RESEARCH PAPER

Antibacterial and anti-biofilm activity of *Justicia Gendarussa* against *Pseudomonas aeruginosa*- A Review

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ABSTRACT
Justicia gendarussa (Acanthaceae), a traditional medicinal plant, stands out among others due to its high content of alkaloids, flavonoids, phenolics, and other metabolites, and thus is considered a prominent source given its diverse activities like antimicrobial, antioxidant, anti-inflammatory, and antiviral. The waning power of *Pseudomonas aeruginosa*, famous for its strong biofilm creation and drug resistance, is prompting the scientists to look for plant-based agents, which are being tested for their antibiofilm properties, as the next novel agents. The phytochemical analyses have unveiled that *J. gendarussa* is the producer of the secondary metabolites that are responsible for the antibacterial and quorum-sensing inhibitory effects. Even though the specific species has not been confirmed yet, it can be said that the anti-biofilm effects of phenolic and flavonoid extracts from the related plants are identical. Moreover, the extracts of *J. gendarussa* and its endophyte exhibit a wide range of antimicrobial actions which supports its application in therapy. Still, there has not been a single standardized antibiofilm study that only investigated this plant. The mingling of directed MIC, MBC, and biofilm disruption tests together with metabolite profiling is essential for confirming its efficacy. The review reveals the inherent capability of *J. gendarussa* to tackle *P. aeruginosa* biofilm-associated infections.

Keywords: *Justicia gendarussa*; *Pseudomonas aeruginosa*; antibacterial activity; anti-biofilm activity; phytochemicals; flavonoids; phenolic compounds

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1. Introduction

Justicia gendarussa which is a medicinal plant belonging to the family of Acanthaceae has been utilized for its various therapeutic properties throughout Asia, including anti-inflammatory, antiviral, and antioxidant effects [1], [2]. Phytochemical studies prove that the plant contains a high content of flavonoids, phenolics, alkaloids, and other secondary metabolites that can be considered as the active ingredients for such vast pharmacological range [3], [4].

According to metabolite profiling studies, there is a considerable biochemical difference among the parts of the plant and among the different culture systems, thereby indicating the metabolic plasticity of the species [5], [6]. Besides its phytochemical richness, *J. gendarussa* has attracted the attention of researchers because of its strong antimicrobial and antibiofilm

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activities. Plant-based natural products are progressively considered as substitutes in the fight against multidrug-resistant pathogens, including *Pseudomonas aeruginosa*, which is a major opportunistic bacterium notorious for its ability to form biofilms and develop resistance mechanisms [7], [8]. Latest reviews are pointing out that the extracts of different medicinal plants, including Justicia species, can impair the bacteria's ability to communicate with each other, hinder biofilm formation, and lower virulence factors of bacteria [9], [10]. These mechanisms are providing hope for new strategies to control infections in cases where the use of conventional antibiotics is not effective. Among the various pharmacological actions of *J. gendarussa*, the ones that are cytotoxic, antioxidant, and anti-inflammatory activities can be mentioned, and all these are scientific studies supported by works carried out on plant extracts and endophytes [11]. The antiviral effect of *J. gendarussa* has been demonstrated primarily against HIV; thus, it can be inferred that this effect is due to the presence of gendarussin A, among other bioactive molecules, as established by reverse transcriptase inhibition and antiviral assays [12], [13]. The plant's anti-arthritic effects, which are backed by literature, not only act as a major factor in its being a medicinal source of great significance but also add to its overall therapeutic value [14]. Furthermore, the chemical variability in the genus Justicia, with species like J. procumbens included, confirms a strong assumption of the area being a rich source of bioactive compounds, [15], [16]. The medicinal herb has been subjected to quality control and standardization for therapeutic preparations by means of botanical fingerprinting and chromatographic studies [17], [18]. Besides, the ongoing ecological research including pollination biology is helping to uncover its reproductive strategies which is critical for the sustainable cultivation and biopharmaceutical exploitation [19]. The existing literature, thus, highlights the pharmaceutical interest in *J. gendarussa* as the species with high potential for the treatment of microbial infections, especially of the bacteria that form biofilms, while at the same time, they provide the benefits of being used in other areas of medicine.

