

Focus on Estimation and Antioxidant Studies of *Salacia* Species

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Abstract - *Salacia* is a valuable medicinal genus found to be composed of various secondary metabolites. Hence, the present study was aimed to estimate three important secondary metabolites such as phenol, flavonoid and tannin. Antioxidant assays such as DPPH assay and ABTS assay and Total Antioxidant Capacity were also studied for this medicinal genus. Seven species of *Salacia* were considered for this present study. They are *Salacia beddomei* Gamble, *Salacia chinensis* L, *Salacia fruticosa* Heyne ex Lawson, *Salacia gambleana* Whiting & Kaul, *Salacia macrosperma* Wight, *Salacia malabarica* Gamble, and *Salacia oblonga* Wall. Various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were taken for all studies and the values are entered in terms of Mean±SD. In case of DPPH assay and ABTS assay, IC₅₀ values are calculated using ANNOVA and Total Antioxidant Capacity was calculated using a calibration curve.

Key Words - *Salacia*, phenol, flavonoid, tannin, Total Antioxidant Capacity, DPPH, ABTS.

I. INTRODUCTION

Salacia is a medicinally important genus belongs to the family Hippocrateaceae, which was formerly known as Celastraceae, Class Magnoliopsida and Order Celastrales [1]. It comprises of about 407 different species and they are widely distributed in South-West India, Peninsular region of India, Sri Lanka, Vietnam, China, Indonesia, Brazil, South Africa, Malaysia, Thailand and Philippines [2,3]. The rich biodiversity of medicinal plants make them a treasure house for obtaining novel compounds either themselves as drugs or pilot molecules for invention of new drugs with different mechanism of action.

Medicinal plants are found to be composed of various phytochemicals also known as secondary metabolites which are used to treat many diseases [4]. The secondary metabolites such as alkaloids, flavonoids, terpenoids, saponins, tannins, glycosides, coumarins and phenolic compounds are important natural chemical constituents [5,6,7]. These medicinal plants with phytochemicals can be used as a supplement for human diseases by acting as a natural antioxidants [8].

Saurai 2015 [9], Lopez-Alarcon *et al* 2013 [10] and Martin-Sanchez *et al* 2014 [11] were studied that polyphenols, such as flavonoids, hydroxycinnamic acids and proanthocyanidins, are considered as a powerful antioxidants and these antioxidants are found to be

distributed in various plants especially in forest trees. The flavonols (e.g., quercetin) and hydroxycinnamic acids (e.g., Caffeic and Ferulic acids) were determined to be more potent antioxidants than ascorbic acid. Hence, this present study was mainly concentrated on estimation of phenol, flavonoid, tannin, total antioxidant capacity and antioxidant assays such as DPPH and ABTS.

The present work was mainly aimed to estimate three important secondary metabolites such as Phenols, Flavonoids, Tannins, Total Antioxidant Capacity etc, also it was focused on the Antioxidant study by carrying out two assays like DPPH and ABTS assays.

II. MATERIALS AND METHODS

Extraction of Plant Materials

Leaves of seven species of *Salacia* such as *S. beddomei*, *S. chinensis*, *S. fruticosa*, *S. gambleana*, *S. macrosperma*, *S. malabarica* and *S. oblonga* were collected from Wayanad district of Kerala. They are washed thoroughly, and shade dried. Plant materials were coarsely powdered and soaked in 300ml of Methanol for 48hrs. The extract was filtered using Whatmann No.1 filter paper and the filtrate was concentrate under reduced pressure in vacuum at 40° C for 25min using a rotary evaporator and kept into lyophilisation to remove the traces of solvent.

Estimation Studies

Total Phenolic Content

Total phenols were assayed according to Dewanto *et al* 2002 [12] with some modifications. Various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were taken and an aliquot of diluted extract was added to 0.5 mL of distilled water and 0.125 mL of Folin–Ciocalteu reagent. The mixture was shaken and allowed to stand for 6 min, before addition of 1.25 mL of 7% Na₂CO₃. The solution was then adjusted with distilled water to a final volume of 3 mL and mixed thoroughly. After incubation in the dark, absorbance at 760 nm was read versus a prepared blank. The total phenol content of plant parts was expressed as milligrams of gallic acid equivalents per gram of dry weight (mg GAE/g DW) from a calibration curve with gallic acid. All samples were analyzed in three replicates.

Total Flavonoid Content

Total flavonoid content in leaf extracts of *S. molesta* was determined by aluminum chloride colorimetric method [13]. 0.5 ml of extracts of at a various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were taken and the volume was made up to 3 ml with methanol. Then, 0.1 ml AlCl₃ (10%), 0.1 ml of potassium acetate and 2.8 ml distilled water were added sequentially. The test solution was vigorously shaken. Absorbance was recorded at 415 nm after 30 minutes of incubation. A standard calibration plot was generated at 415 nm using known concentrations of quercetin. The concentrations of flavonoid in the test samples were calculated from the calibration plot and expressed as mg quercetin equivalent (QE)/g of sample. All samples were analyzed in three replicates.

