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Exploration of the Antimicrobial Mechanism of the Bioactive Components of Tulsi Extract Against Various Microbes and its Future Scope of Development for Application in Textiles

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ABSTRACT

This study delves into the significance of antimicrobial finishes on textiles, focusing mainly on the ancient use of Tulsi and advocating for its preference over other herbal antimicrobial finishes. Tulsi, derived from the Ocimum tenuiflorum plant, has been historically recognized for its potent antimicrobial properties, making it a compelling choice for textile applications. A comprehensive exploration provides insights into various species of tulsi and their geographical significance in shaping the nature and properties of Tulsi oil.

The diverse composition of Tulsi essential oils imparts robust antimicrobial capabilities by targeting bacterial cell membranes. Mechanisms, including lipid bilayer disruption and interference with basic cellular processes, inhibit bacterial proliferation. This understanding is crucial for leveraging Tulsi oils' therapeutic potential as natural antimicrobial agents, with future research aiming to enhance efficacy against diverse bacterial strains for innovative therapeutic interventions.

An in-depth analysis of the primary bioactive compounds found in Tulsi oil illuminates its potent antimicrobial capabilities, showcasing its efficacy in inhibiting various pathogens. Moreover, this study elucidates the methods and techniques for applying Tulsi oil in textiles, considering its implications across different fiber substrates.

This review underscores the promising potential of Tulsi oil as a versatile antimicrobial agent in textile manufacturing by evaluating its current applications and future prospects. The study contributes to a deeper understanding of the antimicrobial properties and scope of Ocimum tenuiflorum, paving the way towards innovative, sustainable, and functional textile solutions that meet emerging consumer demands while promoting health, hygiene, and environmental responsibility.

Keywords: Antimicrobial mechanism, Bioactive, Essential oil (EO), Herbal, Pathogens, Sustainable, Tulsi, Textile

Introduction

With increasing health and hygiene-related concerns in today's world, the textile industries have emphasized incorporating antimicrobial finishes into fabrics due to the growing concerns surrounding health and hygiene. Microbial contamination on textile surfaces promotes the growth of bacteria, fungi and viruses, which leads to a significant increase in risk across diverse settings, healthcare facilities, everyday apparel, and household textiles.

The application of antimicrobial finishes is essential in restraining the proliferation of harmful microorganisms, fostering a hygienic environment and improving functional textiles.

By incorporating antimicrobial finishes, industrialists can effectively eliminate unpleasant odours, unwanted discoloration and health hazards, mainly skin infections associated with microbial growths (Bairagadar and Patil, 2021). These finishes work by disrupting the cell structure or metabolism of microorganisms, thus extending the life of garments and providing a safer experience for users. This is especially important in healthcare conditions where textiles can serve as reservoirs for these infectious agents.

Adopting natural antibiotics derived from plant extracts or essential oils offers attractive advantages over synthetic chemicals (Thulagavthi and Rajendrakumar, 2005; Bairagadar and Patil, 2021). Natural agents provide a safer alternative and a sustainable and environmentally friendly approach to textile finishing, as natural antimicrobials possess a wide spectrum of efficacy against a wide variety of microorganisms, making them suitable for various applications on different materials (Raj Kumar *et al.*, 2005).

Tulsi extract stands out for its potent antimicrobial properties and traditional medicinal uses among natural antimicrobial agents. Revered for centuries in Ayurveda, Tulsi, it possesses antimicrobial, anti-inflammatory, and immunomodulatory properties. Recent studies have highlighted its effectiveness as an antibacterial agent in the textile industry and its ability to prevent the development of different microorganisms (Sathianarayanan *et al.* 2010). However, research on using Tulsi extract in textiles has primarily focused on cotton bases, leaving a gap in understanding its compatibility and effectiveness with other fibers such as silk, wool, jute, polyester, etc. Further, the study on Tulsi extract and its diverse fabric-based properties endorses exploiting the full advantages of its benefits and extending its applicability to a wider range of materials. Natural antibacterial agents, such as Tulsi extract, offer a promising solution to improve textile performance and hygiene (Mahmood, 2008). For thousands of years, the utilization of medicinal herbal extracts in traditional healthcare has been documented. Ayurvedic medical books written during the Vedic period (3500-1600 BC) of Indian civilization describe practices, including the use of medicinal herbs, which were the basis for all other medical sciences developed in the Indian subcontinent (Jiang *et al.*, 2014; Mandave, 2014; Sun *et al.*, 2014; Jiang *et al.*, 2015; Chang *et al.*, 2016; Yamani *et al.*, 2016).

Plants are the main source of therapeutic agents, and every part of a plant, including seeds, roots, stems, leaves and fruits, contains potential bioactive components (Sathianarayanan *et al.* 2010). Medicinal plants, particularly herbs like Tulsi or Holy Basil, offer numerous benefits due to their rich combination of bioactive compounds. Its versatility is evident in its traditional uses, which include treating poisoning, colic, colds and heart disease. Tulsi aqueous extract and oil are widely used for their therapeutic properties, such as their expectorant, analgesic, and anti-inflammatory effects. With a history deeply embedded in ancient traditions (Yamani *et al.*, 2016).

Phytomorphology

Tulsi plant is divided into different parts like stems, leaves, roots, seeds, fruits, etc., as shown in Figure 1. A small herbaceous plant grows up to 3 m with opposite phyllotaxy and slender, pubescent petioles. Its light green and silky leaves are acute and oblong with entire or serrate margins, bearing minute glands and stomata mainly on the lower surface. The purple to pink flowers are collected in terminal racemes 5 to 30 cm long, with hermaphroditic flowers arranged in whorls of 6 to 10 flowers. Flowering begins after 136 days and lasts for 195 days, and seeds ripen after 259 days (Sembulingam *et al.*, 1997; Monga, 2017).

