

## ETHNOBOTANICAL SURVEY OF *SMILAX ZEYLANICA* L. PLANT AND ANALYSIS FOR ANTIMICROBIAL AND ANTICANCER ACTIVITY

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### ABSTRACT

*Smilax zeylanica* L. is a medicinal climbing shrub; many phytochemicals, such as sarsaparilla, smilagen, diosgenin, etc., have been isolated from this plant. From leaf to root, the plant exhibits several medicinal properties, including anti-diabetic, antimicrobial, anticancer, antioxidant, and anti-inflammatory. The objective of the present study is to survey the ethnobotanical importance of the *Smilax zeylanica* L. plant and evaluate in vitro antimicrobial and anticancer activities. Surveys reveal that the plant is used as medicine, food, fodder, dyes, etc. The methanol extract of leaf, stem and root parts was tested for antimicrobial activity by the Agar-Well Diffusion method on eight microorganisms and showed a good inhibition. Fungi: *Aspergillus flavus*, *Penicillium chrysogenum*, *Helminthosporium gramineum*, *Mucor plumbeus* and Bacteria: *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas syringae* pv. *lisi*, *Basillus*

*subtilis*. The anticancer activity of the plant was checked by the MTT assay method on the Breast cancer cell line (MCF7) and yielded good results. All the samples of plant, root, stem and leaf showed a good inhibition potential on microorganisms' growth and cancer cells proliferation.

**KEYWORDS:** Ethnobotanical, Antimicrobial, MIC, Anticancer, and MTT assay.

## INTRODUCTION

According to the Botanical Survey of India (BSI), India is one of the mega-diverse countries with a complex topography comprising about 7 to 8% of the total global species. The major floral diversity of India is mainly concentrated in the Himalayas, Western Ghats, & Andaman & Nicobar Islands, the four biodiversity hotspots out of thirty-four identified “global biodiversity hotspots”. Before selecting and starting the study on any plant, it is very important to survey its ethnobotanical importance and its preliminary work done (Firdos & Roopa, 2025).



**Figure 1: leaves of *S. zeylanica*.**

About 300-350 species containing the monocotyledon family Smilacaceae include the genus *Smilax* (LakshyaJeet et al., 2023). One species, *Smilax zeylanica* Linn., is an endemic plant to Asian countries like India, Bangladesh, Myanmar, Nepal, Sri Lanka, & Indonesia, distributed in various Indian forests (Anjana & Vijaya, 2022). Commonly known as *Jangli aushbha*, *Chobchini* in Hindi, *Ghotvel* in Marathi, *Ushava*, and *Chopachinee* in Sanskrit, *Kummeritheega*, and *Konda* in Telugu, *Kumarika* in Bengal, *Karivilanti* in Malayalam, *Ayadi*, and *Malaittamarai* in Tamil (V Madhavan et.al. 2010). According to the SMPB (State Medicinal Plants Board) Kerala, the *Smilax zeylanica* L. is under the vulnerable category in conservation status.

*Smilax zeylanica* L. is a dioecious, perennial climbing shrub that was first noted by Linnaeus in his book *Species Plantarum* (1788) later it was placed in the family Liliaceae by some taxonomists mainly Bentham and Hooker Classification system & Engler and Prantl Classification system but, later the Genus excluded by Hutchinson in 1959 from Liliaceae

and formed a separate family Smilacaceae that is accepted by many of the botanists (Anjana & Vijaya, 2022).

The plant phytochemicals have potential as an anticancer drug, since 1940 to 2024, half of all licensed anticancer drugs were produced directly or indirectly from plants (Chandra et. al. 2023). *S. zeylanica* roots and rhizomes revealed the presence of secondary metabolites like glycosides, saponins, tannins, flavonoids, protein and amino acid, and phenolic compounds (Firdos & Roopa, 2025). Quantitative estimation revealed the total tannin content is 4.14% w/w, alkaloids are 0.12% w/w, saponins are 54.4% w/w, and no volatile oils and a good antibacterial activity against Gram-negative and Gram-positive bacteria (Uthaman et.al. 2024).

The *S. zeylanica* is importantly used as a source of Sarsaparilla, a phytoconstituent reported in the root and leaf of the plant, which is a steroidal saponin glycoside like Smilagenin, Sarsapogenin, and Diosgenin (V Madhavan et.al. 2010). The plant is used ethnomedicinally to cure many diseases, such as rheumatism, dysentery, skin-related diseases, etc. (Sabari et. al. 2018). Many research studies provide scientific reasons to use *Smilax* z. Traditionally, in asian countries, as the number of bioactive chemicals is discovered from plant parts, smilagenin, diosgenin, alkaloids, hydroxytyrosol, etc (Anjana & Vijaya, 2022).