Total Tannin Content

The tannins were determined by Folin - Ciocalteu method. Various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were taken. About 0.1 ml of the sample extract was added to a volumetric flask (10 ml) containing 7.5 ml of distilled water and 0.5 ml of Folin-Ciocalteu phenol reagent, 1 ml of 35 % Na₂CO₃ solution and dilute to 10 ml with distilled water. The mixture was shaken well and kept at room temperature for 30 min. A set of reference standard solutions of gallic acid were prepared in the same manner as described earlier. Absorbance for test and standard solutions were measured against the blank at 725 nm with an UV/Visible spectrophotometer. The tannin content was expressed in terms of mg of GAE /g of extract [14].

Antioxidant Study

Total Antioxidant Capacity

Phosphomolybdenum assay (PM)

Total antioxidant activity was estimated by phosphomolybdenum assay [15]. Various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were used. Preparation of Molybdate Reagent Solution 1ml each of 0.6

M sulfuric acid, 28 mM sodium phosphate and 4 mM ammonium molybdate were added in 20 ml of distilled water and made up volume to 50 ml by adding distilled water. The Chloroform and ethanolic extracts of *Pouzolzia zeylanica* and *Tribulus terrestris* were taken in different concentration ranging from 100 l to 500 l were added to each test tube individually containing 3 ml of distilled water and 1 ml of Molybdate reagent solution. These tubes were kept incubated at 95 C for 90 min. After incubation, these tubes were normalized to room temperature for 20-30 min and the absorbance of the reaction mixture was measured at 695 nm. Total Antioxidant activity was expressed in terms of Mm of Ascorbic acid/g of extract and was calculated using calibration curve.

a. Scavenging effect on 1,1 - diphenyl- 2-picrylhydrazyl radicals (DPPH)

The free radical scavenging activity of methanolic leaf extract of selected species of *Salacia* was analysed with stable 1,1 - diphenyl - 2 - picryl hydrazyl radical (DPPH) (Sigma - Aldrich) photo spectrometrically as stated by [16]. Stock solution was prepared with Methanol solution for each plant extract at a concentration of 1 mg/ml. From the stock solution. Various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were taken and mixed with equal volume of Methanolic solution of DPPH (0.1 mM). 0.5 ml of each sample in Methanol solution was mixed with 2.5 ml of 0.5 mM of Methanolic DPPH solution. The mixture was vortexed vigorously and incubated for 30 minutes in a dark room under room temperature. The absorbance was measured at 517 nm against a blank using a UV Spectrophotometer (DU 800; Beckman Coulter, Fullerton, CA, USA). Ascorbic acid was used as the positive control. The experiments were conducted in triplicates.

b. Scavenging effect on ABTS radical cation (2,20 azinobis-(3-ethylbenzthiazoline -6-sulphonic acid)

The ABTS cation radical scavenging activity of the extracts was determined according to the modified method of Re *et al.* (1999) [17]. A stock solution of ABTS was produced by mixing 7 mM aqueous solution of ABTS with 2.45 mM of potassium persulfate in the dark at ambient temperature for 12–16 h before use. Various concentrations such as 20µg, 40µg, 60 µg, 80 µg and 100 µg were taken. The radical cation solution was further diluted until the initial absorbance value of 0.7± 0.005 at 734 nm was reached. For assaying test samples, 0.98 mL of ABTS solution was mixed with 0.02 mL of the plant extracts. The decrease in absorbance was recorded at 0 min. and after 6 min. Ascorbic acid was used as the positive control. The experiments were conducted in triplicates.

Statistical analysis

The data was statistically analyzed by one-way ANNOVA using SPSS 17.0. The difference was considered significant when p<0.005. Triplicate assays were performed for each set of test conditions. All the values were expressed

as Mean \pm SD (Standard Deviation). IC₅₀ Value is also calculated for all test conditions.

III.RESULTS AND DISCUSSION

Total Phenolic Content

Total phenolic content of various species of *Salacia* such as *Salacia beddomei*, *Salacia chinensis*, *Salacia fruticosa*, *Salacia gambleana*, *Salacia macrosperma*, *Salacia malabarica* and *Salacia oblonga* was estimated at 84.45 mg/g, 85.38 mg/g, 64.96 mg/g, 68.51 mg/g, 81.84 mg/g, 49.40 mg/g and 52.19 mg/g respectively. Gallic acid was taken as positive control which has 47.90 mg/g of total phenol. Phenols and polyphenols found in plants are two secondary metabolites considered as natural antioxidants. They are usually quantified by using Folin's reagent. They are found to be an effective hydrogen donors widely distributed through out the plant kingdom. There is a wide difference between phenolic compounds in the effectiveness as antioxidants [18] (Table 1 & Fig 1).

Total Flavonoid Content

Total flavonoid content of various species of *Salacia* was estimated as *Salacia beddomei* (54.96 mg/g), *Salacia chinensis* (72.37 mg/g), *Salacia fruticosa* (61.33 mg/g), *Salacia gambleana* (75.91), *Salacia macrosperma* (84.96), *Salacia malabarica* (50.74 mg/g) and *Salacia oblonga* (52.73 mg/g) whereas standard positive control quercetin has total flavonoid of 46.94 mg/g which was lesser than *Salacia* species. Lee 2007 [19] defined flavonoids are the plant pigments which are responsible of the colour of floral parts. Flavonoids are ketonic compounds which can induce anti inflammatory activity and inhibits the oxygen compounds and pro inflammatory activity of an enzyme cyclooxygenase. Moreover, flavonoids possess a potent anti inflammatory activity as they inhibit prostaglandin synthesis (Table 2 & Fig 2).

