GSC Biological and Pharmaceutical Sciences
eISSN: 2581-3250 CODEN (USA): GBPSC2
Cross Ref DOI: 10.30574/gscbps
Journal homepage: https://gsconlinepress.com/journals/gscbps/

(RESEARCH ARTICLE)

Check for updates

Phytochemical profiles and antibacterial activities of the methanolic extracts of Avartaki, *Senna auriculata* leaf and flower

Pushpalatha Veerappan [1], Saravana Bhavan Periyakali [1,*], Manjula Thangaraj [1] and Jayakumar Thanasekaran [2]

[1] *Department of Zoology, Bharathiar University, Coimbatore - 641046, Tamil Nadu, India.*
[2] *Department of Ecology and Environmental Sciences, Pondicherry University, Puducherry – 605014, India.*

GSC Biological and Pharmaceutical Sciences, 2025, 32(01), 274-290

Publication history: Received on 15May 2025; revised on 13July 2025; accepted on 15July 2025

Article DOI: https://doi.org/10.30574/gscbps.2025.32.1.0253

Abstract

The phytochemical profiles of *Senna auriculata* revealed the presence of crude protein, 15.60% and 16.56% in the leaf and flower, respectively. The methanolic extracts of *S. auriculata* showed the presence of secondary phytochemicals, such as alkaloids, flavonoids, terpenoids, saponins, tannins and quinones in the leaf, and alkaloids, flavonoids, polyphenols, saponins, cardiac glycosides and quinones in the flower. There were different secondary metabolic compounds (totally 27 in leaf and 25 in flower) were detected through the GC-MS analyses. The methanolic extracts of *S. auriculata* showed antibacterial activity against Pseudomonas *aeruginosa*, *Escarichia coli*, *Bacillus subtilis* and *Staphylococcus aureus* better than that of the positive control, Ampicillin. The methanolic extracts of *S. auriculata* leaf and flower inhibited the growth, produced leakage of intracellular compounds and apoptosis of these bacteria.

Keywords: *Senna auriculata*; Crude Protein; Phytochemicals; Secondary Metabolites; Anti-Bacterial Activity; Apoptosis

1. Introduction

There are 40 million people affected worldwide due to non-communicable diseases, such as stroke, heart attack, peripheral artery diseases, coronary artery disease, deep vein thrombosis, congenital heart disease etc. These are typically long-lasting, often caused by a combination of genetic, physiological, environmental, and behavioral factors. These are collectively responsible for 74% of all deaths worldwide [1, 2]. Therefore, the extensive use of medicinal plants to discover new therapeutics or active pharmacological compounds is the urgent need to tackle challenging non-communicable diseases. India has a great heritage of traditional systems of medicines like Ayurveda, Siddha, and Unani, where hundreds of medicinal plants are being used to treat various diseases with their known ethnopharmacological evidence.

Avartaki, *Senna auriculata* (L.) Roxby, synonym, *Cassia auriculata* (L.), Family, Fabaceae or Leguminosae. It is commonly known as Tanner's (Senna/Cassia) and Mature Tea Tree in English. This shrub is widely spread in India, covering its southern, westerns, and central dry zones and also in Sri Lanka [3-5]. It is a common traditional and Asian beverage nutritional plant which is widely used in Indian traditional medicine (Ayurveda, Siddha and Unani systems). Almost all the parts of this plant, such as leaves, flowers, unripe fruits, seeds, barks, and roots have been reported for their medicinal uses. Traditionally, it has been used in the treatment of diabetes, asthma, rheumatism, dysentery, skin disease, and metabolic disorders [6-8].

In India, the mixture various parts of *S. auriculata* are used as ''Avarai Panchaga Choornam" which is employed for the management of blood glucose level, urinary infections, conjunctivitis, and ophthalmia [9, 10]. Another preparation of

*Corresponding author: P. Saravana Bhavan

beverage is called ‘‘kalpa herbal tea” containing *S. auriculata* (dried flowers) as its major component which is widely consumed by people who are suffering from diabetes, urinary tract diseases, and constipation [10, 11]. Likewise, *S. auriculata* leaves have been used for treating ulcers, skin disease, leprosy, and herpetic eruption. The roots and flowers of this plant have shown alexeteric and anti-diabetic properties too [7, 12].

The principal phytochemicals in *S. auriculata* are alkaloids, anthraquinone, flavone glycosides, sugar, saponins, phenols, terpenoids, triterpene, flavonoids, tannins, steroids, palmitic acid, linoleic acid, benzoic acid 2-hydroxy methyl ester, 1-methyl butyl ester, resorcinol, α-tocopherol-β-D-mannosidase, epicatechin, ferulic acid, quercetin-3-O-rutinoside, quercetin, proanthocyanidin B1 etc. [13]. The extracts from its different parts and their isolated compounds possess a wide range of pharmacological activities such as antidiabetic, antioxidant, anti-inflammatory, antihyperlipidemic, hepatoprotective, nephroprotective, cardioprotective, anti-atherosclerotic, anticancer, antimutagenic, antimicrobial, antiulcer, antipyretic, anthelmintic, immunomodulatory, antifertility, anti-venom, and anti-melanogenesis.

The secondary phytochemicals of plants have numerous pharmacological benefits [14, 15]. Phytoconstituents such as alkaloids, glycosides, flavanoids, and triterpenes have been reported in methanolic extract of *S. auriculata*. The ethanolic extract of *S. auriculata* flowers and buds exhibited in vitro antioxidant and anti-diabetic properties [16]. However, reports on some other important biological properties of *S. auriculata* are limited [17]. In the present study, we aimed to investigate the primary (the proximate composition) and secondary phytochemicals, the secondary metabolites and their bioactive principles, and antimicrobial capacity of the methanolic extract of flowers and leaves of *S. auriculata*.

2. Materials and methods

2.1. Collection of plant material and authentication

The plant material (Figure 1) taken from Bharathiar University campus was identified as *Senna auriculata* by the Botanical Survey of India, Coimbatore (BSI/SRC/5/23/2024-25/Tech/191). The leaves and flower samples of *S. auriculata* were separated and dried under shadow. The fully dehydrated (well-dried) leaves and flower were ground well using a mechanical blender into fine powders and kept in airtight containers with proper labeling for further use.

Scientific classification
Kingdom: Plantae
Phylum:Tracheophyta
Class: Equisetopsida
Order: Fabales
Family: Fabaceae
Genus: *Senna*
Specie: *auriculata* (L.) Roxb

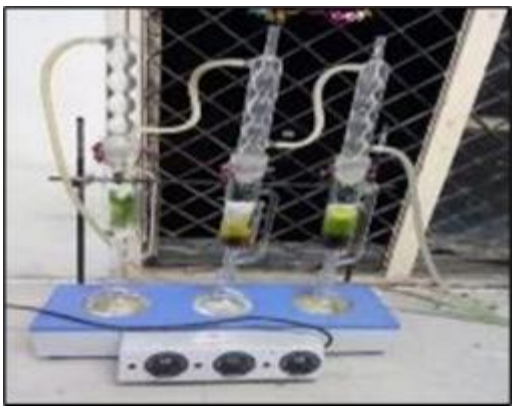
Figure 1 Specimen of *Senna auriculata* with leaves and flowers

2.2. Analyses of primary phytochemicals (the proximate compositions) of *S. auriculata* leaves and flowers

The powdered samples of *S. auriculata* were subjected to proximate composition analysis (crude protein, crude fat, crude fiber, total ash, moisture, sand and silica, calcium, phosphorous, total salt, and gross energy [18-20].

2.3. Methanolic extractions of *S. auriculata* leaf and flower

The powdered samples of *S. auriculata* leaf and flower (75g each) were taken and packed in whatman No. 1 filter paper and put into soxhlet apparatus. The extracts were successively soaked with 300 ml (1:4 w/v) of a polar solvent, methanol (99.9% purity, Changshu Yangyuan Chemicals, China), individually and separately for 6-9 h each (30 to 36 cycle) until a clear colorless solution was obtained. The extracts were filtered by using double layered muslin cloth and concentrated at 40-50 °C using rotary vacuum evaporator (ROTAVAP). The concentrated extracts obtained were vacuum-dried under 40°C and used for further investigation. The vacuum-dried extracts obtained appeared as dark green, gummy solid.



Soxhlet extraction

Rotary evaporator

Ultra-Cryostat

Gummy solid of *S. auriculata* leaf and flower

Figure 2 Preparation of solvent extractions of *S. auriculata* leaf and flower

2.4. Qualitative analysis of preliminary secondary phytochemicals

The qualitative tests were used to identify the presence of specific constituents. The extracts were subjected to analysis to detect the presence of following biomolecules, such as Alkaloids, Flavonoids, Terpenoids, Polyphenols, Saponins, Tannins, Cardiac Glycosides, and Quinones using the standard qualitative procedures [21].

