Journal of Medicinal Plants Studies
www.PlantsJournal.com

ISSN (E): 2320-3862
ISSN (P): 2394-0530
Impact Factor (RJIF): 5.94
www.plantsjournal.com
JMPS 2025; 13(6): 52-59

Received: 22-08-2025
Accepted: 26-09-2025

Maitri M Thakor
Ph.D. Scholar, Department of Biology, Municipal Arts and Urban bank Science College, Mehsasna, Gujarat, India.(Affiliated with Hemchandracharya North Gujarat University, Patan, Gujarat, India

Ashok H Patel
Associate professor, Department of Biology, Municipal Arts and Urban bank Science College, Mehsasna, Gujarat, India. (Affiliated with Hemchandracharya North Gujarat University, Patan, Gujarat, India

Corresponding Author:
Maitri M Thakor
Ph.D. Scholar, Department of Biology, Municipal Arts and Urban bank Science College, Mehsasna, Gujarat, India.(Affiliated with Hemchandracharya North Gujarat University, Patan, Gujarat, India

Phytochemical analysis and disc diffusion-based antibacterial evaluation of leaf and stem extracts of *Ocimum sanctum* L.

Maitri M Thakor and Ashok H Patel

DOI: https://www.doi.org/10.22271/plants.2025.v13.i6a.1976

Abstract
Herbal plants have long been used to treat various maladies. This study investigated the phytochemical constituents of *Ocimum sanctum* L. (stem and leaves) collected from the city area of Mehsana, Gujarat and subjected to extraction by maceration method with two different solvents *viz.* methanol and aqueous. Phytochemical estimation revealed that the methanolic extracts of *O. sanctum* L. yielded highest extractive value than aqueous and contained alkaloids, phenols, flavonoids, tannin etc. The methanolic extract of *O. sanctum* L. (leaves) yielded highest flavonoid content of 19.55±0.58 mg/g with quercetin. Antibacterial activity studied by disc diffusion method against *S. aureus* and *E. coli* bacteria, showing significant potential. These findings strongly supports its widespread use in folk medicine for various chronic disease. The results indicate that medicinally bioactive compounds possesses promising therapeutically value and could be further explore for formulation development and its application in foods, nutrition, pharmaceutical industries and human health.

Keywords: Antibacterial, disc-diffusion methods, pharmaceutical, phytochemical

Introduction
Human health is the main concern because it affects how well people function and has an impact on the economic status of individuals, families, societies, and nations. Plants have provided humans with medicinal agents since the beginning of time. In fact, natural products were once the source of all drugs. *Ocimum sanctum* L. is an aromatic plant in the Lamiaceae family. Ocimum spp. is a grassy, annual medicinal plant with oval leaves that have a sharp tip. It is native to Iran, Afghanistan, and India. This plant belongs to the mint family, Lamiaceae, and is commonly called “holy basil” in English and “Tulasi” in Hindi and Sanskrit. There are various types of tulasi. Some have purple or dark leaves, known as “krishna tulasi” (*Ocimum teniflorum*L.). Others have green leaves, known as “rama tulasi” (*Ocimum sanctum* L.). Another type is “vana tulasi” (*Ocimum gratissimum*L.) [1]. Medicinal plants contain many secondary metabolites. These compounds can be valuable sources of drugs and essential oils that have therapeutic benefits [2]. Secondary metabolites show a wide range of chemical structures and types, and their specific functions are mostly unclear. They have significant uses in human medicine, veterinary practices, agriculture, scientific research, and various other areas [3].

Bioactive compounds such as tannins, alkaloids, carbohydrates, terpenoids, steroids, and flavonoids, which energize direct physiological effects on the human body, are synthesized by primary or secondary metabolic processes in medicinal plants [4-5]. Flavonoids, a subclass of polyphenols renowned for their ability to scavenge free radicals, have been shown to inhibit hydrolytic and oxidative enzymes, lower blood glucose and lipid levels, demonstrate anti-inflammatory effects, and bolster immunity in humnan. Because of these properties, flavonoids have gained interest as a potential solution to several health problems, particularly those linked to oxidative stress [6]. This plant produces several types of volatile oils, such as terpenes, phenol, and aldehydes. It is also believed to contain tannins, alkaloids, saponins, and glycosides [7].

This plant is known for its numerous medicinal properties, including antioxidant, antidiabetic, anti-inflammatory, anticancer, antinociceptive, antifertility, anthelmintic, cardioprotective,

and antimicrobial effects, with various Ocimum species utilized in the treatment of ailments such as diabetes, dysentery, hemorrhoids, gastrointestinal issues, tuberculosis, eye and ear issues, respiratory infections, and other conditions affecting the central nervous system [8-10]. The plant, enriched with natural products, exhibits a wide range of biological activities, including cardioprotective, anti-diabetic, antimicrobial, hepatoprotective, antifertility, anti-fungal, anticancer properties, as well as effects on stomachaches, headaches, common colds, inflammation, analgesics, antiemesis, antipyresis, and stress reduction [11-13]. Recently, Bacterial infections caused by antibiotic-resistant strains continue to pose a significant public health issue. To tackle this problem, we need to exploration and development of new, highly effective antimicrobial agents [14].

The growing presence of multidrug-resistant bacterial strains and the emergence of those with reduced antibiotic susceptibility underscore the urgent need for innovative infection-fighting strategies, particularly as resistance to newly developed antibiotics continues to escalate despite advancements in the pharmaceutical industry over the past three decades.The Tulsi plant contains main chemical components such as eugenol, carvacrol, methyl eugenol, and caryophyllene, and is recognized for its potential medicinal properties, particularly its stress-reducing effects, attributed to its rich content of essential oils and antioxidants.Therefore the aim of present study was toevaluate phytochemical analysis and the antibacterial activity of extracts from the different parts *Ocimum sanctum* L. plant against two pathogenic bacteria *Staphylococcus aureus* and *Escherichia coli* [15].

