FTIR Spectrum, Phytochemical Assessment and Biological Properties of *Solanum Surrattense*

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Abstract

Solanum surattense are the most important medicinal plants that can be useful for the treatment of different diseases. The Seed, flower, root, leaf, and stem of the plant were selected, collected, washed, dried, converted into a fine powder, and stored for phytochemical analysis. The finding of the present study indicated that the roots, stem, leaves, flowers, and seeds showed a significant amount of phytochemicals including alkaloid, flavonoid, tannin, glycoside, terpenoid, steroid, and antioxidant content and the FTIR Spectroscopic characteristic peak values between 4000-500 cm-1 was measured for the detection of the various functional group including aromatic compounds, glycoside, tannin, flavonoid and saponin, amino acids and protein, glycogen, collagen, and DNA. This study showed that of rich sources of proximate composition including Carbohydrate, protein, and ash whereas the presence of the appreciable amount of flavonoid, phenolic compound, and tannin. The seeds showed the appreciable sources of alkaloid and leave showed antioxidant content. The richest sources of principle essential minerals including Ca, K, Na, Mg, Fe, Zn, Mn, Co, Cu, Cr, Ni whereas toxic minerals such as Pb and Cd are investigated in permissible limit. Antibacterial activity indicated in the present studies that the Bacillus cereus, Escherichia coli were showed. The growth of Bacillus Cereus and Escherichia Coli is inhibited by mostly all parts of plants. Solanum surattense is the richest source of phytochemical and possessed biological properties that can be beneficial for the synthesis of new plant-derived for the treatment of different diseases.

Key Words: FTIR Spectrum, proximate, phytochemical, minerals, antibacterial and antioxidant activity

1. Introduction

Medicinal Plants have been used as the bioresources of traditional medicine forthe treatment of various types of diseases(Chandana C B et al., 2015).*Solanum surattense* belongsSolanaceae family locally known as Nightshade yellow berried and widely grows as a perennial herbaceous weed in the various regions of the subcontinent Indo-Pakistan up to very high altitude. The different parts have

been used in the treatment of the diseases including cough, fever, constipation, asthma, toothache, bronchitis, sore throat, laxative, rheumatism, tuberculosis, gonorrhea and kidney diseases. It is also used for antipyretic activity, and antioxidant activity. In the Indian Materia Medica described that leaves are used for the treatment of different diseases including, cough, fever, digestive problem, bronchitis, stomachic, pharyngitis, hypertension, carminative, arthritis, appetizer, rheumatoid, inflammatory, amenorrhea, hemorrhoids, epilepsy, dysmenorrhoea, cardiac disorders, asthma, rhinopathy, urolithiasis, lumbago, anthelmintic, and catarrh(Vaidya Ratnam PSV et al., 1994).Thefruit, stem and flower are used for the treatment of dengue fever, fever, burning sensation, chest infection, acute bronchitis, phytotoxic, haemagglutination, fungal and bacterial diseases(Ahmad H et al., 2012). The entire plants are used for joint pain, gas trouble, diuretic, leprosy, acute bronchitis asthmatic, fever, chronic cough, blood purifier, severe headache, and abdominal(Malik S et al., 2015).

2. Materials and Methods

Sample Preparation:Leave, root, flower, stem and fruit were being selected, and collected from Nasirabad, district Kamber, Sindh, Pakistan. Plant materialshad been dried about 15-30 days under shade and converted into powdered form after complete dryness. Further stored in the airtight bags.Total moisture content identified through the oven, and total ash contentwas determined through muffle furnace(Umadevi KJ et al., 2012).

2.1 FTIR Spectrum: Infrared spectroscopy was used for the detection of the functional groups of phytochemicals in the medicinal plant sample mixture. The plant powder was put into an IR spectrometer. The results were recorded on an FTIR spectrometer between the range 4000-500 cm-1. The wavelength of light absorbed was characteristic of the chemical bonds which can be seen in the annotated spectrum(Lanjwani A H et al., 2015).

2.3 TotalCarbohydrate:Reagent:200 mg of the reagentAnthrone was taken and dissolved in the 100 ml of ice-cold concentrated H_2SO_4 .