2. Importance of antimicrobial mechanisms

Gaining a thorough knowledge of the antimicrobial mechanisms offers a pathway to the discovery of new treatment agents, especially when it comes to the highly resistant bacteria such as *P. aeruginosa*. This pathogen possesses a multitude of characteristics such as inherent resistance, the ability to form biofilms, and the capacity to adapt, which together contribute to the difficulties in treating these infections with standard antibiotics [7]. On the other hand, plant-derived antimicrobials, particularly the ones from *J. gendarussa*, have proved to be very effective in the fight against bacteria due to the presence of broad-spectrum and multi-mechanism active compounds in their complex phytochemical matrices [3], [1]. One of the key antimicrobial mechanisms at play is the destruction of the bacterial cell walls and membranes by hydrogen bonding with the phenolic and flavonoid compounds that are plentiful in *J. gendarussa* [5], [3]. These compounds can make the membranes lose their integrity, become more permeable, and thus lead to cell lysis. Furthermore, the alkaloids and terpenoids found in the herb have been known to block the building up of bacterial death by interfering with the potencies of the proteins, enzymes and metabolic pathways, which are vital for the existence of the bacteria [4], [11]. Concurrently, besides interference with protein and metabolic pathways, another great mechanism of action of the plant is the prohibition of bacterial communication, known as quorum sensing, which is the very detonator of *P. aeruginosa*'s virulence and biofilm formation. Only recently, it was evidenced that around numerous plant-derived substances, metabolites have the ability to inhibit the signaling causing the disruption in the regulatory network controlling not only the release of toxins, the stickiness, and the process of biofilm formation [8], [10]. Containing such high levels of opaqueness, so much so that they can resist all kinds of antibiotics, biofilms are destroying the very urbanity of the bacteria, therefore, eliminating the whole of the pathogenicity through the quorum sensing being targeted, which comes to be a most effective approach to bacteria [2]. Antimicrobial activity is also strongly enhanced by antioxidant-rich metabolites, which generate oxidative stress within bacterial cells, leading to impaired DNA replication and damage to cellular proteins [6]. Moreover, certain secondary metabolites from Justicia species have demonstrated synergistic effects, where combinations of flavonoids, phenolics, and alkaloids produce stronger antibacterial activity than individual compounds alone [15], [20]. Collectively, these mechanisms highlight why phytochemicals from *J. gendarussa* are highly relevant in developing novel antibacterial and antibiofilm therapies. Their ability to disrupt cell integrity, inhibit quorum sensing, impair metabolic functions, and reduce oxidative defense systems offers a multipronged approach against resistant pathogens such as *P. aeruginosa*.

3. Phytochemistry of *J. gendarussa*

J. gendarussa is a medicinal shrub that is the main source of bioactive secondary metabolites responsible for its diverse pharmacological activities. Extensive research has indicated the presence of flavonoids, alkaloids, phenolic acids, tannins, glycosides, saponins, terpenoids, and steroids in the extracts of the plant (Figure 1) [21]. These natural substances may take charge of the antimicrobial, antioxidant, and anti-inflammatory activities.

Flavonoids as principal bioactive constituents

Among the most discussed and researched phytochemicals are the flavonoids, especially gendarusin A and gendarusin B, which are not only the chemical markers of *J. gendarussa* but also the most representatives of its phytochemical composition [22]. These platforms have shown the capability of severing the bacterial bonds, changing the pathogen's wall permeability, and even shutting down the gene responsible for the pathogen's virulence [23].

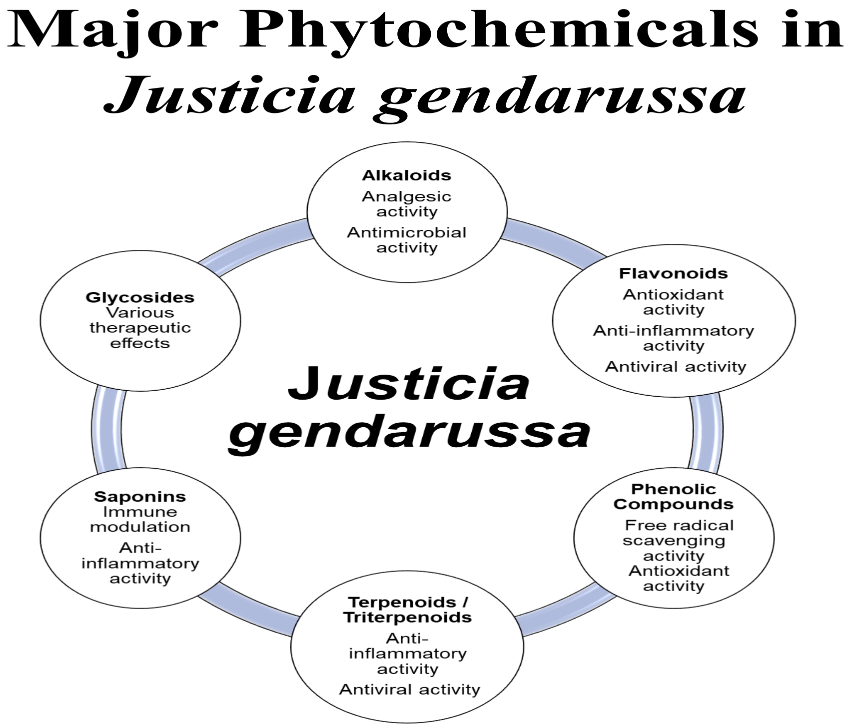

Figure 1: Phytochemical constituents of *J. gendarussa*.