Total Tannin Content

The Tannins form a major group of polyphenols in our diets and are classified into two groups namely hydrolysable tannins and condensed tannins [20]. Total Tannin Content of various species of *Salacia* was estimated as *Salacia beddomei* (62.42 mg/g), *Salacia chinensis* (51.41 mg/g), *Salacia fruticosa* (65.21 mg/g), *Salacia gambleana* (80.15 mg/g), *Salacia macrosperma* (83.97 mg/g), *Salacia malabarica* (64.98 mg/g) and *Salacia oblonga* (69.17 mg/g) whereas standard positive control Gallic acid contained 52.34 mg/g of total tannin (Table 3 & Fig 3).

Antioxidant Study

Antioxidants are thus useful in protecting cells from such oxidative damage. Synthetic antioxidants, such as butylated hydroxyanisole (BHA) and butylated

hydroxytoluene (BHT) are often used as food additives [21]. Natural antioxidants are an interesting alternative in view of their variety of structures and chemical interactions, as well as the numerous biological activities they can perform. Intensive research activities are currently being carried out on plant antioxidants to meet this challenge. Several studies have suggested that polyphenols, such as flavonoids, hydroxycinnamic acids and proanthocyanidins, act as powerful antioxidants.

Total Antioxidant Capacity

Methanolic leaf extract of *Salacia* species shows good Total Antioxidant Activity (TAA). Maximum activity was recorded in *Salacia macrosperma* (72.36mg/g) and *Salacia beddomei*, *Salacia fruticosa*, *Salacia gambleana*, *Salacia macrosperma*, *Salacia malabarica*, and *Salacia oblonga* showed activity of 37.75 mg/g, 51.19 mg/g, 54.06 mg/g, 65.66 mg/g, 68.62 mg/g respectively. Ascorbic acid was taken as a positive control which showed 30.21 mg/g of activity. In this study, all seven species of *Salacia* showed Maximum total antioxidant activity than Ascorbic acid (Positive Control) (Table 4 & Fig 4).

a. Scavenging effect on 1,1 - diphenyl- 2-picrylhydrazyl radicals (DPPH)

According to Huang *et al* 2005 [22], DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) free radical method is one of the antioxidant assay which is based on transfer of electrons that produces a violet colour solution on treating with ethanol. DPPH is a free radical and is stable at room temperature, get reduced in the presence of an antioxidant molecule results in colourless ethanol solution. This assay is an easy and rapid method to evaluate antioxidants by spectrophotometric method.

DPPH Radical Scavenging Activity of various concentrations of methanolic extracts of *Salacia* species such as *Salacia beddomei*, *Salacia chinensis*, *Salacia fruticosa*, *Salacia gambleana*, *Salacia macrosperma*, *Salacia malabarica* and *Salacia oblonga* was estimated as 105 μ g/ml, 108 μ g/ml, 102 μ g/ml, 100 μ g/ml, 104 μ g/ml, 110 μ g/ml, 107 μ g/ml, respectively. Butylated Hydroxy Toluene (BHT) was taken as positive control which showed activity of 195 μ g/ml (Table 5).

Many protocols have been carried out for DPPH antioxidant assays which are results in variation of results in different laboratories. Sharma and Bhat (2009) [23] have studied various protocols carried by different workers with incongruity in their results and found out a standard protocol under sensitivity range of spectrophotometry besides sensitivity of DPPH to light, pH and solubility of the compound..

b. Scavenging effect on ABTS radical cation (2,20 azinobis-(3-ethylbenzthiazoline -6-sulphonic acid)

ABTS radical scavenging assay employs a specific absorbance of 734nm at a wavelength ranging from the visible region and also requires short reaction time. It can be used as an index which reflects the antioxidant activity of the test samples [24]. ABTS assay of various species of *Salacia* was estimated as *Salacia beddomei* (153µg/ml), *Salacia chinensis* (167µg/ml), *Salacia fruticosa* (146µg/ml), *Salacia gambleana* (138µg/ml), *Salacia macrosperma* (151µg/ml), *Salacia malabarica* (183µg/ml) and *Salacia oblonga* (162µg/ml). Ascorbic acid was used as a positive control which showed 201µg/ml of assay activity (Table 6). This method is used for investigating hydroxybenzoic and hydroxycinnamic acid and it can get an advantage of the fact that they probably act as radical scavengers owing to their hydrogen and electron donating capacity and their ability to delocalize/stabilize the resulting phenoxyl radical within the structure [25].

IV. CONCLUSION

The present study investigates the estimation of total phenol, flavonoid, tannin, total antioxidant capacity and antioxidant assays such as DPPH and ABTS assays of seven species of *Salacia*. The inhibitory property of this extract may be attributed to the presence of phytochemicals such as saponins, phenols and flavonoids. Hence, this valuable genus should be protected in all aspects.