Materials and Methods

Sample collection of *O. sanctum* L. plants and Processing

O. sanctum, a perennial herb, was obtained from the Mehsana city area., Gujarat, India. Mehsana is located at the GPS coordinates of 23°35′12.34’’N and 72°22′11.82’’E.This region types vary like in different parts of the city and its outskirts, depending on the level of urbanization and industrial development. The plants were identified and confirmed by the Biology Department, Municipal Arts and Urban Bank Science College, Mehsana.The collected plant materials (Leaves and stem) were initially washed with running tap water then washed with distilled water and, cut into small pieces, air dried, homogenised to fine powder with mechanical grinder and powdered material was stored in sterilized airtight glass container.

Extract Preparation of Plant Material

The plant samples (Leaves and stem) were extracted with maceration extraction method [16]. 1 gm of each sample powder was soaked in conical flasks containing 10 ml of water and methanol as a solvent and put on the shaker for 24 hours. The extracts were filtered through Whattman no.1 filter paper. The supernatants were collected, covered, labelled, stored at 4 °C and used for the screening of various phytochemicals. These extracts were then analysed qualitatively and quantitatively for the presence of different phytochemicals. This extract also used with high concentration for antibacterial activity.

Phytochemical Analysis

Qualitative tests

Qualitative Phytochemical tests were carried out using an aqueous and methanolic extracts to identify various constituents using standard qualitative methods as described previously by Harborne and Baxter [17], Phukan and Shil [18], Sahira Banu and Cathrine [19], Velu and Baskaran [20]. The Components analysed for phytochemicals were alkaloids, phenols, flavonoids, tannins, glycosides, terpenoids, steroids, carbohydrates, amino acids and saponins.

Quantification of Total Phenolic Content [21, 27]

The total phenolic content of the extracts was assessed using the Folin-Ciocalteu reagent, involving a mixture of 0.2 ml of extracts add 3.8 Distilled water (Finalise 4 ml). add 0.5 ml of Folin-Ciocalteu reagent (10% W/V). After 6 minutes incubation add 1 ml of sodium carbonate (2% W/V), and 3 ml of distilled water, After Incubation for 2 hours of reaction at room temperature, the absorbance at 765 nm using UV-visible spectrophotometerand it was measured to determine phenolic content based on gallic acid as a standard.

Quantification of Total Flavonoid Content [22]

Total flavonoid content was estimated using the aluminium chloride method, where 0.1 ml of plant extract was combined with 0.3 ml of distilled water and 0.03 ml of 5% NaNO2, followed by 0.03 ml of 10% $AlCl_3$. After another 5 minutes at 25°C, then treated with 0.2 ml of 1 mM NaOH, diluted to 1 ml with distilled water, when appear its blue colour, measured for absorbance at 510 nmusing UV-visible spectrophotometer.All procedures performed in triplicate and flavonoid contents calculated using a quercetin standard curve.

Quantification of Total Tannin Content [23]

Quantitative analysis of total tannin content in the extract of *O. Sanctum* was conducted using the Folin - Ciocalteu reagent method with some changes. Different concentrations of tannic acid were prepared using various solvents. The reaction mixture contained 0.2 ml of extracts and add 0.8 ml methanol finalise 1 ml., add 0.5 ml of Folin-Ciocalteu reagent mix with 1 ml of sodium carbonate, and 7 ml of pure water (finalise 7 ml with it). After Incubation for half an hours of reaction at room temperature, the absorbance at 765 nm was measured to determine phenolic content based on gallic acid as a standard using UV - visible spectrophotometer.

Quantification of Total Alkaloids Content [24]

The total alkaloids contentwas estimated using standard galanthamine. A total of 69.8 mg of bromocresol green was dissolved in 3 mL of 2 N NaOH and 5 mL of distilled water, then heated and further diluted to 1000 mL with distilled water; subsequently, 0.1 mL of plant extract, 5 mL of bromocresol green, and 5 mL of sodium phosphate buffer (pH 4.7) were combined, extracted with 2 mL of chloroform, and the absorbance of the chloroform layer was measured at 470 nm using UV-Visible Spectrophotometer. Galanthamine was used as a standard, and the content was reported as mg of galanthamine equivalent (GE) per gram of extract.

Antibacterial activity based on disc - diffusion method

The methanolic and aqueous extracts of *O. sanctum* (leaves and stem) were used to study antibacterial activity by the disc-diffusion method in Muller Hinton agar [25]. The cultures used for antibacterial activity with pathogenic isolates were *Escherichia coli* as gram negative and *Staphylococcus aureus*

as gram-positive bacteria from the clinical laboratory. The sterile discs (Himedia, 10 mm) were used in present study. The three different concentration of extracts (75µl, 100µl, 120µl) were poured on the disc using sterile micropipette agar surfaces along with streptomycin and erythromycin as positive control and Distilled water and Methanol as negative control. These plates were incubated for 24 hour at 37 °C. The zone of bacterial inhibition was observed and measured in millimeter.

Results: Qualitative Phytochemical analysis

The results of the ten tests for the qualitative phytochemicals are depicted in Table 1. *Ocimum sanctum* L. (leaves and stem) were observed to reflect the maximum positive result in leaves extracts and stem extracts was observed with equal positive results in methanol extract and water extract. Thus, the majority of the solvents gave positive results and confirmed the presence of primary metabolites in this herbal plant. The results revealed the presence of medicinally active compounds in the studied plant.

Table 1: Detection of phytochemicals from two parts of *O. sanctum* L.