Taxonomically, the classification of Ocimum tenuiflorum from kingdom down to species is shown in Table 1 (Joshi *et al.*, 2011).

Table 1. Taxonomically classification of Ocimum tenuiflorum (Sawangiaroan *et al.*, 2005; Tewtrakul *et al.*, 2019)

Kingdom	**Plantae**
Family	Lamiaceae
Genus	Ocimum
Species	tenuiflorum
Scientific Name	Ocimum tenuiflorum

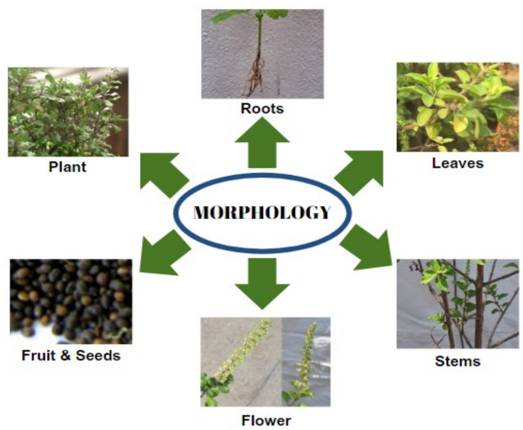


Figure 1. Different morphological parts of Tulsi Plant (adopted and modified from Kumar, 2019)

Comparison of Tulsi compounds/ species cultivated in different geographical locations

The composition and amount of different Tulsi compounds can vary. These differences may be due to factors such as the geographical origin of the plant variety and environmental conditions, which play an important role in determining the volatile composition and proportions. Studies on Tulsi plants growing in different locations have shown that environmental factors significantly impact their chemical composition, as shown in Table 2.

Table 2. Examples of the crucial compounds found in the essential oil obtained from Tulsi plants grown in various geographical locations

Geographical Location	Source of Essential oil	Major compounds
India (Kothari *et al.*, 2005; Helen *et al.*, 2011; Naquvi *et al.*, 2012)	Leaves, inflorescence	Methyl eugenol, cyclooctene Eugenol, bornyl acetate, Camphor, Methyl eugenol, Beta-caryophyllene
Brazil (Machado *et al.*, 1999)	Leaves, inflorescence	Eugenol, Beta-caryophyllene
Australia (Brophy *et al.*, 1993)	Leaves	Methyl chavicol, Camphor, beta caryophyllene
Cuba (Pino *et al.*, 1998)	Leaves, inflorescence	Eugenol elemene, beta caryophyllene
Germany (Laakso *et al.*, 1990)	Leaves	Eugenol methyl, chavicol eucalyptol, beta bisabolene, alpha bisabolene
Australia (Victoria) (Prakash and Gupta, 2005)	Leaves, inflorescence	Camphor, eucalyptol, eugenol, alpha bisabolene, beta bisabolene, beta caryophyllene

The chemical constituents and their respective percentage of several *Ocimum* species varied depending upon the origins and cultivars, as stated in Table 3 (Mondawi *et al.*, 1984; Sajjadi, 2006; Awasthi & Dixit, 2007; Mohhiuddin *et al.*, 2013).

Table 3. The chemical constituents of the essential oils of various Ocimum species

Plant Species	Major constituents (%)
O. basilicum L. (Mondawi *et al.*, 1984)	Methyl chavicol (70%), Linalool (25%), Eugenol (5%)
O. gratissimum L.387E (Ntezuriubanza *et al.*, 1987)	Thymol (35.4%), p-Cymene (18.3%), Eugenol (10.7%)
O. gratissimum 345C (Akgul,1989)	Thymol (46.6%), γ-Terpinene (22.4%)
O. basilicum (Akgul, 1989)	Linalool (45.7%), Eugenol (13.4%), Methyl eugenol (9.57%), Fenchyl alcohol (3.64%)
Ocimum utricifolium 298E (Janseen *et al.*, 1998)	Methyleugenol (73.6%)
O. basilicum (Khatri *et al.*, 1995)	Linalool, Methyl chavicol, Eugenol
O. basilicum (Marotti *et al.*, 1996)	Methyl chavicol (87.3%), Linalool (5.4%), Methyl eugenol (1.5%), β-Caryophyllene (2.4%), α-Pinene (1%), β-Pinene (0.8%), Limonene (0.5%), Camphene (0.2%)
O. basilicum var. minimum (Silva *et al.*, 2003)	Linalool (16%), Estragole (52.2%), 1,8-Cineole (7.4%)
O. basilicum var. purpurascens (Mohhiuddin *et al.*, 2013)	Methyl cinnamate (59.95%), Linalool (16.4%), tau-Cadinal (4.37%)

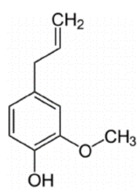
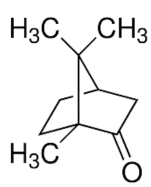
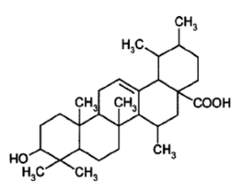
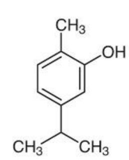
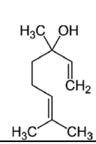
Bioactive components of Tulsi

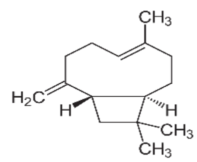
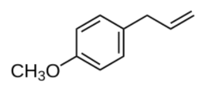
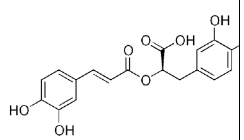
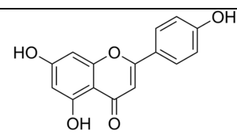
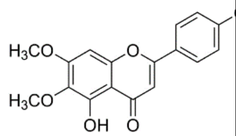
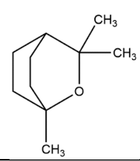
The primary active constituents in the aqueous extract of Tulsi leaves include eugenol, urosolic acid, carvacrol, rosmarinic acid, α & β-caryophyllene, Linalool, euginal (also known as eugenic acid), β-element, geraneol and ocimene (Ahmed *et al.*, 2002).