**Figure 2: leaves of *S. zeylanica*.**

In this present study, *S. zeylanica* is analysed for the antimicrobial activity, as the microorganisms that are very dominant throughout the world are responsible for causing various infections and diseases that lead to death in severe conditions, like Invasive

Aspergillosis (IA) by a virulent fungus *Aspergillus flavus* (S Rudramurthy et. al. 2019). Fungal infections are one of the toughest diseases to cure, and they affect all humans, animals, plants, etc. No doubt, a variety of fungi are beneficial at an industrial level and for the economy of the world. Despite a variety of fungi known to cause serious health issues that lead to malfunctioning of metabolism (J Kohler et. al. 2015).

According to the National Cancer Registry Programme in India, the estimated number of cancer incidents for the year 2022 is 14,61,427 (crude rate: 100.4 per 100,000). Among children (0-14 yrs.), it is 29.2% in boys and 24.2% in girls. Lung and Breast cancer are the leading sites of cancer in males and females. And it is predicted that the incidence can be increased by 12.8 per cent in 2025 compared to 2020 (K Sathishkumar et.al. 2023).

Researchers working world wide to explore the medicinal potential of plants by discovering herbal chemical compounds and using them in medicine. By extracting bioactive compounds from plant parts and their analysis, we can define their medicinal potential to treat diseases like cancer, diabetes, inflammation, infections, etc (G Kumar et. al. 2019).

This study will provide an ethnobotanical survey, antimicrobial and anticancer potential of *Smilax zeylanica* L. plant parts. Selected microorganisms are tested for antimicrobial activity by the Agar Well Diffusion method, and anticancer activity is checked on the Breast cancer cell line (MCF7) by the MTT assay method.

## MATERIALS AND METHODS

### Collection of Plant Material

Figure 3 shows, the plant parts, Leaf, stem, and roots are collected during the rainy season in July in bulk quantities from the Kudal, Sindhudurga Dist. Maharashtra. After the collection of plant material is cleaned and washed with water, and kept for drying under shade on the terrace and in rooms, it requires nearly 25 days for complete drying.



**Figure 3: Collection of *S. zeylanica* plant parts.**

The dried material was brought into the laboratory for further processing. Root and stem are first ground in a mortar pestle, and all materials are then finely ground with the help of an electric mixer.

### External Morphology of a Plant

*Smilax zeylanica* L. belongs to the monocotyledon family Smilacaceae. It is a medicinal climbing shrub with a woody stem and is wild in its habitat. The stem is smooth, woody, greenish, angular, and has small prickles for defence. Stipules are modified into tendrils that are long, narrow and pale green. Root is a Rhizome and reddish. Leaf is simple, alternate, 5-20 × 3-10 cm, green, oval to oblong, coriaceous in texture, petiole approximately. 1 cm, entire margin, reticulate venation 5 ribbed from the base (a typical character of the plant as it lies in monocotyledon but has reticulate venation), shortly Emarginate to cuspidate leaf apex, base rounded. Flowers in clusters, umbel inflorescence from the axil of leaves, unisexual, stalked, bract oblong, pedicellate, actinomorphic, greenish-white, hypogynous, tepals 6, male flower; stamens 6 free, female flower; ovary superior, syncarpous, 3 celled, ovule 1 to 2 per cell, style 3, stigma 3. Fruit is a berry, green and turns red on ripening, with 1-2 seeds.

### Ethnobotanical Survey

- As a food

Through the centuries in the Asian countries, young twigs of *Smilax z.* used as a source of food and consumed by making its sabji. In Sindhudurga dist. In Maharashtra, the young leaves and twigs are cooked and used as food, and believed that it has good nutritional value.

- As a fodder

Cattle and herbivorous animals love to consume the leaves of the plant.



- As a dye

The fruit obtained a reddish colour, used as a dye for several purposes, but very rarely in local crafts and traditional textile dyeing.

- As a medicine

It is an important herbal drug widely used in Ayurveda. It is among the red list and in India's top 20 highly traded medicinal plants (Jyothi et.al. 2012). Studies showed that the plant has medicinal properties for Syphilis, Rheumatism, Skin diseases, Gout (Jyothi et.al. 2012), anti-Alzheimer's activity (Sabari et. al. 2018).

The plant is used worldwide as a medicinal supplement to cure various diseases, either singly or in combination with other herbs. All parts, stem, root, leaf, fruits, twigs, etc, are used traditionally to cure diseases in certain formulations like paste, decoction or powder.