Sr. No.	Phytochemical constituents	Aqueous extracts of Leaves	Methanolic extracts of Leaves	Aqueous extracts of Stems	Methanolic extracts of Stems
1	Alkaloids	+	+	+	+
2	Phenols	+	+	+	-
3	Flavonoids	+	+	+	+
4	Tannin	+	+	+	-
5	Glycosides	+	+	+	+
6	Terpanoids	+	+	+	+
7	Steroids	-	+	+	-
8	Carbohydrates	-	-	-	-
9	Amino acids	+	+	+	-
10	Saponins	+	+	-	-

+ indicates presence and - indicates absence

Quantitative phytochemical analysis

Table 2 shows the proximate phytochemical constituents of *Ocimum sanctum* L.

Table 2: Phytochemical constituents of *Ocimum sanctum* L. (Data±SE, n = 3)

Parts of *Ocimum sanctum* L.	Type of Extract	Total Phenolic Content (mg/g)	Total Flavonoid Content (mg/g)	Total Tannin Content (mg/g)	Total Alkaloid Content (mg/g)
Leaves	Methanolic	178.02±1.32	19.55±0.58	122.04±0.94	39.43±0.56
	aqueous	142.34±2.22	15.69±0.64	99.48±1.64	38.04±0.74
Stem	Methanolic	110.76±0.97	12.43±0.64	66.97±1.73	27.48±0.46
	aqueous	89.42±1.99	8.56±0.58	55.62±2.07	25.41±0.50

Quantification of Total Phytochemical Contents:

In this study, the dried leaf extract of *O. sanctum*L. obtained from the polar solvent, which is aqueous and methanol, contained a high amount of totalphenolic content as compared to an extract of dried stem extracts. Among the two polar extracts of methanolic and aqueous, the methanolic extract has the highest amount of phenolic compounds compared to the aqueous extract. The total phenolic content of methanol extract and aqueous extract was 178.02±1.32mgg-1dry wt. (GAE/g) and142.34±2.22 mgg-1dry wt. of dried leaves, respectively. The lowest totalphenolic content was found in the stemmethanolic extract,which contains only 110.76±0.97mgg-1dry wt. extract than themgg-1dry wt. methanolic extracts.The flavonoid content was recorded maximum amount in 19.55±0.58 mgg-1dry wt. (QE/g) of methanolic leaves extracts followed by aqueous leaves extracts 15.69±0.64mgg-1dry wt. and maximum amount in 12.43±0.64 mgg-1dry wt of stem methanolic extracts followed by stemaqueous extracts 8.56±0.58 mgg-1dry wt. Of *O. sanctum* L.The tannin content was recorded maximum amount in 122.04±0.94 mgg-1dry wt. (TE/g) of methanolic leaves extracts followed by aqueous leaves extracts 99.48±1.64 mgg-1dry wt. and maximum amount in 66.97±1.73 mgg-1dry wt of stem methanolic extracts followed by stem aqueous extracts 55.62±2.07 mgg-1dry wt..The alkaloid content was recorded maximum amount in 39.43±0.56 mgg-1dry wt. (GE/g) of aqueous leaves extracts followed by methanolic leaves extracts 38.04±0.74 mgg-1dry wt. and maximum amount in 27.48±0.46 mgg-1dry wt of methanolic stem extracts followed by aqueous stem extracts 25.41±0.50 smgg-1dry wt.

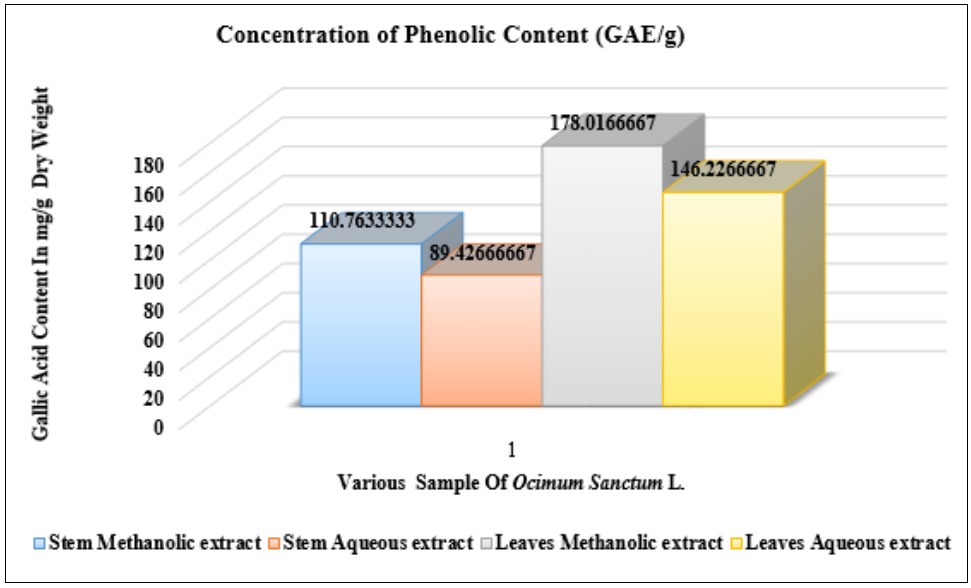

Fig 1: Total phenolic content in *Ocimum sanctum* L.