2.5. Analysis of secondary phytochemical metabolites

The methanolic extracts of *S. auriculata* leaf and flower were subjected to Gas Chromatography-Mass Spectrophotometric (GC-MS) analysis (Thermo GC-trace ultra ver-5.0; Thermo MS-DSQ-II; ZB 5-MS capillary standard non-polar column (30 mts, 0.25 mm id, 0.25 µm film) for identification of different secondary phytochemical metabolic compounds, at South India Textile Research Association (SITRA), Coimbatore, Tamil Nadu, India.

The relative percentage constituent was expressed as a percentage with peak area normalization. Peaks resolved with the relative abundance of 0-100 were considered major compounds. To show the minor peaks, the chromatogram was magnified. Identification of various components present in these extracts was assigned by the comparison of their retention indices and mass spectra fragmentation patterns with those stored in the computer library and also with published literature. National Institute of Standards and Technology (NIST4) and WILEY9 [22] online library sources were used for matching the identified components.

2.6. Antibacterial efficiency of methanolic extract of *S. auriculata* leaf and flower

2.6.1. Antibacterial activity

Agar well diffusion assay was applied to determine the antibacterial activity of methanolic extract of *S. auriculata* leaf and flower. Bacterial culture was uniformly spread using an autoclaved cotton swab on a sterile Luria-Bertani agar (Himedia, India), and wells (7 mm diameter) were punched with a sterile cork-borer. The methanolic extract of *S. auriculata* (1mg/ml) was added into the wells and incubated at 35 ± 1 °C for 24 h. Inhibition zone was measured in mm with a ruler from the well to the end of the clear zone. Ampicillin (100µg/ml) was used as a positive control.

2.6.2. Growth curve analysis

The effect of methanolic extract of *S. auriculata* leaf and flower on the growth kinetics of *Pseudomonas aeruginosa*, *Escherichia coli*, *Bacillus subtilis*, and *Staphylococcus aureus* was monitored by optical density measurements [23]. The newly prepared *S. auriculata* leaf and flower extracts were diluted into 1mg/1ml in the 96 microtiter plates using nutrient broth. Each well was inoculated with 10 µL of overnight grown bacterial cultures, and then incubated at 37 °C for 20 h. The optical density was measured at 620 nm with an interval of 2 h in a microplate reader. Growth curves were plotted by taking the average of three replicates of each culture at each concentration of *S. auriculata*.

2.6.3. ROS production

The intracellular ROS production in bacterial cell was determined by staining the cell with DCFH-DA (Dichloro-dihydro-fluorescein diacetate), which oxidized into fluorescent dichlorofluorescein (DCF) [24]. The cells were inoculated and growth in 96-wells plate for 24h. After treatment with a concentration (1mg/ml leaf/ flower extract of *S. auriculata*, the cells were given 100µL of DCFH-DA for 10 min under dark condition. Then the cells were gently washed with phosphate buffer solution (PBS) and the intensity of green fluorescence was estimated based on the excitation (485 nm) and emission (520nm) wavelengths using spectro-spectrofluorometer. The generated ROS was observed under fluorescence microscopy equipped with excitation sources, appropriate filters for fluorescein isothiocyanate (FITC) and a digital camera.

2.6.4. Live/dead bacterial (BacLight) assay

The bacterial strains (106 CFU/mL) were treated with the concentration (1mg/ml) of methanolic extract of *S. auriculata* and incubated for 60 min. The cells were centrifuged for 6000 ×g at 5 min at 4 °C and the pellet was rinsed with PBS. The cells were stained with SYTO 9 and PI for 15 min in the dark and images were acquired by fluorescence microscope. SYTO 9, a green fluorescent dye, enters all cells, while PI, a red fluorescent dye, only enters cells with damaged membranes. Baclight L-7012 kit (Invitrogen, Carlsbad, CA, USA) was used.

3. Results

3.1. Preliminary phytochemicals of *S. auriculata* leaf and flower

Table 1 Proximate composition of primary phytochemicals and mineral contents of *S. auriculata* leaf and flower

Parameter (% on dry wt. basis)	Leaf	Flower
Moisture	9.99	7.70
Crude Protein	15.60	16.56
Crude Fibre	8.63	8.94
Ether Extract (crude fat)	2.66	2.45
Total Ash	4.84	4.95
Total nitrogen free content	58.28	59.40
Acid Insoluble Ash (Sand and Silica)	0.13	0.16
Calcium	0.60	1.79
Phosphorus	0.20	0.37
Salt	0.33	0.47
Gross Energy (kcal/kg)	3809	3901

The present study revealed that the leaves and flowers of *S. auriculata* contain a significant amount of primary phytochemical constituents (Table 1). The flower contains more quantity of crude protein 16.56% than that of the leaf 15.60%. The content of calcium was also found to be higher in the flower (1.79%) than that of the leaf (0.60). The energy level was higher in flowers (3901 kcal/kg) than that of the leaf (3809 kcal/kg). The total nitrogen free content was also higher in the flower (59.40%) than in the leaf (58.28%) of *S. auriculata*. In contrast the moisture content was higher in the leaf over the flower.

3.2. Quality of secondary phytochemicals of *S. auriculata* leaf and flower

Table 2 Qualitative detection of secondary phytochemicals present in the methanolic extract of *S. auriculata* leaf and flower

Secondary Phytochemicals	Leaf	Flower
Alkaloids	+++	++
Flavonoids	++	+++
Terpenoids	+++	-
Polyphenols	-	+
Saponins	++	++
Tannins	+	-
Cardiac Glycosides	-	+
Quinones	++	+

+, Poor presence; ++, Moderate presence; +++, Luxurious presence; -, Absence

The methanolic extract of leaf powder of *S. auriculata* showed occurrence of six primary phytochemical compounds, such as alkaloids, flavonoids, terpenoids, saponins, tannins and quinones. Among these, alkaloids and terpenoids were luxuriantly present. The flavonoids, saponins and quinones were found to be moderately present. Tannins were poorly present (Table 2). The methanolic extract of flower powder of *S. auriculata* also showed occurrence of six primary compounds, such as alkaloids, flavonoids, polyphenols, saponins, cardiac glycosides and quinones. Of which, flavonoids alone were found to be present luxuriantly. The alkaloids and saponins were found to be moderately present. The polyphenols, cardiac glycosides and quinones were poorly present (Table 2).

3.3. Secondary phytochemical metabolic compounds of *S. auriculata* leaf and flower

The GC-MS analysis of the methanolic extract of *S. auriculata* leaf showed presence of twenty-seven secondary phytochemical metabolic compounds (Fig. 3 and Table 3). Similarly, in the flower extract, there were twenty-five secondary phytochemical metabolic compounds (Fig. 4 and Table 4). All secondary phytochemical metabolic compounds present in the leaf and flower of *S. auriculata* possessed bioactive properties (Tables 3 and 4).

