

# ANTIBIOFILM ACTIVITY OF *JATROPHA MAHESHWARII*- AS AN ENDEMIC PLANT

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## Abstract

Bacterial biofilms can harm humans in hospital environments, the food industry, and drinking water systems. The study aimed to examine the antibiofilm activity of *Jatropha maheshwarii* as an endemic plant. Biofilms were formed by *Staphylococcus aureus* and *Pseudomonas aeruginosa* on the surface of medical biomaterials. Moreover, time-dependent eradication of biofilms performed in polystyrene 96-well culture microplates was examined and expressed as minimal biofilm eradication concentration. The tested staphylococcus aureus formed the highest antibiofilm activity against biofilm at 77% in aqueous extract and the ethanol extract at 73% in different concentrations ( $\mu\text{g/ml}$ ). The application of antibiofilm activity spans various fields, including medicine, the food industry, and environmental management.

**Keywords:** *Jatropha maheshwarii*, Antibiofilm activity, Gram-positive and Gram-negative bacteria

## INTRODUCTION

Biofilms, which are self-produced matrices of various organic chemicals, provide a significant problem (Simões et al., 2010). Biofilms, which are frequently seen in several situations such as industrial and clinical settings, Food processing conditions and drinking water distribution systems, generally create a sticky gel comprised of polysaccharides, proteins, and other organic components on a moist surface (Kavanaugh & Ribbeck, 2012; Oral et al., 2010). According to Gilbert et al. (2011), biofilm-associated cells can attach permanently to a range of surfaces, such as living tissues and indwelling medical devices such as valves, prostheses, catheters, etc. A biofilm is a population of microorganisms adhered to a surface: within the biofilm, bacteria are cocooned in a self-produced extracellular matrix made up of extracellular polymeric substances that, along with carbohydrate-binding proteins, pili, flagella, other adhesive fibres, and extracellular Deoxyribonucleic Acid (DNA), serve as a stabilizing scaffold for the three-dimensional (3-D) biofilm structure. By shielding biofilm bacteria from innate immune responses and antibiotic therapies, the matrix facilitates close cell-to-cell communication and DNA exchange that aid in spreading virulence

factors and medication resistance. As a result, biofilm-forming microorganisms persist, causing chronic and resistant infections. Biofilm-forming pathogen infections can be disastrous, causing severe symptoms and, in many cases, death (Clinton & Carter, 2015; Fisher et al., 2010; Kostakioti et al., 2013).

*Staphylococcus aureus*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Escherichia coli* are some of the most prevalent biofilm-forming bacteria in clinical settings (Wolcott et al., 2010; Sanchez et al., 2013; Donlan et al., 2002). Human *Pseudomonas aeruginosa* is an opportunistic bacteria that has emerged as a significant source of nosocomial infections, including infections of artificial implants, contact lenses, and catheters for the urinary tract (Kipnis et al., 2006; Davey et al., 2003). (Davey et al., 2003). The Gram-positive bacterium *Staphylococcus aureus* develops on the addition; it can play a role in various illnesses such as meningitis, epidural abscesses, urinary tract infections, toxic shock syndrome, and septic thrombophlebitis (Gajd' acs, 2019). Asfar et al. (2014) found that bacteria protected by biofilm exopolysaccharides are up to 1,000 times more susceptible to antibiotics than cells in plankton, which has significant implications for therapy and dramatically complicates treatment options (Russell et al., 2008). Microbial biofilms are responsible for a wide range of issues in various medical, environmental, and industrial contexts. These issues include the colonization of artificial medical implants, fouling of ship hulls, and clogging of industrial piping (Costerton et al., 1999; Kumar & Anand, 1998), among many other issues. This study examined the antibacterial, anti-biofilm, anti-quorum sensing, and synergistic properties of 23 plant extracts.

*Jatropha* belongs to the family Euphorbiaceae and is distributed in India at 13 sp (Abdul Kader, 2014). It is widely distributed in the red clay soils of the southeast coast of Tamil Nadu (Kanyakumari, Thoothukudi, and Tirunelveli), India, and it is related to the petroplant *Jatropha curcas*. It is commonly known as 'Athalai' (Ahmedullah & Nayar, 1986), 'Vel-thali' (Ben et al., 2014), and 'Kattamannaku' (Maria et al., 2014) in Tamil. It is an evergreen shrub that is 200cm in size with a thick stem and dark green leaves with ovate-lanceolate. The aerial parts of this plant have some traditional medicinal properties, and it is used by natives as an insecticide to treat ringworms, eczema, and rheumatism (Ben et al., 2014). Maria Sumathi and Uthayakumari, 2014 found a light green viscid fluid (latex) in the aerial part of the stem, and the rural folk use it for curing numerous ailments like skin diseases, tooth infections, and haemorrhages.