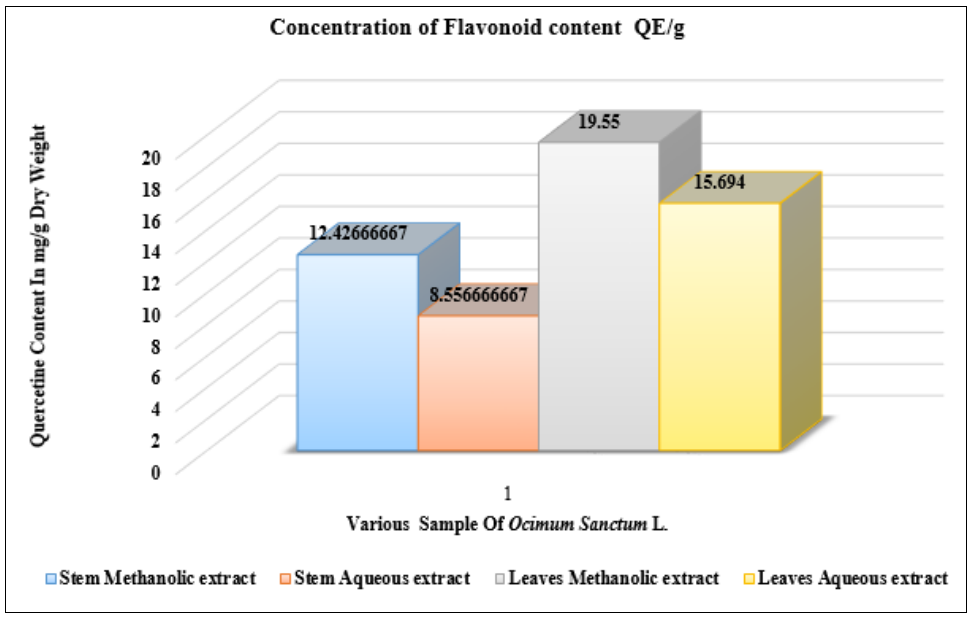

Fig 2: Total flavonoid content in *Ocimum sanctum* L.

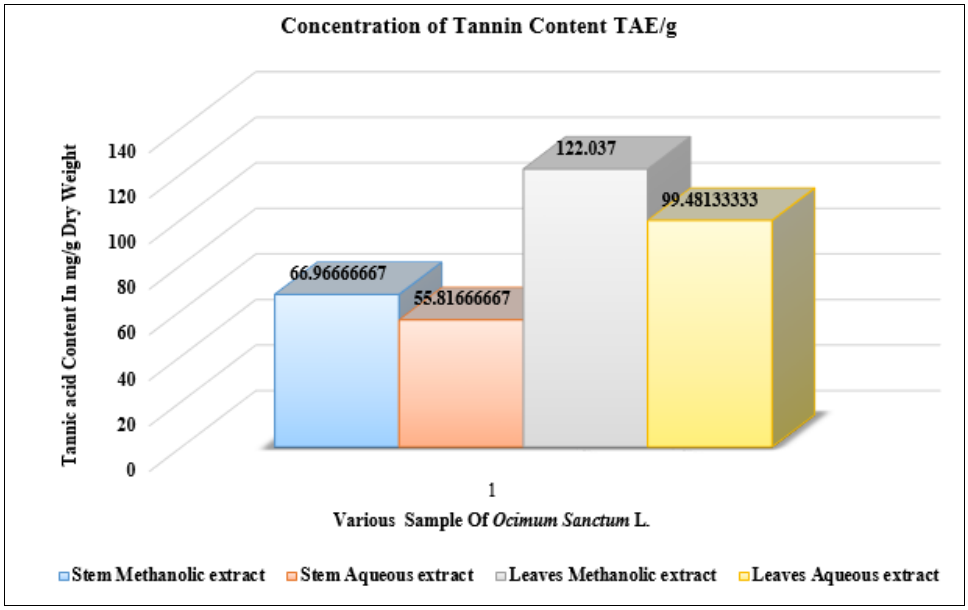

Fig 3: Total tannin content in *Ocimum sanctum* L.

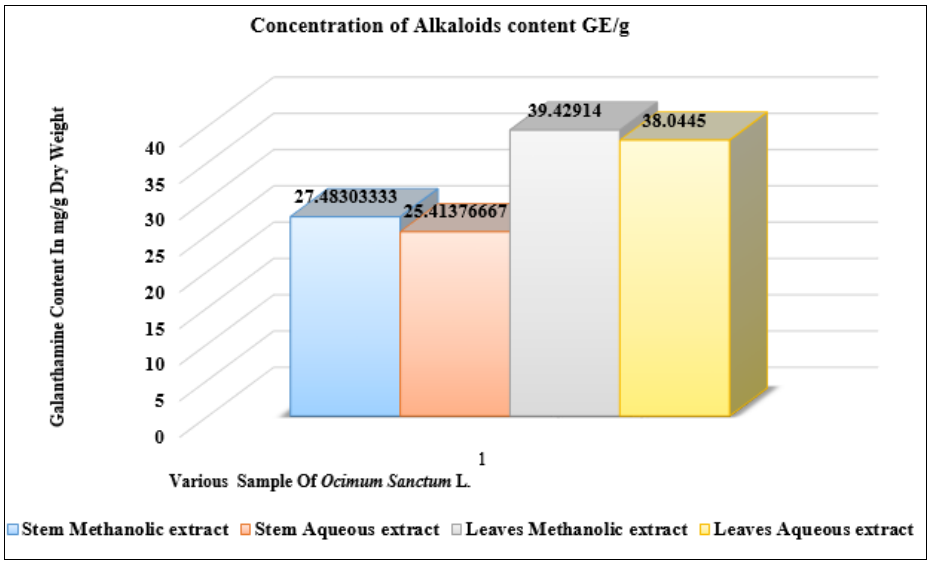

Fig 4: Total Alkaloids content in *Ocimum sanctum* L.

