

# Traditionally used edible Solanaceae plants of Mizoram, India have high antioxidant and antimicrobial potential for effective phytopharmaceutical and nutraceutical formulations

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## Research

**Keywords:** Solanaceae, ethnobotany, bioactive phytochemicals, antimicrobial, FT-IR, Mizoram

**Posted Date:** May 10th, 2021

**DOI:** <https://doi.org/10.21203/rs.3.rs-501387/v1>

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## Abstract

## Background

Solanaceae plants are incredible sources of proteins and minerals; some even have high medicinal value recognized traditionally. The present study was designed to explore and document the ethnobotany, phytochemical and mineral nutrient composition, antimicrobial properties, antioxidant potential and identify functional groups from edible species of Solanaceae from Mizoram, India.

## Methods

Field surveys and samples collection were conducted from Aizawl District, Mizoram, India. All the studied samples were extracted using Soxhlet apparatus for analysis of bioactive compounds. The total phenol, total flavonoid and total anthocyanin content were determined using standard methods. The antioxidant activity was done using DPPH free radical scavenging activity, Ascorbate peroxidase (APX), Catalase (CAT) and Superoxide dismutase (SOD) activities. The proximate analysis and mineral contents were also determined. The antibacterial potential was determined by agar well diffusion method. The functional groups present in plants were analyzed using Fourier Transformed Infrared Spectroscopy (FTIR). All the results were reported as the mean  $\pm$  standard deviation. The linear regression coefficient ( $R^2$ ) for total flavonoid and phenolic content with antioxidant activity was then analysed using Graph Pad Prism Version 5.

## Results

The phytochemical screenings showed the presence of alkaloids, tannins, flavonoids, terpenoids and saponins. The highest total phenolic content was found in *Solanum anguivi* Lam. (29.51 mg GAE/g), and *Capsicum annum* L. contained the highest total flavonoids ( $35.15 \pm 0.03$  mg/g). Proteins and carbohydrates contents were found to be the highest in *Solanum melongena* L. (28.49 mg/g) and *Physalis angulata* L. (35.64 mg/g) respectively. Elemental analysis showed the presence of Calcium (Ca), Copper (Cu), Iron (Fe), Manganese (Mn), Zinc (Zn), Potassium (K), Magnesium (Mg) and Sodium (Na) in high proportion in all the studied samples. All the plants extracts showed effective antibacterial activities against *Bacillus subtilis*, *Escherichia coli* and *Pseudomonas aeruginosa*. FTIR spectra revealed the presence of multiple functional groups, which could be used to identify bioactive compounds that can be subsequently utilized as herbal remedies for various ailments.

## Conclusions

Our findings suggest that considerable amount of nutrients, biologically active and therapeutic compounds are present in the studied samples and these plants could be potential sources for new phyto-pharmaceutical and nutraceutical preparations.

## Background

Plants are a rich source of nutrients and beneficial chemical compounds that can be used for the development of medicines. In recent years, traditional/herbal medicines are gaining special attention due to the increase concerns about safety, availability and negligible side effects as compared to synthetic medicines. In western countries more than 40% of the pharmaceutical industries relies on medicinal plants [1]. Plants also produce secondary metabolites that provide nutritional values and are involved in defense mechanisms against biotic and abiotic stresses to aid in their survival [2]. According to WHO, medicinal plants are the best sources for obtaining high quality drugs and nearly 80% world's population rely on traditional medicine for their health and well-being [3] making them the preferred source of compounds for pharmaceutical and healthcare products [4]. High activity profile drugs have already been developed from biologically active compounds from medicinal plants [5]. Crude extracts from medicinal plants are more biologically active than isolated compounds because of their synergistic effect [6].

Solanaceae is one of the biggest plant families among the angiosperms with a great potential for providing food and medicinal security in the world. It comprises of about 2300 species and is reported to be significant sources of phytochemical and nutritional compounds in pharmaceutical and food industry [7, 8]. Some work on anatomical and phytochemical characterization of *Physalis angulata* [9]; elemental, proximate and phytochemical analysis of *Solanum incanum* [10]; preliminary analysis and antimicrobial activity of *Solanum torvum* [11]; phytochemical analysis, antioxidant and anti-inflammatory of *Physalis peruviana* (Toro et al., 2014); phytochemical evaluation of *Solanum sp.* [12]; comparative morphological anatomical, cytological and phytochemical studies of *Capsicum sps.* [13] and phytochemical screening, nutritional and toxicological analysis from leaves and fruits of *Solanum macrocarpon* [14] have been reported.

Mizoram, one of the states of Northeastern (NE) region of India, lies in the Indo-Burman Biodiversity hotspot, and is known for its high ethnic and cultural diversity. It has the highest tribal population (94.8%) among all the NE states [15]. The Mizo tribes are mainly forest dwellers that rely on shifting cultivation for their livelihood. Majority of the population live in rural areas and most of their resources such as timber, food, medicinal plants etc. are obtained from the forest and hence they have a plethora of traditional knowledge on uses of different plant products. However scientific data and documentation on ethnobotany, nutritional and phytochemicals of these plants is lacking. Considering the importance of Solanaceae species, the ultimate aim of this research is to improve the knowledge about these species. So, the study was designed to investigate the ethnobotanical uses; evaluate and analyse the bioactive phytochemicals, mineral nutrient compositions, antioxidant and antimicrobial potential of methanolic extracts of edible plants of Solanaceae form Mizoram. The outcome of the study will add to the potential use of these plants in nutraceutical and pharmaceutical formulations.

## Methods

### Plant material

Ten edible Solanaceae plants- *Capsicum annuum* L., *C. frutescens* L., *Lycopersicon esculentum* Mill., *Physalis angulata* L., *Solanum americanum* Mill., *S. anguivi* Lam., *S. incanum* L., *S. melongena* L., *S. torvum* Sw. and *S. betaceum* Cav., (Fig. 1) regularly consumed by the Mizos, were collected from the wild, cultivated areas, roadsides and home gardens of Aizawl district of Mizoram. The collected plants were brought to the Department of Botany, Mizoram University for further analysis. Identification and confirmation of the collected specimen were done following published literature [16, 17]. The specimens were also deposited to the Herbarium of Department of Botany, Mizoram University. The ethnobotanical survey was conducted in Aizawl district of Mizoram, and was based on personal interviews with local herbal medicine practitioners and other knowledgeable local people and published literature.

### Bioactive compounds analysis

#### Samples preparation for extraction and Phytochemical analyses

A 50g powdered sample of the edible parts from the selected Solanaceae plants, was extracted with 500mL of methanol using a Soxhlet apparatus for 25 cycles. The extract was then concentrated at 50°C until it formed a paste. The concentration of each sample was adjusted to 100µg/mL using methanol. Presence of various phytochemicals: alkaloids, saponins, flavonoids, tannins and terpenoids from these methanolic extracts were estimated using the procedure proposed by [18].