The decoction of the leaf and root is consumed in the morning as a natural blood purifier, for urinary tract infection, to kill harmful bacteria, to treat fungal infection and to detox the body internally. The fresh leaves paste is used to apply to the swollen and inflamed area. Besides the leaf and root, the stem also exhibits medicinal potential as it is hard and is used as a toothbrush to cure and clean the teeth. Fruits are consumed to improve digestion.

Several literatures are reviewed, the Rhizome used in Venereal diseases in Tiruchirappalli district, Tamil Nadu, Stems are used as a toothbrush in Rewa district, Madhya Pradesh and Makawanpur district, Central Nepal, to cure toothache. In Hazaribagh, Jharkhand, the fruit pulp is mixed with lime and consumed as a remedy for dysentery. In Kadapa district, Andhra Pradesh, Root paste is applied externally for body swellings (Prashith et. al. 2018). In Sri Lanka, the plant parts, mainly the root and leaf used to cure a variety of cancer diseases (Anchala et.al. 2019). It has been utilised as a tonic in folk medicine against rheumatism, used for relief in burning sensation in the feet caused by vesicular watery eruptions and in antifertility, analgesic, etc. (Uthaman et.al. 2024)

### **Methanol extraction**

The collected three parts of the plant, leaf, stem and root, were dried and completely ground into a fine powdered material in an electric mixture jar and stored in an air-tight container for further use. The methanol extract is prepared with the help of a Soxhlet extraction instrument. About 25 g of powdered material is packed in a filter paper and poured into a thimble. It requires about 250 mL of methanol solvent. About 10 to 15 cycles of extraction are done for

complete extraction until the solvent becomes colourless. During leaf extraction, it takes 15 cycles; for root and stem, 10 to 13 cycles are sufficient. A rotatory evaporator is used for the evaporation of solvent after extraction. Within this extracts a 100  $\mu$ g extract is diluted in 1 ml of DMSO solvent for Antimicrobial activity.

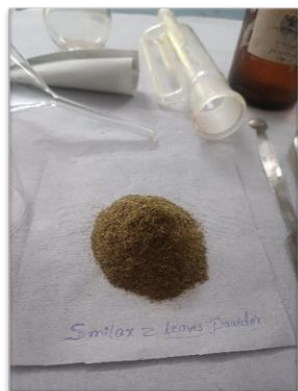


Figure 4 (A)



Figure 4 (B)



Figure 4 (C)

**Figure 4: Extraction of *S. zeylanica* Leaf; (A) Dried Leaf powder for extraction. (B) Extraction in Soxhlet extractor instrument. (C) Collected solid extract in bottles.**

### Antimicrobial analysis

The plant extracts are analysed for antibacterial and antifungal activity.

### Determination of Minimum Inhibitory Concentration (MIC)

For the Determination of Minimum Inhibitory Concentration (MIC), an Agar Well Diffusion method is used. It is a widely used method to assess the antimicrobial activity of plant extracts (Lokman et.al. 2022). In this method, a microorganism suspension is spread on agar medium and a well of selected size is created on the solidified medium plate with the help of a cork borer. A given plant extract in the minimum quantity is poured into the well with a standard chemical (antifungal & antibacterial) solution beside the test extract to observe the inhibition zones, and compare between plant extracts and standard chemicals.

### Test organisms

Fungi: *Aspergillus flavus* 873, *Penicillium chrysogenum* 1996, *Helminthosporium gramineum* 1070, *Mucor plumbeus* 984.

Bacteria: *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas syringae* pv. *lisi*, *Basillus subtilis*.

### Antifungal analysis

The antifungal activity of the methanol extract was carried out using an agar well diffusion method on Martin Rose Bengal Agar media. About 20 ml of media was poured into each sterile petri plate and allowed to cool and solidify. 250  $\mu$ l of the test fungal suspension was inoculated on the media plates and spread evenly with the help of a glass spreader, and four equidistant holes of 8 mm diameter were made with the help of a cork borer to pour liquid. 100  $\mu$ l and 150  $\mu$ l (100  $\mu$ g/ml or 0.1 mg/ml) of sample extracts were poured into two holes. 100  $\mu$ l solution of a standard antifungal drug (150 mg Fluconazol+10 ml DMSO) was used as a standard or positive control, and 100  $\mu$ l DMSO (Dimethyl sulfoxide) solvent as a negative control. After the inoculation and pouring, the plates were kept at 37°C for 3 days to observe the inhibition zone. The experiment was carried out in triplicate.