There are two categories of antimicrobial agents: bactericides and bacteriostatics. The former kills microorganisms (bactericide) and later stops their growth (bacteriostatic agent) (Bairagadar and Patil, 2021). Tulsi has the properties of both bactericides and bacteriostatic (Sawangjaroen *et al.*, 2006).

Other active constituents present in Tulsi extract include α-thujene, isothymusin, octane, α-pinene, oleanolic acid, cirsimaritin, toluene, apigenin, nonane, β-pinene, isoborneol, camphene, α-guaiene, sabsinene, ethyl benzene, borneol etc. as shown in Table-4 (Kelm *et al.*, 2000; Chiang *et al.*, 2005; Douglas *et al.*, 2005; Mondal *et al.*, 2009; Pattanayak *et al.*, 2010; Rathnayaka, 2013; Devi *et al.*, 2015; R. B. and S. P. B., 2018).

Table 4. Primary bio-active constituents of Ocimum Species--Tulsi

Name of Active Constituents	Structure of Active Constituents	Properties
Eugenol (1-hydroxy-2-methoxy-4-allylbenzene		Antimicrobial, Anti-cancer agent, anti-inflammatory, Antidiabetic, Cardioprotective, Hypolipidemic, Hepatoprotective agent.
Camphor		Antibacterial and Antifungal.
Urosolic acid		Antitumor, Antimicrobial, Anti-viral activities, Hepatoprotective, Anti-inflammatory (oral & topical), Anti-ulcer and Anti- hyperlipidaemic.
Carvacrol (5-isopropyl-2-methyl phenol)		Anti-cancer agent, Antioxidant, Antibacterial.
Linalool(3,7-dimethylocta-1,6-dien-3-ol)		Antibacterial, Anti-viral, Antifungal, Anti-cancer agent.

Caryophyllene (4,11,11-trimethyl-8-methylene-bicyclo [7.2.0]undec-4-ene)		Anti-cancer agent, Antioxidant, Antimalarial, Antiviral.
Estragol (1-allyl-4-methoxybenzenes		Antidiabetic, Anti-stress.
Rosmarinic acid - {(3,4-Di-hydroxy- phenyl)-1-oxo-2-propenyl]-3-(3,4-di-hydroxyphenyl) propan oic acid)		Immunomodulatory, Antimicrobial, Anti-cancer, Anti-inflammatory.
Apigenin (5,7-dihydroxy-2- (4-hydroxyphenyl)-4H-I-benzopyran-4-one)		Anti-viral, Antibacterial Antioxidant, Anti-inflammatory.
Cirsinaritin (5,4' dihydroxy-6.7-dimethoxy flavones		Cardioprotective, Anti-ulcer, Anti-stress.
Eucalyptol (1,8-cineole)		Antimicrobial properties.

The major bioactive components identified in Tulsi (Ocimum tenuiflorum) essential oil include:

- **Camphor**
  Camphor was found to be the most abundant volatile compound in Tulsi, constituting about 31.52% of the total (Magiatis *et al.*, 2002; Yamani *et al.*, 2016). Camphor has demonstrated antimicrobial activity against various bacterial species, including E. coli, S. aureus, Bacillus cereus and P. aeruginosa (Magiatis *et al.*, 2002; Yamani *et al.*, 2016).

- **Eugenol**
  Although eugenol is present in a smaller proportion than camphor and eucalyptus, it comprises about 13.8% of the essential oil's cumulative volatile compounds. It is well-known for its healing properties and is recognized for its medicinal value, the main component exhibiting antibacterial properties. (Prakash and Gupta, 2005; Pattanayak *et al.*, 2010, Singh *et al.*, 2010; Yamani *et al.*, 2016).

- **Caryophyllene**
  Caryophyllene, a sesquiterpene, makes up a small amount (about 1.2% of the oil, 4.9% of the inflorescence, and 1.5% of the leaves). Despite its lower concentration, β-caryophyllene

possesses antimicrobial properties and is effective against bacterial species such as S. aureus, E. coli, and P. aeruginosa (Alma *et al.*, 2003; Legault *et al.*, 2003, Xiao-Yu *et al.*, 2012; Yamani *et al.*, 2016).

The above discussion represents the percentage of the bioactive compounds present in different parts of Tulsi. Therefore, these bioactive components are reported to contribute to the antibacterial properties of Tulsi essential oil and may have potential applications in treating various skin infections.

Pharmacological applications of Tulsi

Tulsi is important in Ayurvedic medicine as it treats diseases such as headaches, inflammation, heart disease, poisoning rhinitis, gastrointestinal disorders and malaria (Panchal and Parvez, 2019). The water and alcohol-soluble extracts of the leaves have antimicrobial, anti-inflammatory, antidiabetic, anti-cancer, anti-ulcer, antifungal and wound-healing effects (Skaltsa *et al.*, 1987; Mathew *et al.*, 1999; Kothari *et al.*, 2004; Babu and Maheswari, 2005; Pandey and Madhuri, 2010; Dev *et al.*, 2011). In particular, Tulsi has demonstrated efficacy against various pathogens, including multidrug-resistant strains (Khosla, 1995; Kicel *et al.*, 2005; Lavanya and Kumar, 2011). These results highlight the therapeutic potential of Tulsi as a natural broad-spectrum antibacterial agent against a variety of pathogens and warrant further exploration for pharmaceutical and therapeutic applications (Pramod and Saini, 2018). In addition to these pharmacological actions, Tulsi offers a variety of health benefits, including relieving coughs, improving eye health, treating skin diseases, lowering cholesterol levels, reducing stress and supporting oral health (Rajasekharan *et al.*, 1993; Maurya, 2007; Mishra *et al.*, 2013).