#### Determination of Total Phenolic Content (TPC)

Total phenol was determined using Folin-Ciocalteu reagent method [19] with slight modifications. A 100 µl plant extract sample was mixed with 0.1 ml Folin-Ciocalteu reagent (1N) and incubated at room temperature. Then, 5ml of Na<sub>2</sub>CO<sub>3</sub> was added and incubated at room temperature for 30 minutes. Total phenolic content was determined using a UV-VIS spectrophotometer (Biospectrometer, Eppendorf, Germany) at 760nm. Gallic acid was used as standard and total phenol was expressed as gallic acid equivalent (mg/g of extracted compound).

#### Determination of Total Flavonoid Content (TFC)

The total flavonoid content was determined using the Aluminium chloride calorimetric method [20] with some modifications. Briefly, 1ml methanolic extract was mixed with 1ml methanol, 0.5ml aluminium chloride (1.2%) and 0.5ml Potassium acetate (100mM) and incubated at room temperature for 30 mins. The absorbance was measured at 415nm, and quercetin was used as standard. The total flavonoid content was expressed as quercetin equivalent (mg/g of extracted compound).

#### Determination of Total Anthocyanin Content (TAC)

The total anthocyanin content was measured using method proposed by [21]. The methanolic extracts were mixed with acidified methanol (Methanol and 1N HCl, 85:15 v/v, pH1) and the absorbance was measured at 535nm against reagent blank. Cyanidin 3-Glucoside was used as a standard. Total anthocyanin content was calculated as:

$$\text{TAC } (\mu\text{g/g}) = (A / \epsilon) \times (\text{vol}/1000) \times \text{MW} \times (1/\text{sample wt}) \times 10^6$$

Where A is absorbance,  $\epsilon$  is molar absorptivity of Cyanidin 3-Glucoside, vol is total volume of anthocyanin extract and MW is the molecular weight of Cyanidin 3-Glucoside.

### Evaluation of Antioxidant Activity

#### DPPH radical scavenging activity

Antioxidant activity of the extract was determined with 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging method [22]. To 50µl of 10–100 µg/mL plant extract, 2 ml DPPH was added and kept in dark at room temperature for 30mins. Then, 1ml methanol and 2 ml DPPH was used as positive control while methanol solution was used as negative control. Then the absorbance was measured at 517 nm. The percentage DPPH radical scavenging activity (%RSA) was calculated as:

$$\% \text{RSA} = 100 \times (\text{absorbance of control} - \text{Absorbance of sample}) / \text{Absorbance of control}$$

#### Catalase (CAT)

CAT activity was determined following [23]. Briefly, 0.1ml of the extract was mixed with 1.9ml of 25mM H<sub>2</sub>O<sub>2</sub> in 50mM potassium phosphate buffer (pH 7). Then the absorbance was measured at 240nm. The enzyme activity was defined as the amount of H<sub>2</sub>O<sub>2</sub> (mM) decomposed per minute.

#### Ascorbate peroxidase (APX)

APX activity was determined by using [23]. About 2ml of the extract was mixed with 0.5ml of 100mM potassium phosphate buffer (pH 7), 0.5ml of 1mM ascorbic acid, 0.5ml of 0.4mM EDTA and 0.02ml of 10mM H<sub>2</sub>O<sub>2</sub>. Then, absorbance was measured at 290nm. The enzyme activity was defined as the amount of H<sub>2</sub>O<sub>2</sub> (mM) decomposed per minute.

#### Superoxide dismutase (SOD)

SOD activity was determined following [24]. About, 3ml of the extract was mixed with 1.5M sodium carbonate, 0.1ml of 3mM EDTA, 0.2ml of 200mM methionine, 0.1ml of 2.25mM NBT, 1.5ml of 100mM potassium phosphate buffer, 0.95ml of distilled water and 0.5ml of extract. The tube without the extract was taken as control. The reaction was started by adding 0.1ml riboflavin (60uM) under light for 15mins. The absorbance was measured at 560nm and 1 unit of enzyme activity was defined as the quantity of enzyme which reduced the absorbance reading of samples by 50% in comparison with the control.

## Nutrient Determination

### Proximate Analysis

For the estimation of protein and carbohydrates, 500 mg of edible parts were homogenized with phosphate buffer (50mM, pH 7.6). The extract was centrifuged at 8000 rpm for 10 min at 4°C. The supernatant was then used for estimation of protein content following Lowry's method [25] and Carbohydrate content using [26] method (Anthrone reagent) with glucose as standard.

### Determination of mineral ion content

The standard protocol proposed by [27] was used for the determination of mineral ion contents in Solanaceae plants. One gram of air-dried sample was crushed and digested using Nitric Acid (HNO<sub>3</sub>) and Hydrogen Peroxide (H<sub>2</sub>O<sub>2</sub>) in a 5:1 ratio until it became crystal clear. The clear sample was cooled and diluted with distilled water to make up to 50ml. The diluted solution was filtered using 0.2-micron membrane filter and analysed for detection of elements using Atomic Absorption Spectroscopy (Shimadzu AA-7000, Japan) and Microwave Plasma Atomic Emission Spectroscopy (4100 MP-AES, Agilent Technologies, USA).

### Antimicrobial Activity

The antimicrobial activities of the methanolic extracts from 10 Solanaceae species were tested against three bacterial strains- *Bacillus subtilis* ATCC11774, *Pseudomonas aeruginosa* ATCC9027 and *Escherichia coli* ATCC1229 using agar well diffusion method.

### FT-IR Analysis

Functional groups present in the studied samples were identified using Fourier transformed infrared spectroscopy (Shimadzu IR Affinity-1S, Japan) for frequency ranging from 400–4000 cm<sup>-1</sup> following manufacturer's instruction.

### Statistical analysis

All the results were reported as the mean ± standard deviation. The linear regression coefficient (R<sup>2</sup>) for total flavonoid and phenolic content with antioxidant activity was analyzed using Graph Pad Prism Version 5. P-value < 0.05 was considered significant.

## Results

Ethnobotanical uses of 10 Solanaceae species used in the study are summarized in Table 1. All the different plant parts are used for treatment of various ailments as traditional medicines. The qualitative phytochemical analysis revealed the presence of alkaloids, flavonoid, saponins, tannins and terpenoid (Table 2) in all the plant extracts. The bioactive compounds; phenol, flavonoid and anthocyanin contents varied significantly among the samples (Table 3). TPC ranged from 9.87 to 29.51 mg GAE/g. Among the studied plants, *S. anguivi* had the highest phenolic contents while *S. torvum* had the lowest. Flavonoids exhibited noticeable variations among the plant extracts which ranged from 8.82 mg QE/g in *C. annuum* to 35.15 mg QE/g in *S. betaceum* (Table 3). The TAC varied from 0.069 to 0.91 mg/g in which *P. angulata* showed highest and *C. annuum* had the lowest (Table 3).

Table 1  
Ethno-botanical uses of Solanaceae plants species used in the study.