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Table 1. ESTIMATION OF TOTAL PHENOLIC CONTENT OF VARIOUS SPECIES OF SALACIA

S.No	Concentration (µg)	<i>S.beddomei</i> (Mean ± SD)	<i>S.chinensis</i> (Mean ± SD)	<i>S.fruticosa</i> (Mean ± SD)	<i>S.gambleana</i> (Mean ± SD)	<i>S.macrosperma</i> (Mean ± SD)	<i>S.malabarica</i> (Mean ± SD)	<i>S.oblonga</i> (Mean ± SD)	Gallic Acid (Mean ± SD)
1.	20	0.391± 0.009	0.313± 0.009	0.380± 0.008	0.319± 0.007	0.376± 0.009	0.324± 0.008	0.372± 0.009	0.125± 0.006
2.	40	0.782 ± 0.006	0.626± 0.006	0.761± 0.010	0.776± 0.007	0.783± 0.007	0.642± 0.009	0.773± 0.009	0.250± 0.010
3.	60	1.174 ± 0.010	0.939± 0.007	1.141± 0.009	0.961± 0.009	0.947± 0.008	1.153± 0.009	1.246± 0.010	0.374± 0.009
4.	80	1.565 ± 0.008	1.253± 0.010	1.522± 0.007	1.541± 0.008	1.387± 0.009	1.543± 0.009	1.462± 0.007	0.499± 0.008
5.	100	1.957 ± 0.011	1.566± 0.007	1.902± 0.008	1.647± 0.009	1.926± 0.007	1.934± 0.009	1.876± 0.007	0.624± 0.006

Table 2. ESTIMATION OF TOTAL FLAVONOID CONTENT OF VARIOUS SPECIES OF SALACIA

S.No	Concentration (µg)	<i>S.beddomei</i> (Mean ± SD)	<i>S.chinensis</i> (Mean ± SD)	<i>S.fruticosa</i> (Mean ± SD)	<i>S.gambleana</i> (Mean ± SD)	<i>S.macrosperma</i> (Mean ± SD)	<i>S.malabarica</i> (Mean ± SD)	<i>S.oblonga</i> (Mean ± SD)	Gallic Acid (Mean ± SD)
1.	20	0.325± 0.007	0.376± 0.008	0.358± 0.010	0.383± 0.008	0.387± 0.009	0.263± 0.007	0.321± 0.006	0.135± 0.009
2.	40	0.650 ± 0.009	0.752± 0.006	0.717± 0.009	0.766± 0.006	0.775± 0.006	0.526± 0.010	0.643± 0.008	0.270± 0.008
3.	60	0.976 ± 0.008	1.128± 0.011	1.076± 0.011	1.149± 0.010	1.162± 0.008	0.790± 0.009	0.964± 0.007	0.406± 0.006
4.	80	1.301 ± 0.010	1.504± 0.010	1.435± 0.008	1.532± 0.007	1.550± 0.006	1.053± 0.007	1.286± 0.010	0.541± 0.007
5.	100	1.627 ± 0.009	1.881± 0.009	1.794± 0.013	1.916± 0.010	1.938± 0.010	1.316± 0.008	1.607± 0.007	0.676± 0.008

Table 3. ESTIMATION OF TOTAL TANNIN CONTENT OF VARIOUS SPECIES OF SALACIA

S.No	Concentration (µg)	<i>S.beddomei</i> (Mean ± SD)	<i>S.chinensis</i> (Mean ± SD)	<i>S.fruticosa</i> (Mean ± SD)	<i>S.gambleana</i> (Mean ± SD)	<i>S.macrosperma</i> (Mean ± SD)	<i>S.malabarica</i> (Mean ± SD)	<i>S.oblonga</i> (Mean ± SD)	Gallic Acid (Mean ± SD)
1.	20	0.413± 0.007	0.407± 0.008	0.420± 0.006	0.310± 0.007	0.411± 0.009	0.413± 0.010	0.409± 0.010	0.152± 0.003
2.	40	0.826 ± 0.008	0.814± 0.006	0.841± 0.009	0.622± 0.009	0.822± 0.009	0.826± 0.009	0.818± 0.009	0.304± 0.005
3.	60	1.240 ± 0.006	1.220± 0.009	1.261± 0.008	0.932± 0.010	1.233± 0.008	1.239± 0.011	1.227± 0.007	0.456± 0.002
4.	80	1.653 ± 0.009	1.627± 0.007	1.682± 0.010	1.243± 0.008	1.644± 0.006	1.652± 0.008	1.636± 0.006	0.609± 0.003
5.	100	2.066 ± 0.010	2.034± 0.010	2.102± 0.008	1.554± 0.006	2.055± 0.010	2.065± 0.012	2.045± 0.009	0.761± 0.004