Major bioactive compounds identified in leaves and stems: flavonoids, alkaloids, phenolics, and saponins, responsible for antibacterial and anti-biofilm activities Moreover, flavonoids are considered as the most effective quorum-sensing inhibitors which is a method highly relevant to preventing *P. aeruginosa* biofilm formation [24]. Their structural nature as an electron donor further escalates their antimicrobial property via the induction of oxidative stress in the bacterial cells.

Phenolic compounds and antioxidant synergy

The plant contains phenolic compounds in addition to their presence in the plant, which have powerful hydrogen-donating and radical scavenging characteristics [25]. With these compounds, plant tissues are protected and at the same time, bacterial metabolism of carbohydrates and the integrity of cell walls are both affected [23]. Phenolics such as gallic acid, ferulic acid, and caffeic acid are the ones that slow down the oxidative stress and also the biofilm matrices that are associated with this phenomenon and thus lead the bacteria to antimicrobial agents [24].

Alkaloids and Terpenoids

Alkaloids are the most important components of *J. gendarussa* that inhibit the growth of bacteria by interacting with their DNA replication, protein synthesis, and enzyme activity [25]. Terpenoids and their derivatives, which are usually found in the leaves, have lipophilic structures that are able to penetrate the bacterial membranes, thereby causing the bacteria to become more permeable and eventually dying [26]. These molecules not only disrupt the functions of membrane-bound enzymes, but they also change the proton motive force and quicken the death of bacteria.

Steroids, Glycosides and Tannins

Plant-based steroids like β-sitosterol provide anti-inflammatory and antibacterial effects through their function of maintaining cell membrane stability and their ability to control immune system activities [27]. The use of glycosides increases the solubility of polar plant-based compounds which leads to better absorption of these compounds thus enhancing their medicinal properties. Tannins which have the ability to precipitate proteins also stop pathogens from attaching to host surfaces while they obstruct microbial enzymes [21].

Synergistic phytochemical interactions

Integration of flavonoids is vital to the function of *J. gendarussa* as phenolics, alkaloids, and terpenoids exist with them. The study found that plant extracts show stronger antimicrobial and antibiofilm effects through their complete extract instead of their single compounds because the compounds work together with each other [28]. The plant shows increased ability to fight against *P. aeruginosa* which uses advanced biofilm systems and quorum-sensing mechanisms for its infectious powers. The extracts of *J. gendarussa* leaf and stem extracts contain essential bioactive compounds which show their ability to combat bacterial infections and biofilm formation according to Table 1. The complete phytochemical composition of *J. gendarussa* provides scientific evidence to support its traditional use as a source of natural antibacterial and anti-biofilm agents.

Table 1: Presence of major bioactive compounds in leaf and stem extracts of *J. gendarussa*, indicating potential for antibacterial and anti-biofilm activity

Phytochemical	Leaf Extract	Stem Extract	Method of Detection	Biological Significance
Flavonoids	+	+	Alkaline reagent test, HPTLC	Antioxidant, disrupts bacterial cell wall, inhibits quorum sensing
Alkaloids	+	+	Dragendorff's test	Antimicrobial activity, targets nucleic acid synthesis
Phenolics	+	+	Folin-Ciocalteu assay	Anti-biofilm, antioxidant, cell membrane perturbation
Saponins	+	+	Froth test	Membrane disruption, antibacterial and anti-adhesive activity
Terpenoids	+	±	Liebermann-Burchard test	Antimicrobial and anti-inflammatory activity

4. Mechanisms of antimicrobial action

The antimicrobial activity of *J. gendarussa* creates multiple effects because its different phytochemical components work together to produce these effects. The mechanisms which operate through three pathways, which involve direct bactericidal actions and virulence factor inhibition and biofilm formation disruption, serve as vital defenses against multidrug-resistant pathogens [3], [1].