The antimicrobial properties of Tulsi extract

The fragrance of the Tulsi plant purifies the air, making the environment sacred. The various active ingredients and essential oils in Tulsi give it a distinctive scent. Because of its unpredictable nature, Tulsi medicinal oil can successfully kill many microorganisms, including microscopic organisms, infections and parasites (Yamani *et al.*, 2016).

Tulsi extract is commonly used for therapeutic purposes in treating a diverse array of illnesses, such as fever, headache, malaria and inflammation (Prakash and Gupta, 2005; Raghav and Agarwal, 2016). Tulsi leaves extract contains a variety of phytochemicals, including aldehyde, terpenes (such as sesquiterpenes and monoterpenes), phenols, saponins, tannins, glycosides, quinone, phlobatanin, flavonoids (orientin and vicenin), steroids, coumarin, alkaloids and anthocyans (Raghav and Agarwal, 2016). Studies have shown that the aqueous extract, seed oil, and alcoholic extract of Tulsi effectively inhibit the growth of enteric pathogens and demonstrate activity against gram-positive and gram-negative bacteria, including multi-drug-resistant strains, as shown in Table 5.

Table 5. Antimicrobial properties of Tulsi extract

Types of extract medium	Microbial Strain	Results
Aqueous extract (Rao and Nigam, 1970; Sinha and Gulati, 1990; Geeta *et al.*, 2001; Williamson E. M., 2002; Dharmagadda *et al.*, 2005; Kaya *et al.*, 2008; Pasha *et al.*, 2009; Vidhani *et al.*, 2016)	**Bacteria:** E. coli, Malassezia furfur, Staphylococcus aureus, Klebsiella aerogens, Klebsiella pneumonia, Proteus mirabiliz. Salmonella typhimurium, Shigella dysentriae, Preudomonas aeruginosa, Staphylococcus cohni Salmonella typhi, Salmonella paratyphi, Salmonella typhimurium **Fungi**: Candida albicans, Fusarium solani, Aspergillus flavus. Aspergillus repens. **Virus:** Rhinotracheitis virus (IBR), White spot syndrome virus (WSSV), Buffalo pox virus (GTPV).	Positive
Aqueous Methanolic and Acetone extract (Devi *et al.*, 2015)	**Bacteria:** Gram-positive and Gram-negative Bacteria.	Positive
Aqueous, Chloroform, Alcoholic extract and oil (Yamani *et al.*, 2016)	**Bacteria:** Gram-positive Bacteria (Listeria monocytogenes) and Gram-negative Bacteria (Salmonella enteritica. Vibrio parahaemolyticus and E. coli).	Positive
Methanolic extract (Kaya *et al.*, 2008)	**Bacteria:** Enterococcus faecalis, Enterobacter cloacae, E. coli, Proteus vulgaris, Klebsiella pruemonine, S. aureus and S. saprophytica. **Fungi:** Candida crimei, Fusarium solani.	Positive
Alcoholic extract (Geeta *et al.*, 2001)	**Bacteria:** Staphylococcus aureus, Pseudomonas aeruginosa, Salmonella typhi and Vibrio cholera. **Fungi:** Candida albicans.	Positive
Essential oil extract (Sharma *et al.*, 2014)	**Bacteria:** Bacillus subtilis, Pseudomonas fluorescence, Staphylococcus aureus, E. coli & Pseudomonas aeruginosa.	Positive

Tulsi essential oils are often rich in terpenes, and terpenoid components like carvacrol, Linalool, beta-caryophyllene, beta-pinene, sabsinene, p-cymene and phenylpropenes like estragol, eugenol, etc., exhibit potent antimicrobial properties. These compounds disrupt microbial cell membranes, leading to the bacteria's cell death.

Essential oils (EOs) have been documented to have diverse antimicrobial mechanisms against various bacterial strains (Burt, 2004; Bajpai *et al.*, 2012; Hyldgaard *et al.*, 2012; Chouhan *et al.*, 2017). Nonetheless, before examining the impact of Tulsi essential oil on microorganisms, it is imperative to thoroughly examine the cell wall architecture both in Gram-negative and Gram-positive bacteria (Hang *et al.*, 2016; Phan *et al.*, 2019; Wang *et al.*, 2020).

- Essential oils (EOs) interact with bacterial membranes due to their lipophilic properties, destroying their integrity. This disruption affects fatty acid synthesis enzymes, changing the composition of bacterial fatty acids and inhibiting ATP (adenosine triphosphate) formation and bacterial mutation processes. Changes in fatty acid composition affect membrane fluidity, disrupt essential cellular functions and inhibit bacterial growth, which leads to cell death (Figure 2) (Deininger and Lee, 2001; Hamoud *et al.*, 2012; Han *et al.*, 2020; Liu *et al.*, 2020; Jeyakumar *et al.*, 2021; Angane *et al.*, 2022).