Sample Preparation:100 mg of the plant powder material was taken 100 mg of powdered plants materials were taken. 5 ml of 2.5 N hydrochloric acid was added. Placed on the boiling waterbath for three hours at 100° C, cooled andfurther solid Na₂CO₃ was added until effervescence ceases. The final volume of 100 ml was made with distilled water.

Procedure: 1 ml of plantsample was diluted with 4ml anthrone reagent. Placed on the water boiling bath for eight minutes. Further rapidly cooled then absorbance was taken at 630 nm. The range as 10, 20,40,60,80 and 100 μ g/ml of the standard was arranged(Rawat A et al., 2012).

2.4 Total Protein: Total protein was determined byLowry Method

Reagent Preparation:Reagent A: $Na_2CO_3(2\%)$, reagent B: $CuSO_4(1\%)$, reagent C: sodium potassium tartrate (2%), reagent D: mixed reagent A, B and C prepared as the ratio 100:1:1 respectively, Standard: 2 mg BSA/ml.

Plant Sample Preparation:1 gram of plant sample was dissolved in the 10 ml of phosphate buffer; the solution was kept on shaking water bath overnight. Filtered and filtrated were collected for further

Procedure: 100 μ l of plant sample or standard was taken, 100 μ l 2N NaOH was diluted and placed on the boiled water bath for ten minutesat 100 ⁰C. Further cooled and freshly prepared 1 ml of

reagent D was added, and the solution was allowed to stand for ten minutes at room temperature. 100 μ l of reagent folin was added, well mixed, allowed to stand for 30-60 minutes at room temperature then absorbance wastaken at 550 nm(Naidu MP et al., 2013).

2.5 Total Phenolic Compound: Total phenolic compound was estimated byfolin ciocalteu reagent method.

Preparation of Sample:10 g of plant sample was taken and dissolved in the 100 ml of (80%) aqueous methanol was added. Kept 24 hours on shaking water bath at room temperature. Further filtered through Whatmann filter paper no 1. The solution was centrifuged for 20 minutes at 6000 rpm. The filtered materials were and stored at -20 °C for further analysis of total phenolic compounds, flavonoids, and tannins.

Standard Solution: Gallic acid 100 μ g/ml in the 80 % an aqueous methanol. The range (10, 20, 40, 60, 80 and 100 μ g/ml) was arranged.

Procedure:0.5 ml of sample or standard was taken and 2.5 ml of reagent folin-ciocalteu (10 fold diluted in the distilled water). Further 2 ml of 7.5% Na₂CO₃ was added. Kept for thirty minutes at room temperature in the incubator and absorbance was taken at 760 nm(Maurya S, Singh D, 2010).

2.6 Total flavonoid: It was estimated by the Aluminum chloride method

Standard Solution:Quercetin 500 μ g/ml in the 80 % aqueous methanol. The range(100, 200, 300, 400, 500 μ g/ml).

Procedure:The 1 ml of extract or standard was taken and dissolved in the 4 ml of the distilled water. 0.3 ml of 5 % sodium nitrate (NaNO₂)was added and allowed to stand for five minutes. 0.3 ml of the 10% aluminum chloride (AlCl₃)was added and allowed to stand for six minutes. 2 ml of 1M sodium hydroxide was added.Final volumeof 10 ml in the double distilled water was made, and then reading was measured at 510 nm(Damodar K et al., 2011).

2.7 Total Alkaloid: Total alkaloidwas estimated by Dragendorff's method.

Dragendorff'sReagent:Solution A: for this solution, 0.8 gram of thebismuth nitrate pentahydrate was taken. 40 ml of water distilled was addedand then furtherglacial acetic acid 10 ml was added.

Solution B: for this, 8 gram potassium iodide was dissolved in the 30 ml of water distilled and then further solutions B and A were mixed.Standard Solution:In this, 10 mg of Bismuth Nitrate Pentahydrate was dissolved in the 5 ml of concentrated nitric acid then final volume of 100 ml was made in the double distilled water. The range of standard (200, 400, 600, 800 and 1000 μ g/ml) were arranged.