Table 4. TOTAL ANTIOXIDANT CAPACITY (TAC) OF VARIOUS SPECIES OF SALACIA

S.No	Concentration (µg)	<i>S.beddomei</i> (Mean ± SD)	<i>S.chinensis</i> (Mean ± SD)	<i>S.fruticosa</i> (Mean ± SD)	<i>S.gambleana</i> (Mean ± SD)	<i>S.macrosperma</i> (Mean ± SD)	<i>S.malabarica</i> (Mean ± SD)	<i>S.oblonga</i> (Mean ± SD)	Ascorbic Acid (Mean ± SD)
1.	20	0.184± 0.006	0.717± 0.009	0.268± 0.010	0.276± 0.006	0.217± 0.008	0.328± 0.009	0.297± 0.009	0.026± 0.007
2.	40	0.328± 0.009	0.757± 0.007	0.357± 0.008	0.378± 0.010	0.348± 0.010	0.467± 0.010	0.524± 0.005	0.068± 0.009
3.	60	0.672 ± 0.008	0.976± 0.006	0.475± 0.007	0.527± 0.009	0.525± 0.006	0.674± 0.007	0.776± 0.007	0.136± 0.008
4.	80	0.924 ± 0.007	1.028± 0.010	0.616± 0.007	0.875± 0.006	0.656± 0.007	0.725± 0.007	0.815± 0.007	0.326± 0.007
5.	100	1.078 ± 0.010	1.145± 0.006	0.937± 0.008	1.026± 0.007	0.925± 0.006	1.096± 0.006	1.117± 0.009	0.578± 0.010
Total Antioxidant Capacity (TAC)		37.75 mg/g	36.71 mg/g	51.19 mg/g	54.06 mg/g	72.36 mg/g	65.66 mg/g	68.62 mg/g	30.21 mg/g

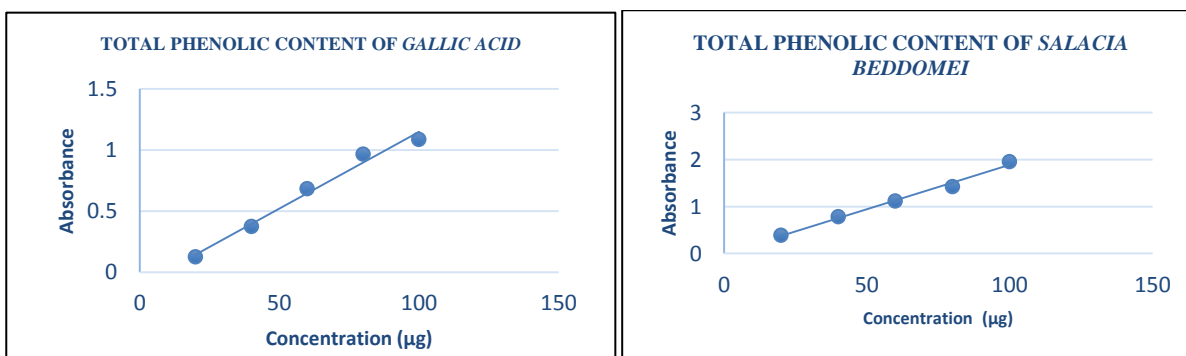
Table 5. DPPH ASSAY OF VARIOUS SPECIES OF SALACIA

S.No	Concentration (µg)	<i>S.beddomei</i> (Mean ± SD)	<i>S.chinensis</i> (Mean ± SD)	<i>S.fruticosa</i> (Mean ± SD)	<i>S.gambleana</i> (Mean ± SD)	<i>S.macrosperma</i> (Mean ± SD)	<i>S.malabarica</i> (Mean ± SD)	<i>S.oblonga</i> (Mean ± SD)	BHT (Mean ± SD)
1.	20	41.25± 0.064	46.12± 0.073	51.23± 0.031	43.21± 0.029	41.19± 0.065	39.82± 0.072	43.23± 0.046	23.96± 0.083
2.	40	41.53± 0.034	49.74± 0.037	54.01± 0.056	45.63± 0.038	43.25± 0.043	42.71± 0.051	44.41± 0.040	28.49± 0.071
3.	60	44.67 ± 0.037	54.06± 0.058	56.23± 0.158	50.01± 0.031	52.38± 0.032	45.08± 0.049	46.81± 0.037	33.61± 0.089
4.	80	52.54 ± 0.037	56.49± 0.160	57.31± 0.063	52.29± 0.042	55.63± 0.057	49.18± 0.032	49.02± 0.033	35.02± 0.068
5.	100	56.70 ± 0.099	57.55± 0.061	60.12± 0.052	59.89± 0.069	57.01± 0.049	55.13± 0.031	53.72± 0.041	38.80± 0.056
IC₅₀ Value		105 µg/ml	108 µg/ml	102 µg/ml	100 µg/ml	104 µg/ml	110 µg/ml	107 µg/ml	195 µg/ml
P - Value		0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Table 6. ABTS ASSAY OF VARIOUS SPECIES OF SALACIA

S.No	Concentration (µg)	<i>S.beddomei</i> (Mean ± SD)	<i>S.chinensis</i> (Mean ± SD)	<i>S.fruticosa</i> (Mean ± SD)	<i>S.gambleana</i> (Mean ± SD)	<i>S.macrosperma</i> (Mean ± SD)	<i>S.malabarica</i> (Mean ± SD)	<i>S.oblonga</i> (Mean ± SD)	Ascorbic Acid (Mean ± SD)
1.	20	29.32± 0.081	28.46± 0.076	32.86± 0.064	31.09± 0.072	29.86± 0.078	21.08± 0.082	32.18± 0.078	18.16± 0.098
2.	40	29.63± 0.063	29.01± 0.081	39.23± 0.048	39.83± 0.059	30.12± 0.043	30.23± 0.064	36.06± 0.063	21.32± 0.086
3.	60	32.98 ± 0.066	42.08± 0.061	41.32± 0.028	41.32± 0.061	39.68± 0.063	38.07± 0.078	40.61± 0.053	29.48± 0.061
4.	80	38.01 ± 0.072	46.14± 0.086	46.03± 0.032	50.89± 0.093	42.86± 0.042	44.18± 0.056	43.20± 0.038	33.04± 0.082
5.	100	43.30 ± 0.053	51.10± 0.053	50.13± 0.041	52.61± 0.081	51.03± 0.061	49.53± 0.036	46.13± 0.043	45.61± 0.059
IC₅₀ Value		153 µg/ml	167 µg/ml	146 µg/ml	138 µg/ml	151 µg/ml	183 µg/ml	162 µg/ml	201 µg/ml
P - Value		0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000

