

Enhancing Agronomic Traits of Food Crops with Biofilm Biofertilizers: Promoting Sustainable Farming in Indonesia - A Systematic Review

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Abstract

Intensive use of inorganic fertilizers causes damage to soil ecosystem and environment. While, the utilization of biofertilizers holds significant potential in enhancing soil health and increasing crop yields as a viable solution for achieving more sustainable agricultural practices. This literature review explores biofilm biofertilizers (BBF) and plant growth promoting rhizobacteria (PGPR), examining their applications in improving soil-plant health, growth, and productivity. Employing Systematic Literature Review (SLR) methodologies, scientific literature from 2012 to 2022 was analyzed following PRISMA guidelines. Reputable journals indexed in Google Scholar and E-Journal Springer were selected, and relevant keywords were used to gather primary references. Results demonstrate that PGPR from various genera (*Serratia* sp., *Bacillus* sp., *Pseudomonas* sp., *Streptomyces* sp., and *Cyanobacteria*) produce growth promoters like indole-3-acetic acid (IAA), gibberellins, ethylene, and cytokinins. Combining BBF with chemical fertilizer can reduce chemical usage by up to 48%, reducing reliance on them. BBF containing PGPR exhibit potential to enhance agronomic of food crops traits and promote sustainable farming practices in Indonesia.

Keywords: Systematic review, Biofilm, Biofilm Biofertilizer, Sustainable farming, Indonesia



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INTRODUCTION

Agriculture plays a pivotal role in driving Indonesia's economy and is a crucial source of livelihood for a substantial portion of its population (Purnamasari & Hartatik, 2018). However, the excessive and indiscriminate use of chemical fertilizers in conventional farming practices has emerged as a significant concern, contributing to environmental degradation and posing potential health risks (Jaffri et al., 2021). The continuous application of chemical fertilizers has resulted in soil nutrient imbalances, water pollution through runoff, and the disruption of natural ecosystems (Firdaus et al., 2019). Efforts are being made to transition towards more sustainable agricultural practices in Indonesia, and the adoption of biofilm biofertilizers has gained attention as a potential solution (Bhardwaj et al., 2014). Biofilm biofertilizers, composed of beneficial microorganisms organized within a biofilm matrix, offer several advantages such as improved nutrient availability, enhanced soil health, and reduced environmental impacts (Parray et al., 2017). These biofertilizers promote the growth and development of crops while minimizing the dependency on chemical inputs (Kumar et al., 2022).

A material called fertilizer is added to farming fields to provide more nutrients for plants and environment. Fertilizers play a crucial role in modern agriculture, providing essential nutrients to plants for optimal growth and maximizing crop yields (Kumar et al., 2018). As the global population continues to grow, the demand for food and agricultural products increases exponentially (Krasilnikov et al., 2022). Synthetic fertilizers have a high concentration of soluble nutrients, which plants can absorb right away. Synthetic fertilizers, however, have the potential to interfere with soil biological processes, for example, by accelerating the degradation of organic matter in soils and deteriorating soil structure (Chittapun et al., 2018).

Since there is a limited amount of land accessible for crop cultivation, it is essential to adopt technology that can increase agricultural production potential while also providing benefits such a sustainable soil environment and economic effectiveness (Khanna et al., 2019).

Alternative environmentally friendly applications that allow for the reduction of chemicals have been used to make agriculture more sustainable. Treatments based on microorganisms are thought to increase the sustainability of food production. When opposed to chemical fertilizers, biofertilizers have less of an adverse effect on the environment, which makes them more appealing (Kholssi et al., 2021). Manufacturing of organic fertilizers has become more innovative every year. The application of biofilm biofertilizer as a biofertilizer decomposer is one of the innovations. Numerous helpful microorganisms, including nitrogen-fixing bacteria, phosphate solubilizing fungi, potassium solvent bacteria, and crop diseases management fungi, are present in biofilm biofertilizer. For usage as a starter or decomposer, the microorganisms are prepared in a unique carrier (Sudadi et al., 2020). Low density biofilms are present in the soil naturally, but they do not have a substantial impact. Therefore, it is crucial to generate biofilms in vitro and apply them as biofertilizers, also known as BBFs, to increase agricultural output in an environmentally benign way (Premarathna et al., 2021).