Preparation of Extract:10 grams of plant fine powder was dissolved in the 50 ml 2% aqueous acetic acid and keptfor 30 minutes on boiling water bath. The solution was filtered withWhatmann paper no 1. The filtrate was collected, and residue were extracted again repeated same respectively. Above both plants extract was mixed, and then 100 ml final volume was made in the aqueous acetic acid final volume of 100 ml was made in the aqueous acetic acid. The pH 2.5of extract was maintained.

Procedure:5 ml of sample or standard was treated with 2 ml of the reagent dragendorff's and allowed to stand. The precipitateswere formed, and then the solutions were centrifuged at 4000 rpm for ten minutes. Precipitates were collected and remaining supernatants were againtreated with reagent dragendorff's forto check the precipitation more, then precipitated were mixed and precipitates were washed with alcohol. The precipitates were dissolved in the 2 ml Disodium sulfide and precipitates

brownish black were formed and remaining solutions were discarded. The precipitates weretreated with 2 ml of HNO_3 and then 10 ml of double distilled water was added. 1 ml was taken outfrom solution and diluted with 5 ml of thiourea. Reading was obtained at 435nm(Sonal P et al., 2011).

2.8 Total Tannin: It was estimated by modified Prussian method

Standard Tannic Acid: For this, tannic acid standardin the 80% methanol were ranged as (200, 400, 600, 800 and 1000 μ g/ml).

Procedure:0.1 ml of plant extract was diluted in the 6.9 ml of water distilled. For this, 1 ml of 0.008M of potassium ferric cyanide was added, 1 ml of 0.2 M of ferric chloride in 0.1 hydrochloric acid was added and mixed well. The blue color was formed. Absorbance was taken at 700 nm by spectrophotometer(Sathishkumar T, Baskar R, 2014).

2.9 Mineral Analysis: Mineral Sample was prepared by wet acid digestion method. 0.5 g of powdered sample was taken in the volumetric flask and then 5 ml of the(Conc.) HNO₃was added and to stand for three hours. The volumetric flask was covered by watch glassand heated at 80-100 °C on a hot plate. Then again 5 ml of concentration of HNO₃were added and heated further 2 ml of H_2O_2 were added and shaken well heated until transparent clear solutions were obtained and heated until near to dryness. 10 ml of deionized water were added and shaken well.Filtered throughwhatmann filter paper 42 and final volume 25 ml was made by the addition of deionizedwater and concentration was taken by atomic absorption spectrometer (AA.800 perkin elmer)(Lanjwani AH, et al., 2016).

2.10 Antimicrobial activity: It was estimated by agar well diffusion method.

Media for bacterial cultures: The medium was neutralized for 30 minutes at 37 $^{\circ}$ C and filtered further then sterilized at 15 lbs for 20 minutes at 121 $^{\circ}$ C.

Procedure:Suspension of 24 hours cultures of *bacillus cereus*, and *Escherichia coli* was made and sterilized with normal saline. Each medium platewas inoculated by test organism and sterilized with swab rolled of cotton in suspension to strip plate surface in the form which lawn growth wasformed. The cork borer of 5 mm diameter was used for the formation of well in the medium plates. 50 µl of plant extract (80 % methanol) were dropped into each well. The agar platewas incubated at 37 °C for 24 hours(Bonjar Bongar GS, 2014).

2.11 Antioxidant Content: It was estimated by ferric reducing antioxidant power method.

Procedure:2.5 ml of extracts were diluted with buffer phosphate (pH 6.6).1 ml (1%) of potassium ferric cyanides was added. The solution was kept for 20 minutes in the incubator at 50°C. It wascooled rapidly and diluted with 2.5 ml of (10 %) Trichloro-acetic acids and then for tencentrifuged. From the above solution, 2.5 ml of aliquots was taken, and 2.5 ml of water distilledwas added, further, 0.5 ml of 0.1% ferric chlorides were added. The abovesolutionscolor were converted to green and to stand for ten minutes. The readings were measured at 593 nmby spectrophotometer(Patel A, et al., 2010).