SI No	Species Name	Local Name	Part Used	Uses
1	<i>Capsicum annuum</i> L.	Hmarcha te	Leaves, Fruits	Fruits used as condiments, spices, improves digestion. Leaves prepared with fermented pork eaten as vegetables. Fruits and leaves juices applied to burn and snake bite. Fruits used as anti-haemorrhoidal, antiseptic, anti-rheumatic.
2	<i>Capsicum frutescens</i> L.	Hmarchapui	Leaves, Fruits	Fruits used as condiments, spices, improves digestion. Leaves prepared with fermented pork eaten as vegetables. Fruits leaves juices applied to burn and snake bite.
3	<i>Solanum betaceum</i> Cav.	Thingtomato	Fruits, Leaves	Fruits eaten as raw, cooked/roasted as vegetables. Also used in inflammatory painful disease, tonsils problem, liver problem. Leaves are heated on low flame and wrapped around the neck for sore throat.
4	<i>Lycopersicon esculentum</i> Mill.	Tomato	Fruits, Leaves	Fruits eaten as raw or cooked, also used as juice. Fruits used as skin care, treatment for sunburn. Leaves grinded in powder form are applied on spotted skin or leprosy spots.
5	<i>Physalis angulata</i> L.	Chal pangpuak	Fruits, Leaves	Fruits eaten as raw or cooked. Leaves are used as analgesic, antiseptic, asthma, diarrhoea. Fruits used for treatment of malaria, liver ailment, rheumatism, indigestion.
6	<i>Solanum americanum</i> Mill.	Anhling	Whole plant	Young shoot and leaves eaten as cooked. Decoction of whole plants are used as antispasmodic, anti-inflammatory blood purification, ulcers, anti-cancer, skin disease.
7	<i>Solanum anguivi</i> Lam.	Tawkte	Fruits, root, Leaves	Green fruit eaten as cooked or raw. Leaves are grinded and applied on skin disease, rash and spots. Fruits used as medicine for high blood pressure, asthma and stomach ache. Roots grounded to powder used as toothache, insect bites.
8	<i>Solanum incanum</i> L.	Samtawk	Fruits, roots, snake bites and wounds.	Green fruits eaten as cooked or raw. Fruits used as analgesic, medicine against high blood pressure, menstrual problem, sore-throat, stomach ache, liver problem, rheumatism, conjunctivitis. Roots or fruit rubbed on gums for toothache.
9	<i>Solanum melongena</i> L.	Bawkbawn	Fruits, Leaves, roots	Fruits cooked or roasted. Fruits used for lowering blood cholesterol level, high blood pressure, antihaemorrhoidal, antidote to poisonous mushrooms. Leaves as narcotics, skin disease, treatment for burns and bites. Decoction of leaves and roots used as toothache, bleeding and antiasthmatic.
10	<i>Solanum torvum</i> Sw.	Tawkpui	Fruits	Young fruits cooked or raw. Fruits used for treatment of fever, sore throats, stomach ache, chest pain. Used as antidiuretic, antidiabetic.

Table 2  
Phytochemical screening of Selected Solanaceae plants species.

SI No.	Species Name	Parts tested	Alkaloids	Flavonoids	Saponin	Tannins	Terpenoids
1	<i>Capsicum annuum</i> L.	Fruits	+	+	+	+	+
2	<i>Capsicum frutescens</i> L.	Fruits	+	+	+	+	+
3	<i>Solanum betaceum</i> Cav.	Fruits	+	+	+	+	+
4	<i>Lycopersicon esculentum</i> Mill.	Fruits	+	+	+	+	+
5	<i>Physalis angulata</i> L.	Fruits	+	+	+	+	+
6	<i>Solanum americanum</i> Mill.	Leaves	+	+	+	+	+
7	<i>Solanum anguivi</i> Lam.	Fruits	+	+	+	+	+
8	<i>Solanum incanum</i> L.	Fruits	+	+	+	+	+
9	<i>Solanum melongena</i> L.	Fruits	+	+	+	+	+
10	<i>Solanum torvum</i> Sw.	Fruits	+	+	+	+	+

Table 3  
Quantitative phytochemical analysis of Solanaceae plants species.

Sl. No	Species Name	Total Carbohydrate Content (mg/g)	Total Protein Content (mg/g)	Total Flavonoids Content (mg/g)	Total Phenolic Content (mg/g)	Total Anthocyanin content(mg/g)
1.	<i>Capsicum annum</i> L.	19.12 ± 0.004	24.75 ± 0.005	35.15 ± 0.034	20.03 ± 0.006	0.069
2.	<i>Capsicum frutescens</i> L.	24.49 ± 0.009	22.95 ± 0.058	32.24 ± 0.001	19.14 ± 0.004	0.075
3.	<i>Solanum betaceum</i> Cav.	18.19 ± 0.012	16.93 ± 0.004	8.82 ± 0.002	12.30 ± 0.008	0.45
4.	<i>Lycopersicon esculentum</i> Mill.	25.27 ± 0.041	14.09 ± 0.004	16.56 ± 0.001	12.52 ± 0.007	0.91
5.	<i>Physalis angulata</i> L.	35.64 ± 0.011	17.05 ± 0.013	30.50 ± 0.002	21.57 ± 0.004	0.75
6.	<i>Solanum americanum</i> Mill.	16.48 ± 0.022	19.18 ± 0.038	23.20 ± 0.003	16.27 ± 0.005	0.5
7.	<i>Solanum anguivi</i> Lam.	26.95 ± 0.217	12.04 ± 0.007	16.56 ± 0.001	29.51 ± 0.004	0.38
8.	<i>Solanum incanum</i> L.	20.41 ± 0.011	21.76 ± 0.055	21.21 ± 0.002	14.95 ± 0.008	0.35
9.	<i>Solanum melongena</i> L.	18.54 ± 0.019	28.49 ± 0.058	19.66 ± 0.002	15.61 ± 0.006	0.25
10.	<i>Solanum torvum</i> Sw.	15.19 ± 0.012	6.1 ± 0.011	11.92 ± 0.037	9.87 ± 0.006	0.15

The antioxidant capacity of plant extracts had significant scavenging activities on DPPH that increased with increase in concentration (10–100µg/ml) as shown in Fig. 2. The IC<sub>50</sub> value was calculated to determine the concentration of the sample required to inhibit 50% of free radicals and lower the IC<sub>50</sub> value, higher the antioxidant activity [28]. The present study shows that free radical scavenging activities of the extracts are concentration dependent and comparable to ascorbic acid. The IC<sub>50</sub> of the extracts and ascorbic acid observed in the study are detailed in Fig. 3 & Table 4. Among the extracts, *L. esculentum* (34 µg/ml) showed the strongest IC<sub>50</sub> value which was almost comparable to that of ascorbic acid. CAT, APX and SOD activities are shown in Table 4. The H<sub>2</sub>O<sub>2</sub> decomposed per minute for catalase activity ranged from 0.32 mM to 6.31 mM. *S. anguivi* had the highest decompose rate at 6.31 mM H<sub>2</sub>O<sub>2</sub> per minute while *S. melongena* decomposed the least amount of H<sub>2</sub>O<sub>2</sub> per minute 0.32 mM H<sub>2</sub>O<sub>2</sub>. H<sub>2</sub>O<sub>2</sub> decomposed per minute for APX activity ranged from 0.89 mM to 7.29 mM. *S. betaceum* decomposed 7.29 mM H<sub>2</sub>O<sub>2</sub> per minute showing highest APX activity and *C. annum* (0.89mM H<sub>2</sub>O<sub>2</sub> per minute) showed the lowest APX activity. The SOD activity of plant samples ranges from 0.23U to 1.63U. *L. esculentum* showed the highest SOD enzymatic activity while *S. torvum* possessed the lowest SOD enzymatic activity.