Antibacterial activity

The zone of inhibition of different samples of *O. sanctum* L. and antibiotics against *E. coli* and *S. aureus* were seen after incubation of overnight and measured in millimetre shown in table 3 and figure 5-8. All the extracts showed best result in 120 µl concentration of sample. All methanolic extracts showed the higher antibacterial property against both organisms exclusive of leaves aqueous extract against *S. aureus*. Leaves Methanolic extracts of *O. sanctum* L. was showed zone of Inhibition against both *E. coli* and *S. aureus* 22.17±0.73 mm and 13.4±0.30 mm of sample respectively. Leaves aqueous extracts was found sensitive against both *E. coli* and *S. aureus* 20.62±0.31 mm and 17.99±0.57 mm of sample respectively.Stem Methanolic extracts of *O. sanctum* L. was showed zone of Inhibition against both *E. coli* and *S. aureus* 15.1±0.66 mm and 17.87±0.46 mm of sample respectively. Stem aqueous extracts was found sensitive against both *E. coli* and *S. aureus*14.62±0.31 mm and 17.23±0.34 mm of sample respectively.These discs of antibiotics were showed zone of sensitivity 24.43±0.29 and 24.25±0.45 mm for streptomycin, 27.4±0.30 and 21.5±0.29 mm for erythromycin against *E. coli* and *S. aureus* respectively. The extract of negative control distilled water and Methanol were found resistant against both organisms.

Table 3: Antibacterial activity of various extracts of *Ocimum sanctum* L.(Data±SE, n = 3)

Plant extract	Zone of inhibition (mm)					
	S. aureus			*E. coli*		
	75 µl	**100** µl	**120** µl	**75** µl	**100** µl	**120** µl
Stem Methanoilc extract	13.12±0.33	14.73±0.37	17.87±0.46	12.1±0.41	13.62±0.31	15.1±0.66
Stem Aqueous extract	13.95±0.53	14.94±0.17	17.23±0.34	11.88±0.48	13.28±0.83	14.62±0.31
Leaves Methanoilc extract	11.8±0.51	12.16±0.24	13.4±0.30	18.58±0.29	20.13±0.46	22.17±0.73
Leaves Aqueous extract	13.96±0.49	15.02±0.24	17.99±0.57	17.82±0.43	18.58±0.29	20.62±0.31
Streptomycin	20.69±0.53	22.06±0.37	24.25±0.45	20.97±0.25	22.21±0.41	24.43±0.29
Erythromycin	17.88±0.48	19.23±0.23	21.5±0.29	25.28±0.36	26.46±0.29	27.4±0.30
Methanol	R	R	R	R	R	R
Distilled water	R	R	R	R	R	R

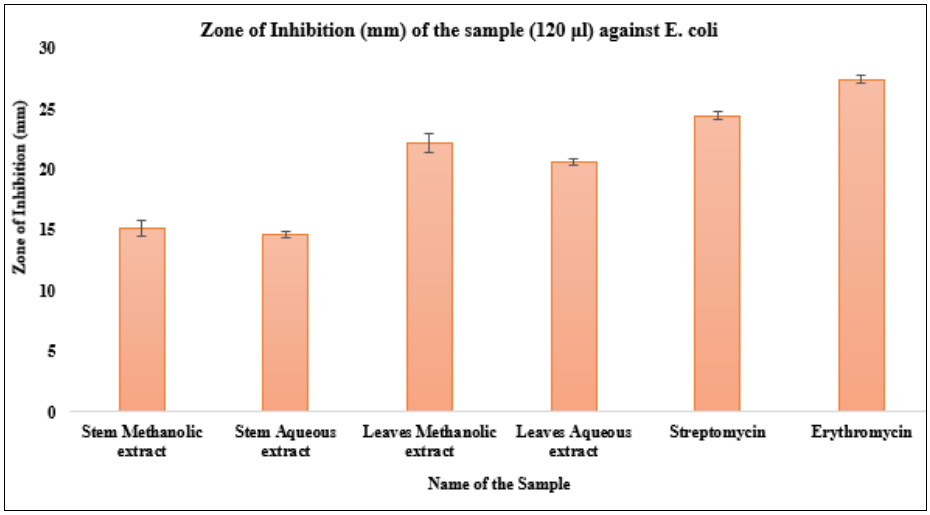

Fig 5: Antibacterial activity with zone of Inhibition of *Ocimum sanctum* L. (leaves and stem) and positive control (120 µl) against *E. coli*.

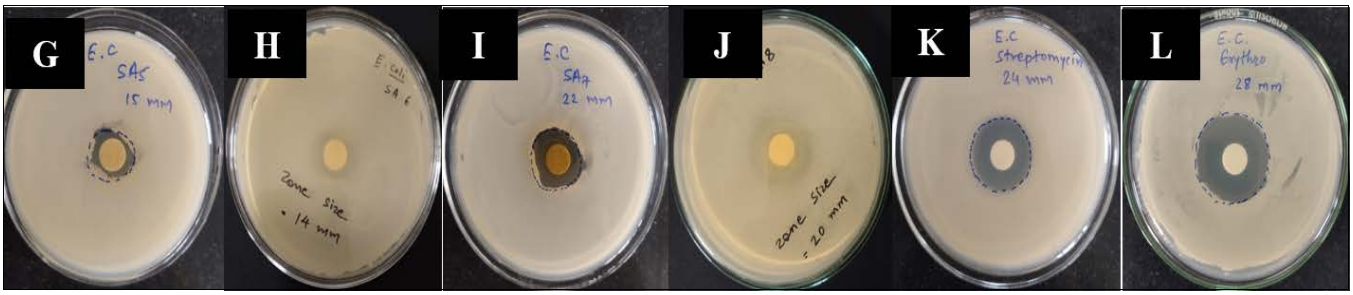

Fig 6: Antibacterial activity of *Ocimum sanctum* L. (leaves and stem) and positive control (120 µl) against *E. coli*; G = Methanolic extracts of *Ocimum sanctum* L. stem, H = Aqueous extracts of *Ocimum sanctum* L. stem, I = Methanolic extracts of *Ocimum sanctum* L. leaves, J = Aqueous extracts of *Ocimum sanctum* L. leaves, K = Streptomycin, L = Erythromycin