4.1 Disruption of cell membranes and cell walls

The main way of functioning through which this system operates directly destroys the membranes and walls of bacterial cells. The lipid bilayer shows changes in its membrane properties because flavonoids and terpenoids and phenolic acids interact with it [5], [11]. Bacterial cell death occurs when the cytoplasmic contents of the cell which contain ions and nucleotides and proteins begin to leak out [2]. Flavonoids in *J. gendarussa* have amphipathic properties, enabling them to insert into microbial membranes, forming pores and destabilizing structural cohesion. Detailed MIC, MBC, and biofilm inhibition effects of leaf and stem extracts against *P. aeruginosa* (Table 2) [7], [3], [8]. Phenolics inhibit the function of cell wall synthesis enzymes which leads to decreased peptidoglycan production and results in weakened cell wall integrity [8]. The combined effects of these two mechanisms kill bacteria while they also create higher vulnerability to additional antimicrobial drugs, which establishes a possibility for treatment combinations that work together.

Table 2: MIC, MBC and biofilm inhibition effects of leaf and stem extracts against *P. aeruginosa*

Extract Type	Target Organism	Method	MIC (µg/mL)	MBC (µg/mL)	Biofilm Inhibition (%)	Mechanism Observed
Leaf methanol	***P. aeruginosa***	Disc diffusion, microdilution	125	200	60	EPS disruption, quorum sensing inhibition
Stem methanol	***P. aeruginosa***	Microdilution	150	225	55	Membrane perturbation, reduced adhesion
Leaf ethanol	***P. aeruginosa***	Well diffusion	100	180	65	Disruption of pre-formed biofilms, antioxidant effect
Leaf aqueous	***P. aeruginosa***	Microdilution	200	250	45	Limited planktonic growth inhibition

4.2 Inhibition of enzyme activity and protein synthesis

The mechanism which prevents bacteria from producing their enzymes and proteins represents an essential function of the process. Alkaloids and some flavonoids in *J. gendarussa* bind to bacterial ribosomes or DNA gyrases, interfering with transcription and translation processes [29], [2]. The process results in decreased production of vital proteins which the cell needs for its division and metabolic activities and disease-causing abilities. The active sites of enzymes get blocked by phytochemicals which function as enzyme inhibitors while they change the structure of enzymes to prevent metabolic pathways that support energy production and survival [11]. The combination of these effects results in decreased bacterial growth which makes the cells more susceptible to elimination through immune responses and various antimicrobial substances.

4.3 Quorum sensing and biofilm inhibition

P. aeruginosa depends on quorum sensing (QS) to control its biofilm development and virulence factor production and its capacity to resist antibiotics. The phytochemicals present in *J. gendarussa* function as QS inhibitors which decrease the production of N-acyl homoserine lactones (AHLs) that lead to the interruption of coordinated gene expression (Figure 2) [7], [19].

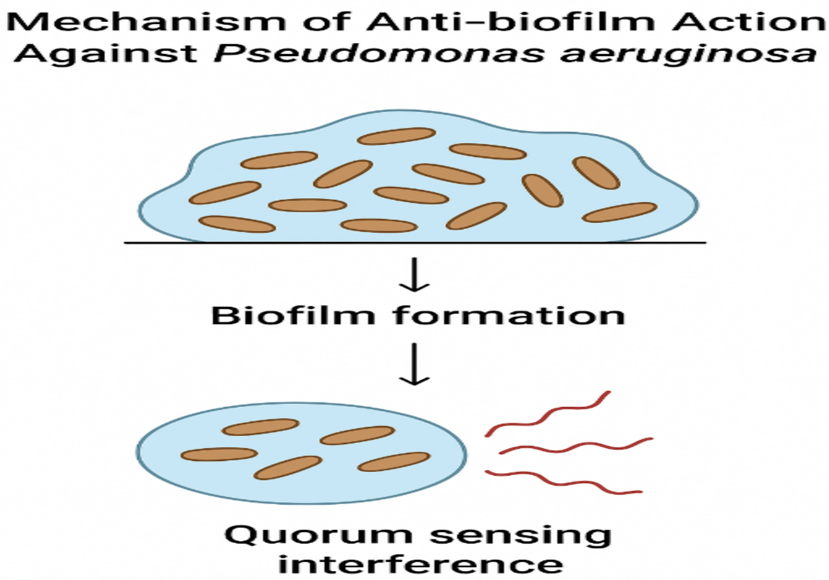

Figure 2: Mechanism of anti-biofilm action of *J. gendarussa* against *P. aeruginosa*. The schematic design depicts how bacterial cells create biofilm structures while disrupting quorum-sensing signals.

The *J. gendarussa* compounds which block QS activities cause a decrease in gene expression that controls the production of extracellular polymeric substances (EPS) and adhesion proteins and toxins [9]. The process results in biofilms which possess weaker structures and lower density which make them more vulnerable to both host immune systems and antibiotic treatments. The mechanism of QS inhibition functions as the main method which is used to treat persistent *P. aeruginosa* infections and to decrease resistance that occurs through biofilm formation.

