



Phytochemical screening, antimicrobial and radical scavenging activity of *Senna siamea* (Lam.)

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Abstract

Crude extract from *Sennasiamea* was screened for their in vitro antioxidant and antimicrobial properties. Phytochemical analysis as well as (GC-MS) Gas Chromatography and Mass Spectroscopy analysis of crude leaves extract of *Senna siamea* was conducted, with a view to identifying compounds with biological activities. Antimicrobial activity was determined by using Agar well diffusion assay and MIC (Minimum inhibitory concentration) micro dilution method. The antioxidant capacities of *Senna siamea* extract was evaluated using ferric reducing antioxidant power, 1,1-diphenyl-2-picrylhydrazyl (DPPH⁺) free radical scavenging, 2,2-azinobis 3-ethylbenzothiazoline 6-sulfonate (ABTS⁺) radical scavenging and the total phenolic contents of these plants was measured by the Folin–Ciocalteu method. From the results it is observed that glycosides, phenols, flavonoids and saponins was present while Tannins, alkaloids, proteins and carbohydrates was absent in extracts. The extract was shown to inhibit all test bacteria with marked inhibitory effect against *K. pneumoniae*. The Minimum inhibitory concentration and minimum bacterial concentration was ranges from 0.312 to 2.5mg/ml. The extract scavenged 1, 1-diphenyl-2-picrylhydrazyl and 2, 2-azinobis 3-ethylbenzothiazoline 6-sulfonate radical scavenging radicals with an (IC₅₀) inhibitory concentration value of 20.63µg/ml and 11.05 µg/ml, respectively. Total phenolic content was (19.84) mg/g of Gallic acid equivalents. In ferric reducing antioxidant power assay extract are increased in reducing powers with increasing concentration. Compounds in crude extracts was identified by GC-MS. A total of 34 compounds was detected from the methanol extract respectively. The results provided evidence that the studied plant might indeed be potential sources of natural antioxidant and antimicrobial agents.

Keywords: *Senna siamea*, Antimicrobial activity, Antioxidant activity, Gas Chromatography and Mass Spectroscopy.

1. Introduction

Medicinal plants have long been utilized in traditional medicine and worldwide ethno medicine. So these plants are also known as “backbone” of traditional medicine. Before the introduction of chemical medicines, man relied on the healing properties of medicinal plants which means more than 3.3 billion people in the less developed countries utilize medicinal plants on a regular basis. The World Health Organization also recognized the use of plant extracts or their phytochemical components in the name of folk medicine as traditional therapies in 80% of the world's population for healthcare needs (Yadav *et al.*, 2011).

Antibiotic resistance is a serious problem that continues to challenge the healthcare sector in both developing and developed countries. The emergence and spread of multidrug resistant pathogens have substantially threatened the current antibacterial therapy. This has necessitated a search for a new source of antimicrobial substances isolated from extracts of medicinal plants as they produce a variety of bioactive compounds of known therapeutic properties (Dahiru *et al.*, 2013). Medicinal plants contain some phytochemical compounds which provide antimicrobial, antioxidant and definite physiological action on the human body and these bioactive substances include carbohydrates, terpenoids, steroids, tannins, alkaloid and flavonoids. These compounds are synthesized by primary or rather secondary metabolism of microorganisms (Vaou *et al.*, 2021). The antimicrobial activity of different medicinal plant extracts against some of the human pathogen such as *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Candida albicans* and *Aspergillus* spp responsible for production of some of the diseases it is determined by agar well diffusion method (Dahiru *et al.*, 2013).

Antioxidants from plant material terminate the action of free radicals thereby protecting the body from various diseases. Antioxidant activity of plants might be due to their phenolic compounds

and Flavonoids. Phenolic compounds from medicinal plants possess strong antioxidant activity and may help to protect the cells against the oxidative damage caused by free-radicals. They are well known as radical scavengers, metal chelators, reducing agents, hydrogen donors, and singlet oxygen quenchers. Antioxidants from plant materials terminate the action of free radicals thereby protecting the body from various diseases (Narayanaswamy *et al.*, 2011). Flavonoids are a group of polyphenolic compounds with known properties which include free radical scavenging, inhibition of hydrolytic oxidative enzymes and anti-inflammatory action. The major natural antioxidants include vitamin E, vitamin C (ascorbic acid), polyphenols, bioflavonoids and carotenoids (Kumar *et al.*, 2008).

Most of the common plants have been reported to have antimicrobial activity, since a number of unexplored wild plants are available in the forests having good ethnic values as traditional medicine. So, there is need for the screening of bioactive compounds present in such plants and the antimicrobial activities of their extracts. Antimicrobial resistance to commercially available medications has become a worldwide problem in recent years. Similarly, several species are developing resistance to currently available antifungal medications. It shows that there is a need to investigate other options. So medicinal plants have become the focus on significant research such as phytochemical screening, antimicrobial and radical scavenging activity of *Senna siamea*. The present study elaborates on the phytochemical analysis and antioxidant activity of *Senna siamea* methanolic extracts. It is clear that due to the complex nature of the different phytochemical classes in the plants, the antioxidant capacities of plant extracts cannot be evaluated using a single method so work on the FRAP, Total phenolic content, ABTS⁺, DPPH⁺ methods was used to assess the antioxidant activities of plant extracts.