Fig 1. TOTAL PHENOLIC CONTENT OF SALACIA SPECIES



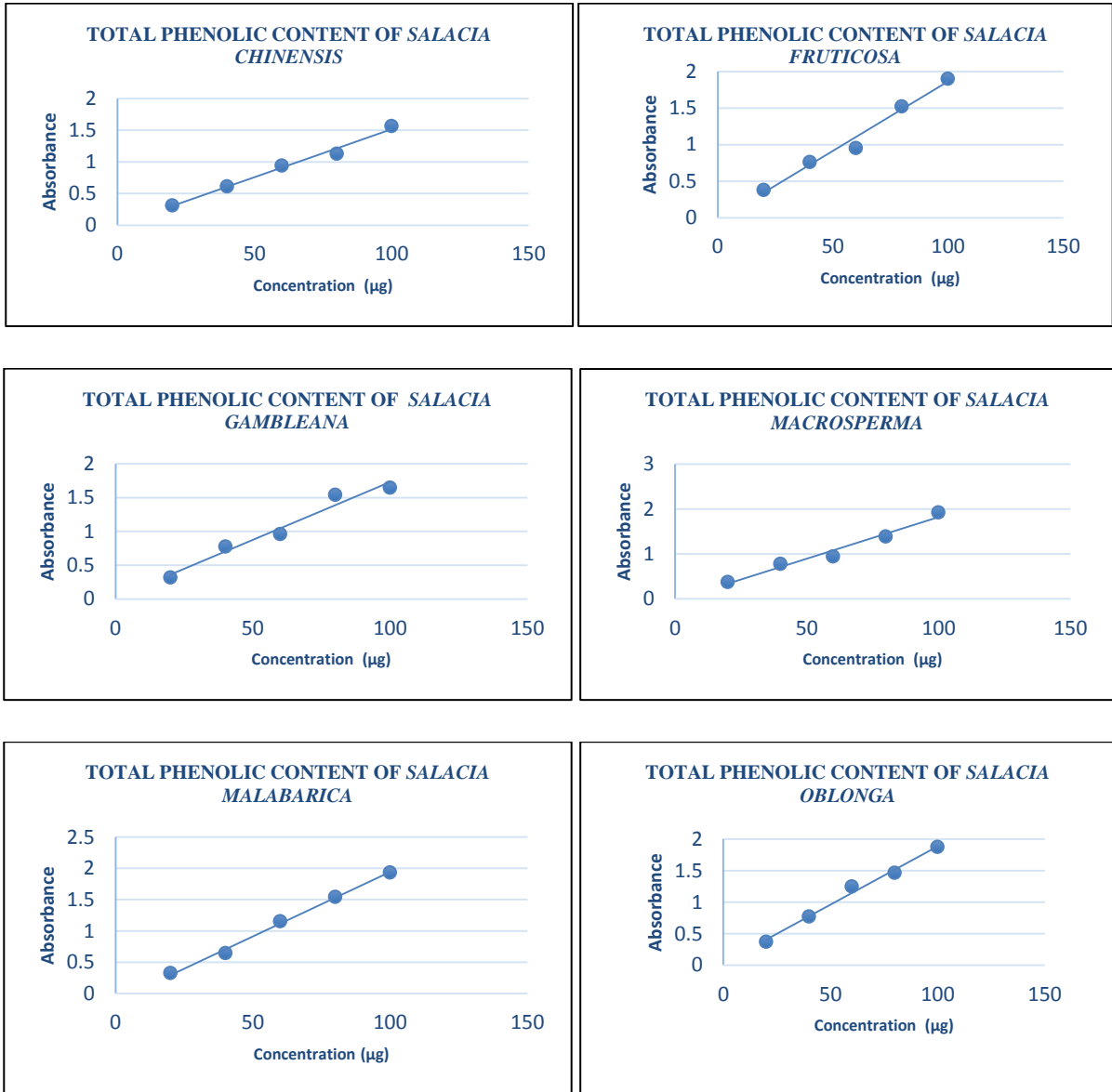
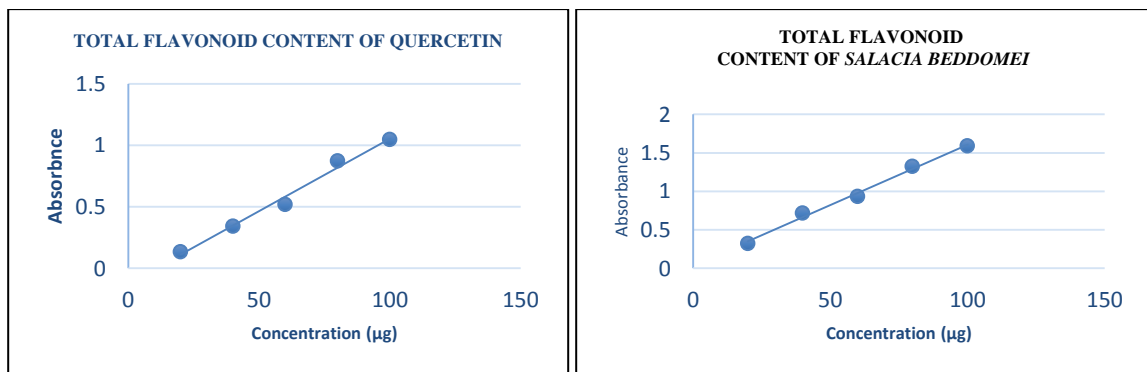