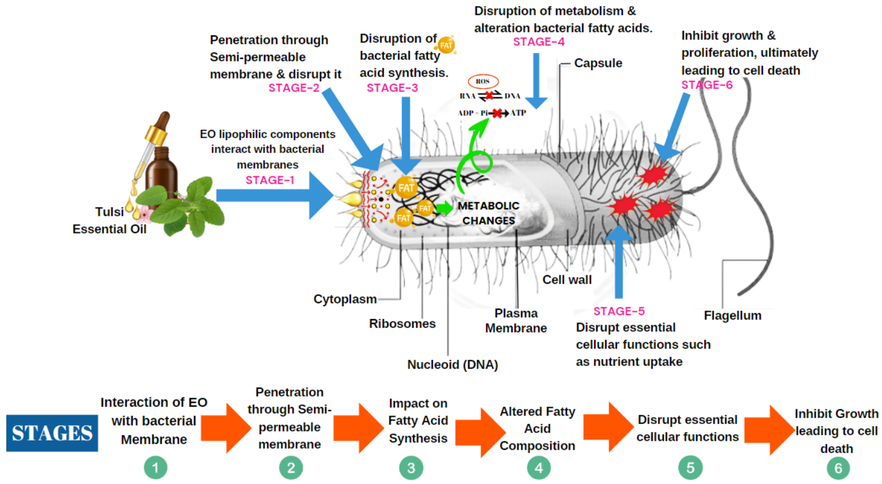


Figure 2. Alteration of the fatty acid composition (self-drawn)

- Essential oils (EOs) interact with the membrane in bacteria, disrupting their structure and increasing their mobility. This support allows potassium ions (K+) and protons (H+). It leaks from the cell and disrupts the electrochemistry and pH balance. This disruption leads to cell failure and, eventually, cell death due to interference with vital cell processes (Figure 3) (Akiyama *et al.*, 2002; Togashi *et al.*, 2008; Togashi *et al.*, 2010; Angane *et al.*, 2022).

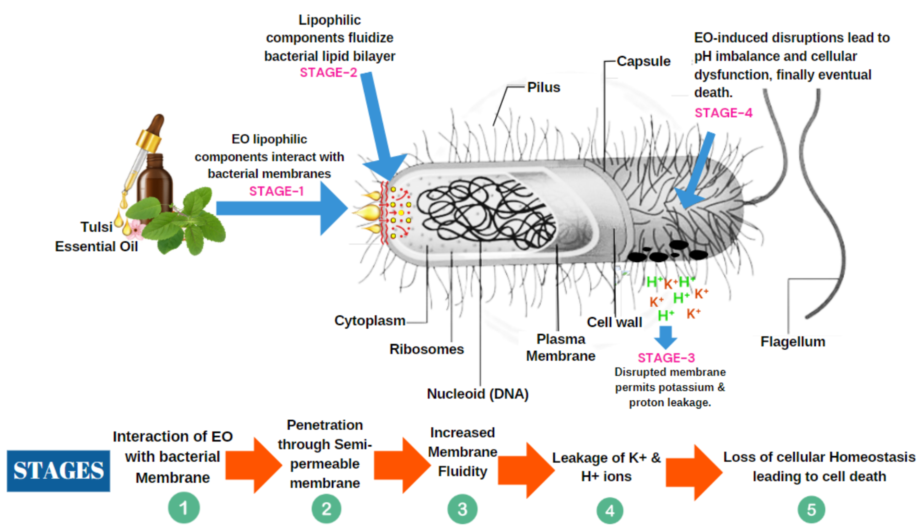


Figure 3. Increase in membrane fluidity resulting in leakage of potassium ions and protons (self-drawn)

- Essential oil (EO) interacts with bacteria, disrupting glucose transporters in the cell membrane. This effect prevents the Increase of glucose and affects energy metabolism. Without sufficient glucose, bacteria experience cell stress and dysfunction, hindering growth and development. This process ultimately limits bacterial growth by depriving the bacteria of essential nutrients, resulting in poor growth and possible cell death (Figure 4) (Angane *et al.*, 2022).

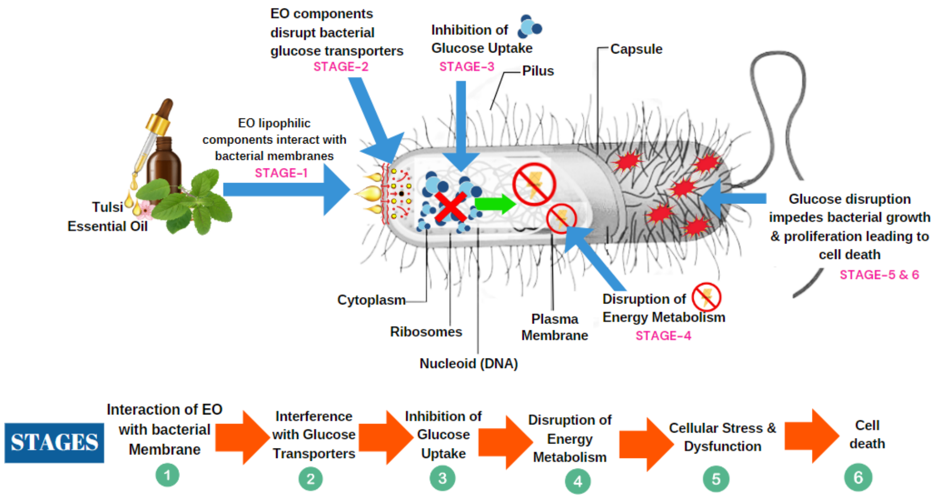


Figure 4. Interference with glucose uptake (self-drawn)

- Essential oils (EOs) and bacterial cells interact, targeting enzymes essential for survival. This effect inhibits enzyme activity and disrupts important cell processes. Some EO products can affect the integrity of bacterial cell walls, causing cell lysis. Therefore, the loss of cellular components, including essential proteins and nucleic acids, causes cellular dysfunction and death, ultimately leading to cell death (Figure 5) (Fisher *et al.*, 2008; Angane *et al.*, 2022).