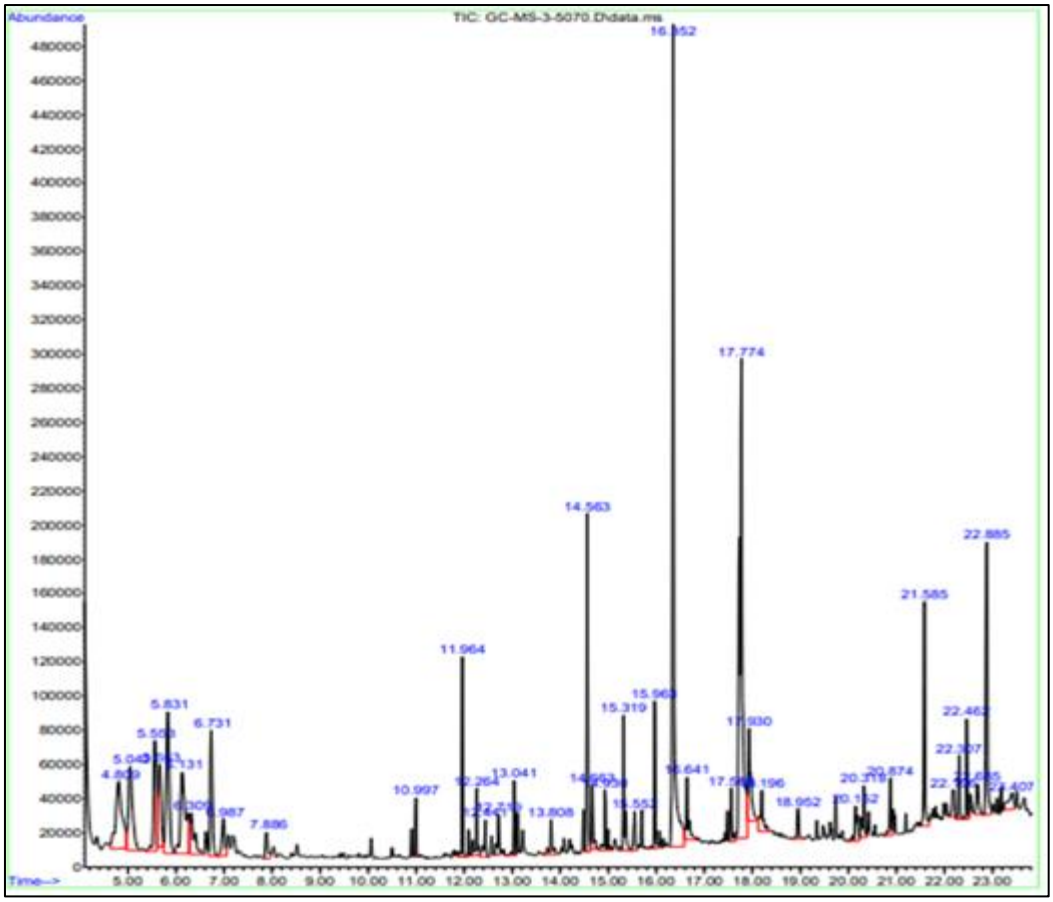


Figure 3 GC-MS chromatogram of the methnolic extract of *S. auriculata* leaf

Table 3 Secondary phytochemical metabolic compounds detected in the methanolic extract of *S. auriculata* leaf

Sl. No.	RT	Compounds	Molecular Formula	Biological Properties (by literature only)
1.	4.809	Cyclohexene, 1-methyl-	C_7H_{12}	Anti-inflammatory and antimicrobial activity
2.	5.042	Benzaldehyde	C_7H_6O	Antioxidant and antimicrobial activity
3.	6.131	Benzyl Alcohol	C_7H_8O	Antimicrobial and antifungal activity
4.	6.987	Cyclohexane, 1,4-dimethyl-	C_8H_{16}	Antimicrobial activity
5.	10.997	Tetradecane	$C_{14}H_{30}$	Antimicrobial and antifungal activity
6.	11.964	Benzene, 1-(1,5-dimethyl-4-hexenyl)-4-methyl-	$C_{15}H_{22}$	Antioxidant, anti-inflammatory, and antimicrobial activities
7.	12.264	Di-epi-. alpha. -cedrene-(I)	$C_{15}H_{24}$	Antimicrobial activity
8.	12.441	Dodecane	$C_{12}H_{26}$	Antibacterial and antifungal activity
9.	13.041	2-Tetradecene, (E)-	$C_{14}H_{28}$	Antimicrobial activity
10.	13.808	Ar-tumerone	$C_{15}H_{20}O$	Anti-inflammatory, antioxidant, and anti-angiogenic effects
11.	14.663	2-Propenoic acid, 3-(4-methoxyphenyl)-, ethyl ester	$C_{12}H_{14}O_3$.	Antioxidant and anti-inflammatory
12.	14.930	Bromoacetic acid, hexadecyl ester	$C_{18}H_{35}BrO_2$	Antimicrobial and Antibacterial activity
13.	15.552	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester	$C_{16}H_{22}O_4$	Antioxidant and anticancer Activities

14.	16.352	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	Antioxidant and anti-inflammatory
15.	16.641	3-Eicosene, (E)-	$C_{20}H_{40}$	Antimicrobial activity
16.	17.518	Eicosane	$C_{20}H_{42}$	Antifungal, neuroprotective, and anti-inflammatory effects
17.	21.429	Phytol	$C_{20}H_{40}O$	Antimicrobial, cytotoxic, antioxidant, autophagy-and apoptosis-inducing, anti-inflammatory activity
18.	17.774	9,12,15-Octadecatrienoic acid, (Z, Z, Z)-	$C_{18}H_{30}O_2$	Antioxidant, Anti-inflammatory, anti-asthma, antimicrobial activity
19.	17.930	Octadecanoic acid	$C_{18}H_{36}O_2$	Antimicrobial, anti-inflammatory, and antioxidant activities
20.	18.952	Docosane, 11-decyl-	$C_{32}H_{66}$	Antioxidant and antimicrobial activities
21.	20.318	Heptadecane	$C_{17}H_{36}$	Anti-inflammatory and antimicrobial activity.
22.	20.874	Dodecanoic acid, phenylmethylester	$C_{19}H_{30}O_2$	Antioxidant, and antimicrobial activity.
23.	21.585	3-Eicosene, (E)-	$C_{20}H_{40}$	Antimicrobial, antiviral and anti-inflammatory
24.	22.196	Nonadecane	$C_{19}H_{40}$	Antioxidant, Antimicrobial and Anti-diabetic activity
25.	22.307	Squalene	$C_{30}H_{50}$	Antioxidant and antitumor activity
26.	22.885	1-Hexacosanol	$C_{26}H_{54}O$	Anti-inflammatory and antimicrobial activity.
27.	23.407	1,2-Bis(trimethylsilyl)benzene	$C_{12}H_{22}Si_2$	Antioxidant, antimicrobial activity

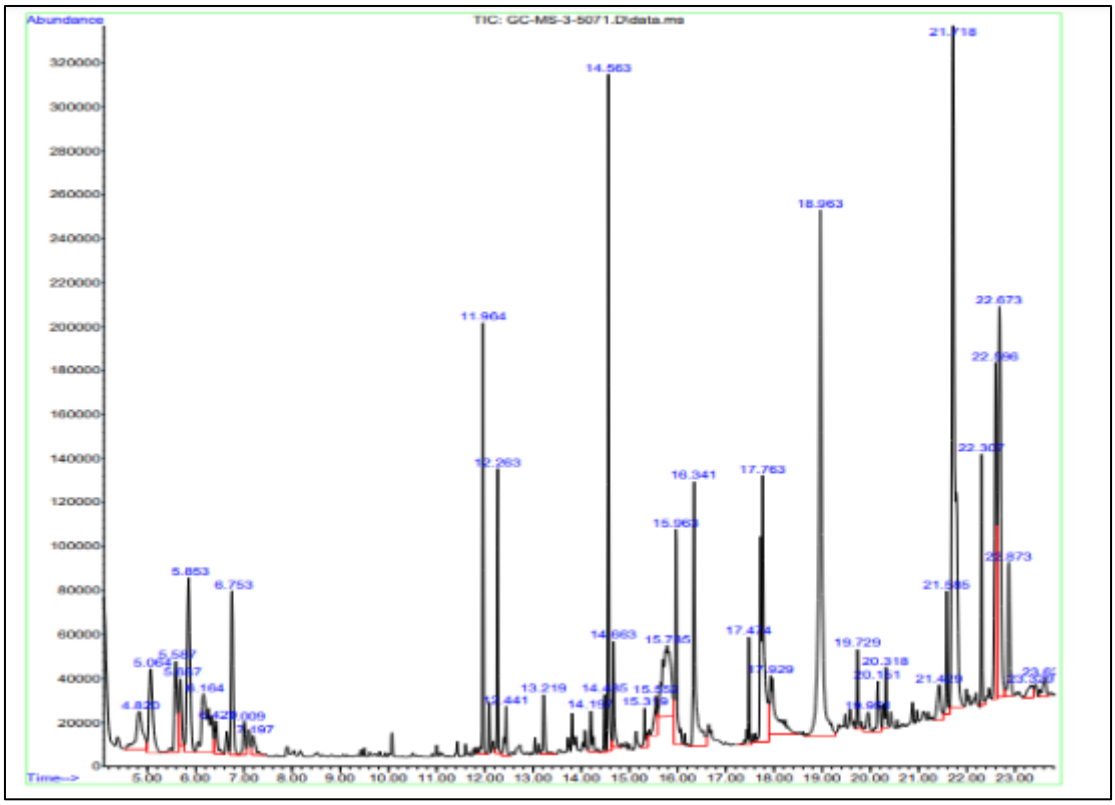


Figure 4 GC-MS chromatogram of the methnolic extract of *S. auriculata* flower

Table 4 Secondary phytochemical metabolic compounds detected in the methanolic extract of *S. auriculata* flower