Fig 2. TOTAL FLAVONOID CONTENT OF SALACIA SPECIES



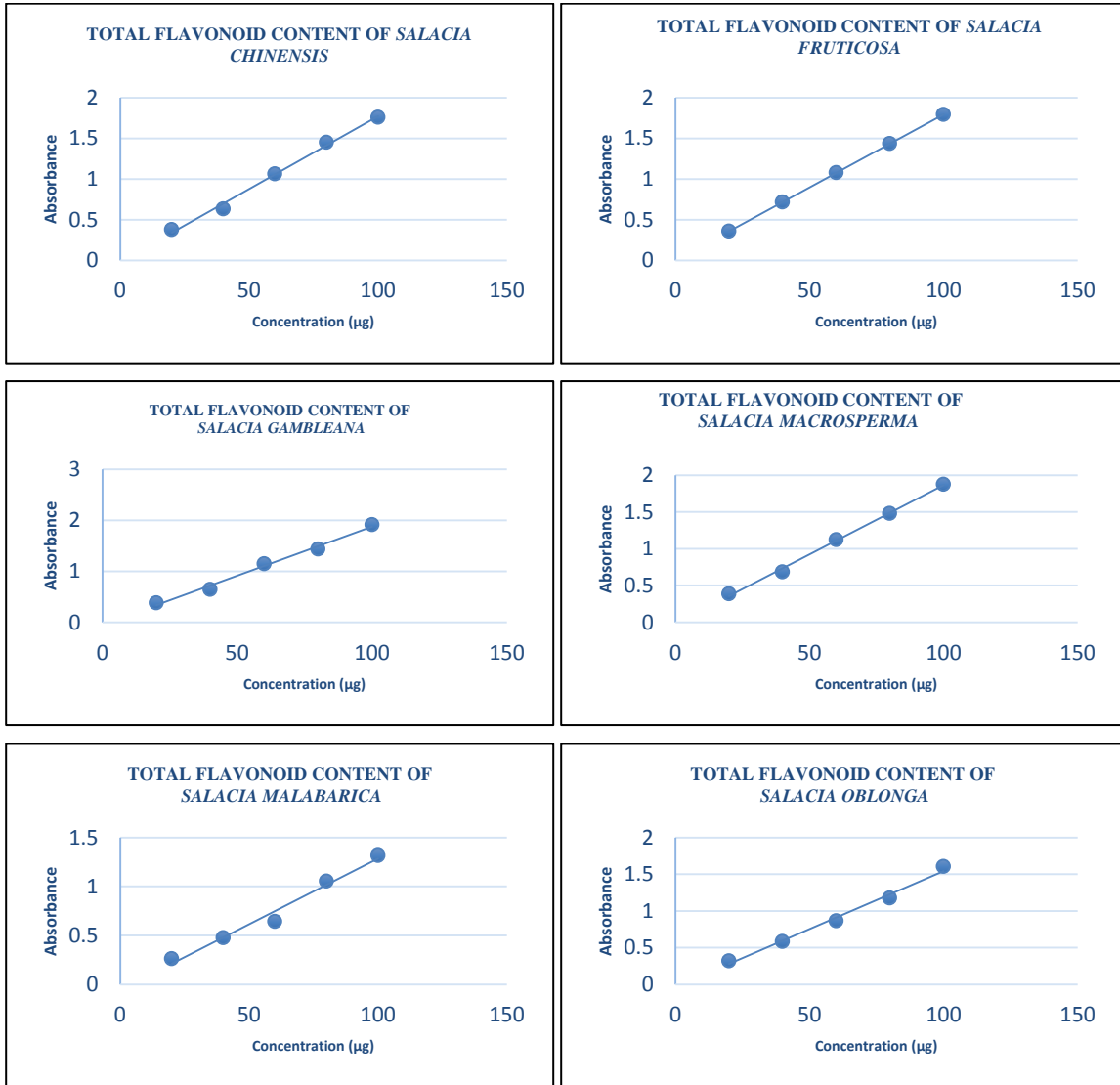
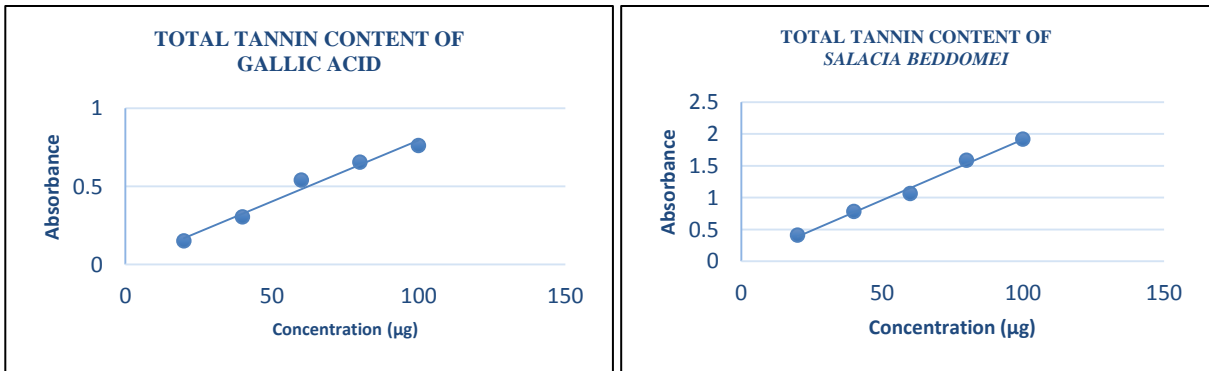