Table 4  
Enzymatic antioxidant activity of Solanaceae plants species.

Species Name	Catalase (mM H2O2 decomposed/min)	APX (mM H2O2 decompoed/min)	SOD (Unit)	Total Antioxidant IC50 (µg/ml)
<i>Capsicum annum</i> L.	4.21	0.93	1.46	49.51
<i>Capsicum frutescens</i> L.	3.72	0.87	1.38	45.72
<i>Solanum betaceum</i> Cav.	4.2	7.94	1.32	54
<i>Lycopersicon esculentum</i> Mill.	0.89	0.98	1.63	34
<i>Physalis angulata</i> L.	0.52	1.24	0.94	41.81
<i>Solanum americanum</i> Mill.	5.61	1.4	0.23	53.4
<i>Solanum anguivi</i> Lam.	6.31	3.26	1.3	49.46
<i>Solanum incanum</i> L.	3.73	2.8	0.56	44.24
<i>Solanum melongena</i> L.	0.32	3.27	0.84	35.67
<i>Solanum torvum</i> Sw.	5.89	4.29	0.29	36.09
Ascorbic acid				29.23

The nutrient composition of edible parts of Solanaceae plants are presented in Table 3. The protein content of the edible parts ranged from 6.1 mg/g in *S. torvum* to 28.49 mg/g in *S. melongena*. The carbohydrate content varied from 15.19 mg/g in *S. torvum* to 35.64 mg/g in *P. angulata*. The mineral compositions found in the study are presented in Table 5. High values of Na, Mg, Ca and K were found all the samples and considerable amount of Fe, Mn, Cu, Zn were also observed. The toxic mineral ions such as Pb and Ni were absent in the studied samples.

Table 5  
Element analysis of Solanaceae Plants Species.

Element Concentration (mg/kg)											
Sl No.	Species Name	Ca	Cu	Fe	Mn	Zn	K	Mg	Na	Ni	Pb
1	<i>Capsicum annuum</i> L.	1.39	0.019	0.23	0.05	0.072	1.4	0.02	1.4	0	0
2	<i>Capsicum frutescens</i> L.	1.68	0.02	0.41	0.05	0.17	0.9	6.56	1.71	0	0
3	<i>Solanum betaceum</i> Cav.	1.95	0.021	0.36	0.02	0.077	3.4	5.97	34.3	0	0
4	<i>Lycopersicon esculentum</i> Mill.	2.59	0.014	0.39	0.02	0.058	4.2	1	44.7	0	0
5	<i>Physalis angulata</i> L.	1.65	0.019	0.27	0.04	0.12	4	5.7	45.1	0	0
6	<i>Solanum americanum</i> Mill.	1.89	0.018	0.28	0.1	0.15	3.5	6.54	6.78	0	0
7	<i>Solanum anguivi</i> Lam.	2.79	0.02	0.26	0.04	0.12	2	4.05	2.23	0	0
8	<i>Solanum incanum</i> L.	2.34	0.039	3.24	0.12	0.11	2.2	1.5	34.1	0	0
9	<i>Solanum melongena</i> L.	2.73	0.039	0.43	0.12	0.12	1.3	2.29	9.24	0	0
10	<i>Solanum torvum</i> Sw.	5.46	0.045	0.4	0.1	0.1	2.3	1.14	43.4	0	0

The microbial growth inhibition of the methanolic extracts is summarized in Table 6. The antibacterial activities of extracts show strong effective inhibition activity against *Escherichia coli*, *Bacillus subtilis* and *Pseudomonas aeruginosa*. The maximum antibacterial activity was shown by *C. annuum* and the least by *S. torvum*. The FTIR analysis showed the presence of different peaks indicating the presence of different functional metabolite groups in the plant extracts (Fig. 4a & Fig. 4b).

Table 6  
Antibacterial Activity of Solanaceae Plants Species.

Species Name	<i>Escherichia coli</i> Inhibition Zone (mm)			<i>Bacillus subtilis</i> Inhibition Zone (mm)			<i>Pseudomonas areuginosa</i> Inhibition Zone (mm)		
	20 mg/ml	40 mg/ml	60 mg/ml	20 mg/ml	40 mg/ml	60 mg/ml	20 mg/ml	40 mg/ml	60 mg/ml
Streptomycin (Positive control)	15.44 ± 0.58			20.44 ± 0.78			22.73 ± 0.69		
<i>Capsicum annuum</i> L.	7.33 ± 0.33	9.97 ± 0.54	11.9 ± 0.58	10.67 ± 0.34	14.67 ± 0.89	16.67 ± 1.76	9.33 ± 1.21	13 ± 0.57	14 ± 0.78
<i>Capsicum frutescens</i> L.	6.12 ± 0.46	8.2 ± 0.74	10 ± 1.31	9.45 ± 0.72	15.2 ± 0.42	16.62 ± 1.19	8.21 ± 0.67	11.06 ± 1.12	15 ± 0.21
<i>Solanum betaceum</i> Cav.	3.45 ± 0.39	7.97 ± 0.89	10.89 ± 0.42	5.67 ± 0.38	8.88 ± 1.12	11.43 ± 0.47	6.33 ± 0.19	9.78 ± 0.49	12.22 ± 0.29
<i>Lycopersicon esculentum</i> Mill.	9.01 ± 0.54	11.74 ± 0.23	13 ± 0.36	8.5 ± 1.07	12.1 ± 0.22	14.2 ± 0.44	6.4 ± 0.56	9.2 ± 0.67	16.34 ± 0.67
<i>Physalis angulata</i> L.	5.03 ± 1.42	7.41 ± 0.44	10.2 ± 0.22	4.07 ± 0.33	6.51 ± 0.81	9.5 ± 0.61	5.6 ± 0.88	7.8 ± 1.41	10.7 ± 0.74
<i>Solanum americanum</i> Mill.	5.76 ± 1.12	7.45 ± 0.67	9.01 ± 1.21	7.22 ± 0.11	10.67 ± 0.96	12.45 ± 0.11	8.56 ± 0.68	11.33 ± 0.33	12.22 ± 0.29
<i>Solanum anguivi</i> Lam.	6.33 ± 0.33	8.22 ± 0.11	10.66 ± 0.48	7.66 ± 0.11	11.33 ± 0.44	14.55 ± 0.58	7.44 ± 0.56	9.27 ± 0.63	11.67 ± 1.02
<i>Solanum incanum</i> L.	4.27 ± 0.11	7.01 ± 0.41	7.92 ± 0.34	4.22 ± 0.64	8 ± 0.49	9.3 ± 0.24	5.1 ± 0.97	7.2 ± 0.87	8.9 ± 0.52
<i>Solanum melongena</i> L.	5.89 ± 0.22	8.43 ± 0.29	10.66 ± 0.19	6.22 ± 0.22	9.77 ± 0.39	12.33 ± 0.19	5.1 ± 0.55	6.67 ± 0.38	11.17 ± 0.19
<i>Solanum torvum</i> Sw.	4.44 ± 0.56	6.27 ± 0.63	7.44 ± 0.29	3.5 ± 0.40	5.76 ± 0.34	6.9 ± 1.45	3.9 ± 0.89	6.2 ± 1.12	7.1 ± 0.92