3. Results

The finding of the present study indicated that the roots, stem, leaves, flowers, and seeds showed a significant amount of phytochemicals including alkaloid, flavonoid, tannin, glycoside, terpenoid, steroid, and antioxidant content. The FTIR Spectroscopic the characteristic peak values between 4000-500 cm-1 for identification of the various functional group. FTIR analysis indicated that the

similarity in the selected parts including leaves, flowers, and seed based on the functional group presence and infrared spectrum. The peak at 2926 wave number cm-1. This peak indicated the presence of glycoside, tannin, flavonoid, and saponin. The peak at 1030 cm-1 is due to organosulfur compounds. The very strong band at the between region 2933–2922 cm-1 due to N-H stretching. The C=O stretching vibration band is due to the presence of saturated aliphatic ester. The bands at 900-1350cm-1, 1030 cm-1, in the leaves, and 1027 cm-1 in the seed and flowers are attributed to phosphodiester stretching bands region which peak due to collagen, glycogen, and DNA. The functional group C–O stretch is due to the presence of the glycogen, phosphate, and oligosaccharides PO-2 stretching modes. The strong absorption band is investigated between 1600 - 1660 cm-1 region which is due to the presence of amino acid and protein. The 1601 cm-1, 1605, and 1615 were observed in the selected parts of the plant. FTIR Peak Values of leave are shown in (Figure 1). The high peak at 3292.72 wave number cm-1 may be the presence of the O-H group of hydrogen bounded alcohols and phenols. The peak at 2919 wave number cm-1 is due to the presence of an asymmetric stretching of the C-H group of the aromatic compounds which is indicated the presence of the flavonoid, tannin, glycoside, and saponin. The strong peak at 1615 wave number cm-1. It is due to the presence of amino acids. The peak at 1375.01 and 1315.98 is due to C-N bonds showed Alkyl ketone, amide, and amines. The highest band absorption at 1030 cm-1 in the leave is due to the phosphodiester stretching bands region which is indicated the presence of collagen, glycogen, and DNA. FTIR Peak Values of Solanum Surattense flowers are shown in (Figure 2). The high peak at 3288.10 wave number cm-1 may be the presence of the O-H group of hydrogen bounded alcohols and phenols. The peak at 2919.19 wave number cm-1 is due to the presence of the asymmetric stretching of the C-H group of aromatic compounds which is indicated the presence of the flavonoid, tannin, glycoside, and saponin. The strong peak at 1605 wave number cm-1. It is due to the presence of amino acids. The peak at 1409.39 wave number cm-1 due to the C-H group due to Alkanes whereas, 1316.48 and 1235.87 are due to C-N bonds showed Alkyl ketone, amide, and amines. The highest band absorption at 1019.81 cm-1 in the flower is due to the phosphodiester stretching bands region which is indicated the presence of collagen, glycogen, and DNA. FTIR Peak Values of Solanum Surattense seed are shown in (Figure 3). The strong peak at 3322.86 wave number cm-1 may be due to the O-H group of hydrogen bounded alcohols and phenols. The peak at 2926.37 wave number cm-1 is due to the presence of an asymmetric stretching of the C-H group of aromatic compounds which is indicated the presence of the flavonoid, tannin, glycoside, and saponin. The strong peak at 1601 wave number cm-1. It is due to the presence of amino acids. The peak at 1378.07 wave number cm-1 due to the C-H group due to Alkanes whereas, 1315.08 and 1254.44 are due to C-N bonds showed Alkyl ketone, amide, and amines. The strong band absorption at 1027.61 cm-1 in the seed is due to the phosphodiester stretching bands region which is indicated the presence of collagen, glycogen, and DNA. Proximate composition is shown in (Figure 4). Roots showed the highest amount of ash that is a good sign because ash possessed minerals deposition in the plants. The seeds and roots of plants showed good sources of carbohydrates; whereas the highest percentage of protein was observed in the flowers. These parts can play a key role against malnutrition because Protein is the building block of cells and the body. Phytochemical content and antioxidant content are shown in (Figure 5). The leaves and fruits showed appreciable sources of phenolic compound,