### Antibacterial analysis

The antibacterial activity of the extracts was tested using an agar well diffusion method on Nutrient Agar medium. About 20 ml of media was poured into each sterile petri dish and allowed to cool and solidify. 250  $\mu$ l of the test bacterial suspension was inoculated onto the media plates and spread evenly with a glass spreader. Four equidistant holes of 8 mm diameter were made with a cork borer to pour the liquid. 50  $\mu$ l and 100  $\mu$ l (100  $\mu$ g/ml or 0.1 mg/ml) of sample extracts were added into two holes. 100  $\mu$ l of a standard antibiotic drug, Cefpodoxime, was used as a positive control, and 100  $\mu$ l of DMSO (Dimethyl sulfoxide) as a negative control. After inoculation and pouring, the plates were incubated at 37°C for 24 hours to observe the inhibition zone. The experiment was conducted in triplicate.

### Anticancer analysis

#### MTT assay

This assay is the most common and reliable method for detecting the anticancer potential of drugs, whether plant-based or synthetic (Shilpee et al., 2015). By this assay, we evaluated the potential of *S zeylanica* plant extracts to inhibit cancer cell proliferation.

MCF7 (Breast cancer) cells at  $1 \times 10^4$  cells/ml were incubated in culture medium for 24 hours at 37°C and 5% CO<sub>2</sub>. 100  $\mu$ L cells were seeded at  $10^4$  cells/well in 100  $\mu$ L culture medium and 20, 40, 60, 80, 100  $\mu$ g/ml of plant extracts into microplates, respectively (tissue culture grade, 96 wells). Control wells were incubated with DMSO (0.2% in PBS) and the cell line. Controls were maintained to determine the control cell survival and the percentage of live cells after culture. All samples were incubated in triplicate. Cell cultures were incubated for



24 h at 37°C and 5% CO<sub>2</sub> in a CO<sub>2</sub> incubator. After incubation, the medium was completely removed, and 20µl of MTT reagent (5mg/ml PBS) was added and incubated for 4 hours at 37°C in a CO<sub>2</sub> incubator. Observed under the microscope, the wells for formazan crystal formation. The yellowish MTT was reduced to dark coloured formazan by viable cells only. Triplicate samples were analysed by measuring the absorbance of each sample with a microplate reader at a wavelength of 550 nm.

## RESULT AND DISCUSSION

### Antifungal activity

From the above study, it is evaluated that the plant has a great antifungal activity, as it showed inhibition on fungal growth to a great extent. As Figure 5 indicates, two concentrations of plant extracts are taken, 100µl and 150µl, and it is seen that as the concentration increases inhibition zone increases and at 150µl extract, it showed higher inhibition. Table 1 indicates, the SL showed a 20 mm inhibition zone for an *A. flavus*, 23 mm for *P. chrysogenum*, 10 mm for *H. gramineum* and 17 mm for *M. plumbeus*. The SS showed 23 mm for *A. flavus*, 15 mm for *P. chrysogenum*, 12 mm for *H. gramineum* and 18 mm for *M. plumbeus*. The SR showed 21 mm for *A. flavus*, 18 mm for *P. chrysogenum*, 12 mm for *H. gramineum*, and 14 mm for *M. plumbeus* at the highest concentration of 150µl. Compared to these, the standard drug showed high inhibition with an average of 20 mm. A negative control, DMSO, shows negligible inhibition.



Figure 5 (A)

Figure 5 (B)

Figure 5 (C)

**Figure 5:** Some plates of Antifungal activity on Martin Rose Bengal Agar media by Agar Well Diffusion Method showing four inhibition zones of DMSO, Control (Standard antifungal drug), 100µl and 150µl plant extract. (A) SR with *H. gramineum* (B) SS with *A. flavus* (C) SR *A. flavus*.

### Antibacterial activity

From the above study, it is evaluated that the plant has a great antibacterial activity as it showed inhibition on bacterial growth to a great extent. Figure 6 shows, two concentrations are taken, 50 $\mu$ l and 100 $\mu$ l, and it is seen that as the concentration increases inhibition zone increases and at 100 $\mu$ l extract, it shows higher inhibition. Table 2 and Table 3 indicate, SL showed 18 mm for *E. coli*, 17 mm for *S. aureus*, 24 mm for *P. pisi* and 15 mm for *B. subtilis*. SS showed 13 mm *E. coli*, 14 mm *S. aureus*, negative for *P. pisi* and *B. subtilis*. SR showed 20 mm for *E. coli*, 24 mm for *S. aureus*, 17 mm for *P. pisi* and 13 mm for *B. subtilis* at the highest concentration of 100 $\mu$ l. Compared to these, the standard drug showed high inhibition with an average of 20 mm. A negative control, DMSO, shows negligible inhibition.