## Correlation between Antioxidant DPPH Scavenging Activity, Total phenolic and Flavonoid Content

A correlation analysis was performed for total phenol, flavonoid content against antioxidant activities detected in Solanaceae plants species (Fig. 5). A significant correlation was observed between total phenol, total flavonoid content and antioxidant potential ( $y = 0.515x$ ,  $R^2 = 0.73$  and  $y = 0.411x$ ,  $R^2 = 0.68$ ,  $p \leq 0.05$  respectively). The strong correlation means that the phenolic and flavonoid contents contributed significantly to the antioxidant activity [28]. The present

analysis indicates that the antioxidant activity of studied samples is strongly correlated with high content of total phenolic and flavonoid that can play as reductones by donating electrons and reacting with free radicals thereby converting into more stable products [28].

## Discussion

The present study shows that the Mizo people harbor significant knowledge on traditional use of medicinal plants. In India, particularly in northeastern region, Asteraceae is the most dominant family of medicinal plants [29]. However, members of Solanaceae family are very important medicinal plants used by the people of Mizoram [30]. Local people not only collect medicinal plants but also collect large number of wild edible fruits and vegetables to supplement their domestic nutritional requirements. They use different parts of these plants as medicine for different ailments where leaves and fruits are used for medicine preparation, in the form of a decoction, or as powders. The traditional knowledge of ethnobotanical uses of plants among the Mizo people requires documentation for preserving it for the future generations. The present investigation will add significantly to the knowledge on the importance of Solanaceae plants that are used for various purposes.

The preliminary qualitative phytochemical analysis of edible plants of Solanaceae revealed the presence of various bioactive compounds which are reported to have different biological and therapeutic properties. Alkaloids are nitrogenous compounds having antioxidant potential and have been used in folk medicine [31]. Saponin is commonly used as natural antioxidant and also promotes apoptosis in tumor cells [32, 33]. Tannins are well known antimicrobial agents [34], have antioxidant potential and have been used as active ingredients in medicine and beverages [35]. Likewise, flavonoids have antioxidant properties and prevent cell damage, providing anticancer and anti-inflammatory activities [36, 37]. Similarly, it has been reported that the presence of terpenoids influence antimicrobial properties [38], and have been used as a protective agent against oxidative stress-induced diseases [39].

Plants are the diverse source of phenolic compounds with different functions and majority are bioactive compounds with anti-cancer, anti-viral, antioxidant and anti-bacterial potentials [40]. The total amount of phenol observed in the extracts was in comparison with the previous reports by Elekofehinti et al. [41], Oyeyemi et al. [42], Yousaf et al. [43]. Among the extracts, *S. anguivi* has the highest amount of phenol and this might be the reason that the plant is being used for the treatment of various skin diseases. Flavonoids are bioactive compounds belonging to polyphenolic class and constitute the major antioxidant in fruits, plants and have advantageous effects on human health. Due to their high antioxidant properties, flavonoids are important sources of human diet [44]. They have high potential in antimicrobial, anticancer, anti-inflammatory and anti-allergic activities due to their ability to scavenge reactive oxygen species (ROS) consisting free radicals [45]. In our study, total flavonoid obtained was slightly higher than previous report [46–48]. Even a positive correlation of flavonoid and phenol content with high antioxidant potential of the extracts was recognized (Table 4). Thus, the extracts, filled with high phenol and flavonoids, could be good sources of antioxidants thereby lowering the risk of diseases triggered by oxidative stress and also improving overall antioxidant capacity. Anthocyanins are involved in enzymatic reaction in the flavonoid biosynthesis pathway [49]. Anthocyanins also provide protection against certain chronic diseases such as hyperglycemia [50], inhibit growth of tumour cells in human [51, 52], and also improve vision [53]. Anthocyanin have high antioxidant potential, antibacterial properties and are used as natural food colorants [54]. The total anthocyanin content was found the highest in *L. esculentum* (0.91 mg/g) followed by *P. angulata* (0.75 mg/g). *C. annuum* showed the lowest total anthocyanin content (Table 3). In our findings, the TAC was found higher than previous work in *S. nigrum*, *S. tuberosum*, *S. lycopersicon*, *S. melongena*, *N. tabacum*, *P. hybrida* and *Withania somnifera* extraction [55, 56]. Recent reports have suggested that Solanaceae plants are promising resources for anthocyanin extraction [49]. The demand for anthocyanins is increasing in commercial industries and in pharmaceuticals for treatment of various diseases and also in beverage industries [57]. So, Solanaceae plants could be good sources of anthocyanins for various pharmaceutical and other commercial industries.

Antioxidants present in food are gaining prominence due to their significant function in maintaining human health by preventing diseases through inhibiting free radicals that are responsible for the spread of various diseases such as cancer, neurodegenerative disorders etc. The  $IC_{50}$  for DPPH of *L. esculentum* was lowest among the studied plants indicating strong antioxidant potential while *S. torvum* showed the highest DPPH. The phenol and flavonoids are multifunctional bioactive compounds which are antioxidant, antimicrobial, anti-inflammatory and anti-cancer agents. Several studies concluded that these multifunctional bioactive compounds are the major contributors on antioxidant potential of plant extracts [58]. Hence, free radical scavenging capacity observed in our study could be due to high levels of phenol and flavonoid in the extracts. This is in agreement with a report, showing higher free radical scavenging activity with higher overall phenolic and flavonoid content [59]. Hence, the present study reveals that *L. esculentum* has a strong antioxidant potential. This property may be due to higher phenol, flavonoid and anthocyanin content, which are required for scavenging activity, in *L. esculentum*. It is also known that the amount of phenolic and flavonoid content in plants are responsible for the free radical scavenging activity. Our study suggests that the extracts of edible plants of Solanaceae display high antioxidant capacity. Environmental conditions like extreme temperature, water stress, high light intensity can cause oxidative damage by over-production of toxic ROS [60]. However, plants can protect themselves against oxidative damage by antioxidant system such as anti-oxidative enzymes and non-enzymatic compounds [61]. Plants contain various anti-oxidative enzymes including SOD, CAT, APX etc. [62]. SOD converts superoxide radicals into hydrogen peroxide, APX uses ascorbate as an electron donor to reduce hydrogen peroxide to water, CAT dismutates hydrogen peroxide into water and oxygen [62]. Living organisms are able to protect themselves from toxic effects of ROS. SOD, APX and CAT are enzymes that help in detoxifying ROS. Increased level of SOD, APX and CAT can clearly lead to enhanced oxidative stress protection [63]. Previous reports have also shown that Solanaceae plants have potential activities of SOD, APX and CAT [64, 65, 55]. Our investigation thus indicates that the Solanaceae plants are good sources of SOD, APX and CAT that have significant value in reducing stress oxidative reaction. Owing to their high antioxidant capacity, these plants can serve as good sources of antioxidants in pharmaceutical and nutraceutical formulations.