Traditionally *Senna siamea* is used for the treatment of typhoid fever, jaundice, abdominal pain, menstrual pain, and is also used to reduce

sugar level in the blood. Ethno medicinally *S. siamea* is used as laxative, blood cleaning agent, cure for digestive system and genitourinary disorders, herpes and rhinitis. The leaves of *Senna siamea* are locally used as antimalarial drug especially when decocted. In traditional medicine, the fruit is used to charm away intestinal worms and to prevent convulsion in children (Nas *et al.*, 2018).

2. Materials and Methods

2.1. Study area and identification of plant sample

The healthy plant samples was collected aseptically with the help of secateurs, photographed, documented, and placed in paper bags. The collected plant was identified based on their taxonomic features by referring the standard floras and manuals (Uddin *et al.*, 2008).

2.2. Preparation of plant material

The collected plant samples was washed thoroughly with the running tap water to remove the adhered soil and peripheral dust particles. The completely dried plant material was pulverized in mechanical grinder to get the fine powder. The pulverized plant material was stored in zip lock polythene covers at room temperature until subjected for the extraction process (Safari *et al.*, 2019).

2.3. Extraction of Phyto-compounds from plants

The crude compound from plants was extracted using cold maceration. The pulverized plant material was immersed in methanol and agitated for 48-72 hours. The solvent-containing phyto-compounds were separated and filtered. The solvent was evaporated, and the crude extract was dried to obtain a persistent weight. The physical properties of the extract were examined, and the percentage yield was calculated. The dried extract was stored in a refrigerator for further use (Gautam *et al.*, 2020).

$$\text{Yield of crude extract (\%)} = \frac{\text{Weight of the plant crude extract}}{\text{Weight of the pulverized plant material taken}} \times 100$$

2.4. Qualitative screening of secondary metabolites in plants extract

The groups of secondary metabolites in plants extract was determined by implementing the standard protocol of qualitative tests of Phyto-chemicals (Horborne *et al.*, 1998).

2.4.1. Test for Phenols (Ferric Chloride Test)

About 2ml of plant extract was treated with 5% of FeCl₃ solution, the formation of deep blue or black colour indicates the presence of phenols.

2.4.2. Test for Tannins (Gelatin Test)

About 2 ml of aqueous solution of plant extract was mixed with 1% of gelatin solution to this mixture around 10% NaCl solution was added. The formation of white precipitation specifies the presence of tannins.

2.4.3. Test for Alkaloids (Mayer's Teat)

To the 2 ml of extract, 2 ml of Mayer's reagent was added. An organic dull white or cream color precipitate indicates the presence of alkaloids.

2.4.4 Test for Flavonoids (Alkaline Reagent Test)

About 2 ml of plant extract was treated with 20% NaOH solution. A formation of intense yellow coloration confirms the presence of flavonoids.

2.4.5. Test for Terpenoids (Salkowski's Test)

To 2 ml of extract, 2 ml of chloroform was added. To this mixture around 2 ml of concentrated H₂SO₄ was added gently along the sides of test tube. The formation of reddish-brown monolayer coloration at the interface, confirms the presence of terpenoids.

2.4.6. Test for Steroids (Liebermann-Burchard Test)

Approximately 2 ml of acetic anhydride was added to 0.5gm of plant extract and then added 2 ml of concentrated H_2SO_4 . The colour change from violet to blue or green indicates the presence of steroids.

2.4.7. Test for Saponins (Foam Test)

The plant extract was agitated with 20ml of distilled in a graduated cylinder for 15minutes. The formation of foam layer about 1cm indicates the presence of saponins.

2.4.8. Test for Glycosides (Keller-Killiani Test)

Around 2 ml of plant extract was treated with 2 ml of glacial acetic acid to this mixture 2-3 drops of 5% $FeCl_3$ solution was added to this 1ml concentrated H_2SO_4 was added gently. Development of dark brown color ring at the interphase indicates the presence of glycosides.

2.4.9. Test for Protein and Amino acids (Ninhydrin Test)

To 2 ml of plant extract, 3-5 drops of freshly prepared 2% ninhydrin reagent was added and heated on water bath. The reaction mixture turns to blue colour confirms the presence of proteins and amino acids.

2.4.10. Test for Carbohydrates (Fehling's Test)

To 2 ml of extract, equal volume Fehling's solution A and Fehling's solution of B was added and then heated on boiling water bath. The formation of brick red precipitate indicates the presence of carbohydrates.

2.5. Antimicrobial activity of methanol extract of plants

2.5.1. Preparation of test pathogenic microorganisms

The assay was performed according to the Clinical and Laboratory Standards

Institute (CLSI) guidelines. The active test bacterial cultures was inoculated into the tubes containing Mueller Hinton Broth (MHB) medium aseptically and incubated at $37^\circ C \pm 2^\circ C$ for 24 hours. The test pathogenic yeast (*Candida albicans*) culture broth was prepared by inoculating the cells in Sabouraud Dextrose Broth (SDB) tubes and incubated for 48 hours at $30^\circ C \pm 2^\circ C$. The test fungal pathogen (*Aspergillus brasiliensis*) culture broth was prepared by inoculating the spores in to the sterile SDB and incubated at $28 \pm 2^\circ C$ for 72 hours. These pathogenic broth cultures was used for the assay.