Compared to traditional biofertilizers, biofilm fertilizer has a stronger capacity to improve crop yields and is more resilient to environmental stresses, predators, and antagonists. The generation of plant growth chemicals (such IAA) and the suppression of plant diseases are all improved by biofilm-based biofertilizers relative to those that are not. Various nations have created and used biopesticides and biofertilizers to boost agricultural yields, fertilizer effectiveness, resource conservation, and food safety (Sudadi et al., 2018). In summary, biofilms are structured microbial communities that are attached to surfaces (Muhammad et al., 2020). Biofilms have unique architecture, are heterogeneous, and are resistant to physical and chemical stresses. They use quorum sensing to coordinate gene expression and regulate the formation and dispersal of biofilms (Bhatia et al., 2021).

The results and discussion of this review article offer several studies that show the beneficial effects of fertilization with biological agents that show the effects of using biofilm biofertilizers on agronomic traits and sustainable farming. The primary purpose of this study is to present scientific data on the potential use of to explore and to investigate the potential of biofilm biofertilizers as a sustainable solution to enhance the agronomic traits of food crops and promote sustainable farming practices in Indonesia.

RESEARCH METHODS

The study utilized a systematic literature review (SLR) methodology, following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow guidelines. The data for this study were sourced from various databases and indexes accessible through Google Scholar and Science Direct. The search was conducted on April 16, 2022, using specific keywords such as "Biofilm Biofertilizer" and "enhancing traits" and "food crops" and "sustainable farming, in Google Scholar. Additionally, an Science Direct search was performed using the same keywords to gather relevant literature on the topic of biofilm biofertilizers and their potential for enhancing agronomic traits in sustainable farming.

The screening criteria are based on the title and abstract of scientific articles that were published in research journals between the years of 2012 until 2022. Data on crop reactions to Biofilm Biofertilizer treatment is presented in the contents of all examined publications, together with results of tests on crop attributes. Control soil was used in the investigation, which was carried out in a greenhouse and field test. These modifications result in more treatments, bioagent fertilizers with various compositions, including single biological agents and consortia, as well as a combination of biological agents and chemical fertilizers.