The carbohydrate content was much higher than the previous work reported by Akoto et al. [66] in *S. torvum* (7.033 mg/g). The protein content in the plant extracts was also found to be higher than a previously reported value of 2.32 mg/g of the plant extract [67]. High values of protein and carbohydrate indicate rich in essential nutrients that could be utilized for enhancing nutrition. The mineral ion compositions of the plants were also relatively high in all the studied samples. Dietary intake of potassium has significant effect on coronary heart diseases by reducing blood pressure [68]. Calcium is essential mineral ion for human diet and is involved in cell differentiation, muscle and bone formation [69]. Sodium is required for physiological processes, body fluid balance and



cellular homeostasis [70]. Magnesium is essential for circulatory system and is important for metabolism [71]. Our study also showed the presence of micronutrients such as Fe, Cu, Mn, Zn. These micronutrients are required for metabolic processes like respiration and DNA synthesis [72]. Thus, the findings also suggest the effective utilization of these plants as source of minerals or nutrient supplement.

Antibiotic resistance is an epidemic that continues to plague the healthcare system in both developing and developed countries around the world. The appearance and dissemination of multidrug-resistant pathogens has significantly jeopardized conventional antibacterial therapy. This has led to a hunt for new antimicrobial sources preferably plants that contain various bioactive compounds with established therapeutic properties. The present study was undertaken to assess the antimicrobial efficacy of edible plants of Solanaceae against multi-resistant bacterial strains- *B. subtilis*, *E. coli* and *P. aeruginosa*. Results indicated that the plant extracts exhibited significant antibacterial activities towards the tested bacterial isolates. *L. esculentum* extract showed maximum activity against all the three pathogens. The inhibition was even higher than one reported on methanol extracts of other Solanaceae plants [73]. One of the most serious challenges to humanity is the rise of multidrug resistance by pathogens. Application of effective plant extracts might be a valuable option in combating this phenomenon. Hence, the plants under current investigation could be useful in combating antidrug resistance in the tested bacterial strains. However, further investigations are sought to evaluate anti-viral, anti-fungal and anti-parasitic activities in order to harness the potentials of the plants.

Another important aspect of our study was to identify the functional groups found in the plant extracts using FTIR. This analysis helps in the identification of chemical composition, elucidation of chemical structure and to understand the importance of functional groups as bioactive compounds for phyto-pharmaceutical formulations. The plants have shown similar infra-red spectrum and some intense bands at various frequencies which define the presence of O-H (hydroxyl), O-H stretch (carboxylic acid), O-H bend (phenol or tertiary alcohol) C-H stretch (alkanes), C = C-C (aromatic compounds), C = C stretch (ketone), N-O (nitro compound), C-O (ether), C-N (aromatic primary amines), N-H (amines), C  $\equiv$  C (carbonyl), C-Br (aliphatic bromo compounds) (Table 7) groups. The presence of these functional groups indicates different metabolites such as aldehydes, alkanes, alkenes, alkynes, alkyl halides, aliphatic amines, primary and secondary amines, alcohols, aromatics, carboxylic acids, esters, ethers, glycogen, hydroxyl, lipid, organic halogen compounds, nitro compounds, phenols and tryglycerides, that are integral parts of most of the secondary metabolites such as alkaloids, flavonoids, tannins, terpenoids and polyphenol [74]. Functional groups in the plants can be used in different pharmaceutical products such as for anti-cancers, anti-ulcers, jaundice, headache, stomach ache and anti-inflammatory drugs; or as sources of antimicrobial, antioxidant compounds etc. [75–77]. This may also be the reason why traditionally these plants are used by the locals in treatment of stomach ache, as anti-inflammatory medicine etc. (Table 1). The phytochemical screening and FTIR analysis showed that various bioactive compounds were found in the plant extracts that can be used as active antioxidant and anti-microbial agents of plant origin. The current study also revealed clear discrimination between the plants parts tested (leaf, fruit whole plant etc.), displaying significant heterogeneity for identification of bioactive phytochemicals that can be used as herbal medicines. However, further studies are necessary to evaluate *in vivo* biological activities of the bioactive phytochemicals for designing effective phyto-pharmaceutical formulations.

Table 7  
Evaluation of FT-IR spectra of Solanaceae plants.

Frequency range (cm-1)	Peak wavenumber (cm-1)										Functional group
3870 – 3550	<i>C. annuum</i> L.	<i>C. frutescens</i> L.	<i>L. esculentum</i> Mill.	<i>P. angulata</i> L.	<i>S. Americanum</i> Mill.	<i>S. anguivi</i> L.	<i>S. betaceum</i> Cav.	<i>S. incanum</i> L.	<i>S. melongena</i> L.	<i>S. torvum</i> L.	O-H stretch alcohol
3500 – 3200	3750	3672	3672.5	3865	3742	3865	3834	3741.9		3672	O-H stretch vibration presence of alcohols, phenols
3300 – 2850	3441	3441	3387	3441	3449	3649	3487	3417.9	3364	3325	O-H stretch vibration, carboxylic acids
2500 – 2300		2916		2924	2924	3225		3302.1	2924		C-H stretch vibration, alkenes
2260 – 2100			2330	2307			2446			2484	C = C stretch vibration, alkynes
1990 – 1739		2160	2152.6	2137	2207	2160		2237.4		2237	Ester C = O stretch, lipid, triglycerides
1700 – 1600		1836	1743.7		1983	1921	1844	1975.1		1975	C = C stretch vibration, alkenes
1550 – 1475		1605			1643	1651			1620		N-O asymmetric stretch, nitro compounds
1470 – 1400					1520			1543.1			C-C stretch vibration, aromatics
1400 – 1320			1458.2		1458	1458	1420			1458	N-O stretch vibration, nitro compounds
1300 – 1290		1319		1319					1319	1319	C-O stretch vibration, alcohol, carboxylic acids, esters, ether
1275 – 1150											C-H wag stretch vibration, alkyl halides
1020 – 1000		1219	1219	1219	1219	1219		1219	1242	1219	C-N stretch vibration, aliphatic amines
990 – 800		1026			1034	1034			1034	1034	N-H wag stretch vibration, primary & secondary amines
790 – 690											C(triple bond)C-HC-H bend stretch vibration, alkynes

Frequency range (cm-1)	Peak wavenumber (cm-1)										Functional group
680 – 510	772	772		771.5		77.5	741.53	779	772		C-Br stretch vibration, alkyl halides, glycogen
490 – 400	556	517	640.37	671.2	594.1	617	664	616.92	556	617	Halogen compound

Conclusions

Ethnobotanical uses, bioactive compound compositions, antioxidant activities, nutrient compositions and antimicrobial potential of edible plants of Solanaceae from Mizoram, India were analysed. These Solanaceae plants contain various bioactive phytochemicals, antimicrobial agents with various functional groups and has promising nutritional and antioxidant potential. Results demonstrated that these plants could be used as an easily accessible source of natural bioactive compounds with antioxidant and antimicrobial potentials and can also substitute synthetic drugs. To the best of our understandings, this is the first complete study of edible Solanaceae plants from Mizoram to investigate bioactive compounds, mineral nutrient contents, antimicrobial potential, antioxidant determination and identifying functional groups. Further studies on these plant species could open new perspective for developing novel health-promoting agents in pharmaceutical and nutraceutical industries.