2.5.2. Agar well diffusion assay

The potential of methanolic extract of the plants to inhibit pathogenic microorganisms was determined by Agar well diffusion assay. In this method, the prepared culture broth of the test bacterial and fungal pathogens was swab inoculated on sterile Mueller Hinton Agar and Sabouraud Dextrose Agar (SDA) plates respectively. By using 6mm diameter sterile gel borer the wells was perforated into swab inoculated plates and labeled. A 100 μ l of plants extract of different concentrations (20mg/ml, 10mg/ml, 5mg/ml and 2.5mg/ml prepared in 10% DMSO) along with positive and negative control was transferred aseptically into the labeled wells. MHA and SDA plates were incubated for 18- 24 hours at $37^\circ C \pm 2^\circ C$ and $30^\circ C$ for 48 hours respectively. After incubation the zone of inhibition around the wells was the sign of antimicrobial activity of the compound was measured by using zone gauge. The increased dimension of zone indicates the proficiency of the plants extract to inhibit the test pathogens (Sathiyavimal *et al.*, 2023).

2.5.3. Minimum Inhibitory Concentration (MIC) of plants extract

The study tested the effectiveness of plants extracts in inhibiting pathogenic microorganisms using a modified resazurin 96-well microtitre plate broth dilution assay. The extract was diluted in a concentration range of 5000 to $9.76 \approx 10 \mu g/mL$ for bacterial pathogens and 10,000

to 19.53 \approx 20 μ g/mL for fungal pathogens. The MIC value of the pigment extract was determined by incubating the extract with resazurin dye for 1-2 hours. The lowest concentration without a color change was considered the MIC value (Vora *et al.*, 2018).

2.6 Antioxidant activity of plants extract

2.6.1. DPPH⁺ assay

The 1, 1-Diphenyl-2-picrylhydrazyl (DPPH⁺) radical scavenging activity of plants extract was performed as described by (Zahin *et al.*, 2009). The assay involved dissolved plant extract and ascorbic acid in methanol, with DPPH radical solution added to each tube. The reaction mixture was incubated at 37°C for 30 minutes, with methanol used as a control. The effectiveness of plant extract to scavenge the DPPH⁺ radicals was determined by using the bellow equation.

$$\text{DPPH}^+ \text{ radical scavenging activity (\%)} = \frac{(A_c - A_s)}{A_c} \times 100$$

Where, A_c is absorbance of the control and A_s is the absorbance of the tested sample.

2.6.2. ABTS⁺⁺ assay

The 2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid (ABTS⁺⁺) radical scavenging activity of the plants extract was performed as described in the method of (Rajurkar *et al.*, 2011). In this assay, 0.2 ml of plant extract of concentration ranging from 10 to 100 μ g/ml prepared in methanol was mixed with 1.8ml of the ABTS⁺⁺ radical solution. Ascorbic acid (10-100 μ g/ml concentration) was used as reference standard. The optical density of reaction mixtures was read at 734nm. The percentage of ABTS⁺⁺ free radicals scavenged was calculated by using following formula.

$$\text{ABTS}^{++} \text{ radical scavenging activity (\%)} = \frac{(A_0 - A_1)}{A_0} \times 100$$

Where, A_0 is absorbance of the control and A_1 is the absorbance of the tested sample.

2.6.3. FRAP assay

Ferric reducing power Assay (FRAP) of plants extract was determined as described by (Al-Laith *et al.*, 2019). A mixture of plant extract, phosphate buffer, and potassium ferricyanide was mixed and incubated at 50°C for 20 minutes. After cooling, trichloroacetic acid and ferric chloride were added, and absorbance was measured at 700 nm against a blank with Ascorbic acid will be used as standard. An increase in the absorbance with increase in concentration of plant extract/standard indicates the increasing capacity of ferric ion reducing power of plant extract.

2.6.4. Total antioxidant activity

The total antioxidant activity of plants extract was determined by phospho-molybdenum assay as described by (Bayliak *et al.*, 2016). Plant extract was mixed with phospho-molybdenum reagent, mixed vigorously and incubated at 95°C for 90 minutes, then the tubes was allowed to cool and the absorbance was measured at wavelength of 765nm against methanol (blank). Ascorbic acid was used as standard.

2.6.5. Total phenolic content in the plants extract

The total phenolic content of the plants extract was determined by using Folin-Ciocalteu (FC) method. Plant extract with FC reagent, adding sodium carbonate solution, was added and vortexed vigorously and tubes were incubated at room temperature for 30 minutes and the absorbance was read at 765 nm. Gallic acid was used as standard. The total phenolic content of the plants extract will be express as gallic acid equivalent in mg/g (GAE mg/g extract) (Al-Laith *et al.*, 2019).

2.7.GC-MS (Gas Chromatography and Mass Spectroscopy) analysis

The array of Phyto-compounds present in the plants extract was determined by GC-MS analytical technique. The Bio Centre in Shivamogga conducted a GC-MS analysis on plants extract samples using a Shizuzu QP 2010 Plus instrument. The RtxR-5 column was used, and the sample was filtered using a Whatman filter. The ion sources were maintained at 250°C, and the mass spectrum of compounds was obtained by ionization of electrons at 70eV. The procedure took 22.5 minutes, including a three-minute solvent delay, and involved a total of 30

and 500 Da atomic units. A comparison was made between the extracted spectrums of plants extract with the NIST (National Institute of Standards and Technology) library version 17 database(Daskum *et al.*, 2020).