alkaloid, flavonoid, tannin, and antioxidant content. Mineral Composition is shown in (Table 3). It is investigated that appreciable sources of principle essential macrominerals including calcium, iron, magnesium, sodium, and potassium. The leaves showed top nutritional values as compared to other parts. The content of minerals in the plants depends on the abundance of the soil, including the intensity of fertility. This result shows this plant is the richest source of calcium ranging from (1840-17360 mg/Kg) and sodium ranged from (504-1760 mg/Kg) whereas most parts of solanum surrattense are rich sources of calcium and sodium. Calcium and sodium will be beneficial to consumers because of their high values in human health. Irons and potassium content ranged from (19.1-437.8 mg/Kg);(1221.6-1349.6 mg/kg) were investigated. The lead, cadmium, and cobalt were showed around the permissible limit in most parts. It is beneficial for the consumers because of its high toxicity. Antibacterial activity (Inhibition Zone in mm diameter) is indicated in (Figure 6). The present study that the Escherichia coli, Bacillus cereus were showed to be the maximum sensitive. The Bacillus cereus and Escherichia coli growth are inhibited by mostly all parts of plants. The Bacillus cereus growth is inhibited zone of inhibition measured as (10 mm) in the flower, (9 mm) in the root, fruit, and stem, (8 mm) in the leave. The growth of Escherichia Coli is inhibited zone of inhibition measured as (27 mm) in the flower, (22 mm) in the leave, (21 mm) in the fruit, (8 mm) in the root, (7 mm) in the stem.

4. Discussion

Pakistan is diverse in medicinal plants. There is a broad range of plant parts possessing a variety of pharmacological properties. The present findings showed that The FTIR Spectroscopic characteristic peak values between 4000-500 cm-1 for identification of the various functional group. FTIR analysis indicated that the similarity in the selected parts including leaves, flowers, and seed based on the functional group presence and infrared spectrum. The peak at 2926 wave number cm-1. This peak indicated the presence of glycoside, tannin, flavonoid, and saponin. The peak at 1030 cm-1 due to organosulfur compounds including alliin, allicin, and diallyl disulfide

(Shivani Ghildiyal). The very strong band at the between region 2933– 2922 cm–1 due to N–H stretching. The C=O stretching vibration band is due to the presence of saturated aliphatic ester. The bands at 900–1350cm–1, 1030 cm–1, in the leaves, and 1027 cm–1 in the seed and flowers are attributed to phosphodiester stretching bands region which peak due to collagen, glycogen, and DNA. The functional group C–O stretch is due to the presence of the glycogen, phosphate, and oligosaccharides PO–2 stretching modes(Vijay Amirtharaj L, et al., 2015). The strong absorption band is investigated between 1600 - 1660 cm-1 region which is due to the presence of amino acid and protein. It is noticed that there is no evidence of cyanide in the between the region 2220-2260 cm-1 in all parts whereas cyanide have a toxic effect on consumer(Manju Sharma et al., 2013).Roots showed the highest amount of ash that is a good sign because ash possessed minerals deposition in the plants. The seeds and roots of plants showed good sources of carbohydrates; whereas the highest malnutrition because Protein is the building block of cells and the body. The leaves and fruits showed appreciable sources of phenolic compound, alkaloid, flavonoid, tannin, and antioxidant content. It is reported that phytochemicals have adverse beneficial effects on human beings. For

example, phenolic compounds, flavonoids, and tannins possessed medicinal properties including antioxidant, anti-inflammatory, antihypertensive hypocholesterolemia, and anticancer, hypoglycemic, properties(Oluwole, S, et al., 2013). Alkaloids have huge medicinal properties including antitumor, antihypertension, antimalarial, analgesic, antiprotozoal, anticholinergic, cough medicine etc(Egbuna Chukwuebuka and Ifemeje Jonathan Chinenye, 2015). Phytochemicals including phenolic compound, alkaloid, flavonoid, tannin, and lignin possessed biological activities including antioxidant, flavonoid, phenolic compound, and tannin, etc can scavenge the H2O2 and into water.(Annapandian VM, Rajagopal SS, 2017). The present study that the Escherichia coli, Bacillus cereus were showed to be the maximum sensitive. The Bacillus cereus and Escherichia coli growth are inhibited by mostly all parts of plants. The Bacillus cereus growth is inhibited zone of inhibition measured as (10 mm) in the flower, (9 mm) in the root, fruit, and stem. A similar antibacterial finding was reported that inhibited Escherichia coli, Bacillus cereus. (Sheeba E, 2010). This plant is richest biosources of antibacterial phytochemicals that can be beneficial for preventing the movement of life ominous pathogenic including Escherichia coli leads hemorrhagic colitis Syndrome and hemolytic uremic syndrome, and (Zhou B, et al., 2017). Previously various studies reported that medicinal plants are most valuable antibiotics(Basile A, et al., 2000) and the chemical compounds have an effective role against antibiotic resistant strain of bacteria that is produced by traditional remedies(Kone WM, et al., 2014).