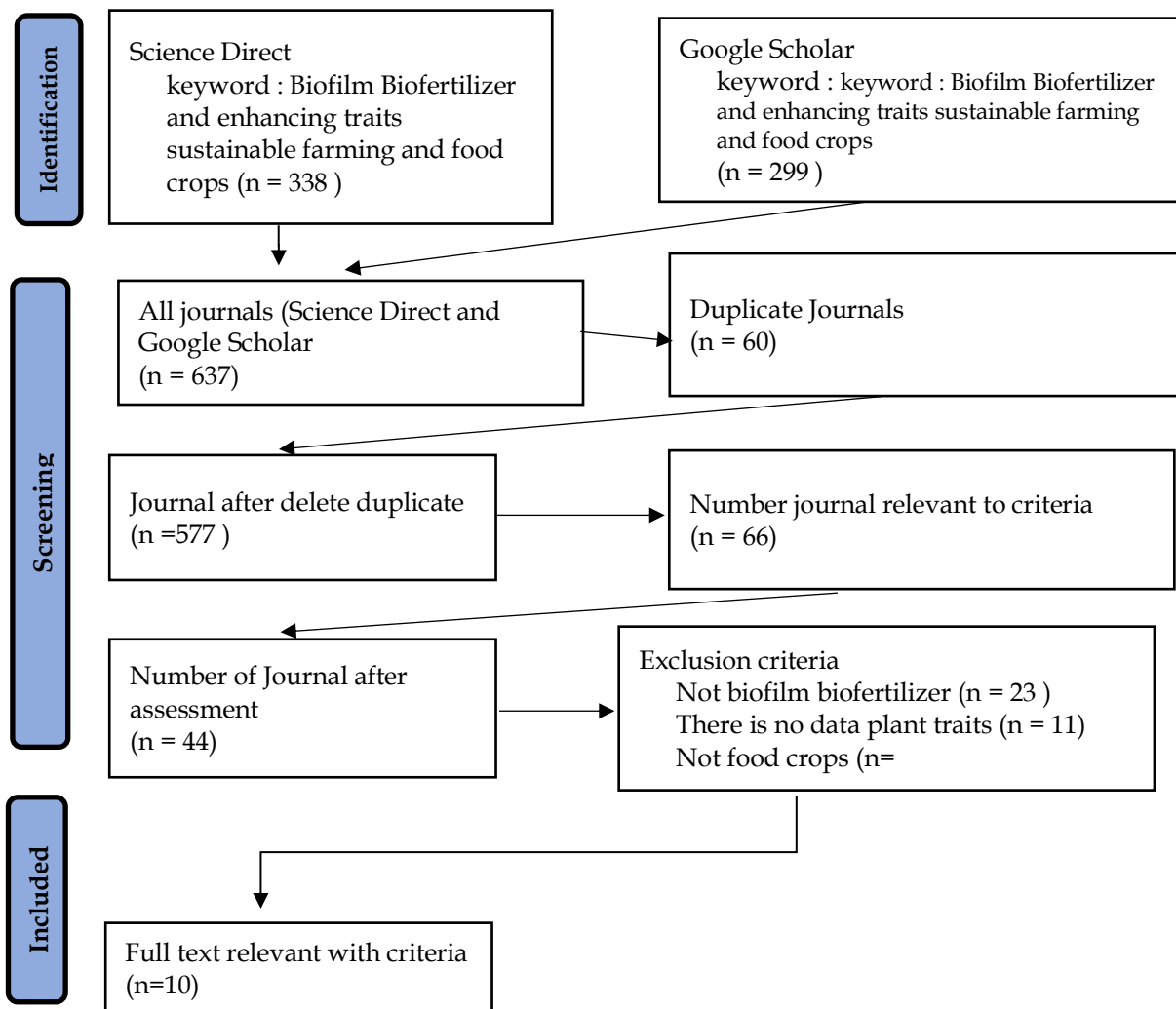


Figure 1. Flow chart Systematic Literature Review: Biofilm biofertilizers for enhancing the agronomic traits of food crops to promote sustainable farming

RESEARCH RESULT AND DISCUSSION

The results of a manual search for journals on Google Scholar yielded 1,610 journal titles, which were then vetted for relevance to the required theme to yield 338 journal names. Following a screening for the journal's publication date, the type of article (research article), and the type of access used (open access), a total of 299 research papers were found from the 1,093 results from the ScienceDirect search. Screening from the title method using plants resulted in 66 journal titles, followed by specific titles using sustainable agriculture, which resulted in 44 appropriate journal titles. The journals with information meeting the requirements were acquired in 10 titles after being fully screened. Every journal is an international publication. The findings demonstrated that using the biofilm biofertilizer responded favorably to the plant plant height, root length, and shoot length growth parameters. Biofertilizers are substances that include several bacteria helpful cells for agriculture they can also be referred to as manufactured products that include living or dead cells of effective microbe strains that enhance agricultural plants in absorbing different nutrients through their contact with the rhizosphere (Khanna et al., 2019). A biofilm is a colony of microorganisms and the extracellular polymeric substances (EPS) that the local bacterial population releases to defend the community. In addition to increasing crop output and improving soil quality, the use

of biofilm fertilizer can restore the livability of soil that has been compromised by conventional farming systems (Rathnathilaka et al., 2022)

There are two main mechanisms through which bacteria promote plant growth: direct and indirect mechanisms (Figure 2). The direct approach involves the bacteria's activities inside the plant, which impact hormone balances and enhance the host plant's cell proliferation, adaptive capacity, and nutrient absorption. These factors are crucial for optimal plant development, growth, and physiological processes.

On the other hand, the indirect mechanism of plant growth promotion occurs outside the host plant. This is achieved through the production of various substances by the plant growth-promoting bacteria, including antibiotics, hydrolases, and volatile chemicals. These substances play a role in inhibiting the growth of competing or pathogenic bacteria, thus creating a favorable environment for the host plant's growth and development (Soni & Keharia, 2021).