3. Results and Discussion

3.1. Study area and collection of plant sample

The plant sample was collected from the Kuvempu University at Shankaraghatta (LongitudeE75.62 and LatitudeN13.72). Location and geographical area of sampling plots are shown in Figure 1.



Fig.1 Location and geographical area of sampling plots

3.2. Identification of plant samples based on taxonomic characters.

Senna siamea (Figure 2) belongs to the family Apocynaceae, commonly called as cassia tree. Medium-size, evergreen tree, 10-12 m tall, occasionally reaching 20 m. The bole is short;

Taxonomy of *Senna siamea*

Taxonomic Rank	Taxon
Kingdom	Plantae
Class	Tracheophyta
Order	Fabales
Family	Fabaceae
Sub family	Caesalpinaceae
Genus	<i>Senna</i>
Species	<i>Siamea</i>

crown dense and rounded at first, later becoming irregular and spreading; young bark grey and smooth, later with longitudinal fissures. Leaves alternate, 15-30 cm long, compound, with 6-14 leaflets each ending in a tiny bristle. Flowers bright yellow, in large, up to 60 cm long, upright, pyramid-shaped panicles(Joker, 2000).



Fig. 2 *Senna siamea*

3.3. Physical properties and yield of the extract

The colour of the obtained crude methanolic extract of *Senna siamea* was yellowish-brown in

colour shown in Figure 3, yield and percentage of the obtained crude extract was 2.42g and 9.68% for the dried plant powder.



Fig.3 Dried Methanolic extract of *Senna siamea*

3.4. Qualitative screening of secondary metabolites in plants extracts.

Qualitative phytochemical studies identified the presence of different secondary metabolites and their derivatives. The phytochemicals groups detected in the methanolic extracts of *Senna siamea* was summarized below in Figure 4. Phenols, steroids, flavonoids, saponins and glycosides was present and tannins, alkaloids, protein was absent in the plants extract. These findings were earlier reported on. Phytochemical screening of the methanolic extracts showed that *Senna siamea* sample contain, Anthraquinone, saponin, phenol, steroid, flavonoid, terpenoid and glycosides by (Nas *et al.*, 2018).

3.5. Antimicrobial activity of methanol extracts of plants:

3.1. Agar well diffusion assay

The antibacterial and antifungal activity of the plant *S. siamea* extracts was evaluated against test

pathogenic microorganisms by agar well diffusion method. The effect of the antimicrobial activity of the compound was measured in diameter of zone of inhibition in mm. All the test bacterial pathogens were more susceptible to standard antibiotic Ciprofloxacin compared to Chloramphenicol and Streptomycin (Figure 5). Fungal pathogens were more susceptible to standard antifungal Clotrimazole compared to Fluconazole (Shown in Figure 7). The *S. siamea* extract showed potential to inhibit all the test bacterial and fungal pathogens (Figure 6 and 8 respectively). Highest zone of inhibition is demonstrated by *E. coli* (15mm) at 20mg/ml. Another fungal pathogen *C. albicans* was susceptible to the plant extract. The result of this study was in conformity with that of [6] on the antibacterial activity of *S. senna* has been previously reported by *E. coli* was found to be the highest susceptible organisms with average zone of inhibition of 14.29 ± 1.12 mm, followed by *S. aureus* (13.61 ± 1.23 mm) and *Salmonella typhi* (13.56 ± 1.89 mm).