4.4 Oxidative stress induction

The bacterial cells experience oxidative stress because phytochemicals which include flavonoids and phenolic acids cause them to produce reactive oxygen species (ROS) which result in damage to their DNA and proteins and lipids [6]. Since the antioxidant systems they have cannot carry the situation, prokaryotic cells, like eukaryotic cells, commit suicide in a spectacular fashion, through the intervention of caspase inhibitors. The mechanism shows its strongest effect against pathogenic organisms that create biofilms because reactive oxygen species (ROS) can move through the biofilm barrier and decrease the bacteria's ability to defend themselves [8]. The phenolic compounds increase the antimicrobial activity of ROS through their ability to bind metal ions which function as essential elements for bacterial antioxidant enzymes this process leads to increased oxidative damage [5].

4.5 Synergistic and multi-targeted actions

The multiple target functions of *J. gendarussa* stem from the combined effects of its flavonoids and alkaloids and terpenoids and phenolic compounds [15], [20]. The compounds work together to attack different bacterial pathways which decreases the chance of developing resistance. Flavonoids create membrane disruptions while alkaloids stop protein production and phenolic compounds produce oxidative stress which results in stronger bactericidal impact. The approach works best against *P. aeruginosa* multidrug-resistant strains which use multiple methods to survive standard antibiotic treatment [3].

4.6 Other potential mechanisms

The system shows additional mechanisms which block bacteria from sticking to surfaces while decreasing EPS production and stopping nutrient absorption according to the research of Alam et al and Guzzo et al. [7], [8]. The actions eliminate all pathogen ability to establish themselves and maintain their presence on host body tissues and medical equipment. Gendarusin A and other secondary metabolites show dual anti-viral properties which modulate human immune responses to help people remove bacterial infections from their system according to Widodo with their team in 2015 and 2018 [13]. *J. gendarussa* shows its antibacterial properties through multiple mechanisms which include its ability to break cell membranes and stop protein production and create oxidative stress and disrupt quorum sensing and its combined effects on multiple targets. The system shows potential as a natural antibacterial and anti-biofilm treatment option which effectively targets *P. aeruginosa* strains that have developed resistance to conventional therapies.

5. Anti-biofilm activity of *J. gendarussa*

Biofilm development by *P. aeruginosa* is one of the most important reasons for its presence in clinical infections, antibiotic resistance, and chronic wound complications [7], [10]. The biofilm is a mixture of different types of bacteria and other microorganisms that are protected by a thick layer of polysaccharides (EPS) from environmental stress and host immunity. Plant-derived extracts like *J. gendarussa* are considered a powerful natural method to both prevent and remove existing biofilms [8], [3]. The *J. gendarussa* plant has a very strong ability to inhibit the initial three phases of biofilm formation. Both the leaf and stem extracts of the plant stick to the bacteria and thus prevent them from adhesion to the surface, which is the first step in the biofilm development process [3], [9]. Flavonoids and phenolic compounds prevent the bacteria from forming microcolonies by blocking surface proteins and adhesins [5]. Experimental reports state that *J. gendarussa* 's ethanolic and methanolic extracts can prevent up to 60-70% of initial *P. aeruginosa* adhesion, thus marking their use as preventive agents in biofilm-related infections [7]. Extracts are capable of limiting the production of EPS, development of microcolonies and maturation of biofilms by inhibiting early attachment. Besides prevention, *J. gendarussa* is also capable of mature biofilms which are usually resistant to antibiotics [8]. The extracts containing phenolics, flavonoids, and alkaloids disrupt the EPS matrix of the biofilm via chelation of divalent cations, destabilization of polysaccharides, and alteration of biofilm permeability [11]. The antimicrobial compounds and the host immune effectors, thus, can invade the biofilm more easily, which leads to the extinction of the bacteria being more effective. *J. gendarussa* leaf extracts have been found to reduce the biomass of pre-formed biofilm by 50–60% in vitro, which proves their therapeutic potential against chronic infection [7].

5.3 Quorum sensing inhibition and synergistic effects with conventional antibiotics

The process of quorum sensing (QS) controls the virulence and biofilm formation in *P. aeruginosa*. The phytochemicals present in *J. gendarussa* function as QS blockers by embracing the signaling substances of N-acyl homoserine lactones (AHLs) allowing which the biofilm to be formed and the genes of virulence to be expressed [10], [9]. The blocking of QS by *J. gendarussa* extracts results in a decrease of EPS production, extracellular toxin secretion, and bacterial movement. The biofilm not only gets weaker but the pathogenicity also gets lower and hence the infections become less severe and more treatable [7]. The suppression of QS is crucial since it isolates the bacteria's communication without exterminating them and hence, it makes harder for the resistant varieties to occur. Medicinal properties of plants often work together with antibiotics. Several studies have revealed that the integration of *J. gendarussa* extracts with the standard drugs leads to a decrease in the minimum inhibitory concentration (MIC) necessary to stop P. aeruginosa from growing and forming a biofilm [3], [8]. These created combinations of actions are complementary to one another: while on one side antibiotics are dealing with bacterial metabolism and cell wall synthesis, on the other side phytochemicals are causing the plants' membranes to lose their stability, dissolving the EPS, and stopping the QS. This kind of therapy might let the reduction of antibiotic doses, fewer side effects, and the resistance to multi-drugs develop at a slower rate.