Figure 6 (A)

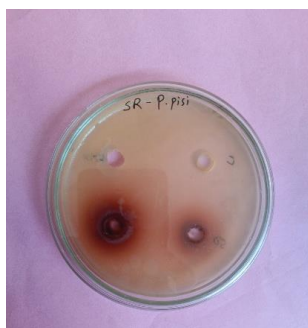


Figure 6 (B)

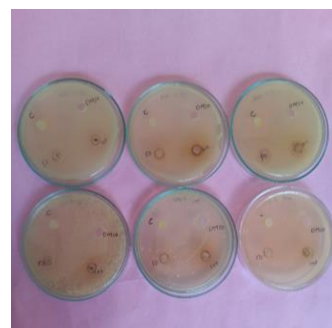


Figure 6 (C)

**Figure 6:** Some plates of Antibacterial activity on Nutrient Agar Medium by Agar Well Diffusion Method showing four inhibition zones of DMSO, Control (Standard antibacterial drug), 50 $\mu$ l and 100 $\mu$ l plant extract. (A) SR with *B. subtilis* (B) SR with *P. pisi* (C) inverted plates showing four zones of; DMSO, Control, 50 $\mu$ l and 100 $\mu$ l.

### Anticancer activity

Based on % of inhibition, it is confirmed that all the extracts showed good anticancer activity. Table 4 and Figure 9, graphical representation indicates that, among the five concentrations, 20, 40, 60, 80, 100  $\mu$ g/ml, the extracts showed the highest inhibition potential at 100  $\mu$ g/ml, i.e. SL showed 63.42 % of inhibition, SS showed 63.49 % and SR showed 64.79 %. The assay is performed in triplicate, and three Optical Densities are taken. As the concentration increases, OD and % of viable cells decrease. The potential of plant extracts is compared with a standard anticancer drug, 5-Fluorouracil, which showed inhibition, more than plant extracts, up to 83.96 %. Figure 10, a pie diagram showing a general comparison between the inhibition potential of samples.

**Table 1: Antifungal Activity of three sample extracts of *S. zeylanica* in two concentrations of 100µl and 150µl.**

Fungi	Sample	Control	DMSO	Zone of inhibition in mm*	
				100µl (in mm)	150µl (in mm)
1) <i>Aspergillus flavus</i>	SL	18 ± 2	10 ± 1	18 ± 0.5	20 ± 2
	SS	16 ± 1	11 ± 1	22 ± 0.5	23 ± 1
	SR	18 ± 1	9 ± 1	18 ± 1	21 ± 1
2) <i>Penicillium chrysogenum</i>	SL	20 ± 0.5	11 ± 1	22 ± 0.5	23 ± 0.5
	SS	18 ± 1	10 ± 1	13 ± 2	15 ± 1
	SR	20 ± 0.5	11 ± 1	15 ± 2	18 ± 1
3) <i>Helminthosporium gramineum</i>	SL	26 ± 1	-	9 ± 0.5	10 ± 1
	SS	25 ± 0.5	-	11 ± 1	12 ± 1
	SR	25 ± 0.5	-	9 ± 0.5	12 ± 0.5
4) <i>Mucor plumbeus</i>	SL	20 ± 0.5	-	15 ± 0.5	17 ± 0.5
	SS	18 ± 0.5	10 ± 1	15 ± 0.5	18 ± 0.5
	SR	16 ± 0.5	-	12 ± 1	14 ± 1

Each value is presented as the mean ± of the antifungal activity of *S. zeylanica* methanol extract

\* Including the diameter of the hole (8mm)

SL (*Smilax* Leaf), SS (*Smilax* Stem) and SR (*Smilax* Root)

**Table 2: Antibacterial Activity of three sample extracts of *S. zeylanica* in two concentrations of 50µl and 100µl for *E. coli* and *S. aureus*.**

Bacteria	Sample	Control	DMSO	Zone of inhibition in mm*	
				50µl (in mm)	100µl (in mm)
1) <i>Escherichia coli</i>	SL	18 ± 2	-	15 ± 1	18 ± 0.5
	SS	18 ± 2	-	12 ± 1	13 ± 1
	SR	18 ± 1	-	17 ± 1	20 ± 1
2) <i>Staphylococcus aureus</i>	SL	18 ± 1	10 ± 0.5	14 ± 0.5	17 ± 1
	SS	18 ± 1	10 ± 0.5	12 ± 0.5	14 ± 1
	SR	20 ± 0.5	10 ± 0.5	18 ± 1	24 ± 0.5

Each value is presented as the mean ± of the antifungal activity of *S. zeylanica* methanol extract

\* Including the diameter of the hole (8mm)