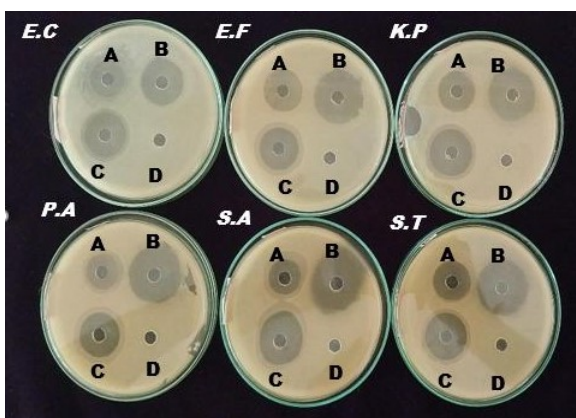


Fig. 5 Antibacterial activity of standard antibiotics

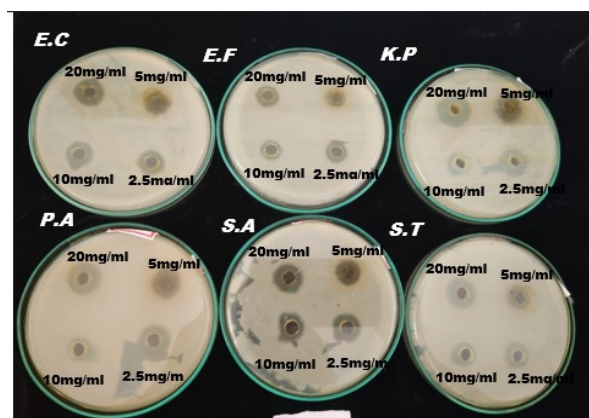


Fig. 6 Antibacterial activity of *S. siamea*

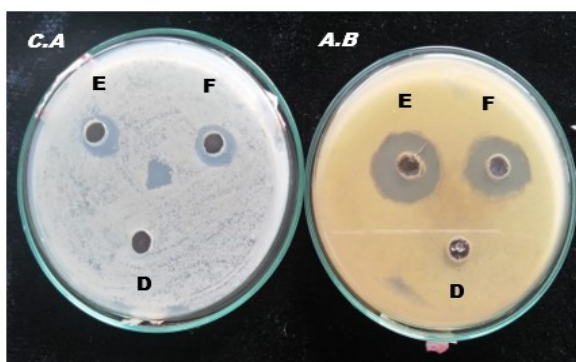
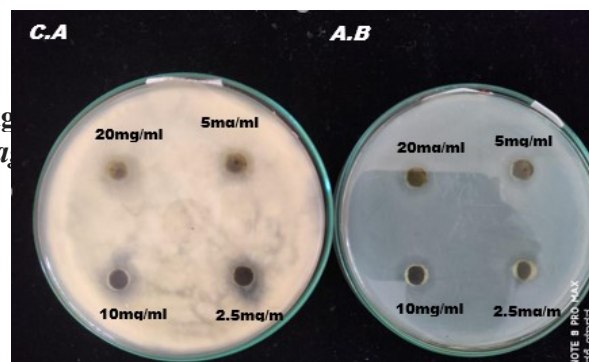


Fig. 7 Antibacterial activity of *S. siamea* against *C. albicans* and *A. brasiliensis*



NOTE: A: Streptomycin; B: Chloramphenicol; C: Ciprofloxacin; D: DMSO(10%); E: Clotrimazole; F: Fluconazole.

E.C: *E. coli*; **E.F:** *E. faecalis*; **K.P:** *K. pneumoniae*; **P.S:** *P. syringae*; **S.A:** *S. aureus*; **S.T:** *S. thypi*; **C.A:** *C. albicans*; **A.B:** *A. brasiliensis*

3.5.2. Minimum inhibitory concentration (MIC) of plant extracts.

The minimum inhibitory concentration (MIC) is the lowest concentration of antimicrobial agent, which prevents visible *in-vitro* growth of bacteria. The MIC was carried out with methanolic extract of *Senna siamea* plants. The MIC of *Senna*

siamea ranged from $0.312 \times 10^3 \mu\text{g/ml}$ to $2.5 \times 10^3 \mu\text{g/ml}$. The result of minimum inhibitory concentration of plants extract shown in Figure 9. The antibacterial activity of *S. siamea* has been previously reported by Nas *et al.*, 2018, on the MIC of the extract's ranges from 3.125 to 50mg/ml of *Senna siamea* plant extract.

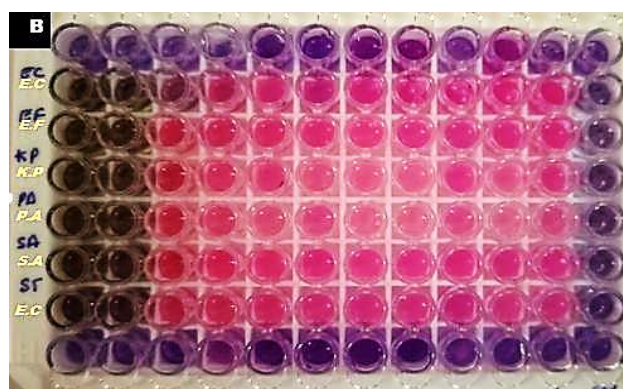


Fig. 9 MIC of methanolic extract of *Senna siamea* against bacterial strains

Note: **E.C:** *E. coli*; **E.F:** *E. faecalis*; **K.P:** *K. pneumoniae*; **P.S:** *P. syringae*; **S.A:** *S. aureus*; **S.T:** *S. thypi*

3.6. Antioxidant activity of plant extracts.

3.6.1. DPPH⁺ assay.

DPPH⁺ radical scavenging assay is one of the most frequently used methods for evaluating the antioxidant potential of plant extracts. This assay is based on the principle the decrease in absorbance of DPPH⁺ caused by antioxidants is due to the reaction between antioxidant molecules

and the radical, which results in the scavenging of the radical by hydrogen donation. In the present study, *Senna siamea* extract showed concentration dependent DPPH⁺ radical scavenging activity. IC₅₀ value is inversely related to the activity. The antioxidant potential of standard ascorbic acid was found to be 10.78 µg/ml while IC₅₀ for metabolic extract of *Senna siamea* was 20.63 µg/ml shown in Figure 10.

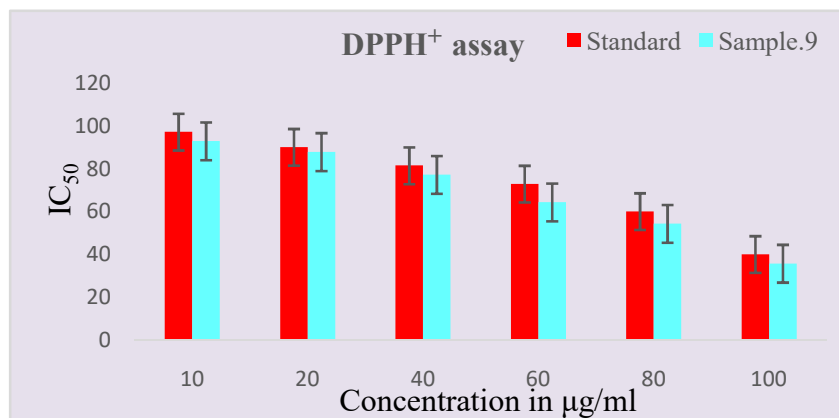


Fig. 10 DPPH⁺ radical scavenging activity of extracts and reference standard

3.6.2. ABTS⁺ assay

In the ABTS⁺ radical scavenging assay (an electron transfer-based assay), the 2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonate) radical cation (ABTS⁺), which has a dark blue color, is reduced by an antioxidant into colorless ABTS⁺. In the present study, evaluated the ability