Sl. No.	RT	Bioactive compounds	Molecular Formula	Biological properties (by literature only)
1.	4.820	Oxirane, 2-methyl-3-propyl-, trans-	$C_6H_{12}O$	Antioxidant activity
2.	6.164	Benzyl Alcohol	C_7H_8O	Antimicrobial, antifungal, and local anesthetic effects
3.	7.009	Cyclohexane, 1-ethyl-1-methyl-	C_9H_{18}	Antimicrobial and Antioxidant activity
4.	14.485	Phenol, 5-(1,5-dimethyl-4-hexeny...	$C_{15}H_{22}O$	Antimicrobial, Antitumorand Antiinflamatory activity
5.	15.319	Bicyclo[3.1.1] heptane, 2,6,6-tri...	$C_{10}H_{18}$	Anti-inflammatory, Antioxidant activity
6.	15.552	1,2-Benzenedicarboxylic acid, bi...	$C_8H_6O_4$	Anticancer and antioxidant activity
7.	15.785	Oxirane, hexadecyl-	$C_{18}H_{36}O$	Anti-inflammatory, and antineoplastic
8.	15.963	7,9-Di-tert-butyl-1-oxaspiro (4,5...	$C_{17}H_{24}O_3$.	Antioxidant, antimicrobial and antiinflamatoryactivity
9.	16.341	n-Hexadecanoic acid	$C_{16}H_{32}O_2$	Antioxidant, antimicrobial and Antiinflamatoryactivity
10.	17.474	Heneicosane	$C_{21}H_{44}$	Antioxidant activity
11.	17.763	9,12-Octadecadienoic acid (Z, Z)-	$C_{18}H_{32}O_2$	Antioxidant, Antimicrobial and Anticancer activity
12.	17.929	9,17-Octadecadienal, (Z)-	$C_{18}H_{32}O$	Antimicrobial, antioxidant and anti-inflammatory activity
13.	19.729	Triphenyl phosphate	$C_{18}H_{15}O_4P$	Antioxidant and genotoxicity activity
14.	19.951	1,19-Eicosadiene	$C_{20}H_{38}$	Antimicrobial activity
15.	20.151	9,12-Octadecadienoic acid (Z, Z)-	$C_{18}H_{32}O_2$	Antioxidant and anticancer activity
16.	20.318	Eicosane	$C_{20}H_{42}$	Antioxidant, antifungal and anticancer activity
17.	21.429	Campesterol	$C_{28}H_{48}O$	Antioxidant, antimicrobial and Anti inflammatoryactivity
18.	21.585	1-Heptadecene	$C_{17}H_{34}$	Antibiotic activity
19.	21.718	1,19-Eicosadiene	$C_{20}H_{38}$	Antimicrobial activity
20.	22.307	2,6,10,14,18,22-Tetracosahexaene...	$C_{24}H_{38}$	Antioxidant and Anticancer activity
21.	22.596	1-Heptacosanol	$C_{27}H_{56}O$	Antimicrobial activity
22.	22.673	gamma. -Sitosterol	$C_{29}H_{50}O$	Anticancer, Anti-inflammatory and Antioxidant activity
23.	22.873	Heptadecane, 2,6,10,15-tetramethyl-	$C_{21}H_{44}$	Antimicrobial and Antioxidant activity
24.	23.329	1,2-Bis(trimethylsilyl)benzene	$C_{12}H_{22}Si_2$	Antimicrobial, Antioxidant, Anti-diabetic and Anti-inflammatory activity
25.	23.629	Silane, 1,4-phenylenebis [trimethyl-	$C_{12}H_{22}Si_2$.	Antioxidant, Anti-diabetic, Anti-inflammatory and Antibacterial activity

3.4. Antibacterial activity of the methanolic extract of *S. auriculata* leaf and flower

3.4.1. Assessment of zone of inhibition of leaf and flower extracts against bacteria

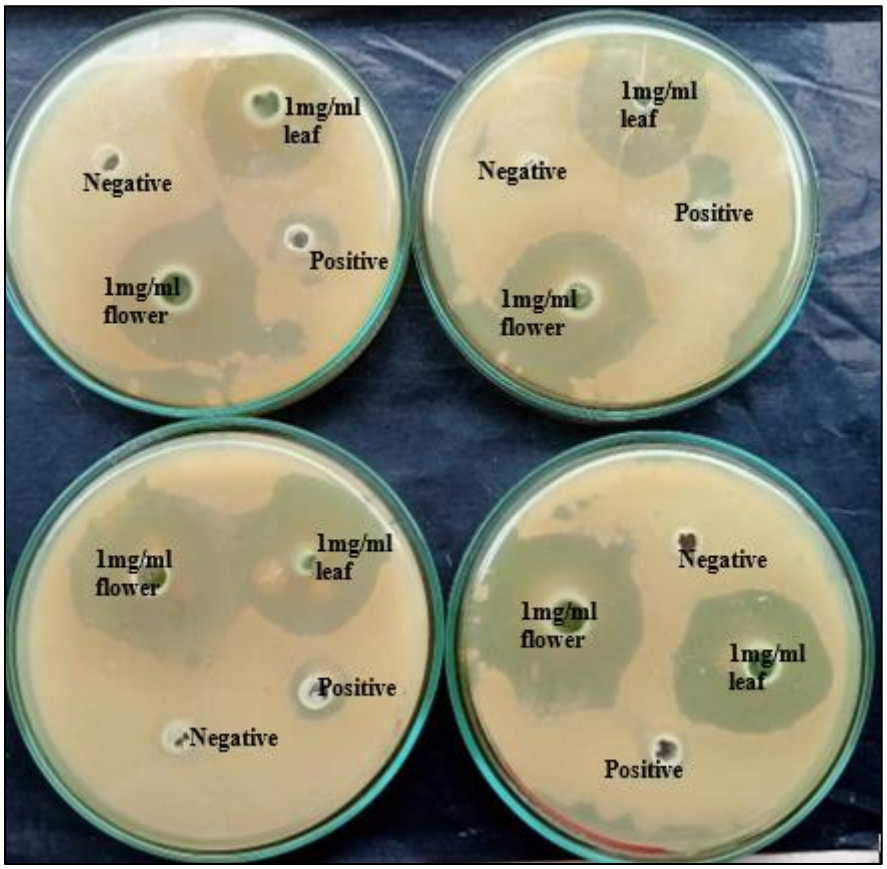


Figure 5 Zone of inhibition against *P. aeruginosa, E. coli*, *B. subtilis*, and *S. aureus* (left to right in top and down) by diffusion of *S. auriculata* leaf and flower extracts (1mg/ml) on agar well plates

Figure 5 revealed that the methanolic extract of *S. auriculata* leaf and flower (1mg/ml) showed greater zone of inhibition than that of the positive control, Ampicillin (10µg/ml), against all the bacterial species tested (*P. aeruginosa*, *E. coli*, *B. subtilis*, and *S. aureus*). The methonolic extract of *S. auriculata* flower extract exhibits greater inhibitory zone than that of the leaf extract, particularly against *E. coli* followed by *B. subtilis, P. aeruginosa* and *S. aureus*.

3.4.2. Growth Kinetics of bacteria against methanolic extract of S. auriculata leaf and flower

Figure 6 revealed bacterial growth regulation by the methanolic extracts of *S. auriculata* leaf and flower, in which the flower extract showed better control of growth of all the four bacteria, such as *P. aeruginosa*, *E. coli*, *B. subtilis*, and *S. aureus* than that of the leaf extract. The control which was not subjected to leaf and flower extracts showed proportionally higher growth rate of all the four bacterial species, duration dependent.