Fig 3. TOTAL TANNIN CONTENT OF SALACIA SPECIES



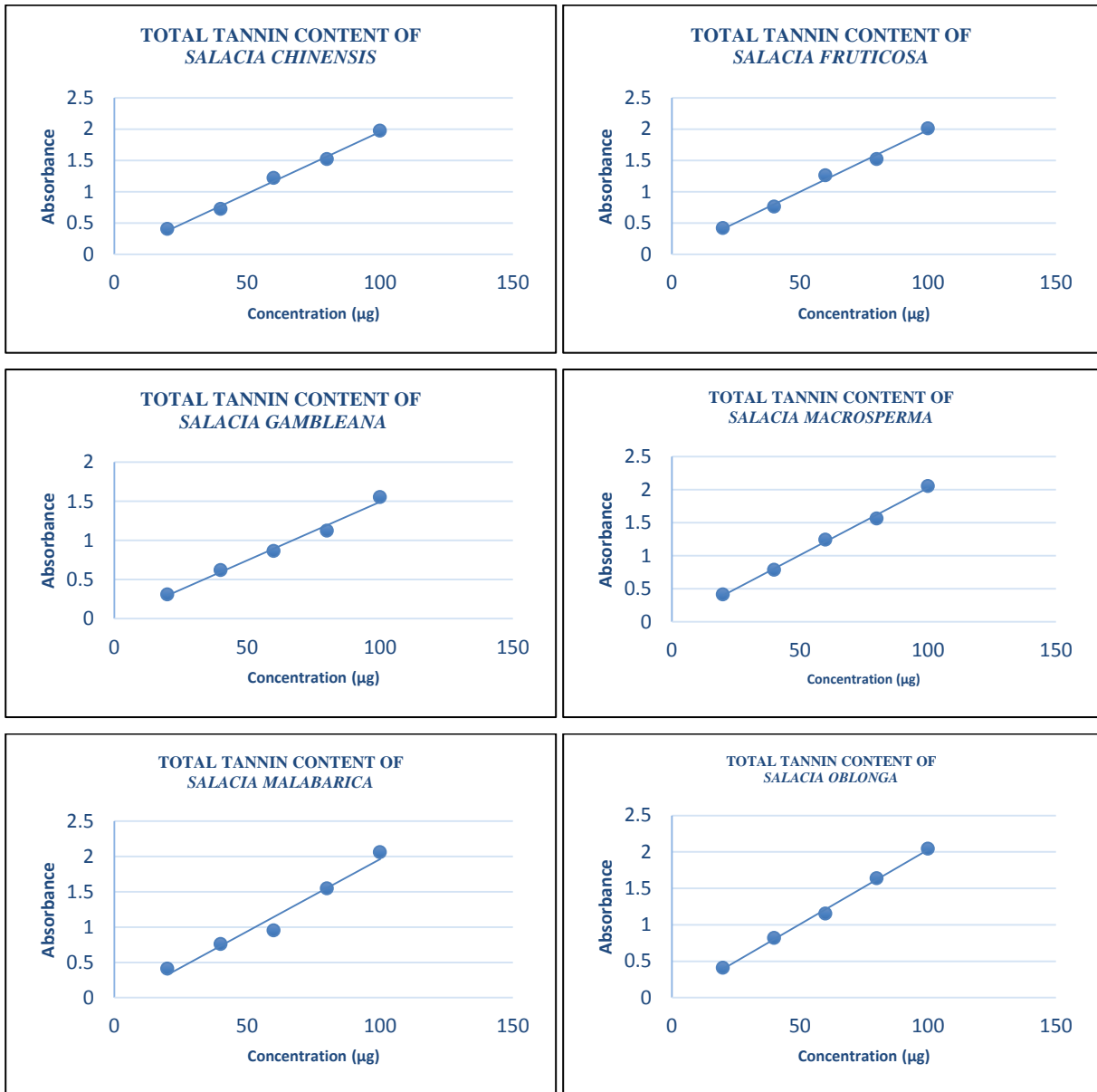


Fig 4. TOTAL ANTIOXIDANT CAPACITY OF SALACIA SPECIES