of plants extract to scavenge ABTS⁺ radicals and the result is represented graphically shown in Figure 16. Reference compound ascorbic acid scavenged ABTS⁺ radicals more efficiently (IC₅₀ value of 5.01 µg/ml) when compared to (S9) *Senna siamea* (IC₅₀ value of 11.05 µg/ml) shown in Figure 11.

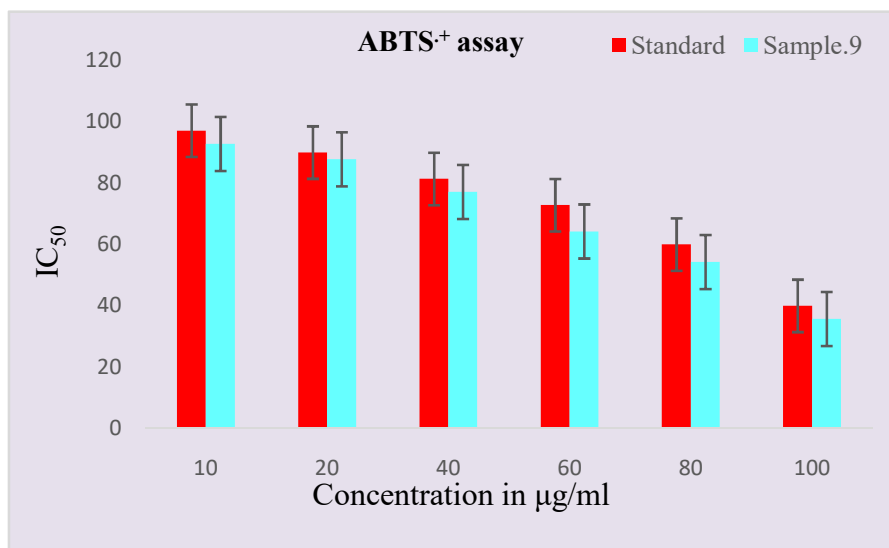


Fig.11 ABTS⁺ Radical scavenging activity of extracts and reference standard

3.6.3. FRAP assay

Ferric reducing antioxidant power (FRAP) assay is a widely used method that uses antioxidants as reductants in a redox-linked colorimetric reaction, wherein Fe^{3+} is reduced to Fe^{2+} . *Senna siamea* extracts and ascorbic acid show different absorbance at different concentration. A higher absorbance indicates higher reducing power due

to formation of reduced intermediate. Extracts are increased in reducing powers with increasing concentration. The exhibited standard higher reducing ability than the extract. The previous investigation reported that the high yield of some plant extracts contained high antioxidant activities and phenolic substances by (Haiphan *et al.*, 2014). The detailed result graphically represented below in Figure 12.

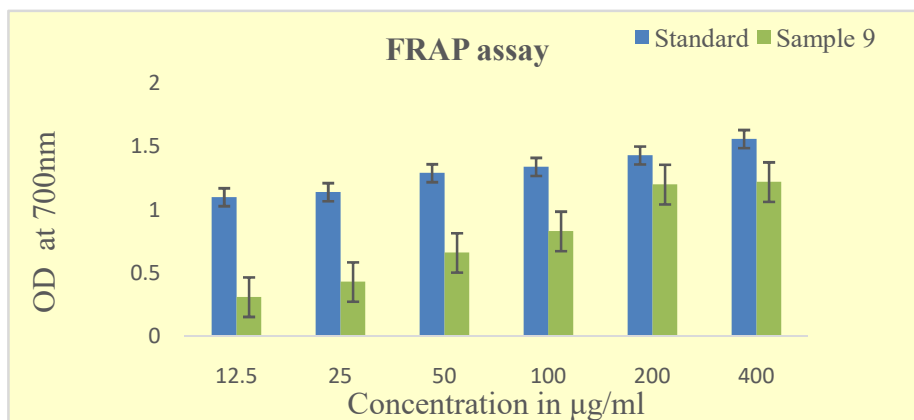


Fig12 FRAP assay values of plant extract and standard

3.6.4. Total antioxidant activity

The phosphomolybdate method has been used routinely to evaluate the total antioxidant capacity of plant extract. In the presence of extracts, Mo (VI) is reduced to Mo (V) and forms a green colored phospho-molybdenum V complex, which shows a maximum absorbance at 765nm. In the

present study, the standard exhibited higher total antioxidant activity than the *Senna siamea* (sample 9) extract. The previous investigation reported that the high yield of some plant extracts and *S. siamea* contained high antioxidant activities and phenolic substances by (Goldson Barnaby *et al.*, 2016). The detailed result graphically represented below in Figure 13.