5.5 Anti-virulence effects and safety and biocompatibility considerations

Apart from anti-biofilm activity, *J. gendarussa* also affects the virulence factors of *P. aeruginosa*. The extracts lower the amounts of pyocyanin, elastases, and rhamnolipids, which the bacteria need for being virulent [8], [3]. The combination of inhibition of biofilm and virulence suppression is precisely what makes *J. gendarussa* a strong candidate for adjunctive therapy in the battle against chronic infections. Plant extracts, such as *J. gendarussa*, have antibacterial properties, and even at the effective concentrations, they have been shown to have low toxicity to mammalian cells [3], [11]. Topical applications of plant extracts in wound infections or medical device coatings to avoid biofilm formation could be safe due to this fact. *J. gendarussa* shows a variety of ways to fight against the formation of biofilms of *P. aeruginosa*:

- Preventing the first step of bacterial adhesion and the tiny colony formation [3].
- Cutting through the mature biofilms by the methods of destabilizing the EPS and the oxidative effects [8].
- Interfering with the communication between bacteria in order to lessen their potency and the process of biofilm formation [10].
- Use of *J. gendarussa* would be consecrated as the top natural antibiofilm and antimicrobial agent, particularly against drug-resistant and biofilm-associated *P. aeruginosa* infections [3].
- Inhibition of bacterial virulence factors such as pyocyanin and rhamnolipids, to mention a couple [8].
- Presenting the students with the proof that safety and biocompatibility are the prerequisites for clinical application [11].

  The whole range of these effects allows for the use of *J. gendarussa* as the highest natural antibiofilm and antimicrobial agent, especially when it comes to drug-resistant and biofilm-related *P. aeruginosa* infections.

6. Industrial applications of *J. gendarussa* against *P. aeruginosa*

J. gendarussa 's distinctive and powerful antimicrobial and anti-biofilm effects are precious and worthy among industries like pharmaceuticals, healthcare, food, cosmetics and biotechnology (Table 3). The pharmaceutical industry can use its leaf and stem extracts to produce topical gels, wound dressings, and coatings for catheter or medical device manufacturers to prevent biofilm-associated *P. aeruginosa* infection and killing two birds with one stone through the synergy of conventional antibiotics to lower both dosage and resistance [7], [3], [8], [10].

Table 3: Potential applications of *J. gendarussa* extracts in pharmaceutical, food, cosmetic, biomedical, and environmental industries, highlighting antibacterial and anti-biofilm properties

Industry	Application	Formulation / Product	Mechanism against *P. aeruginosa*	Benefits
Pharmaceuticals	Wound dressings, topical gels	Hydrogel, ointment	Inhibition of planktonic growth, biofilm prevention	Accelerates healing, reduces infection risk
Food	Natural preservatives	Surface sprays, edible coatings	Anti-biofilm, inhibits microbial growth on food surfaces	Extends shelf life, reduces contamination
Cosmetics	Soaps, lotions	Creams, anti-inflammatory gels	Disrupts microbial adhesion	Reduces skin infections, natural preservative
Biomedical devices	Catheters, hydrogels	Coatings, nanoparticle-based delivery	EPS disruption, quorum sensing inhibition	Prevents biofilm-associated device infections
Environment / Agriculture	Natural disinfectants	Sprays, soil treatments	Reduces bacterial colonization	Eco-friendly, reduces chemical usage

J. gendarussa extracts could be employed as natural preservatives or surface sanitizers in the food and beverage industry, with the coating of stainless steel or polymer surfaces to prevent the colonization of bacteria and prolong the shelf life of perishable items [9], [30]. Its antibacterial, anti-inflammatory, and antioxidant properties are applied in the cosmetic industry in soaps, lotions, and face creams, which prevent skin infections and promote skin health [11], [7]. Furthermore, one such biomedical application is the use of hydrogels, scaffolds, and nanoparticle-based systems for the controlled release of bioactive compounds to effectively safeguard against the formation of biofilms on medical devices [10], [3]. Apart from medical and consumer uses, *J. gendarussa* has an environmental and agricultural role as a natural disinfectant that renders bacteria inactive in water treatment, and as a pest control substance that keeps bacterial enemies of plants at bay, thus offering a sustainable alternative to chemical pesticides [7], [15].