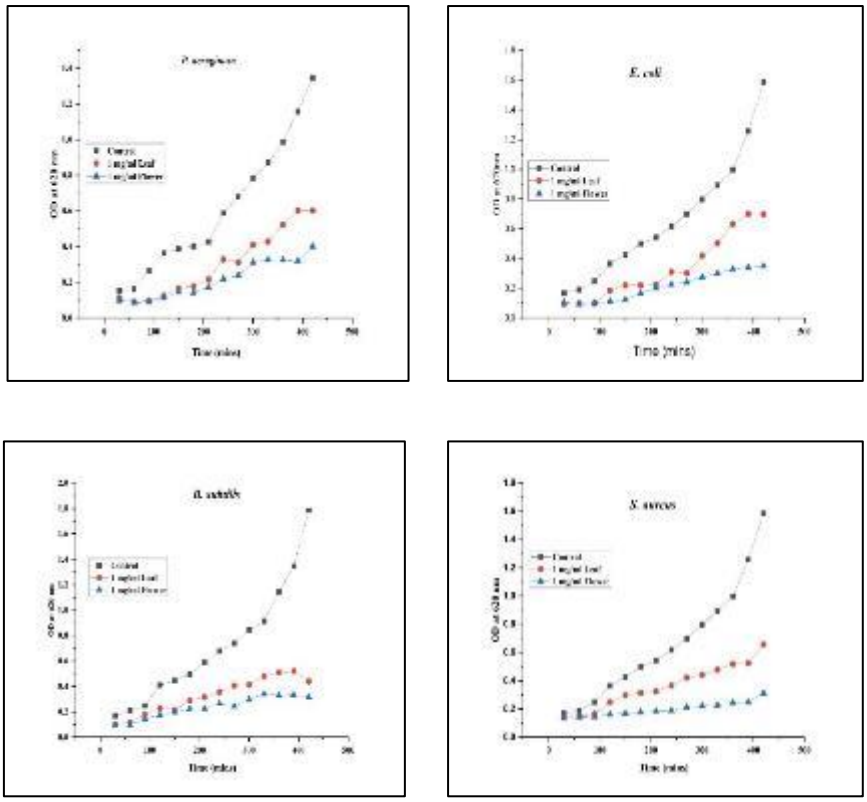
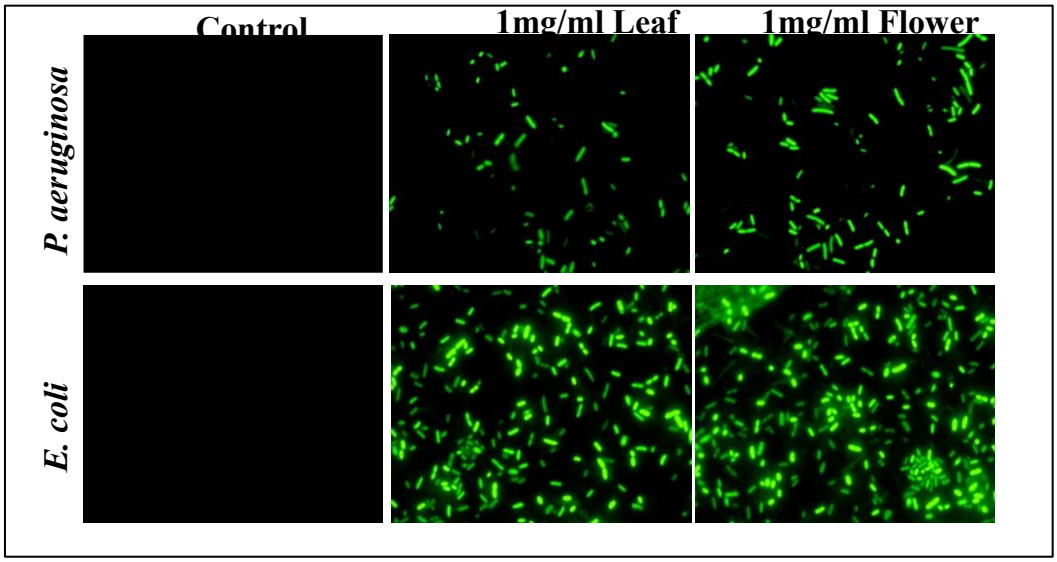


Figure 6 Growth kinetics of *P. aeruginosa*, *E. coli*, *B. subtilis*, and *S. aureus* against methanolic extracts of *S. auriculata* leaf and flower

3.4.3. ROS production in different bacteria treated with methanolic extracts of S. auriculata leaf and flower



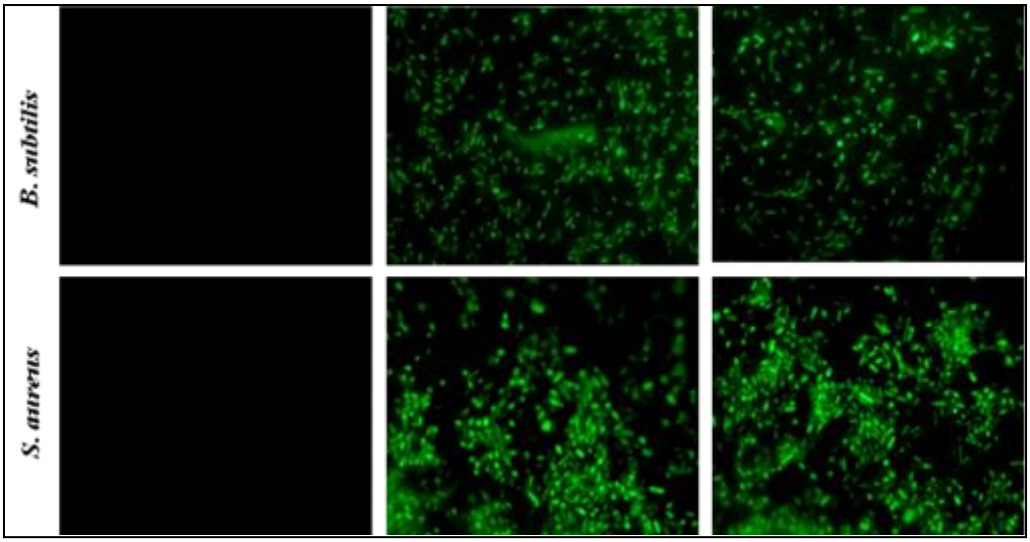
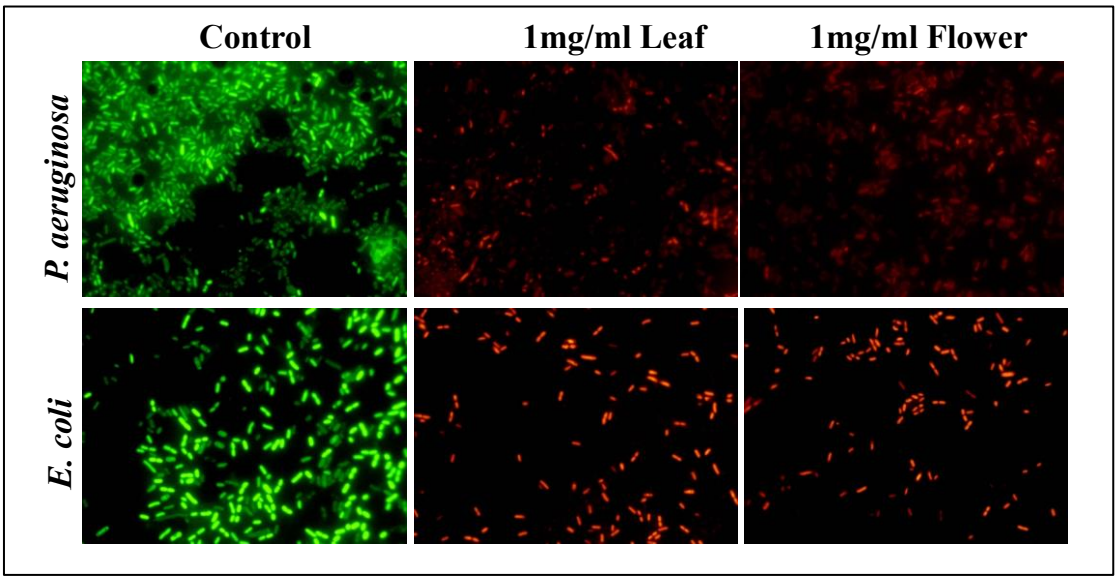


Figure 7 The intracellular ROS production in different bacteria staining with DCFH-DA after treatment with a concentration (1mg/ml) of methanolic extract of *S. auriculata* leaf and flower. The green fluorescence colour indicates generated ROS

Figure 7 depicts ROS production in different bacterial cells treated with methanolic extracts of *S. auriculata* leaf and flower. The ROS production was detected in all four bacterial species treated with leaf and flower extracts of *S. aeuriculata*, which was found to be higher in *E. coli* and *S. aureus* as well. This was followed by *B. subtilis* and *P. aeruginosa*. Among the two extracts, the flower showed more efficient ROS production than that of the leaf. No ROS production was detected in untreated controls.

3.4.4. Live/dead bacterial (BacLight) assay against the methanolic extract of S. auriculata leaf and flower

Figure 8 depicts the methanolic extract of *S. auriculata* leaf and flower resulting in destabilized membrane integrity and production of cell death, which was detected by penetration of PI into the dead or damaged cells with compromised membranes and fluoresces red/brown. The intensity of red/brown colour seems to be greater in flower extract treated bacteria than that of the leaf of *S. auriculata*. SYTO 9 produced fluoresce green in live bacteria, the untreated control.