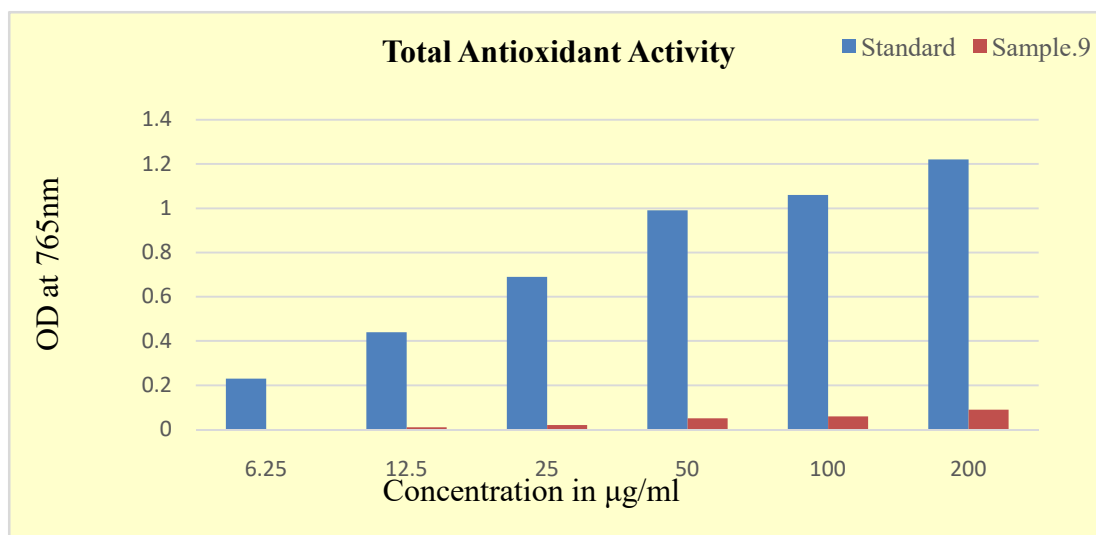


Fig. 13 Comparison of Total antioxidant activity between plant extracts and standard

3.6.5. Total phenolic content in the plant extract

Total phenolic content of the selected plant was measured using the Folin-Ciocalteu method, and the results are shown in Figure 14. Phenolic compounds are plant metabolites characterized by the presence of several phenol groups. Some of

them are very reactive in neutralizing free radicals by donating a hydrogen atom or an electron, chelating metal ions in aqueous solutions. Extract of *Senna siamea* (S9) shown the phenolic content of 18.072mg GAE/g extract. The Total phenolic content of *S. senna* have been previously reported by Kaur *et al.*, 2006, shown the phenolic content of *S. siamea* (79.37 ± 4.46 mg/g gallic acid).

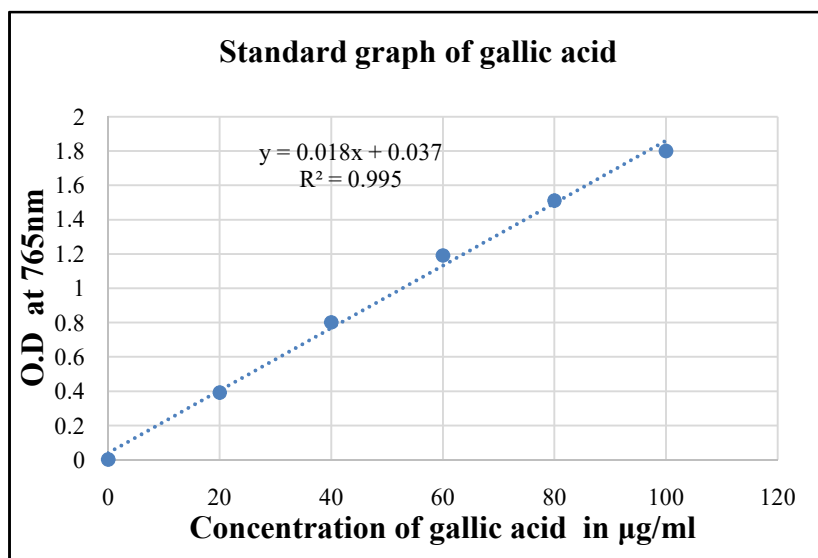


Fig.14 Standard graph of Gallic acid

Table1:Total phenol content of two extracts

Samples	<i>Senna siamea</i> (S.9)
Absorbance of plant extracts at 765nm	1.09
Plant extracts expression in GAE mg/g extract	23.12

3.7. GC-MS (Gas Chromatography and Mass Spectroscopy) analysis

The presence of certain phytochemicals in the medicinal plant crude extracts was detected by Gas Chromatography Mass Spectroscopy (GC-MS) analysis. Compounds produced was compared with those in the National Institute Standard and Technology (NIST) database. Based on the results obtained, a total of 34 compounds was detected from the methanol extract of *Senna*

siamea leaves the obtained chromatogram from GCMS analysis is shown in Figure 15. These findings was earlier reported on GC-MS analysis of *S.siamaea* revealed the presence of Saponins specifically, triterpenoids (Lupeol, α -amyrin), Sesquiterpene (Octadecane), Diterpenes (Eicosane), Esters of phthalic acids (Diethyl phthalate) Squalene, α -Tocopherol (a fat soluble Vitamin E) and hexadecanoic acid butyl ester by (Joker, 2000).

