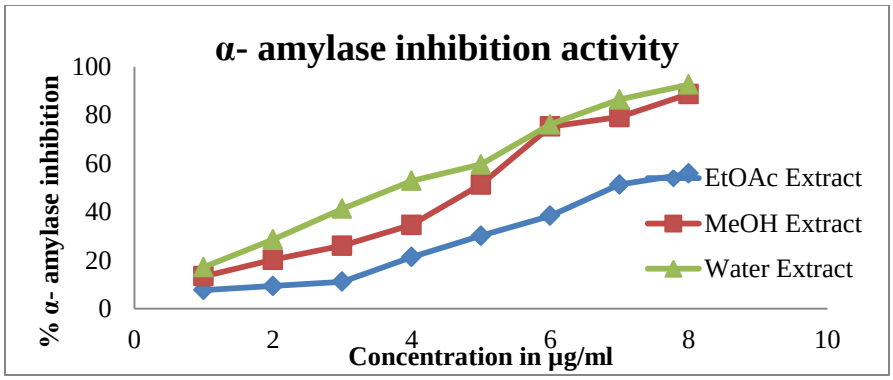
S. No.	Concentration in µg/ml	Ascarbic Acid	EtOAc Extract	MeOH Extract	Water Extract
1	2	13.262	2.246	6.417	3.422
2	4	22.460	4.492	18.930	8.449
3	6	37.005	13.155	33.155	18.930
4	8	56.684	21.283	49.519	28.449
5	10	74.011	33.155	76.898	45.134
6	20	89.519	52.620	78.930	63.422
7	40	90.695	60.535	88.021	75.508



Graph A: Comparative graph of DPPH assay of *S. samarangense* root extracts

Table 4: α- amylase inhibition activity of *S. samarangense* root extracts

S. No.	Concentration of Sample	EtOAc Extract	MeOH Extract	Water Extract
		% of α-Amylase inhibition		
1	5µg/ml	7.709	13.408	17.207
2	10µg/ml	9.385	20.223	28.603
3	15µg/ml	11.173	26.033	41.341
4	20µg/ml	21.341	34.637	52.849
5	25µg/ml	30.168	51.285	59.665
6	50µg/ml	38.436	75.307	76.201
7	100µg/ml	51.285	79.218	86.480
8	200µg/ml	55.866	88.715	92.626



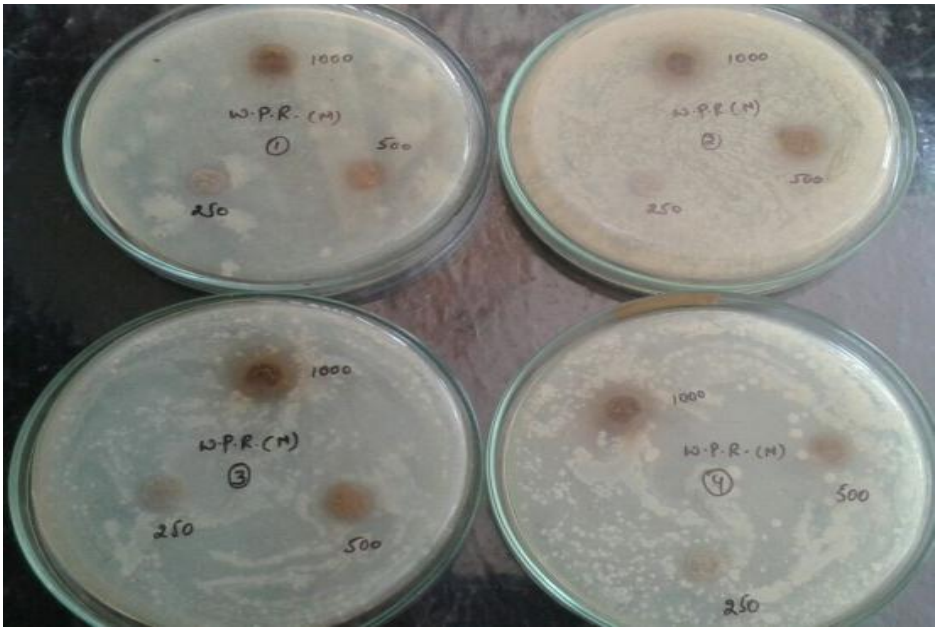
Graph B: Comparative graph of α- amylase inhibition activity of *S. samarangense* root extracts

Table 5: Anti inflammatory activity of *S. samarangense* by Albumin denaturation assay

S. No.	Concentration of Sample	EtOAc Extract	MeOH Extract	Water Extract
		% Albumin denaturation		
1	10µg/ml	5.241	27.724	15.724
2	50µg/ml	13.793	35.034	19.724
3	100µg/ml	23.724	44.000	29.379
4	200µg/ml	36.414	55.448	50.069
5	400µg/ml	54.621	68.828	60.138
6	500µg/ml	60.827	84.552	70.759

Table 6: Anti microbial activity of *S. samarangense* root extracts

S. No.	Test organism	Size of zones (in mm)			
		methanolic	ethyl acetate	aqueous	Standard
		1000 µg/ml	1000 µg/ml	1000 µg/ml	500 µg/ml
1	*Salmonella typhi*	12.7	….	5.6	19.5
2	*Escherichia coli*	19.2	11.2	17.5	28.7
3	*Pseudomonas aeruginosa*	15.3	8.7	11.6	21.2
4	*Bacillus subtilis*	19.5	14.3	16.5	31.5



1) *Salmonella typhi* **2)** *Escherichia coli* **3)** *Pseudomonas aeruginosa* **4)** *Bacillus subtilis*

Fig. 3: Antimicrobial activity results of *S. samarangense* root extracts

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