5. Conclusion

In the present conclusion, it can be noticed that *Solanum surattense* significant resources of phytochemical and strongly emphasize that can be accessible bio-resources plants derived new drugs. This plant can be used for the treatment of bacterial infectious disease and also richest sources of essential minerals including calcium, potassium, and iron which can be beneficial for consumers. These results urge to make the best utilization and scientifically database to find out standardization of new herbal drugs and natural products.

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Conflict of Interest

Author declare no conflict of interest

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Appendix:

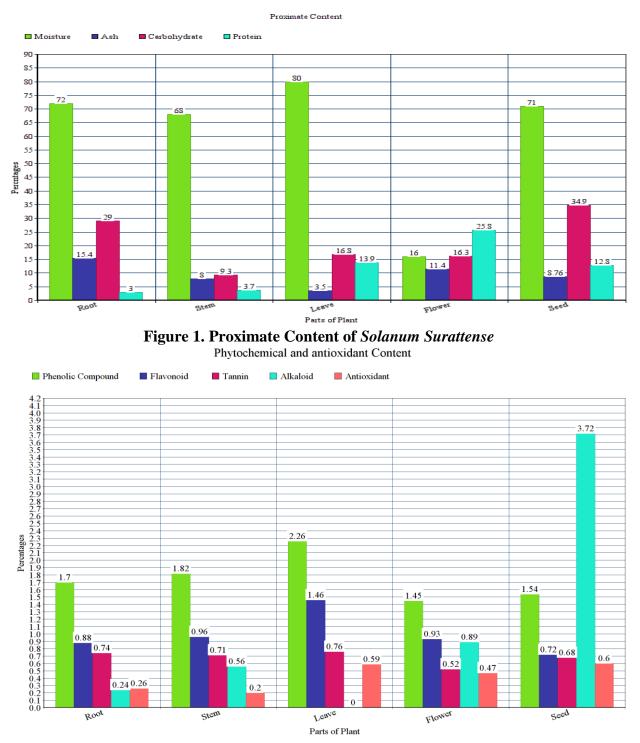


Figure 2. Phytochemical and antioxidant Content

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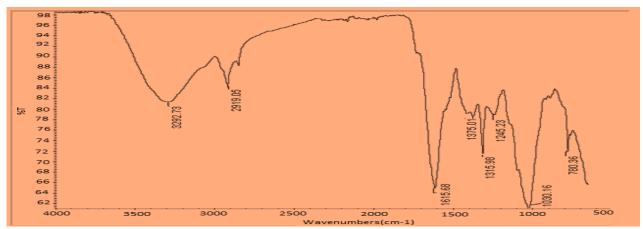