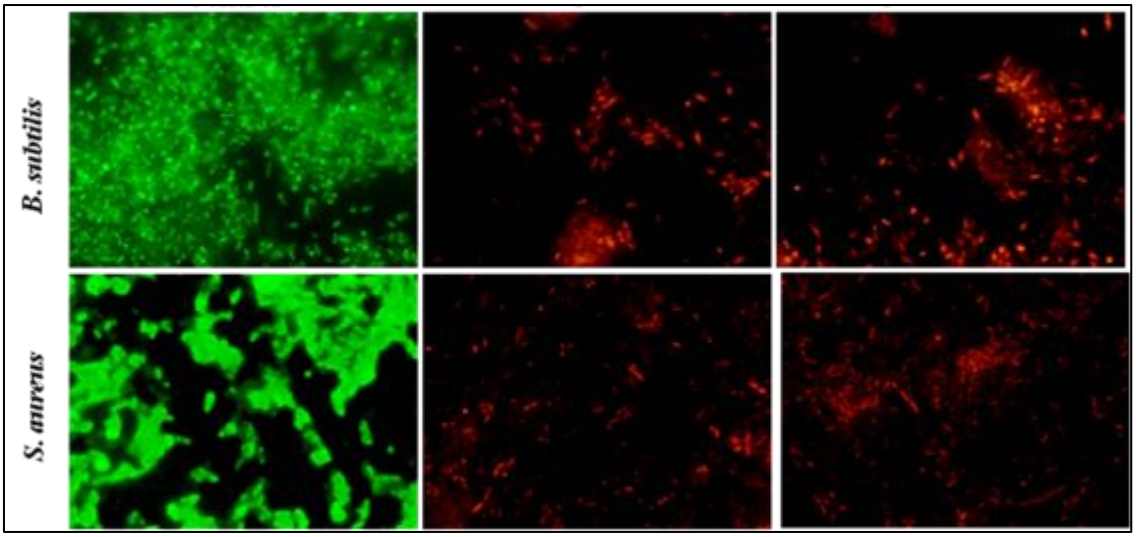


Figure 8 The live and dead bacteria are stained with SYTO 9 and PI after being treated with (tests) and without (control) a concentration (1mg/ml) of methanolic extract of *S. auriculata* leaf and flower. The green fluorescence color indicates live bacteria of untreated control, and the red/ brown colour indicates dead bacteria due to the treatment of leaf and flower extracts

4. Discussion

S. auriculata is one of the plants with great medical properties. It's leaves, flower, root, bark, and seed exhibit various potential biological activities due to presence of proteins, polysaccharides, alkaloids (pyrrolizidine alkaloids, suspected of hepatotoxic properties), flavonoids, saponins, phenols, and tannins [25-27]. In the present study, the leaf and flower of *S. auriculata* possessed such property. It has antioxidant and anti-inflammatory properties due to presence of phenols (rhein, chrysaphanol, kaempferol, aloeemodin, and glycosides), tannins, and terpenoids (sitosterol, stigmasterol, and campesterol) [25]. It has hormonal action also due to the presence of steroids. It possesses laxative effects due to presence of anthraquinones glycosides (sennosides A, B, C, and D), anthraquinones (alatinone and alatonal), cardiac glycosides (sennapicrin), steroids, and terpenoids (sitosterol, stigmasterol, and campesterol). It has fatty acids (oleic, palmitic, and linoleic acids, fatty acid esters and fatty acid amide), amino acids, and other metabolites, such as 3-O-methyl-D-glucose, α-tocopherol-β-D-mannoside, resorcinol, n-hexadecanoic acid, 13-octadecenal, (Z)-, 1,2,3,4-tetrahydroisoquinolin-6-ol-1-carboxylic acid, benzoic acid 2-hydroxy methyl ester, 1-methyl butyl ester, epicatechin, ferulic acid, quercetin-3-O-rutinoside, quercetin, proanthocyanidin B1. Other compounds present in *Cassia*/*Senna* are stigmasterol, lupeol acetate, chrysophanol, emodin, coumarin, proteins, cytoprotective agents, antioxidants, cardio protectants etc., [28, 29]. Supare and Patil [30] have reported the presence of glycosides, flovonoids, saponin, alkaloid, tannin, protein, carbohydrates, gum and phenolic compounds in *Cassia tora*. Various compounds including phenolics, terpenoids (triterpene, and diterpene alcohols), alkaloids and phytol present in plants possessed biocidal activity [31-34]. The presence of 3-O-Methyl-d-glucose (48.50%), α- Tocopherol-β-D-mannoside (14.22%), Resorcinol (11.80%), n-Hexadecanoic acid (3.21%) and 13-Octadecenal, (Z)-(2.18%) was elucidated in the methanolic extract of *C. auriculata* leaf [35].

The following specific plants possessed certain specific bioactive compounds, which worked against specific diseases. The *in vitro* cytotoxic potential of melosuadimins (monoterpenoidbisindole alkaloids) from *Melodinus suaveolens* against three colorectal cancer cell lines exhibited marked anti-proliferative activity, inducing apoptosis and G2/M cell cycle arrest [36]. Nudifloids O and P (diterpenoids) from *Callicarpa nudiflora* showed inhibitory activities on NO and IL-1β production in the LPS-stimulated RAW 264.7 macrophage cells. Nudifloids P showed potent inhibitory activities against IL-1β and NO production [37]. Phenolics present in leafy vegetables having anti-diabetes properties by inhibiting α-glucosidase, α-amylase, lipase, acetylcholinesterase and β-glucuronidase [48]. Pentacyclic Triterpene Acid present in *Cecropia angustifolia* Roots possessed anti-diabetes property [49]. Ethanolic extract of *P. fistulosus* (Tinda) leaves contains phenols, flavonoids, terpinoids, alkaloids, tannins, steroids, and carbohydrates. These compounds have the potential of reducing oxidative stress and have a variety of medical uses, including psychopharmacological activity [40]. Therefore, now-a-days, many modern medications are made from *Senna/Cassia* are used to treat a variety of illnesses and chronic conditions. *S. auriculata* root is used in decoctions against fevers, diabetes, diseases of the urinary system and constipation. The leaves have laxative properties. The dried flowers and flower buds are used as a substitute for tea in case of diabetes patients. It is also useful in the treatment of various ailments and chronic diseases such as

asthma, conjunctivitis, ulcerative colitis, cancer (colorectal carcinoma, renal calculi, larynx cancer, and breast cancer, causing apoptosis in MCF-7 and Hep-2 cells due to triterpene glycosides present in flavonoids and procyanidins, 3-O-beta-D-xylopyranoside), kidney, liver disease, hyper melanosis, inflammation, ulcers, fever, psoriasis, viral infection, helminth infection, hepatitis, rheumatism, dysentery, skin disease, metabolic disorders and infertility [26, 27, 29, 41, 42].

According to Deshpande *et al*., [43], *C. auriculata* possessed significant antioxidant activity due to presence of rich source of phytochemicals such as natural phenol, flavonoids, saponins compounds and vitamins (A, C, and E). Therefore it could be useful for preparation of nutraceuticals to treat various diseases and its complications in animals as well as humans including anti-hyperglycemic and anti-hyperlipidemic [44]. Since *C. auriculata* has potential hypoglycemic properties, it can be used for treating diabetes (insulin-dependent and non-insulin-dependent diabetes, diabetic retinopathy, diabetic peripheral neuropathy etc.) in humans [45, 46]. *S. auriculata* also has antibacterial (*Vibrio cholera*, *Staphylococcus aureus* and *E. coli*), anti-fungal (*Candida albicans*, *Candida tropicalis* and *Aspergillus niger*), anti-inflammatory, and wound healing properties [25, 26], which exhibited positive effects on neuro, renal, and liver support [44]. In the present study, the leaf and flower of *S. auriculata* showed good antibacterial activity, and greatly reduced the growth of bacteria.

The presence of many bioactive phytocomponents in *S. auriculata* might have a very important medicinal value, which has to be proved further through isolation and purification of specific bioactive components. Since *S. auriculata* possessed phytopharmacological properties, it is an ingredient in several formulations of traditional/home remedies including Ayurveda and Siddha [36]. The promising natural cures have advancements therapeutically/immunologically. Therefore *S. auriculata* may be an invaluable asset in complementary and alternative medicine for a range of medical ailments in the forms of ecofriendly artistry and antimicrobial soap to nourish the skin to overcome skin acne, eczema and psoriasis as it has anti-peroxidative, anti-tanning, and anti-ageing activities, and it also provides pleasant natural scents and promoting relaxation through aromatherapy [26, 29, 47].

It has been reported that another species of *Senna* is used in the treatment of typhoid, diabetes, malaria, asthma, ringworms, tinea infections, scabies, blotch, herpes, and eczema. It also displayed anti-bacterial, anti-oxidant, anti-fungal, dermatophytic, anti-cancer, hepatoprotective, anti-lipogenic, anti-convulsant, anti-diabetic, anti-hyperlipidemic, anti-malarial, anthelmintic, and anti-viral activities could be due to the array of secondary phytochemicals and their metabolites, such as tannins, alkaloids, flavonoids, terpenes, anthraquinone, saponins, phenolics, cannabinoid alkaloids, reducing sugars, steroids, volatile oils, 1,8-cineole, caryophyllene, limonene, α-selinene, β-caryophyllene, germacrene D, cinnamic acid, pyrazol-5-ol, methaqualone, isoquinoline, quinones, Ethane,1,1-Diehoxy, Diethyl Phthalate, 3-O-Methyl- D-Glucose, n- Hexadecanoic acid, 1-butanol,3-Methyl, Resorcinol etc., present in different parts of the *Senna* plants [48, 49]. *Senna* plant is also a key ingredient in detergents (foaming and surface-active agents), molluscicides and pesticides [48]. *S. auriculata* seed has been reported to use for biodiesel production [50]. The secondary metabolites of *Senna* contributed to the pronounced wound healing and antibacterial, antifungal and anticancer activities [51-56]. It has been reported that *C. auriculata* derived silver nanoparticles served as a novel male contraceptive agent [57].Therefore, the genus, *Senna* has a wide range of natural pharmaceutical properties. Its application needs studies for specific ailments.