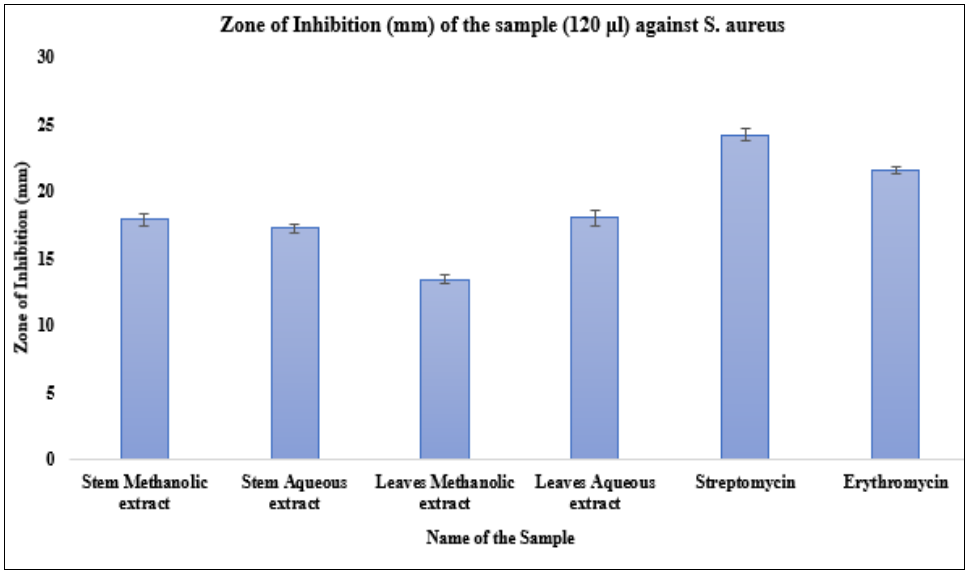

Fig 7: Antibacterial activity with zone of Inhibition of *Ocimum sanctum* L. (leaves and stem) and positive control (120 µl) against *S. aureus*

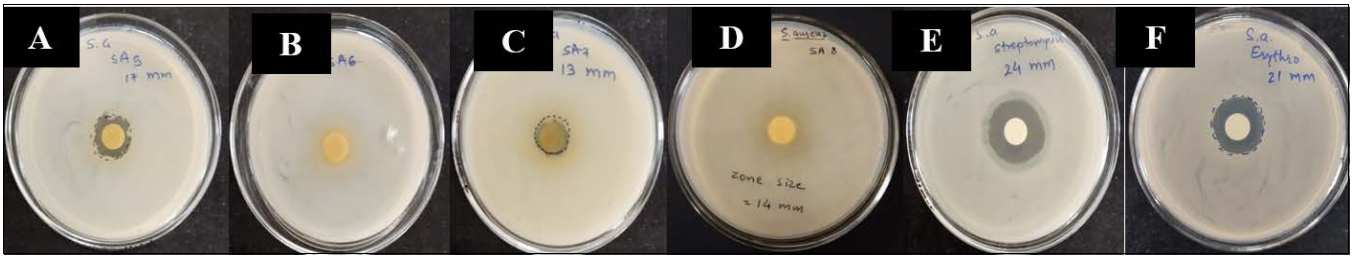

Fig 8: Antibacterial activity of *Ocimum sanctum* L. (leaves and stem) and positive control (120 µl) against *S. aureus*; A = Methanolic extracts of *Ocimum sanctum* L. stem, B = Aqueous extracts of *Ocimum sanctum* L. stem, C = Methanolic extracts of *Ocimum sanctum* L. leaves, D = Aqueous extracts of *Ocimum sanctum* L. leaves, E = Streptomycin, F = Erythromycin

Discussion

The presence of compounds associated with physiological and medicinal benefits was confirmed by phytochemical analysis of the plant extract [26]. In our present study of *Ocimum sanctum* L. the phytochemical investigation on the aqueous & methanolic extract of leaves and stem, which indicates the presence of rich amount of secondary metabolites such as alkaloids, phenols, flavonoids, tannins, glycosides, terpenoids, steroids, carbohydrates, amino acids and saponins. A similar results was erliar given [15,27-28] but, it shosed from minute to rich quantities in some seconady methabolites. As suggested by Divisha R. *et al*., 2018 [27], this disssimilarity in the qualitative phytochemial composition may have resulted due to different environmental conditions and agronomical practices followed.

The varying proportions and characteristics of other biomolecules present in complex mixtures can effect both the extraction efficiency and bioactivity of polyphenols across different solvents. Although the structure activity relationships of phenolic compounds in herbal matrics persist inadequately explored, existing evidence suggests that antioxident properties are closely correlated with the phenolic content of the extracts [6]. There are several flavonoids and phenolic acids found in high amounts in *Ocimum sanctum* L., which also has essential oils. Some of these flavonoids and acids are rosmarinic acid, apigenin, luteolin, quercetin, and cirsimaritin. Due to their strong antioxidant properties, these polyphenolic compounds have anti-aging, neuroprotective, and liver-protective effects [34]. Secondary metabolites like alkaloids which are one of the largest groups of the phytochemicals in plants, which have the most common biological properties of alkaloids is their toxicity against cells foreign organisams like bacteria [15]. Herbal plants are a valuable source of bioactive compounds, including phenolic acids, flavonoids, and tannins. These compounds provide antimicrobial and anticancer effects by scavenging free radicals. Phenolic compounds are extensively distributed across various plant parts, with the aerial parts generally

containing the highest concentrations of total phenols, flavonoids, alkaloids and exhibiting stonger chealating activity than the root and seed part of the plant [33].