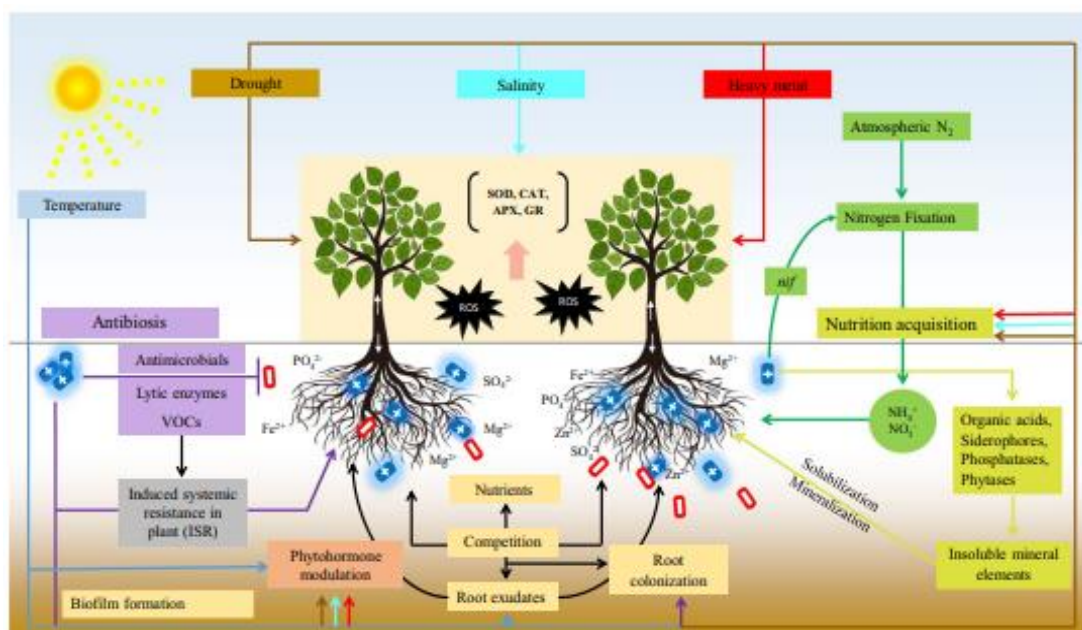


Figure 2. Direct and indirect plant growth-promoting pathways are shown schematically (Soni & Keharia, 2021)

Biofilm Formation

The process of forming a biofilm on roots involves four stages: adhesion to the surface, development of microclusters, maturation, and production of architecture or structure (Figure 3.). Initially the bacteria adhere to the surface of the roots or other plant parts and then multiply and form a thin layer (monolayer) biofilm. Bacteria will produce EPS which is useful for attaching the bacteria to a surface and sticking to each other by forming microcolonies. As time goes by, the colony forms a thickened layer and the microbes attached to the deepest tissues will be deprived of nutrients and then become a fungus that has channels or pores that can be passed by nutrients and metabolic products from all cells. Bacteria communicate with each other using quorum sensing. A mature biofilm has formed and consists of many bacterial species (Timmusk & Nevo, 2011). The biofilm process initiates with the initial attachment of bacteria and progresses to an irreversible adhesion. During these stages, extracellular DNA, proteolytic enzymes, cell membrane proteins, and biofilm-associated proteins are believed to play crucial roles.

The subsequent steps involve the generation of extracellular matrix (ECM) and the maturation of the biofilm (Ajjah et al., 2023). Quorum sensing (QS), a cell-cell communication mechanism, becomes vital in the final two stages of biofilm development. Various autoinducers and their corresponding transcriptional receptors regulate the production of different virulence factors, which contribute to the maintenance of biological control and environmental balance. Lastly, bacteria may detach from the biofilm to explore new habitats and initiate the development of new biofilms, completing the biofilm life cycle. Anti-biofilm strategies primarily target each stage of biofilm development by inhibiting microbial adhesion and matrix production, interfering with ECM synthesis, and disrupting QS signaling (Lu et al., 2019).