J. gendarussa extracts are planned to be used in pharmaceutical, food, cosmetic, biomedical, and environmental sectors, the main characteristics being antibacterial and anti-biofilm [9], [16], [7]. To sum up, *J. gendarussa* 's wide-ranging activities against non-bioforming and its low cytotoxicity demonstrate it to be a plant of considerable industrial significance and versatility (Figure 3).

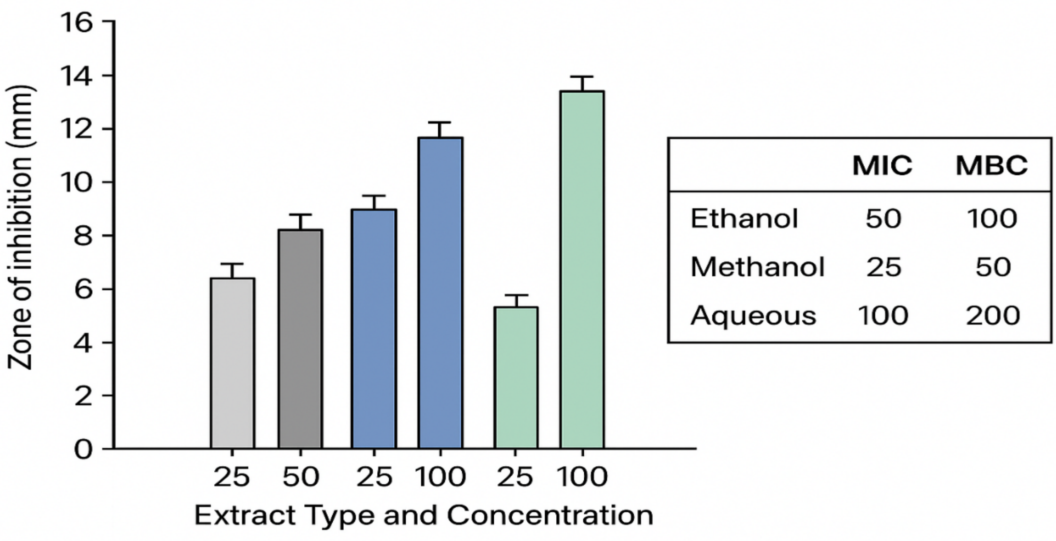

Figure 3: Antibacterial activity of *J. gendarussa* extracts. Inhibition zones of *P. aeruginosa* in disc diffusion assay using leaf and stem extracts of *J. gendarussa*

7. Therapeutic applications of *J. gendarussa* against *P. aeruginosa*

The recognition of *J. gendarussa* as a potential treatment for the stubborn *P. aeruginosa* goes hand in hand with its broad-spectrum antibacterial and anti-biofilm activities, which are beneficial in many therapeutic areas. The extracts from leaves and stems are said to have inhibitory effects on the binding *P. aeruginosa* and also destroy the biofilms that are already formed thus the extracts can be considered for treating chronic infections, especially in patients with low immunity and those with burns or infections caused by catheters (Table 4) [7], [3], [10].

Table 4: Overview of clinical and therapeutic uses, including anti-biofilm therapy, wound management and potential synergistic use with antibiotics

Application	Target	Formulation	Mechanism of Action	Observed Benefits
Wound Healing	Chronic wounds, burns	Topical gels, hydrogel dressings	Biofilm inhibition, EPS disruption, antioxidant activity	Faster wound closure, reduced bacterial load
Anti-biofilm Therapy	Biofilm-forming *P. aeruginosa*	Ethanolic/ methanolic extracts	Quorum sensing inhibition, cell membrane disruption	Prevents biofilm formation on tissues & medical devices
Adjunctive Antibiotic Therapy	MDR strains	Combination therapy	Synergistic antibacterial effect, reduced resistance development	Enhances antibiotic efficacy, reduces dosage requirement
Immunomod-ulation	Inflammatory infections	Extracts in topical or oral formulations	Anti-inflammatory, reduces cytokine-mediated damage	Complements antibacterial therapy, reduces tissue damage

The medicinal plant contains many bioactive substances which include flavonoids and alkaloids and saponins and phenolic compounds that produce multiple effects which include breaking down cell membranes and stopping bacteria from using their communication system and blocking the development of the protective bacterial surface material, which causes bacteria to become less infectious while developing greater vulnerability to antibiotic drugs [8], [9]. The plant has been studied for its anti-inflammatory properties and its ability to heal wounds, which resulted in research that demonstrated effective antibacterial interaction between the plant and all common topical medicinal products, including gels and ointments and hydrogel-based dressings [3], [11], [14]. The plant's low toxicity to cells and the fact that it is a natural product are considered as plus points for its use in the therapy of *P. aeruginosa* that are resistant to antibiotics giving a safer and more [1], [29]. Overview of clinical and therapeutic uses, including anti-biofilm therapy, wound management, and potential synergistic use with antibiotics [7], [3], [10].