The current study's objective was to assess the anti-biofilm activity of a crude extract from the plant *Jatropha maheshwarii* against microbial infections that are clinically significant.

## MATERIALS AND METHODS

### Collection of plant

The aerial parts of *Jatropha maheshwarii* were obtained from Periyathalai (Lat. 8.33690N Long. 77.97240E), Tuticorin District, Tamil Nadu. Typical flora verified the sample. After thoroughly washing the samples with distilled water, they were allowed to air in the shade for two weeks. The dried leaves were finely blended using a mechanical blender before being stored in an airtight container.

## Microbial pathogens

Bacterial strains of *Echerichia coli*, *Psuedomonas aeruginosa*, *Staphylococcus aureus*, *Salmonella typhii*, and *Bacillus sp.* originated from the Department of Microbiology at Bharathidasan University in Trichy, Tamil Nadu, and India. The gained cultures were kept on a nutrient agar medium.

## Extract Preparation

Ten grams of plant powder were blended with 100 millilitres of distilled water at room temperature (28<sup>0</sup> C). Following 48 hours of agitation, the material was filtered using Whatman No. 1. Use filter paper. The filtrate was collected and used for future investigation. The exact process was used to make the ethanol extract.

## Anti-biofilm activity

Fungal and bacterial cultures were grown on a 96-well polystyrene microtiter plate. The autoclave has been utilized to dissolve and sterilize media with a Thermo Magnetic Stirrer. To culture bacteria, 20 µL was applied to each well of a sterile 96-well polystyrene microtiter plate at varied stock solution concentrations (12, 14, 16 µg/mL, and 80 µL nutrient broth) and incubated at 37°C for 24 hours (Brambilla et al., 2017).

## RESULT AND DISCUSSION

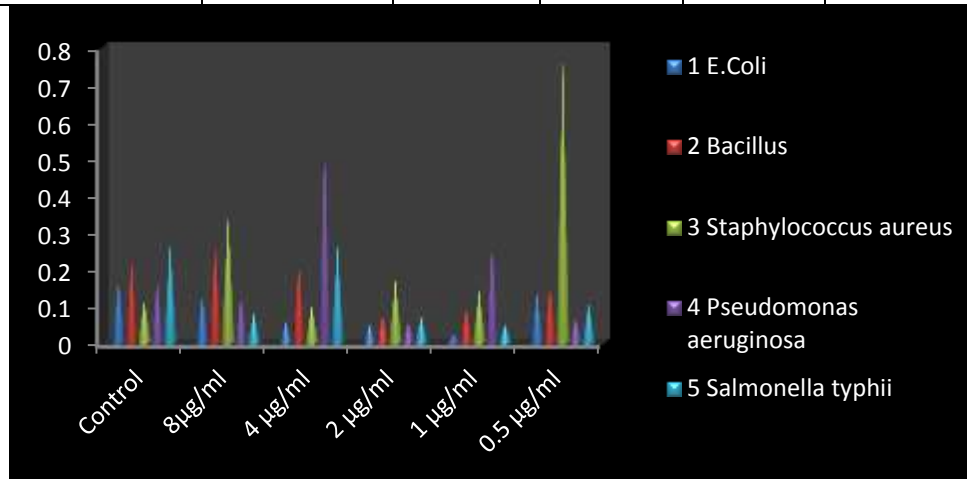
### Antibiofilm activity

The capacity to prevent biofilms from forming and spreading is known as antibiofilm activity. In this study, we looked at the antibiofilm properties of plant extracts from *Jatropha maheshwarii*. The aqueous and ethanol extracts are done on five bacterial strains of gram-negative and positive bacteria. The antibiofilm activity data are shown in Table 1 and Figure 1 for aqueous activity and Table 2 and Figure 2 for ethanol activity. There was also a control with nutritional broth (no bacterial culture). The formula utilized to calculate the biofilm development of extracts in bacteria  $(OD\ CONTROL - OD\ SAMPLE) \times 100$  was applied to five different concentrations of biofilm activity. A value greater than one indicates strong biofilm formation (no inhibition), 0.5 indicates moderate biofilm formation (less inhibition), and less than 0.5 indicates weak biofilm formation (potent inhibition). All plant extracts demonstrated considerable biofilm suppression against selected bacterial strains. As a combination of *Staphylococcus aureus* and *Pseudomonas aeruginosa*, whose biofilm development was severely disturbed by all of the plant extracts, suggesting that the plant extracts are significantly active in inhibiting biofilm forms firmly. All plant extracts severely disturbed the biofilm produced by these bacteria. According to Kuyucuklu and Eryildiz (2021), using molecules and plant extracts to prevent or decrease biofilm formation has become a potential therapy option in recent years. Khan et al. (2017) found that both *Mirabilis jalapa* and *Ajuga bracteosa* extracts had moderate (+++) and mild (+++++) effects on pellicle inhibition against the tested *P. aeruginosa* strains. Pattiyathane et al. (2009) investigated curcumin's effect on *Helicobacter pylori* biofilm both qualitatively and quantitatively using the pellicle test and crystal violet staining, indicating that it could be used as a supplementary medicine to treat *H. pylori* biofilm-related infections. According to Quave et al. (2008), ten different plants prevented *S. aureus* biofilm from forming. *R. officinalis*, one of these plants, prevented the production of biofilms at 8 µg/ml. Differences in antibacterial activity among plant species may be caused by