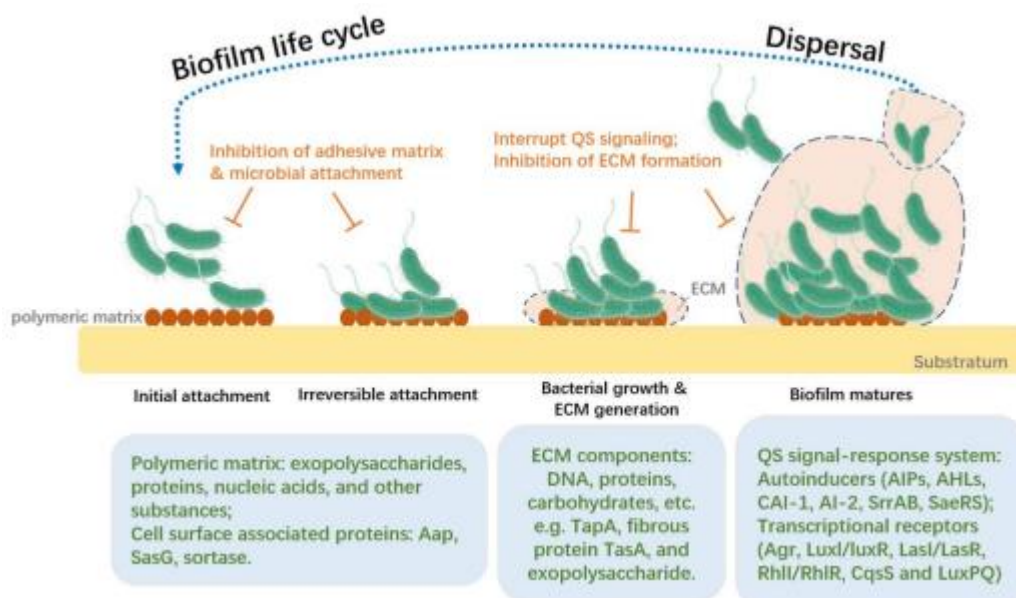


Figure 3. Illustrates the four stages of the biofilm life cycle and highlights the variables involved in quorum sensing, a key process in biofilm development.

Effect of Biofilm Biofertilizer for traits improvement

Rhizobacteria, which colonize the roots of plants, are helpful bacteria. Through the provision of nutrients (like iron, zinc, potassium, zinc, and phosphorus) and the production of phytohormones (such as ethylene, gibberellins, cytokinins, and indole-3-acetic acid (IAA), they are understood to actively encourage plant development.. In addition, they are known to do so indirectly by the synthesis of hydrolytic enzymes and antibiotics (such as amylases, dehydrogenases, chitinases, lipases, pectinases, cellulases, proteases) and inducing systemic resistance, using volatile chemicals (like acetoin, ammonia, hydrogen cyanide, and indole), and using phytopathogens for biocontrol (Haque et al., 2020).

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Numerous instances of the PGPR activity of biofilms are found in the literature. Pseudomonas, Trichoderma, Bradyrhizobium, and Penicillium single and dual species biofilms

outperformed the planktonic inocula in terms of ammonia generation, siderophore synthesis, phosphate solubilization, nitrogenase activity, and/or IAA generation. Additionally, when seeds were infected with biofilms as opposed to planktonic cells, Wheat root and shoot length, soybean dry weights, nitrogen accumulation, maize seedlings, and cotton germination were all higher (Backer et al., 2018).

Plant Growth Promoting Rhizobacteria (PGPR) successfully colonize the plant root as a result of an advantageous plant-microbe interaction, and they later grow into microcolonies or generate biofilm. The host plant is well-protected by the plant-associated biofilms, which also lessen microbial competition, stimulate growth, production, and crop productivity (Kasim et al., 2016). The application of biofilm biofertilizer enhance agronomics traits of food crops (Table 1.).

Tabel 1. Biofilm Microbs Improve Agronomics Traits of Food Crops

Author, Year	Microbs	Plant	Traits Improvement
Prasanna et al., 2015	Cyanobacteria	Maize	Improved plant height, cob weight, days until male blossoming and days until female flowering.
Kasim et al., 2016	Bacillus circulans, Pseudomonas fluorescens, Paenibacillus polymyxa, Bacillus megatherium, Azotobacter chroococcum	Barley	Improved plant height, root height, salinity tolerance
Chittapun et al., 2018	Cyanobacteria	Rice	Improved Length (root and shoot), weights of the root, shoot, root after drying, and shoot after drying.
Sudadi et al., 2018	Aspergillus niger, Penicillium nalgiovense, Azotobacter sp.	Spinach	Improved plant height, stover fresh weight, stover dry weight, increase available phosphate, increase total nitrogen
Ricci et al., 2019	Pseudomonas sp. , Bacillus sp.	Tomato	Height, dry weight of the roots and shoots, root length, leaf area, and leaf count increases
Kushwaha et al., 2019	Bacillus amyloliquefaciens, B. subtilis, B. cereus	Millet	Improve the length and weight of roots, as well as the percentage of disease index and disease compared to the control group.
Karthika et al., 2020	Bacillus sp.	Tomato	Increased total nitrogen, total phosphate, fresh weight, dry weight, improved shoot length, improved root length, and fresh weight
Nookongbut et al., 2020	Rhodopseudomonas palustris	Rice	Extracellular polymeric substance (EPS) stimulated growth (height) and grain yield, induced resistance against fungal pathogens
Bao et al., 2021	Cyanobacteria	Rice	Improved plant height, root length, dry biomass, filled