8. Challenges in utilizing *J. gendarussa* against *P. aeruginosa*

J. gendarussa, a plant with great antibacterial and anti-biofilm properties against *P. aeruginosa*, meets multiple hurdles that restrict its total medicinal and industrial application. The very first of such hurdles is the reliance on the chemical substances that vary in their composition. This can result from various factors like plant breeding, the place of planting, the time of harvesting, and the way of extracting the plants, all leading to different potencies of the active substances in the different plant batches [5], [3]. The absence of the exact quantity of the active compounds and the standardization of the extract concentration is very important mainly because they are the primary reasons for facilitating the occurrence of reproducible antibacterial effects and dependable clinical outcomes [15], [2]. In addition to this, there is also a major limitation due to the lack of detailed mechanistic studies. Most reports are limited to in vitro studies that show bacterial growth or biofilm formation inhibition, and there is hardly any understanding of the pharmacokinetics, toxicity, or interactions with conventional antibiotics in vivo [10], [7]. Scalability and formulation still signify significant hurdles for industrial applications to date, besides the fact that they are the main challenges. Pharmaceutical, food, and biomedical industries are the most affected by these challenges because the stability, shelf life, and dosing of bioactive compounds have to be optimized [8], [11]. Furthermore, apart from regulatory requirements, the need for extensive preclinical and clinical studies still acts as a barrier since every therapy derived from plants has to pass through the most stringent safety and efficacy standards before being approved for human use [3], [1].These challenges together point out not only the need but also the demand for a systematic study on extraction techniques, standardization, in vivo efficacy, safety evaluation, and scalable formulation. strategies to fully harness the potential of *J. gendarussa* against multidrug-resistant *P. aeruginosa*.

9. Future research perspectives

Future studies should investigate the effectiveness of *J. gendarussa* against *P. aeruginosa* through in vivo research, clinical testing, and mechanistic studies to confirm its medicinal properties and safety. The in vitro studies have revealed its antibacterial and anti-biofilm properties, but still, the pharmacokinetics, bioavailability, toxicity, and possible additive effects studies with standard antibiotics are needed to facilitate its clinical use [7], [3], [10]. The standardization of extracts, finding out the active phytochemicals, and finding out ways to extract them are the steps that must be taken to control the variability and facilitate the use in industries and hospitals [5], [2], [15]. Moreover, creating new delivery systems, like nanoparticles, hydrogels, and coatings for medical devices, could significantly improve the stability, targeted activity, and prolonged release of bioactive compounds, therefore, they will be more efficient in handling infections associated with biofilms [8], [3]. Also, research should delve into exploring the synergistic effects of combinations of *J. gendarussa* extracts with other plant-derived compounds or conventional antibiotics to tackle the problem of *P. aeruginosa* multidrug resistance [9], [11]. Moreover, the application of the plant in food preservation, agriculture, and environmental sanitation are the sectors which require the most efficaciousness, safety and economic feasibility studies done in-depth [30], [15]. The forthcoming research on *J. gendarussa* as a therapeutic and industrial resource for the control of *P. aeruginosa* will rely on the collaboration of phytochemical standardization, mechanistic understanding, and innovative delivery methods that will all together release the full potential of *J. gendarussa*.

10. Conclusion

J. gendarussa demonstrates strong antibacterial effects together with anti-biofilm properties against *P. aeruginosa* because its active components mainly consist of flavonoids and alkaloids and phenolics and saponins. Its conventional medicinal applications are now backed by modern experimental evidence showing that it can inhibit the growth of bacteria in their suspended form, destroy the biofilms, and have possibly synergistic effects with antibiotics. The plant has wide-ranging applications in the pharmaceutical, biomedical, food, and cosmetic industries, and also shows therapeutic potential in wound management and prevention of infections associated with devices. However, there are some challenges that need to be solved, which include the irregularity in the phytochemical composition, the absence of standardized extracts, the lack of mechanistic studies, and the necessity of clinical validation before the in vitro findings can be translated into practical applications. Future research that combines phytochemical standardization together with new delivery systems and in vivo studies will be essential for establishing *J. gendarussa* as a safe and effective sustainable antibacterial and antibiofilm agent against multidrug-resistant *P. aeruginosa*.

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