factors such as soil type and harvest time, which can influence the plant's chemical composition (Correa Jr, 1991). Pre-formed biofilms of *S. aureus*, MRSA, *E. coli*, and *C. albicans* were suppressed by *P. granatum* and ellagic acid, one of its ingredients (Bakkiyaraj et al., 2013).

**Table 1: Antibiofilm activity of aqueous extract from *Jatropha maheshwarii***

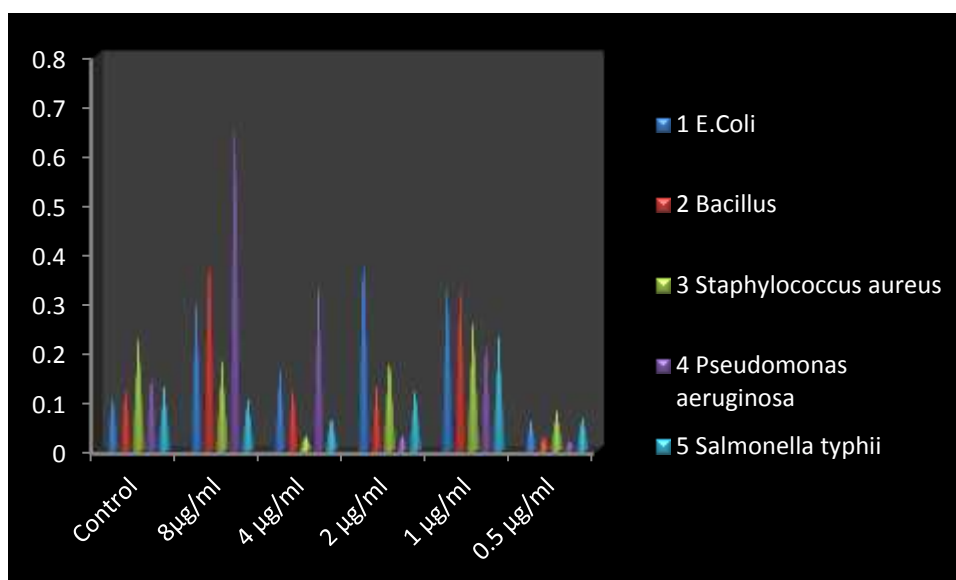
S.no	Name Of The Bacteria	Control	8µg/ml	4 µg/ml	2 µg/ml	1 µg/ml	0.5 µg/ml
1	<i>E.Coli</i>	0.176	13%	6%	5%	2%	13%
2	<i>Bacillus</i>	0.227	27%	22%	8%	10%	16%
3	<i>Staphylococcus aureus</i>	0.117	35%	10%	17%	14%	77%
4	<i>Pseudomonas aeruginosa</i>	0.176	13%	57%	6%	28%	7%
5	<i>Salmonella typhii</i>	0.271	8%	26%	7%	5%	11%



**Figure 1: Antibiofilm activity of aqueous extract from *Jatropha maheshwarii***

**Table 2: Antibiofilm activity of Ethanol extract from *Jatropha maheshwarii***

S.no	Name of the bacteria	Control	8µg/ml	4 µg/ml	2 µg/ml	1 µg/ml	0.5 µg/ml
1	<i>E.Coli</i>	0.117	30%	17%	43%	36%	6%
2	<i>Bacillus</i>	0.132	44%	13%	13%	35%	3%
3	<i>Staphylococcus aureus</i>	0.237	18%	3%	20%	27%	8%
4	<i>Pseudomonas aeruginosa</i>	0.166	73%	34%	3%	24%	2%
5	<i>Salmonella typhii</i>	0.129	11%	7%	13%	23%	7%



**Figure 2: Antibiofilm activity of ethanol extract from *Jatropha maheshwarii***

## CONCLUSION

The study's findings indicate that most plant extracts have antibiofilm efficacy because they can prevent the beginning stage of biofilm development and subsequent growth. While most extracts could prevent cell attachment, it was more challenging to stop the formation of a preexisting biofilm. To incorporate them as alternatives in the control of biofilms, isolating and identifying the ingredients that show antibiofilm capabilities may be necessary.

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