Padukkage et al., 2021	Bradyrhizobium japonicum, B. elkanii, B. diazoefficiens	Rice	grains per panicle and increase nitrogen availability Rice plant development was measured at the seedling stage, two weeks after Bradyrhizobia inoculation, for root, shoot, and whole plant length and dry weight.
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In research conducted by Prasanna et al., 2015 Cyanobacteria inoculant can not only resulted in a reduction of 60 kg N ha⁻¹, but also contributed to an increase of 20-60 kg N ha⁻¹ in the soil. Cyanobacteria have the potential to enhance various aspects of corn plant growth, including increased plant height, cob weight, as well as delaying the onset of male blossoming and female flowering. Biofilm cyanobacteria improve plant growth by stimulating the production of phytohormones, bioactive metabolites, and increasing the content of glomalin-related soil proteins in the soil. The application of biofilm cyanobacteria, specifically *A. variabilis* SCAU30, on rice plants has been shown to have positive effects. According to Bao et al. (2021), this application can lead to improvements in plant height, root length, dry biomass, and the number of filled grains per panicle. Furthermore, it increases the availability of nitrogen, which further enhances the growth and productivity of rice plants. Research conducted by Chittapun et al. 2018 supports the positive effects of using biofilm cyanobacteria, specifically *N. carneum* TUBT04 and *N. commune* TUBT05, as biofertilizers in rice plants. According to their study, the application of these biofilm cyanobacteria resulted in improvements in the length of both the root and shoot, as well as increased weights of the root, shoot, root after drying, and shoot after drying.

By suppressing pathogens and enhancing plant resistance to diseases, there will be a positive impact on the improvement of plant agronomic traits (Jayaraman, 2021). The application of inoculant biofilm biofertilizer in spinach can increase the availability and uptake of phosphorus (P) in plants. This leads to improved growth and yield, ultimately enhancing the overall agronomic performance of the spinach crops (Sudadi et al., 2018). In a recent study conducted in 2021, it was found that the application of biofertilizer biofilm on mustard plants resulted in an increase in soil organic matter content, total nitrogen (N), available phosphorus (P), and exchangeable potassium (K) (Sudadi et al., 2021).

Applying biofilm biofertilizer inoculant on tomato plants can lead to an increase in various growth parameters. Specifically, there can be an increase in plant height by 53.33 ± 2.51 units, fresh weight by 22.66 ± 3.05 units, dry weight by 1.78 ± 0.18 units, and total nitrogen content by 9.60 ± 0.81 units. These improvements can be attributed to the biofilm bacteria isolate KTMA4, which has the ability to produce ammonia, break down complex nitrogenous materials, exhibit catalase activity to alleviate abiotic stress and facilitate nutrient turnover, and synthesize gibberellic acid, which plays a role in germination, stem elongation, sex expression, and fruit senescence (Karthika et al., 2020). In line with the research conducted by Ricci et al. in 2019, the mixture of *Pseudomonas* sp. and *Bacillus* sp. has been found to enhance various growth parameters in plants. This includes an increase in plant height, dry weight of the roots and shoots, root length, leaf area, and leaf count. The combination of these beneficial microbial strains has demonstrated the potential to promote overall plant growth and development.

The utilization of *Pseudomonas stutzeri*, which has the ability to create biofilm on soybean plants, can result in a 10% enhancement in seed germination. This particular strain of *Pseudomonas stutzeri* aids in reducing salt stress in *Brassica napus* L. by activating the plant's antioxidant defense system and prompting the rearrangement of cell walls. Consequently, this stimulation contributes to the advancement of plant growth and overall development (Lami et

al., 2020). In accordance with the research conducted by Kumawat et al. in 2019, on soybean plants, it was found that there is an increase in plant height, shoot and root dry weight, chlorophyll content, as well as the number and dry weight of nodules. This can be attributed to the abilities of Rhizobia and PGPR that can form biofilm, which include the release of extracellular lytic enzymes such as chitinase, β -1,3 glucanase, protease, and lipase. These enzymes play a crucial role in breaking down the cell walls of pathogenic fungi, thereby competing for nutrients with phytopathogens. Additionally, Rhizobia and PGPR occupy colonizing sites on the root surface and other parts of the plant, thereby improving nutrient availability and enhancing nitrogen fixation ability in legume crops.