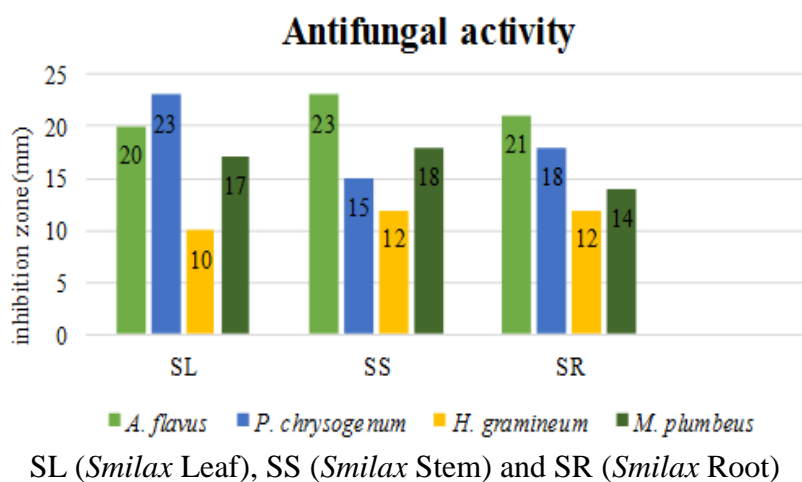
SL (*Smilax* Leaf), SS (*Smilax* Stem) and SR (*Smilax* Root)

**Table 3: Antibacterial Activity of three sample extracts of *S. zeylanica* in two concentrations of 50µl and 100µl for *P. pisi* and *B. subtilis*.**

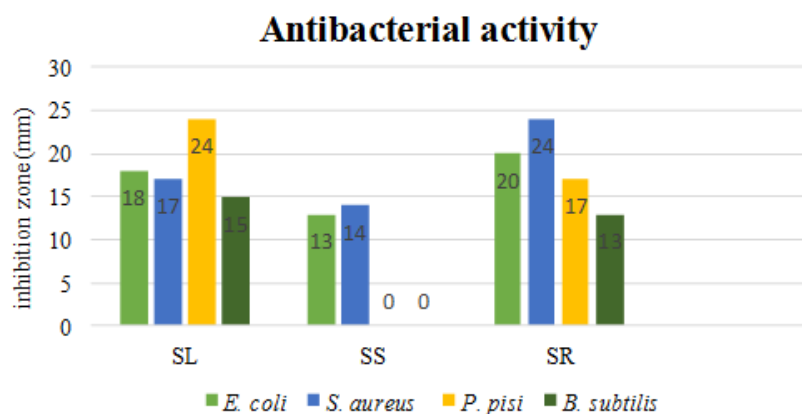
Bacteria	Sample	Control	DMSO	50µl (in mm)	100µl (in mm)
3) <i>Pseudomonas syringae</i> pv. <i>pisi</i>	SL	20 ± 1	-	10 ± 0.5	24 ± 0.5
	SS	24 ± 0.5	-	-	-
	SR	24 ± 0.5	-	14 ± 0.5	17 ± 0.5
4) <i>Basillus subtilis</i>	SL	20 ± 1	-	10 ± 1	15 ± 0.5
	SS	24 ± 0.5	-	-	-
	SR	18 ± 0.5	-	10 ± 0.5	13 ± 0.5

Each value is presented as the mean ± of the antifungal activity of *S. zeylanica* methanol extract

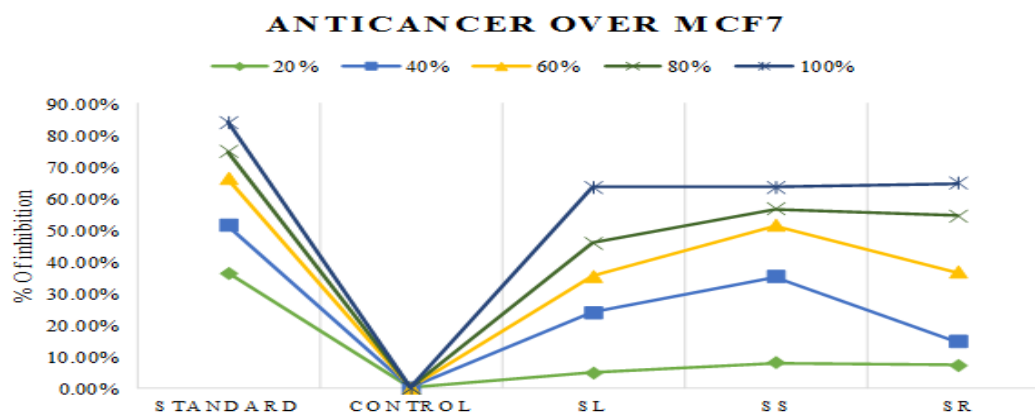
\* Including the diameter of the hole (8mm)