Figure 3. FTIR Peak Values of leave

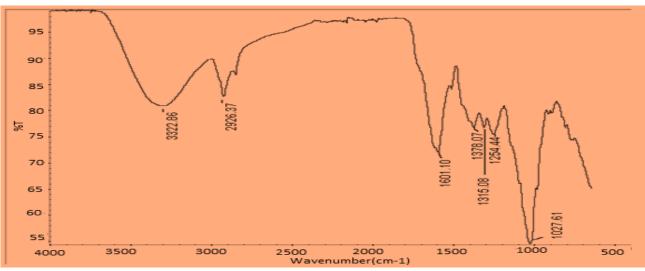


Figure 4. FTIR Peak Values of Flowers

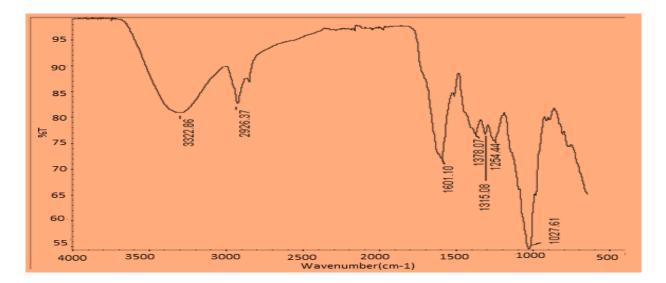


Figure 5. FTIR Spectrum of Seed

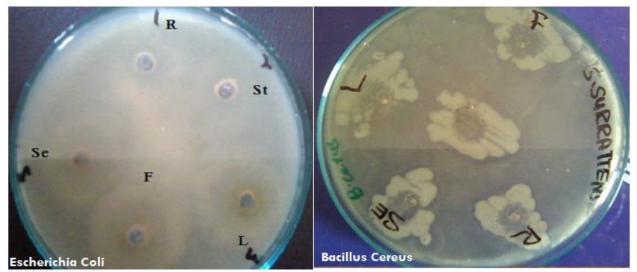


Figure 6. Antibacterial Activity of different parts of Solanum Surattense

Phytochemical	Root	Stem	Leave	Flower	Seed
Phenolic	+++	+++	+++	+	+++
Flavonoid	++	++	+++	+	+
Tannin	+++	+++	+++	+++	+++
Alkaloid	+	++	-	+	+++
Saponin	+++	+++	+++	++	+++
Steroid	+++	+++	++	+++	+++
Glycoside	-	++	+++	++	+++
Carbohydrate	+++	+++	+++	+++	+++
Protein	+++	+++	+++	+++	+++
Amino Acid	+++	+++	+++	+++	+++
Fat and Oil	+	+	+	++	+++
Vitamin C	+++	+	++	++	++

Table 2. FTIR Peak ranges

Bond	Functional Group Assignment	Group Frequency, cm-1
О-Н	Hydrogen bonded alcohols, phenols	3200-3600
C-H	Alkanes	2850-2970
N-H bend	Secondary amine	1550-1650
C-N	Alkyl ketone, Amines, Amides	1215–1325
C-F stretch	Aliphatic Fluoro compounds	1000-1150
С-Н	Alkenes	675-995

Minerals	Root	Stem	Leave	Flower	Seed
Calcium	12640±8.23	16000 ± 15.45	17360 ± 7.96	2656 ± 16.24	1840 ± 18.38
Iron	112.3 ± 0.95	212.8 ± 0.23	437.8± 1.28	289.0 ± 0.34	19.1 ± 0.86
Potassium	1221.6 ± 0.42	1349.6±2.45	1257.6 ± 1.56	1278.4 ± 2.49	1274.4 ± 0.87
Magnesium	870.2 ± 0.39	903.3 ± 0.72	912.8± 1.35	695.4± 2.13	728.1±0.23
Sodium	1760 ± 0.99	1360 ± 3.12	1736 ± 0.72	1600 ± 2.34	504 ± 3.11
Zinc	38.5 ± 2.44	30.2 ± 0.45	$24.5{\pm}0.97$	37.8 ± 0.51	8.5 ± 2.63
Manganese	21 ± 1.52	9.6± 0.12	BDL	BDL	$20.1{\pm}0.68$
Cobalt	7.6± .47	BDL	8.8±1.5	BDL	3.6 ± 0.99
Lead	14.6 ± 0.89	0.3 ± 0.09	25.3±.74	9.1±0.19	21± 1.56
Copper	BDL	BDL	9.1	BDL	BDL
Chromium	4 ± 0.45	3±1.78	BDL	2.5 ± 0.09	4.9 ± 0.56
Nickel	3.5±0.91	3.3±1.41	3.4 ± 0.25	3± 0.49	3.4±0.23
Cadmium	BDL	BDL	BDL	BDL	BDL

Table 3. Mineral Composition (mg/Kg) of Solanum Surattense

Note: (BDL) Below Detection Limit; $\pm =$ Standard Deviation