5. Conclusion

In this study, the leaf and flower of *S. auriculata* showed very good primary and secondary phytochemical properties. It exhibited antibacterial activity, ROS production and apoptotic potential. Therefore, it can be taken to the pharmaceutical industry for the development of natural products to treat different ailments. For which in vitro studies on cell lines and in vivo studies on animal models are warranted.

Compliance with ethical standards

Acknowledgments

The authors gratefully acknowledge Botanical Survey of India (BSI), Southern Regional Centre, Coimbatore, India, for authentication of the Medicinal Plant, *Senna auriculata* (BSI/SRC/5/23/2024-25/Tech/191). South India Textile Research Association (SITRA), Coimbatore, Tamil Nadu, India, is acknowledged for providing GC-MS outsourcing service. The authors sincerely acknowledged Dr. G. Thirupathi, Nematology Lab, Dept. of Zoology, Bharathi University, Coimbatore, India, who is now doing PDF in South Korea, for rendering experimental support.

Disclosure of conflict of interest

The authors declare no conflict of interests.

Data available statement

The data used in this study are provided and included within the article.

Authors' contribution

Designing and formatting: PSB; Manuscript preparation: V.P; Analysis of data: TM; Content evaluation: T.J.

Disclaimer (Artificial intelligence)

Author(s) hereby declares that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during preparation of this manuscript.

References

[1] Nethan, S. Sinha, D. and Mehrotra, R. (2017). Non-communicable disease risk factors and their trends in India. Asian Pac. J. Cancer Prev. 18; 2005–2010.

[2] Devassy, S.M. Benny, A.M. Scaria, L. Nannatt, A. Newlin, M.F. and Joubert, J. (2020). Social factors associated with chronic non-communicable disease and co-morbidity with mental health problems in India: A Scoping Review. BMJ Open 10, e035590. doi:10.1136/bmjopen-2019-035590.

[3] Gupta, S. Sharma, S.B. Singh, U.R. and Bansal, S.K. (2011). Salutary effect of *Cassia auriculata* L. leaves on hyperglycemia-induced atherosclerotic environment in Streptozotocin Rats. Cardiovasc. Toxicol. 11; 308–315.

[4] Nille, G.C. Reddy, K.R.C. and Tripathi, J.S. (2018). Clinical evaluation of talapotakachurna– A poly herbal ayurvedic formulation in type-2 diabetes mellitus. Indian J. Tradit. Knowl. 17 (1); 168–175.

[5] Win, E.T. and Min, A.K. (2018). Investigation of Phytochemical constituents, physicochemical properties and antimicrobial activities from the leaves of *Senna auriculata* (L.) Roxb. J. Myanmar Acad. Arts Sci. 16 (4); 295–314.

[6] Badaturuge, M.J. Habtemariam, S. and Thomas, M.J.K. (2011). Antioxidant compounds from a South Asian beverage and medicinal plant, *Cassia auriculata*. Food Chem.125 (1); 221–225.

[7] Begum, M.A.J. Vijayakumar, A. and Selvaraju, P. (2013). Standardization of seed dormancy breaking treatment in *Senna* (*Cassia auriculata*). J. Plant Breed. Crop Sci. 5; 220–223.

[8] Vijayakumar, R. and Nachiappan, V. (2017). *Cassia auriculata* flower extract attenuates hyperlipidemia in male Wistar rats by regulating the hepatic cholesterol metabolism. Biomed. Pharmacother. 95; 394–401.

[9] Deshpande, S. Kewatkar, S.M. and Paithankar, V.V. (2013). In-vitro antioxidant activity of different fraction of roots of *Cassia auriculata* Linn. Drug Invent. Today 5 (2); 164–168.

[10] Khader, S.Z.A. Ahmed, S.S.Z. Balasubramanian, S.K. Arunachalam, T.K. Kannappan, G. Mahboob, M.R. Ponnusamy, P. and Ramesh, K. (2017). Modulatory effect of dianthrone rich alcoholic flower extract of *Cassia auriculata* L. on experimental diabetes. Integr. Med. Res. 6; 131–140.

[11] Thabrew, M.I. Munasinghe, T.M.J. Senarath, S. and Yapa, R.M.S.C. (2004). Effects of *Cassia auriculata* and *Cardospermum halicacabum* teas on the steady state blood levels of theophylline in rats. Drug Metabol. Drug Interact. 20; 263-272.

[12] Esakkirajan, M. Prabhu, N.M. Manikandan, R. Beulaja, M. Prabhu, D. Govindaraju, K. Thiagarajan, R. Arulvasu, C. Dhanasekaran, G. Dinesh, D. and Babu, G. (2014). Apoptosis mediated anti-proliferative effect of compound isolated from *Cassia auriculata* leaves against human colon cancer cell line. Spectrochim. Acta A. 127; 484–489.

[13] Devi, P.S. Santhi, V. Kannagi, A. and Shobana, J.J. (2014). Antibacterial activity, preliminary phytochemical screening and HNMR analysis of *Senna auriculata* and *Abuliton indicum*. Int. J. Curr. Microbiol. Appl. Sci. 3; 1069.

[14] Dhiman, A. Nanda, A. and Ahmad, S. (2016). A quest for staunch effects of flavonoids: utopian protection against hepatic ailments. Arab. J. Chem. 9; S1813–S1823.

[15] Wang, T.Y. Li, Q. and Bi, K.S. (2018). Bioactive flavonoids in medicinal plants: structure, activity and biological fate. Asian J. Pharm. Sci. 13; 12–23.

[16] Nambirajan, G. Karunanidhi, K. Ganesan, A. Rajendran, R. Kandasamy, R. Elangovan, A. and Thilagar, S. (2018). Evaluation of antidiabetic activity of bud and flower of *Avaram senna* (*Cassia auriculata* L.) in high fat diet and streptozotocin induced diabetic rats. Biomed. Pharmacother. 108; 1495–1506.

[17] Khyade, M. Kamble, S. Waman, M. Padwal, A. and Gunjal, M. (2020). Food potential and antioxidant property of *Cassia auriculata* seed: a nutritionally unexploited legume. Curr. Nutr. Food Sci. 16; 1381–1392.

[18] Castell, J.D. Tiews, K. (1980). Report on the EIFAC , IUNS and ICES working group on standardization of methodology in fish nutrition research , Hamburg , Federal Republic of Germany. Documents techniques de la CECPI (FAO). 36; 24-28.

[19] Valipour, A. Nedaei, S. Noori, A. Khanipour, A.A. and Hoseinifar, S.H. (2019). Dietary *Lactobacillus plantarum* affected on some immune parameters, air-exposure stress response, intestinal microbiota, digestive enzyme activity and performance of narrow clawed crayfish (*Astacus leptodactylus*, Eschscholtz). Aquaculture. 504; 121-130.

[20] AOAC. (1995). Official methods of analysis of association of analytical communities international, 2 Vol. 16th Edn. Arlington. VA, USA.

[21] Trease, G.E. Evans, W.C. (1989). Pharmacology 11th Edn. Bailliere Tindall Ltd. London. 60-75.

[22] Vandendool, H. Kratz, D.J. (1963). A generation of the retention index system including liner temperature programmed gas-liquid partition chromatography. J. Chromatogar. 11; 463-467.

[23] Wawrzyniak, J. and Drożdżyńska, A. (2024). Impact of dried garlic on the kinetics of bacterial growth in connection with thiosulfinate and total phenolic content. Applied Sciences. 14(15); 6756. DOI: 10.3390/app14156756.

[24] Goggin, F.L. and Fischer, H.D. (2022). Reactive oxygen species in plant interactions with aphids. Frontiers in Plant Science. 12; 811105.

[25] Prasathkumar, M. Raja, K. Vasanth, K. Khusro, A. Sadhasivam, S. Sahibzada, M.U.K. Gawwad, M.R.A. Dunia A. Farraj, A. and Elshikh, M.S. (2021). Phytochemical screening and in vitro antibacterial, antioxidant, anti-inflammatory, anti-diabetic, and wound healing attributes of *Senna auriculata* (L.) Roxb. Leaves. Arabian Journal of Chemistry. 14; 103345.

[26] Ghule, S.S. Sutar, S.B. and Patil, S.V. (2021). Review on phytochemical constituents and phytopharmacological ativities of *Senna auriculata* Linn. IJMPR, 5(4); 324-327.