Effect of Biofilm Biofertilizer for Sustainable Farming

The effect of biofilm biofertilizers on sustainable farming is significant. Biofilm biofertilizers, which consist of microbial-produced exo-polysaccharides, offer numerous benefits for plant growth, soil health, and environmental sustainability (Ghiasian, 2020). Plant growth promoting bacteria perform effectively in blocking rival organisms, nutrient absorption, fast responses, and adaptation to changing environmental conditions while they're in biofilm phase. The understanding of PGPR biofilm mechanism and their effects is mostly unknown, and the naturally existence PGPR inside the soil hasn't been adequately investigated (Seneviratne et al., 2011). PGPR biofilms possess anti-pathogenic properties and offer long-lasting benefits through the release of beneficial substances by the microorganisms. Furthermore, PGPR biofilms can help alleviate environmental stresses like salinity, drought, flooding, and pathogenic attack (Bhatia et al., 2021).

Diverse microbial communities in the soil have a significant impact on the functional stability of ecosystems. Interactions between plants and microbes in the soil play a key role in regulating ecosystem processes, and these interactions may serve as a mechanistic link between plant variety and ecosystem dynamics. In addition to acting as a biofertilizer, biofilm biofertilizer are also one of the most potent novel treatment methods currently available. They could be applied to lessen the usage of synthetic fertilizers, increase crop growth, and restore the soil's viability, which has been compromised by traditional agricultural practices (Premarathna et al., 2018). The application of Biofilm Biofertilizer (BFBF) on rice crops (*Oryza sativa* L.) has demonstrated the potential to reduce the dependency on chemical fertilizers by approximately 50%, while simultaneously achieving a 24% increase in crop yield. (Premarathna et al., 2021).

However, excessive use of chemical fertilizers has led to a number of issues, including significant soil degradation, nitrogen leaching, and soil compaction, a decline in organic matter in the soil, and soil carbon loss. Additionally, chemical fertilizers ability to increase agricultural productivity has been less effective over time (Lin et al., 2019). Many research reported that organic and chemical fertilizers both supply equivalent amounts of nutrients. In research conducted by Gangthilaka et al., 2022 showing that the use of biofilm biofertilizer can decrease use of chemical fertilizer, the quantity of chemical fertilizer required could be further reduced to 48% by blending biofilm biofertilizer with chemical fertilizer. Application of biofilm biofertilizer combined with lower chemical fertilizer rates develops plant roots, boosting their ability to absorb water and nutrients. This suggests that the biofilm biofertilizer should be evaluated as a significant factor in improving the chemical fertilizer use efficiency. The additional nitrogen supplied by PGPR efficient nitrogen fixation when they are in biofilm state may also be responsible for the higher production. In agriculture, the PGPR in biofilm mode is essential for promoting plant growth and high-quality production.

A new sustainable strategy to agriculture is required to solve this issue and ensure food security for the growing population. Agriculture that produces the most at the lowest cost is considered to be sustainable (Figure 3.). In order to increase production, beneficial microbes have been effectively used in sustainable agriculture. However, using pesticides and synthetic fertilizers to boost agricultural production was primarily motivated by the growing world population and the limited availability of land resources (Ali et al., 2021). The application of *Bacillus subtilis* inoculant capable of forming biofilm can contribute to a biocontrol effect on tomato wilt disease, resulting in a reduction of disease incidence by 8.1% to 50.9% (Chen et al., 2012). Therefore, the use of *Trichoderma* and *Anabaena* in cotton plants has been shown to enhance plant growth, nitrogen content, and dry weight. The increased activity of PAL, PPO, and PO enzymes induced by this combination plays a crucial role in strengthening the plant cell wall, restricting pathogen penetration, and enhancing the plant's defense response against disease (Triveni et al., 2015).