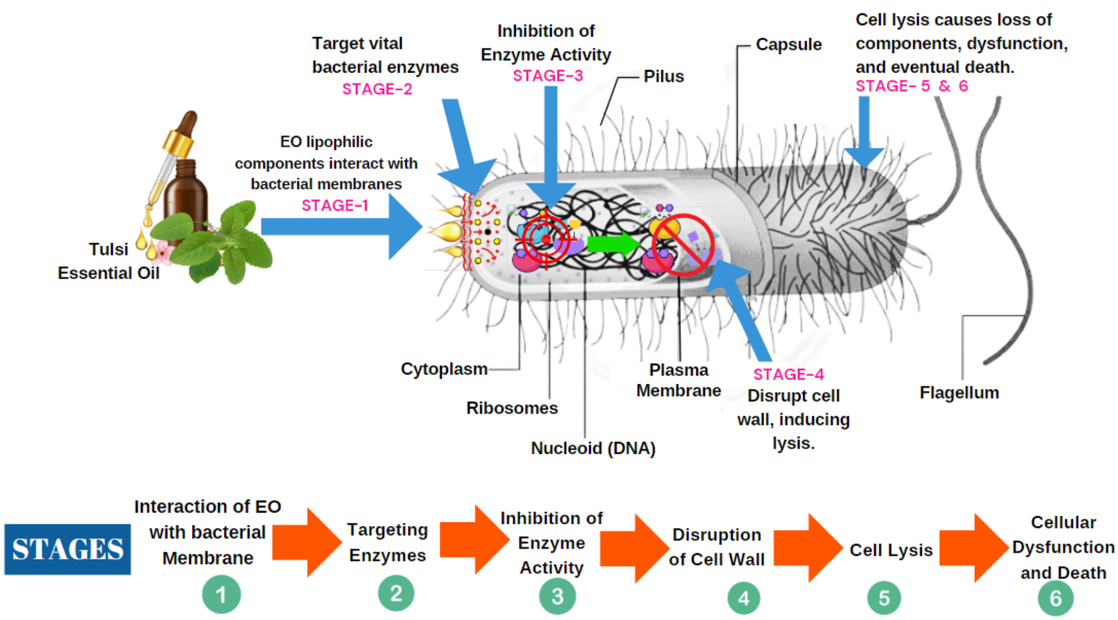


Figure 5. Inhibition of enzyme activity or cell lysis (self-drawn)

Essential Oil Extraction Method

Different oil extraction methods from Tulsi leaves are explained below:

Extraction using supercritical fluid

The leaves of the Tulsi plant are dried to reduce the initial moisture content, which was at about 90% before drying. This drying process was carried out in a recirculating oven with air circulation, keeping it at 50 °C for 5 hours until the humidity was consistently attained. The dried leaves are ground with increased surface area, making it easy for further oil extraction. The ground leaves are then placed in a sealed plastic bag to protect them from light and moisture and stored moisture-free in the refrigerator (Alves De Barros *et al.*, 2013).

Soxhlet Extraction

This method determines the oil content in raw materials. This extraction procedure was performed in triplicate using approximately 5 g of basil and 200 ml of hexane. The extraction time was set to 4 h, and the temperature was approximately 69°C (Alves De Barros *et al.*, 2013).

Hydro-distillation Extraction

It is performed using a Clevenger device for hydro-distillation, with 30 g of crushed basil leaves and 300 ml of water added to the 500 ml bottom glass. The extraction took approximately four hours, and the resulting oil extract was diluted with hexane, separated, filtered, and dried using Na_2SO_4 to remove moisture (Alves De Barros *et al.*, 2013).

Extraction with methanol

Tulsi is collected, washed and dried under the shadow. Rotten leaves are discarded and not included in the experiment. Dried leaves are ground into fine powder. 50 g of fine powder is mixed with 250 ml of 80% conc. Methanol. The solution is put in a container with a lid for 24 hours at room temperature so that methanol is taken into the solution completely. The solution is filtered out by muslin cloth first and then by using filter paper. Finally, it is evaporated to concentrate the extract. The prepared extract is applied to a treated group of fabrics through the pad-dry-cure technique at 110 °C for about 2 minutes and further cured at 120°C for about 3 minutes (Thangamani *et al.*, 2017; Ijaz, 2020).

Application methods and techniques of antimicrobial Tulsi extract

Different methods and techniques of application of Tulsi extracts are explained below:

Direct application

This method treats fabric samples with Tulsi's methanol extract. This involves applying the extract directly to the fabric by padding. The process consists of padding the fabric with extracts for 10 minutes at room temperature with a liquor and material ratio of 20:1 and is further squeezed through a pressure of 20 kg/cm^2 at a speed of 15 m/min. After this, the treated fabric is allowed to dry in air naturally and is further cured at 120°C for 3 minutes. The curing is essential for the fixation of the extract on the textile substrate as well as the effectiveness of the application. This method of impregnating fabrics with plant extracts provides various desired properties such as antibacterial, antioxidant, etc., depending on the type of herb used (Thangamani *et al.*, 2017; Ijaz, 2020).

Micro-encapsulation

This method involves the utilization of micro-encapsulation. This imparts functional properties to textiles, mainly using extracts from herbs as the core material and gum acacia as the wall material. First, 10 g of gum acacia powder is allowed to swell in 100 ml of hot water for 15 minutes. Subsequently, an additional 50 ml of hot water is added, and the mixture is stirred for another 15 minutes, maintaining a temperature of 40°C-50°C. Following this, 1.5 g of the core material (herbal extract) is slowly added to the mixture with stirring. Stirring is maintained for a further fifteen minutes, after which 10 ml of sodium sulfate (20% conc.) and 6 g of citric acid are added to the solution (Williamson, 2002). The stirring process is ceased, and the mixture is subjected to freeze-drying inside a freezer to produce microcapsules. Further, the fabric swatch is immersed in the microcapsule solution and passed through a pneumatic padding mangle at a pressure of 3 psi. The treated fabric is then dried at 80°C for 5 minutes. The whole process aims to encapsulate the herb extract within the gum acacia, which allows for controlled release with durable, functional properties on the surface of the textile substrate (Dridi *et al.*, 2021).