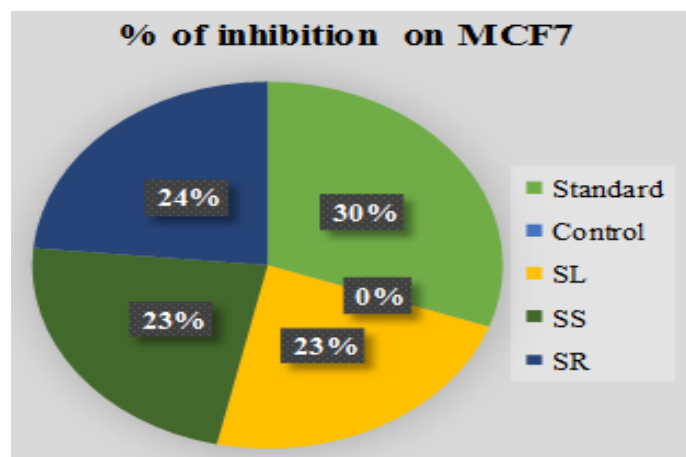
**Figure 7: graphical representation of Antifungal activity, comparison of inhibition zones in between Plant extracts of *S. zeylanica* leaf, stem and root (SL, SS and SR) for four fungi; *A. flavus*, *P. chrysogenum*, *H. gramineum* and *M. plumbeus*.**



**Figure 8: graphical representation of Antibacterial activity, comparison of inhibition zones in between Plant extracts of *S. zeylanica* leaf, stem and root (SL, SS and SR) for four Bacteria; *E. coli*, *S. aureus*, *P. pisi* and *B. subtilis*.**



**Figure 9:** The graphical representation of sample versus % of inhibition on MCF7 cell line. Five concentrations of extracts denoted by five different colors and shapes on line. The negative control DMSO showed a 0% inhibition i.e. complete reverse effect of plant extracts and standard drug.



**Figure 10:** Comparison of inhibition % of sample extracts.

**Table 4:** Anticancer activity of three sample extracts of *S. zeylanica* on MCF7.

Sr.	Sample code	Conc. (µg/ml)	OD			Mean	% of Inhibition	% of Viability	IC50 (µg/ml)
1.	Control		1.534			-	-	-	-
2.	Standard (5-Fluorouracil)	20	0.977	0.975	0.979	0.977	36.31%	63.69%	38.12
		40	0.746	0.744	0.748	0.746	51.36%	48.64%	
		60	0.519	0.521	0.518	0.519	66.16%	33.84%	
		80	0.389	0.389	0.387	0.388	74.70%	25.3%	
		100	0.246	0.248	0.244	0.246	83.96%	16.04%	
3.	SL	20	1.463	1.465	1.461	1.463	4.62%	95.38%	84.26
		40	1.168	1.170	1.166	1.168	23.85%	76.15%	
		60	0.991	0.993	0.990	0.991	35.39%	64.61%	
		80	0.830	0.828	0.833	0.830	45.89%	51.11%	
		100	0.562	0.560	0.561	0.561	63.42%	36.58%	
4.	SS	20	1.416	1.414	1.418	1.416	7.69%	92.31%	58.26



5.		40	0.996	0.998	1.010	0.998	34.94%	65.06%	
		60	0.748	0.744	0.746	0.746	51.36%	48.64%	
		80	0.665	0.667	0.663	0.665	56.64%	43.36%	
		100	0.562	0.560	0.558	0.560	63.49%	36.51%	
	SR	20	1.428	1.426	1.426	1.426	7.04%	92.96%	72.35
		40	1.312	1.314	1.310	1.312	14.47%	85.53%	
		60	0.975	0.973	0.977	0.975	36.44%	63.56%	
		80	0.698	0.700	0.696	0.698	54.49%	45.51%	
		100	0.540	0.542	0.538	0.540	64.79%	35.21%	

Anticancer activity of *S. zeylanica* for three parts, SL, SS, and SR, in comparison to the standard anticancer drug 5-Fluorouracil on the Breast cancer cell line MCF7. The % of inhibition is inverse to the % of viable cells. As the concentration of extracts increases % of inhibition.