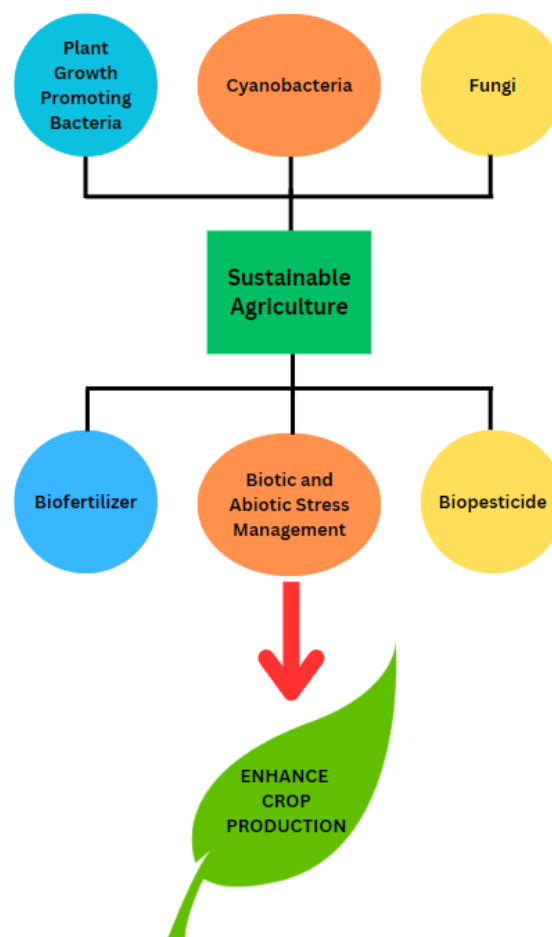


Figure 3. Schematic representation of direct and indirect utilization on the ecosystem (Ali, et al., 2021).

Biofilm biofertilizer has the ability to enhance plant protection against pathogens and diseases. The formation of biofilm enables microorganisms to become more resistant to these harmful agents (Muhammad et al., 2020). Microbial cells form connections with one another by adhering to the surface through a complex matrix composed of different extracellular polymeric substances (EPS), which include exopolysaccharides, proteins, and DNA. These EPS components function as signaling molecules. Bacteria, cyanobacteria, and fungi are among the microorganisms that engage in specific interactions with plant tissues through a process known as quorum sensing (QS). Quorum sensing is a widely observed communication channel among

microorganisms (Lahlali et al., 2022). Moreover, *Azotobacter chroococcum* bacteria and *Trichoderma viride* fungus have the ability to create biofilms as a means of safeguarding diverse crops from pathogens (Triveni et al., 2012). In the case of *B. velezensis* QST713 strains, they can form biofilms on non-living surfaces to impede the proliferation of *T. aggressivum*, the pathogen responsible for green mold disease (Pandini et al., 2019).

Potential In Indonesia

Biofilm biofertilizers have emerged as a promising solution to address the excessive and indiscriminate use of chemical fertilizers in conventional farming practices in Indonesia. By introducing biofilm biofertilizers, farmers can create a conducive environment for beneficial microorganisms, leading to improved nutrient availability, enhanced disease resistance, and overall plant vigor. The success of applying biofilm biofertilizer in increasing the productivity of rice, tomatoes, spinach, barley, and millet through the enhancement of microbial diversity, abundance, and network interactions has been proven by several previous studies. Because many people in Indonesia are cultivating food crops such as rice, tomatoes, and spinach, it creates an opportunity to isolate microorganisms that can form biofilms and multiply them, making them potential biofilm-forming biofertilizers for agricultural development in Indonesia.

CONCLUSION

Potential biofilm biofertilizers are crucial for preserving the productivity and longevity of soil systems, which in help promote in enhancing crops potential for production. It functions as a cost-effective, environmentally friendly, and farmer-friendly input that is simple to utilize in farms for a variety of crops. Furthermore, it can be inferred from the evaluations above that biofilm biofertilizers offer a wide range of advantages. Several studies have demonstrated the synergistic effects of biofilm biofertilizers and reduced chemical fertilizer application. The challenges use of biofilm biofertilizers in Indonesia are include the availability of suitable biofertilizer formulations, farmer awareness and adoption, and the need for further research to optimize biofertilizer application techniques.

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