Tannins are known to "tan" the outermost layer of the mucosa and put up it less permeable and more resistantto chemical and mechanical injury or irritation. Recently, it has beenexplored that these compounds play important roles inhuman health when ingested into the body [32]. Although tannins are commonly found in many plants and are known for their ability to neutralize free radicals and fight different pathogens, the Omani herbal extracts showed relatively low levels of tannins [33]. Secondary metabolites, such as alkaloids, flavonoids, phenols, and tannins, are known for their positive effects in fighting different pathogens [29]. India has a prosperous record associated with applying different capable natural herbs and plant based ingredients relating to management of different diseases. Plants are the basis for many medications. However, only a small number of plant species have been studied for antimicrobial compounds [30].

Plant extracts, therefore, have shown strong activities against the pathogens because of the wide range of phytochemicals. Few in-depth studies on these plants and their possible role as phytochemical sources and in treating infections. Rising antibiotic resistance, harmful side effects, and the high costs of developing synthetic drugs are driving attention toward plant-based medicines. This study identified potential plant species that people have traditionally used to treat various infections, such as tropical diseases, gastrointestinal issues, skin problems, and wounds [31]. The result bring out from our reserch are usually in agreement with the studies associated with the other researchers in the same field [15,25]. Largest zone of inhibition were observed at 120 µl concentration of all the extracts. Methanolic extracts provided more potent antibacterial activity as compared to the aqueous extracts. The leaves extract showed higher antibacterial activity than the stem extracts against both organisms *E. coli* and *S. aureus*.The polarity of the phytochemicals and antibacterial compounds make them more expediently extracted by organic solvents because secondary metabolites are more soluble in organic solvent then water and using both solvents does not give the negatively affect their bioactivity against bacterial species.

Phytochemicals are bioactive molecules found in plants that, along with nutrients and dietary fibres, help to protect humans from diseases. From the findings of the study, the phytochemical substances which have been detected may be the bioactive components, and the plant is proving to be a significant rich source of bioactive chemicals which is more useful for pharmacological investigation with significant therapeutic value.

Conclusion

Our study has pointed out the presence of phytochemicals considered as active medicinal chemical constituents in tulsi extracts indicated the presence of alkaloids, phenols, flavonoids, tannins, glycosides, terpenoids, steroids, carbohydrates, amino acids and saponins etc. In summary, methanol was found to be the best solvent for phytochemical screening from *Ocimum sanctum* L. leaves, it has showed the high amount result among water (decoction) extracts. The phytocompounds present in these plants proved to have potential antibacterial activity against *E. coli* and *S. aureus*. It is therefore, we hope that the important properties identified by our study in the *O. sanctum* L. will be helpful for the treatment of different diseases and suggested that efforts should be made by pharmaceutical companies carrying out more research work in the development of new drugs.

Acknowledgment

The authors would like to acknowledge the valuable support of Biology department, Municipal arts and Urban Bank Science College, Mehsana, Gujarat, India for providing their lab facility for performing the qualitative and quantitative phytochemical analysis and also thankful to Sterling Accuris Diagnostics - Pathology Lab, Mehsana for providing their lab facility for performing antibacterial activity.

References

1. Kothari SK, Bhattacharya AK, Ramesh S, Garg SN, Khanuja SPS. Volatile constituents in oil from different plant parts of methyl eugenol-rich Ocimum tenuiflorum Lf (*syn. O. sanctum* L.) grown in South India. J Essent Oil Res. 2005;17(6):656-665.
2. Kavyamala D, Jayamma P, Usha Rani N, Mahesh Babu M. Phytochemical screening of aqueous, acetone, ethanol and methanol leaf extracts of Ocimum tenuiflorum, Ocimum gratissimum and *Ocimum sanctum*. Int J Econ Plants. 2023;10(2):109-113. doi:10.23910/2/2023.0515b
3. Vasu K, Goud JV, Suryam A, Singara Chary MA. Biomolecular and phytochemical analyses of three aquatic angiosperms. Afr J Microbiol Res. 2009;3(8):418-421.
4. Edoga HO, Okwu DE, Mbaebie BO. Phytochemical constituents of some Nigerian medicinal plants. Afr J Biotechnol. 2005;4(7):685-688.
5. Mann J. Secondary Metabolism. London: Oxford University Press; 1978. p.154.
6. Chaudhary A, Sharma S, Mittal A, Gupta S, Dua A. Phytochemical and antioxidant profiling of *Ocimum sanctum*. J Food Sci Technol. 2020;57(10):3852-3863. doi:10.1007/s13197-020-04417-2
7. Joseph B, Nair VM. Ethanopharmacological and phytochemical aspects of *Ocimum sanctum* Linn - the elixir of life. J Pharm Res Int. 2013;273-292.
8. Ali H, Nguta J, Musila F, Ole-Mapenay I, Matara D, Mailu J. Evaluation of antimicrobial activity, cytotoxicity, and phytochemical composition of *Ocimum americanum* L. (Lamiaceae). Evid Based Complement Alternat Med. 2022;2022:6484578. doi:10.1155/2022/6484578
9. Adela Alemu M, Andargie Y, Sisay W. Antidiarrheal effect of 80% methanol extract and fractions of the leaves of Ocimum lamiifolium in Swiss albino mice. Evid Based Complement Alternat Med. 2022;2022:6838295. doi:10.1155/2022/6838295
10. Venancio AM, Ferreira-da-Silva FW, da Silva-Alves KS. Essential oil of Ocimum basilicum L. and (−)-linalool blocks the excitability of rat sciatic nerve. Evid Based Complement Alternat Med. 2016;2016:e9012605. doi:10.1155/2016/9012605
11. Selvam K, Rajinikanth R, Govarthanan M, Paul A, Selvankumar A, Sengottaiyan A. Antioxidant potential and secondary metabolites in *Ocimum sanctum* L. at various habitats. J Med Plants Res. 2013;7(12):706-712.
12. Yamani HA, Pang EC, Mantri N, Deighton MA. Antimicrobial activity of tulsi (Ocimum tenuiflorum) essential oil and its major constituents against three species of bacteria. Front Microbiol. 2016;7:681. doi:10.3389/fmicb.2016.00681
13. Samanta S, Das NK, Ghosh A. Precious phytomedicinal