[27] Kavita, R.N. Sunagad, R. and Bhandarkavathe, R. (2024). Exploring the therapeutic potential of *Senna auriculata* Linn: A comprehensive review. Int. J. of Pharm. Sci. 2 (12); 2811-2819.

[28] Padmavathi, V. (2018). Phytochemical constituents and their biological aspects of *Senna auriculata*. IAJPS: 05 (07); 6470-6475.

[29] Nille, C.G. Mishra, S.K. Chaudhary, K.C. and Reddy, K.R.C. (2021). Ethnopharmacological, phytochemical, pharmacological, and toxicological review on *Senna auriculata* (L.) Roxb.: A special insight to antidiabetic property. Front. Pharmacol. 12; 647887.doi: 10.3389/.2021.647887.

[30] Supare, S. and Patil, M. (2015). Estimation of phytochemical components from *Cassia tora* and to study its larvicidal activity. Intl. J. Pharmaceutical Sci. Invention. 4 (6); 11-16.

[31] Wink, M. Production and application of phytochemicals from an agricultural perspective. In Phytochemistry and Agriculture; vanBeek, T.A. and Breteler, H. (Eds).; Clarendon Press: Oxford, U.K., 1993; pp 171-213.

[32] Assabgui, R. Lorenzetti, F. Terradot, L. Regnault-Roger, C. Malo, N. Wiriyachitra, P. Sanchez Vindas, P.E. San Roman, L. Isman, M.B. Durst, T. and Arnason, J.T. (1997). Efficacy of botanicals from the Meliaceae and Piperaceae. In: Hedin, P.A. Hollingworth, R.M. Masler, E.P. Miyamoto, J. and Thompson, D.G. (Eds.), Phytochemicals for Pest Control. ACS Symp. Ser. American Chemical Society. Washington DC, USA. 658: pp. 38–48.

[33] Hostettmann, K. and Potterat, O. (1997). Strategy for the isolation and analysis of antifungal, molluscicidal, and larvicidal agents from tropical plants. In: Hedin P.A. Hollingworth, R.M. Masler, E.P. Miyamoto, J. and Thompson, D.G. (Eds.), Phytochemicals for pest control, ACS Symposium Series, American Chemical Society, Washington, D.C. No. 658, pp.14-26.

[34] Kim, S.I. Shin, O.K. Song, C. Cho, K.Y. and Ahn, Y.J. (2001b). Insecticidal activities of aromatic plant extracts against four agricultural insects. Agric. Chem. Biotechnol. 44; 23–26.

[35] Anandan , A. Eswaran , R. Doss , A. Sangeetha , G. and Anand, P.S. (2011). Chemical compounds investigation of *Cassia auriculata* leaves – A potential folklore medicinal plant. Bulletin of Environment, Pharmacology Life Sciences. 1 (1); 20-23.

[36] Jing, Wu. Wei, Wu. Bao, MF. Yu, Y. and Cai, XH. (2025). Aspidosperma-type bisindole alkaloids from *Melodinus suaveolens* and their antiproliferative activities. Phytochemistry. 238; 114556.

[37] Chen, S.Q, Zhang, J.H. Tang, X. Lai, M.Y. Lv, X.Q , Liu, Z.Q. Duan, L.X. Cheng, Y.Y. Wang, G.C. and Wu, P. (2025). Nudifloids O–R, four undescribed diterpenoids from *Callicarpa nudiflora* with anti-inflammatory activity. Phytochemistry. 238; 114563.

[38] Das, S. De, I. Nag, G. Karak, S. Biswas, M. Das, S and De, B. (2025). Phenolic profile and inhibitory activity of leafy vegetables against enzymes related to stress induced diseases: A comparative study. DOI: 10.22271/phyto.2025.v14.i2a.15271.

[39] Diaz-Sanchez, L.M.| Chacon-Patino, M.L.| Blanco-Tirado, C.| Combariza, M.Y. and | Montoya, G. (2025). Comparative metabolic profiling of wild and in vitro cultivated *Cecropia angustifolia* roots fraction reveals conservation of pentacyclic triterpene acid biosynthesis. Phytochemical Analysis. 0; 1–11. DOI: 10.1002/pca.70001.

[40] Chouhan, A.S. and Sharma, K. (2025). Phytochemical investigation on ethanolic extract of *Praecitrullus fistulosus* (Indian Squash) Cucurbitaceae family plant. DOI: DOI: 10.22271/phyto.2025.v14.i2a.15267.

[41] Nandhini, H.H. Punithavathy, S. Lokesh, A. and Hariram, S. (2024). Exploring the therapeutic potential-A comprehensive review of the medicinal properties of *Senna auriculata* Linn. The god gifted plant. DOI: 10.22271/plants.v12.i4c.1713.

[42] Jancy, J.J.V. Kalaichelvan, K.V. and Balakrishnan, N. (2021). Phytochemical evaluation of *Cassia auriculata* using analytical studies. IJPSR, 12(6); 3331-3338.

[43] Deshpande, S. Kewatkar, S.M. and Paithankar, V.V. (2013). In-vitro antioxidant activity of different fraction of roots of *Cassia auriculata* Linn. Drug Invention Today. 5; e164 - e168.

[44] Tietel, Z. Ananth, D.A. Sivasudha, T. and Klipcan, L. (2024).*Cassia auriculata* L.– A mini review of phytochemical compounds and their antidiabetic mechanisms. 9 (1); 374-392.

[45] Mukherjee, P.K. Maiti, K. Mukherjee, K. and Houghton P.J. (2006). Leads from Indian medicinal plants with hypoglycemic potentials. Journal of Ethnopharmacology. 106; 1–28.

[46] Sathasivampillai, S.V. Rajamanoharan, P.R.S. Munday, M. and Heinrich, M. (2017). Plants used to treat diabetes in Sri Lankan Siddha Medicine – An ethnopharmacological review of historical and modern sources. Journal of Ethnopharmacology. 198; 531–599.

[47] Tilak, M. Vijayan, R. Kumari, R.S. Radha, P. Velmurugan, M. and Sivakumar, K. (2024). Herbal soap formulation with *Senna auriculata* (L.) Roxb. Floral extract.Annals of Phytomedicine. 13(2); 963-969.

[48] Oladeji, O.S. Adelowo, F.E. Oluyori, A.P. and Bankole, D.T. (2020). Ethnobotanical description and biological activities of *Senna alata*. Evidence-based Complementary and Alternative Medicine. 13; 1-12. 2580259.

[49] Sindhuja G. and Agnes, A.M. (2025). Combination of selected medicinal plants using ethanol extract by GC-MS analysis. Mass Spectrometry Letters. 16 (1); 22-30.

[50] Kuppan, B. and Ganesan, M.C. (2024). Thermochemical conversion approach for sustainable bioenergy production from *Senna auriculata* biomass source. Arabian Journal for Science and Engineering. 49:11119–11137.

[51] Palanichamy, S. Nagarajan, S. and Devasagayam, M. (1988). Effectof *Cassia alata* leaf extract on hyperglycemic rats.Journal ofEthnopharmacology, 22; 81–90.

[52] Onwuliri, F.C. (2004). Antimicrobial studies of the extracts of Acalypha wllkesiana L. on microorganisms associated with wound and skin infections. West African J. Biological Science. 15; 15–19.

[53] Zhao, Z.X. Zou, Q.Y. Ma, Y.H. Morris-Natschke, S.L. Li, X.Y. Shi, L.C. Ma, G.X Xu, X.D. Yang, M.H. Zhao, Z.J. Li, Y.X. Xue, J. Chen, C.H. and Wu, H.F. (2025). Recent progress on triterpenoid derivatives and their anticancer potential. Phytochemistry. 229; 114257.

[54] Revathi, S. Altemimi, A.B. Sutikno, S. and Cacciola, F. (2024). Phytochemical screening along with in vitro antioxidant, antibacterial and anticancer activity of *Senna auriculata* (L.) bark extracts. Int. J. Environmental Health Research. 34(9); 2395446.

[55] Palanivel, R. Muthumanickam, M. Chinnathambi, A. and Subbiah, S. (2025). GC/MS analysis and green-synthesis of silver nanoparticles using *Senna auriculata* flower extract: Antibacterial, antioxidant effects and anticancer effects. Microbial Pathogenesis. 199; 107274.

[56] Shkir, M (2025). High-performance photocatalyst and antimicrobial potentials of biosynthesized SrO nanoparticles using *Cassia auriculata* leaf extract and evaluating their multifunctional applications. Materials Science and Engineering B 318; 118321.

[57] Yadav, P. Bharti, N. Sharma, P.K. and Mali P.C. (2025). *Cassia auriculata*-derived silver nanoparticles as a novel male contraceptive agent. Pharmacological Research - Modern Chinese Medicine 15; 100626.