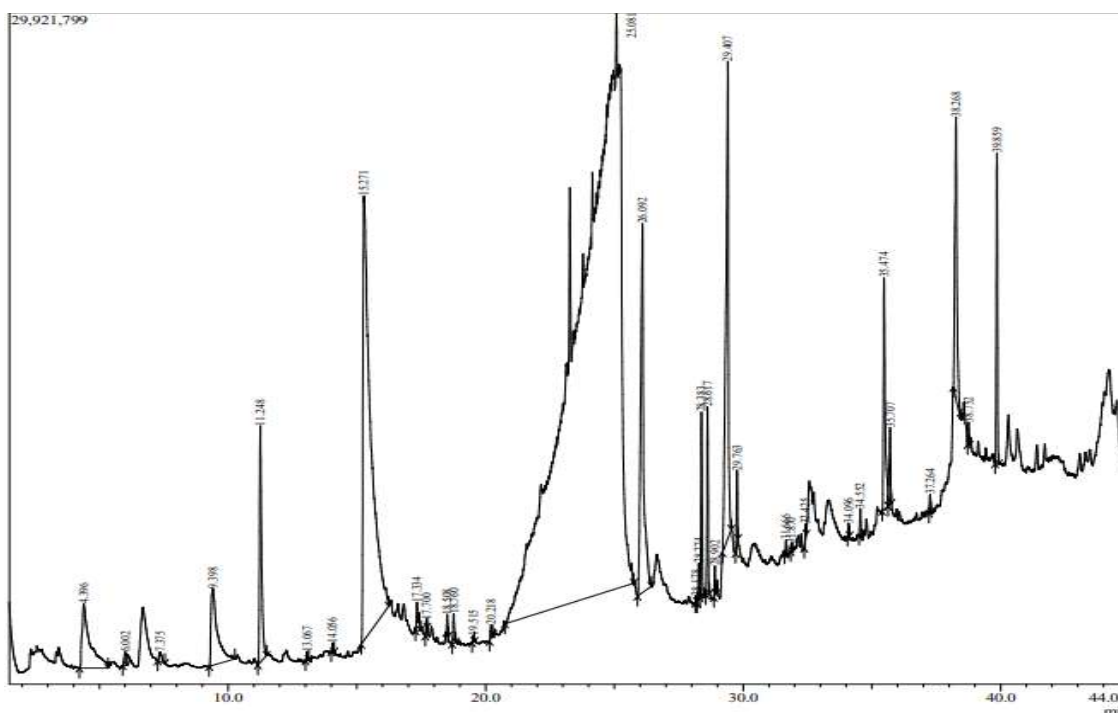
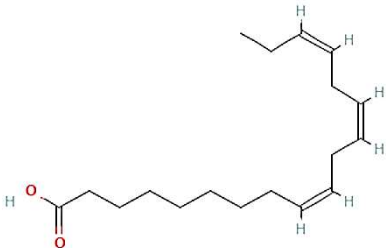
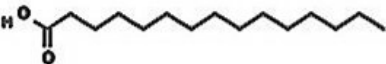
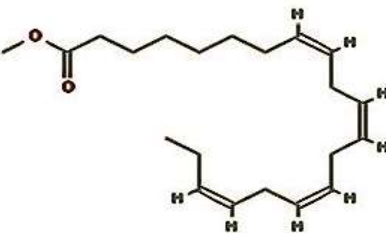


Fig.15 GC-MS chromatogram of methanol extracts of *Senna siamea* leaves.

Table 2: Major bioactive compounds present in Senna siamea extract

Compound name	Structure	Significance
<i>D-Fructose,3-O-methyl-</i>		Used to study blood-brain barrier transport and the distribution spaces of hexoses in brain.
<i>(3-Nitrophenyl) methanol, isopropyl ether</i>		This compound may be used in the pharmaceutical industry for the development of new drugs or for research purposes. It could serve as a precursor or intermediate in the synthesis of pharmaceutical compounds.

9,12,15- Octadecatrienoic acid, (Z, Z,Z)		Compound which inhibits or antagonizes the biosynthesis or actions of androgens. A drug used for the treatment or prevention of cardiac arrhythmias
Pentadecanoic acid		It has a role as a plant metabolite, a food component, a human blood serum metabolite and an algal metabolite.
Omega3, Arachidonic Acid methyl ester		ω -3 Arachidonic acid methyl ester is a neutral, lipid-soluble form of the rare fatty acid ω -3 arachidonic acid. The ω -3 fatty acids, as a group, are associated with decreased and autoimmune activity and a reduction in the thrombosis and platelet activation.

4. Conclusion

The present study demonstrated that the methanolic extract of *Senna siamea* leaves possessed strong antibacterial and antioxidant properties. The results of the present study showed that, *S. siamea* leaves extracts are effective against all tested bacteria pathogens. This work has further confirmed the presence of bioactive principles in the extracts of *Senna siamea* leaf. The methanolic extract of *Senna siamea* exhibited are an effective scavenger of free radicals therefore, a powerful inhibitor of DPPH⁺ and ABTS⁺ due to the high yield of phenolic compounds. Phyto-compounds detected from the leaves of this plant could be used as novel lead compounds to develop new drugs. For further fractionation and purification of bioactive compounds from various fractions have to be detected by GC-MS for further research is

needed to know the more bioactive potential of these plants.

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