components of tulsi (*Ocimum sanctum* Linn.) and their therapeutic importance. Int J Curr Res. 2021;13(10):19119-19141.
14. Sharaf MH, Abdelaziz AM, Kalaba MH, Radwan AA, Hashem AH. Antimicrobial, antioxidant, cytotoxic activities and phytochemical analysis of fungal endophytes isolated from Ocimum basilicum. Appl Biochem Biotechnol. 2021;193(4):1142-1157. doi:10.1007/s12010-021-03702-w
15. Singha AR, Bajaj VK, Sekhawat PS, Singh K. Phytochemical estimation and antimicrobial activity of aqueous and methanolic extract of *Ocimum sanctum* L. J Nat Prod Plant Resour. 2013;3(1):51-58.
16. Gadhvi K, Depani P, Karangiya R, Vyas S. Phytochemical analysis of algal species found in Sutrapada coastal region of Gujarat, India. Eur J Med Plants. 2019;29(1):1-9.
17. Harborne JB, Baxter H. Phytochemical Dictionary: A Handbook of Bioactive Compounds from Plants. London: Taylor & Francis; 1995.
18. Phukan R, Shil D. Preliminary phytochemical screening and *in vitro* anticonvulsant activity of methanolic extract of leaves of Cinnamomum tamala (Buch.-Ham.) T. Nees & Eberm (Lauraceae). Int J Pharm. 2017;4(10):345-351. doi:10.13040/IJPSR.0975-8232.IJP.4(10).345-51
19. Sahira BK, Cathrine L. General techniques involved in phytochemical analysis. Int J Adv Res Chem Sci. 2015;2(4):25-32.
20. Velu S, Baskaran C. Phytochemical analysis and *in vitro* antimicrobial activity of Withania somnifera (Ashwagandha). J Nat Prod Plant Resour. 2012;2(6):711-716.
21. Zhou K, Yu L. Total phenolic contents and antioxidant properties of commonly consumed vegetables grown in Colorado. LWT Food Sci Technol. 2006;39(10):1155-1162.
22. Mandal N, Chudhari D, Ghate NB, Sarkar R. Phytochemical analysis and evaluation of antioxidant and free radical scavenging activity of Withania somnifera root. Asian J Pharm Clin Res. 2012;5(4):193-199.
23. Maitreya BB, Kumarkhaniya HR. Pharmacognostic standardization and phytochemical evaluation of *Phyllanthus maderaspatensis* L. Indian J Nat Sci. 2023;14(79):976-997.
24. Patel SB, Ghane SG. Phyto-constituents profiling of Luffa echinata and *in vitro* assessment of antioxidant, anti-diabetic, anticancer and anti-acetylcholinesterase activities. Saudi J Biol Sci. 2021;28(7):3835-3846. doi:10.1016/j.sjbs.2021.03.050
25. Sikrodia R, Chhabra D, Gangil R, Audarya SD, Sharda R, Mahor SS. Comparative *in vitro* antibacterial efficacy of different common herbs and antibiotics against E. coli and S. aureus isolated from bovine mastitis. Int J Curr Microbiol Appl Sci. 2020;9(2):1069-1075. doi:10.20546/ijcmas.2020.902.125
26. Yadav RNS, Agarwala M. Phytochemical analysis of some medicinal plants. J Phytol. 2011;3(12):10-14.
27. Divisha R, Ranganathan V, Vijayakaran K, Elamaran. Evaluating Ocimum basilicum and Ocimum tenuiflorum leaf extracts for the presence of phenolic compounds. J Pharmacogn Phytochem. 2018;7(6):2453-2456.
28. Naik LS, Shyam P, Marx KP, Baskari S, Devi VR. Antimicrobial activity and phytochemical analysis of Ocimum tenuiflorum leaf extract. Int J PharmTech Res. 2015;8(1):88-95.
29. Usman H, Rahman AFI, Usman A. Qualitative phytochemical screening and *in vitro* antimicrobial effects of methanol stem bark extract of Ficus thonningii (Moraceae). Afr J Tradit Complement Altern Med. 2009;6(3):289-95.
30. Bhandari S, Khadayat K, Poudel S, Shrestha S, Shrestha R, Devkota P, Marasini BP. Phytochemical analysis of medicinal plants of Nepal and their antibacterial and antibiofilm activities against uropathogenic Escherichia coli. BMC Complement Med Ther. 2021;21:1-11. doi:10.1186/s12906-021-03293-3
31. Dubale S, Kebebe D, Zeynudin A, Abdissa N, Suleman S. Phytochemical screening and antimicrobial activity evaluation of selected medicinal plants in Ethiopia. J Exp Pharmacol. 2023;15:51-62. doi:10.2147/JEP.S379805
32. Mustafa AA, El-Kamali HH. Proximate and phytochemical constituents of *Ocimum sanctum* in Sudan. Adv Pharm J. 2020;5(6):201-205.
33. Al-Mutaani J, Zourgui L, Missaoui N. Phytochemicals, bioactive compounds, and antimicrobial activities of Ocimum basilicum, Teucrium polium, Cleome amblyocarpa, and Caralluma arabica extracts: a comparative Omani study. Cell Mol Biol. 2025;71(3):134-45. doi:10.14715/cmb/2025.71.3.16
34. SR SK, Bongale MM, Sarwade PP, Vijayalakshmi K, Goswami M, Khongshei R, Gaisamudre KN. *Ocimum sanctum*: Phytochemistry, therapeutic uses, pharmacological activities and its anticancer activities. Asian J Pharm Res Dev. 2025;13(2):119-125.