Abbreviations

- FTIR
- Fourier Transformed Infrared Spectroscopy
- TPC
- Total Phenol Content
- TFC
- Total Flavanoid Content
- TAC
- Total Anthocyanin Content
- DPPH
- 2,2-diphenyl-1-picrylhydrazyl
- CAT
- Catalase
- APX
- Ascorbate Peroxidase
- SOD
- Superoxide Dismutase
- MP-AES
- Microwave Plasma Eatomic Emission Spectroscopy
- AAS
- Atomic Absorption Spectroscopy

Declarations

Ethics approval and consent to participate: Not applicable

Consent for publication: Not applicable

Availability of data and materials: All data generated or analysed during this study are included in this published article.

Competing interests: The authors declare that they have no competing interests

Funding: No funding was received to undertake this research

Authors' contributions: LR performed the experiments, analysed the data, wrote the manuscript; UB analysed the data, wrote the manuscript; YTS conceptualized the research, analysed the data, wrote the manuscript.

Acknowledgements: LR thanks University Grants Commission, India for UGC-MZU Doctoral Fellowship. Authors thank Department of Chemistry, Mizoram University for AAS and FT-IR facility and Central Instrumentation laboratory, Mizoram University for MP-AES facility.

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## Figures

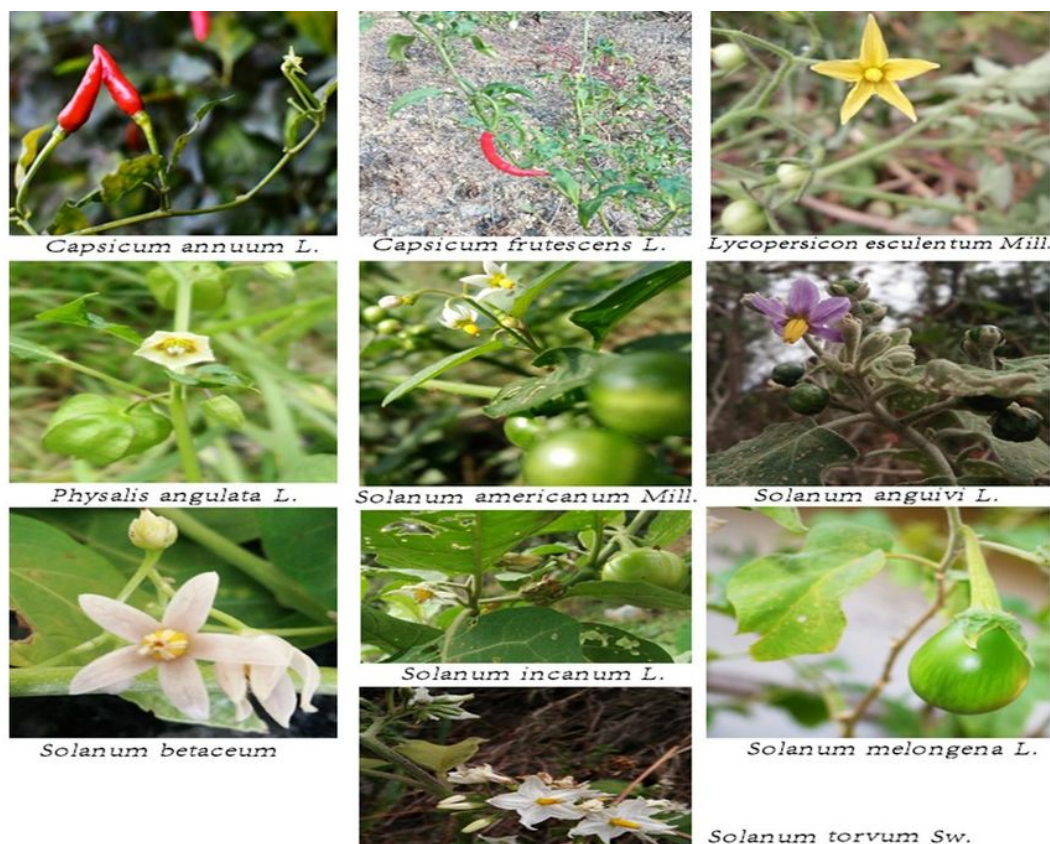


Figure 1

Solanaceae plants species used in the study.

## DPPH radical Scavenging activity

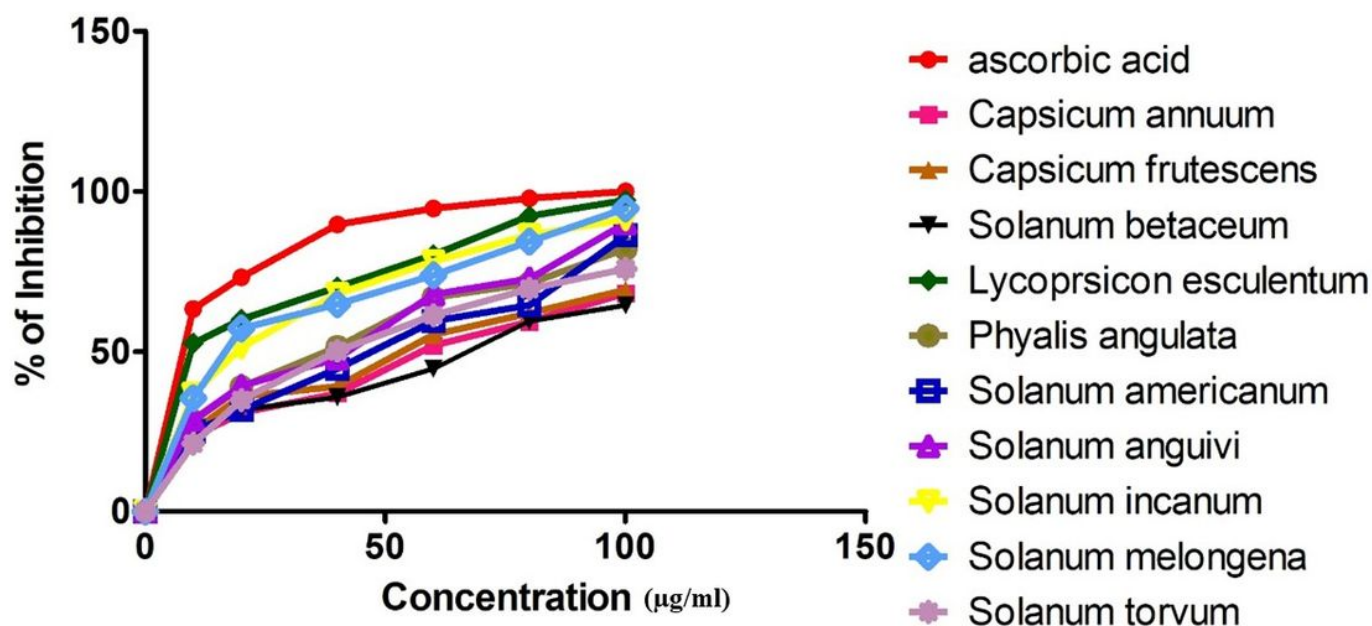


Figure 2

Antioxidant DPPH radical scavenging activity of Solanaceae species.