Cross-linking process

Cross-linking involves mixing 1g of herbal extract with 100 ml of non-formaldehyde-based resin (KRISOF NIL/F) at a concentration of 120 gpl. Further, 2g of $MgCl_2$ is incorporated as a catalyst. The fabric is then immersed in this resin solution and passed through a pneumatic padding mangle. Subsequently, the treated fabric undergoes drying at 80°C for about 5 minutes, followed by curing at 150°C for about 3 minutes (Dridi *et al.*, 2021).

Micro-encapsulation and cross-linking method

Micro-encapsulation and cross-linking are two methods used in combination to apply finishes to textiles. The former is a physio-chemical approach, and the latter is a chemical one. Combining these two techniques is reported to enhance the functional properties of textiles. In micro-encapsulation, the Tulsi extract is used as the core material and acacia gum is used as the wall material. Before applying it to the fabric, a mixture containing 18 g of non-formaldehyde resin and 3 g of $MgCl_2$ is prepared. Subsequently, the fabric undergoes immersion in this solution and passes through a pneumatic padding mangle. The treated fabric is subsequently dried at 80°C for about 5 minutes and cured at 150°C for about 3 minutes (Dridi *et al.*, 2021).

Discussion

The effectiveness of the Tulsi extracts on different fibers, and their comparative analysis with other herb extracts are explained below:

Implication on different fibers

The application of Tulsi extract on cotton, bamboo and soya fabrics showed notable antimicrobial properties, as evidenced by the observed zone of inhibition in the agar diffusion method. While cotton and bamboo fabrics treated with Tulsi extract displayed significant efficacy against both gram-positive and gram-negative microorganisms, the effectiveness on the soya fabric was found to be slightly lower but still exhibited significant antimicrobial effects. This difference in efficiency is probably due to the definite formation and structures of the fabrics, especially with soya fabric, as it is derived from naturally occurring protein (Nagarajan, 2009; Thangamani *et al.*, 2017). Similarly, using the Tulsi extracts on polyester fabrics exhibits considerable antimicrobial effectiveness, resulting in a substantial reduction in the proliferation of bacteria compared to untreated polyester fabrics. Moreover, applying the Tulsi extract improved the mechanical properties of polyester fabrics, increasing tear and tensile strength (Ijaz, 2020). These findings underscore the potential of incorporating Tulsi extract into fabric finishing processes to develop antimicrobial textiles that augment their value and functionality. This study highlights the feasibility of using natural herbal extracts like Tulsi in textile manufacturing as a sustainable and efficient method of imparting antimicrobial properties to fabrics. The effectiveness of Tulsi-treated fabrics may vary based on factors such as the concentration of Tulsi extracts used, the application method and the specific microbial strains targeted (Sathianarayanan *et al.* 2010). Further research and development may be necessary to optimize the antimicrobial properties of these fabrics for commercial applications.

Comparison of Tulsi with other herbal extracts in terms of antimicrobial activities

Table 5 presents a detailed comparison of the antimicrobial efficiency of several extracts of herbs, including Clove, Tulsi (holy basil), Neem, Aloe Vera, Kadukai (Terminalia chebula) and Lemon oil. This comparative analysis emphasizes the diverse antimicrobial properties inherently present in these herbal extracts, each containing distinct active compounds that exhibit varying spectra of antimicrobial activity against different pathogen classes. Remarkably, Clove, Tulsi, Neem, and Lemon oil come out as avenues, showcasing broad-spectrum antimicrobial activity against various pathogenic microorganisms. Other than Tulsi, Neem also possesses a broad spectrum of antimicrobial activities, making it a useful resource in antimicrobial formulations. Lemon oil, containing compounds like limonene and citral, shows excellent antimicrobial properties, contributing to its effectiveness against pathogens.

The comparison provided in Table 5 shows the multifaceted antimicrobial potential of herb extracts, which in turn emphasizes the importance of factors like potency and suitability of application when using these natural remedies for antimicrobial purposes.

Table 5. Comparison of Tulsi with other herbs

Herbal extra	**Active Components**	**Spectrum of activity**	**Potency**	**Application in different fibres**
Clove	Eugenol	Broad-spectrum (bacteria, fungi, viruses)	High	Cotton (Kant *et al.*, 2017; Barajas *et al.*, 2021)
Tulsi	Eugenol, camphor, eucalyptol, beta-caryophyllene	Broad-spectrum (bacteria, fungi, viruses)	Moderate to high	Cotton, polyester, bamboo, and soya fibre (Ali *et al.*, 2014; Thangamani *et al.*, 2017)
Neem	Azadirachtin, nimbin, azadirone	Broad-spectrum (bacteria, fungi, viruses)	Moderate to high	Cotton, bamboo, hemp, silk, and a wide range of other fibres (Khurshid *et al.*, 2015; Thangamani *et al.*, 2017; Abd *et al.*, 2018)
Aloe Vera	Anthraquinones, polysaccharides	Moderate spectrum (mainly bacteria, some fungi)	Moderate	Cotton, bamboo, and a wide range of other fibres (Khurshid *et al.*, 2015; Thangamani *et al.*, 2017; Abd *et al.*, 2018; Inprasit *et al.*, 2018)
Kadukai	Tannins, chebulinic acid, ellagic acid	Moderate spectrum (mainly bacteria, some fungi)	Moderate	Cotton, bamboo (Thangamani *et al.*, 2017)
Lemon grass oil	Limonene, citral	Broad-spectrum (bacteria, fungi, viruses)	Moderate	Cotton, bamboo, and soya (Thangamani *et al.*, 2017)