## DISCUSSION

The ethnobotanical survey of *Smilax zeylanica* L. reveals that the plant is used as food, fodder and for dye, and it is fully medicinal from root to leaf and fruit. As it is edible in its locality, People consume its cooked young twigs; it is nutritious. And in their traditional way practice the plant in their daily lives to cure diseases. The unlimited medicinal properties of a plant are studied through a survey. The preliminary phytochemical analysis reveals the presence of phenol, flavonoid, glycoside, tannin, saponin and protein (Firdos & Roopa, 2025). In a traditional way, the paste of root and leaf is used to control various health issues like swelling and infections. Decoction is consumed as a blood purifier, etc. The Antifungal activity of the 100µl and 150µl methanol extract is analysed at concentrations of 100 µg/ml, compared to the standard antifungal drug Fluconazole (100µl) on four different fungi: *Aspergillus flavus*, *Penicillium chrysogenum*, *Helminthosporium gramineum* and *Mucor plumbeus*. The standard drug for all the fungi showed a high growth inhibition, with an average of 20 mm. All the extracts of SL, SS and SR showed good inhibition on *A. flavus* and *P. chrysogenum* compared to *Helminthosporium gramineum* and *Mucor plumbeus*. But despite this standard drug showed maximum inhibition on *H. gramineum* and *M. plumbeus* ( $21 \pm 0.6$ ) compared to *A. flavus* and *P. chrysogenum* ( $18 \pm 0.3$ ). Among the three extracts, the SS showed higher inhibition on *A. flavus* (23 mm), *H. gramineum* (12 mm) and *M. plumbeus* (18 mm) at 150µl, and the SL showed maximum effect on *P. chrysogenum* (23 mm) at 150µl. With this study, it is seen that *H. gramineum* is a very devastating fungus and is hard to control its growth. The negative control, 100 µl DMSO, showed less inhibition on some fungi (*A. flavus* and *P. chrysogenum*) and none on others (*H. gramineum*). The

antibacterial activity is checked on four different bacteria, two Gram-negative *Escherichia coli* and *Pseudomonas syringae* pv. *lisi* and two Gram-positive *Staphylococcus aureus* and *Bacillus subtilis*. Two concentrations of plant extracts were taken, 50µl and 100µl, respectively. As always, at a higher concentration extract shows maximum effect on bacterial growth. The SR showed its maximum effect on *E. coli* and *S. aureus*, while SL showed maximum effect on *P. lisi*. The SS gave a negative result on the inhibition of *P. lisi* and *B. subtilis*. The standard antibacterial drug Cefpodoxime showed a higher inhibition with an average 20 mm inhibition zone. Negative control, DMSO showed a little inhibition on *S. aureus* and NE in all other bacteria.

By the MTT assay method, the anticancer activity of *S. zeylanica* is analysed on the Breast cancer cell line (MCF7). Breast cancer is the leading cause of death worldwide in males and females (K Sathishkumar et.al. 2023). The plant extracts on their way to kill cancer cells, induce apoptosis by condensation of chromatin and lowering DNA content and increasing G<sub>1</sub> phase cell population, and the caspase 3 activation is also recorded after plant extract treatment, which is the key regulator in apoptosis (Greenwell and Rahman, 2015). All three extracts of *S. zeylanica*, SL, SS and SR, showed positive results of good inhibition percentage on cancer cells. The standard anticancer drug 5-Fluorouracil was taken as a positive control, and DMSO (0.2% in PBS) as a negative control. Five concentrations of extracts are taken: 20, 40, 60, 80, and 100 µg/ml. All samples were incubated in triplicate. Three Optical Densities were taken for each sample extract. As the concentration increases, OD decreases, which indicates the potential of the extracts to kill the cancer cells. Minimum OD was observed for SR, i.e. 0.540, 0.560 for SS and 0.561 for SL at 100 µg/ml. It is observed that, as the concentration of extract increases, the percentage of inhibition also increases, and the percentage of viable cells decreases. Among the three samples, a maximum inhibition was observed in SR at a higher concentration, about 64.79%. 63.49% in SS and 63.42% in SL, respectively. The percentage of viable cells is inversely proportional to the percentage of inhibited cells. The IC<sub>50</sub> value was calculated, 84.26 µg/ml for SL, 58.26 µg/ml for SS and 72.35 µg/ml for SR.

Phytochemicals have a powerful anticancer potential as they have complementary and overlapping pathways for reduction of carcinogenesis by altering free radicals (Chandra et. al. 2023). In this study, it is observed that as the concentration increases, the activity of plant extracts increases, with the highest concentration sample giving maximum activity. All the

extracts showed positive results for antimicrobial and anticancer activity, indicating that *S. zeylanica* has the potential to treat diseases and needs advanced studies.

## CONCLUSION

Based on the above study on the *Smilax zeylanica* L. plant, the ethnobotanical survey concluded that it is truly medicinal from root to leaf and fruit. Its root, stem and leaf parts were collected and certifiably authenticated, dried under shade and made into a fine powder, extracted in methanol and analysed for antimicrobial activity by the Agar Well Diffusion method and anticancer activity by the MTT assay method on breast cancer cell line (MCF7). The extracts give positive results for both activities in a good percentage and indicate that it has antimicrobial as well as anticancer potential properties.

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