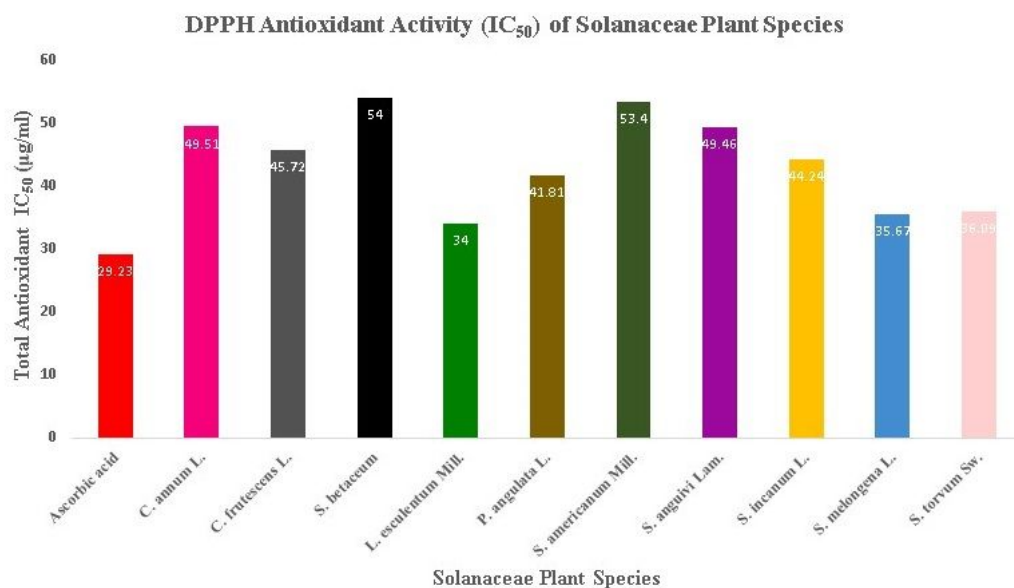


Figure 3

Antioxidant IC50 of Solanaceae plants.

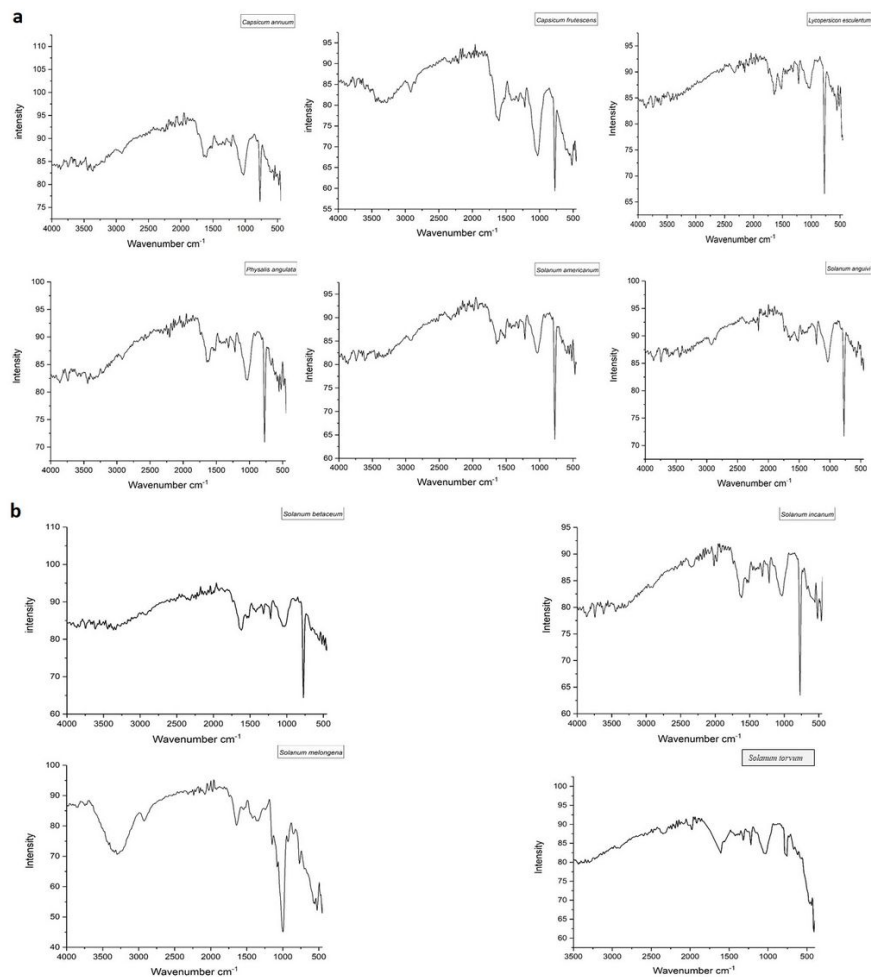


Figure 4

a. FT-IR spectra of Solanaceae plants. b. FT-IR spectra of Solanaceae plants.

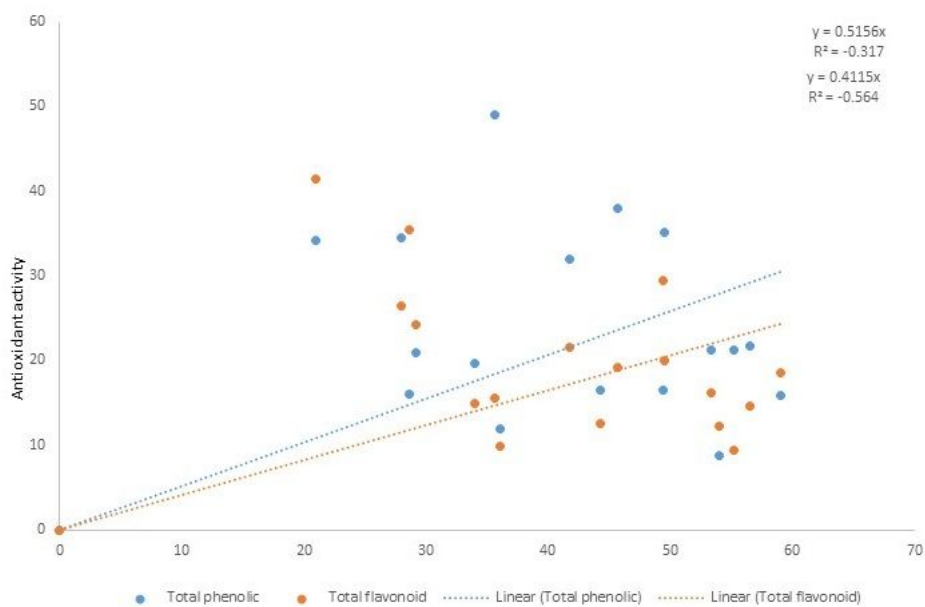


Figure 5