Conclusion

This study also highlights the versatility of Tulsi in textile applications, demonstrating its affinity with different fabric types, including cotton, silk, wool, bamboo, polyester, nylon and polypropylene. Different methods such as direct application, micro-encapsulation and cross-linking offer constructive means of incorporating Tulsi extracts onto fabrics, imparting durable antimicrobial properties. Moreover, the eco-friendly nature of Tulsi extracts aligns with the increasing demand for sustainable textile solutions, endorsing environmentally conscious alternatives to synthetic antimicrobial agents. By embracing Tulsi-based finishes, the textile industry can not only improve the hygiene and functionality of textiles but also contribute to the broader objective of sustainability. The composition of terpenes and terpenoids in Tulsi essential oils presents potent antimicrobial properties by disrupting bacterial cell membranes. Through mechanisms such as lipid bilayer disruption, Alteration of fatty acid composition, intervention with membrane integrity and essential cellular processes, Tulsi essential oils successfully inhibit bacterial growth and proliferation. Understanding these mechanisms is pivotal for harnessing the therapeutic potential of Tulsi essential oils as natural antimicrobial agents. Further research may improve their chemical profiles for enhanced performance against a wide range of bacterial strains, offering promising avenues for developing novel antimicrobial therapies. The antimicrobial efficiency of Tulsi in textiles indicates the need for further research and development to enhance its application techniques and efficacy across diverse textile substrates. By harnessing the potential of Tulsi, the textile industry can pave the way for innovative, eco-friendly and sustainable antimicrobial solutions, meeting the evolving demands of consumers while promoting health, hygiene and environmental responsibility.

When assessing herbal antimicrobial agents like Aloe Vera, Kadukai, Lemongrass and Tulsi, the latter, Tulsi, undoubtedly stands as the premier choice. Tulsi is more effective in treating E. coli than Neem. While each herb boasts its own antimicrobial properties, Tulsi surpasses its counterparts with a broader spectrum of action against bacteria, viruses, fungi, and parasites. Its rich chemical composition, including compounds like eugenol and beta-caryophyllene, ensures potent efficacy. Tulsi's long-standing recognition in traditional medicine systems also underscores its safety and effectiveness.

Future scopes of exploration

Several future scopes can be identified, the antimicrobial properties and application of Tulsi in textiles:

- **Consumer awareness and market adoption**

  Promoting consumer awareness about the benefits of Tulsi-treated textiles and market adoption is essential for commercial success. Marketing campaigns and educational initiatives can help convey the value proposition of Tulsi-based finishes, which drives the demand for sustainable and eco-friendly textile products.

- **Optimization of extraction techniques**

  Further research can be conducted on optimizing the extraction techniques for obtaining Tulsi extracts with maximum antimicrobial efficacy. This includes exploring novel extraction methods, varying solvent systems, and adjusting parameters such as temperature, pressure, and time to enhance the yield and bioactivity of the extracts.

- **Fabric compatibility studies**

  Further studies can help understand the compatibility of Tulsi treatments with different textile substrates. Investigating the effects of Tulsi treatments on fabric properties such as durability, colorfastness, and comfort can provide valuable insights for commercial applications.

- **Synergistic formulations**
  Research can explore the development of various synergistic formulations by combining various natural antimicrobial agents, such as Neem, Aloe Vera, etc., with Tulsi extracts. These can offer enhanced antimicrobial activity while reducing the reliance on single-source extracts, contributing to the development of broad-spectrum antimicrobial finishes on textiles.

- **Functional finishes over antimicrobial**
  While the review focuses on Tulsi's antimicrobial properties, future research can explore its potential for imparting other functional properties to textiles. This includes investigations into UV-protective, antioxidants, anti-inflammatory, and other functional properties by expanding the scope of Tulsi-based finishes for diverse textile applications.

- **Introduction of new application techniques and process optimization**
  Further optimization of application techniques such as direct application, micro-encapsulation, and cross-linking can enhance the efficiency and durability of Tulsi treatments on textiles. Additionally, research can focus on scaling up these processes for industrial applications while minimizing environmental impact. Besides this, research can be done to find suitable application techniques.

- **Lack of quantitative studies**
  More quantitative research is needed to precisely measure the antimicrobial efficacy of Tulsi-treated textiles. Quantitative data can provide clearer insights into the extent of microbial reduction achieved by Tulsi finishes, aiding in the standardization of application methods and dosage for optimal results.

- **Lack of study in wash sustainability**
  There is a gap in understanding the sustainability of these properties over repeated wash cycles. Investigating the durability of Tulsi finishes against laundering and determining the number of washes before efficacy diminishes can help assess the long-term viability of Tulsi-treated textiles in practical use.

- **Studies on fungal growth**
  Several studies have focused on the antibacterial properties of Tulsi, where bacterial growth has been explored widely. Therefore, there is a scope to comprehensively explore its effectiveness against fungal growth. Research examining Tulsi's efficacy against various types of fungi through a quantitative study will benefit the textile industry. It can also provide a more holistic understanding of its potential applications for fungal control in textiles.

- **Clinical validation and regulatory compliance**
  Conducting clinical trials to validate Tulsi-treated textiles' antimicrobial efficacy and safety in the real world is essential for gaining regulatory approval and consumer acceptance with different healthcare institutions and regulatory agencies to facilitate the validation process and ensure compliance with industry standards.

These future scopes can help researchers to further unlock the potential of Tulsi as a natural antimicrobial agent for textiles, contributing to the development of innovative, sustainable and functional textile solutions for diverse applications.

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