

4. Bacterial Endophytes: Recent Developments and Applications

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Abstract:

Endophytic bacteria have been found in virtually every plant studied, where they colonize the internal tissues of their host plant and can form a range of different relationships including symbiotic, mutualistic, communalistic and trophobiotic.

Most endophytes appear to originate from the rhizosphere or phyllo sphere; however, some may be transmitted through the seed. Endophytes can also be beneficial to their host by producing a range of natural products that could be harnessed for potential use in medicine, agriculture or industry.

In addition, it has been shown that they have the potential to remove soil contaminants by enhancing phytoremediation and may play a role in soil fertility through phosphate solubilization and nitrogen fixation. There is increasing interest in developing the potential biotechnological applications of endophytes for improving phytoremediation and the sustainable production of nonfood crops for biomass and biofuel production.

The major focus of the review relies on the isolation and identification of bacterial endophytes from diverse habitats and illustrates their various potential applications in the pharmaceutical, industrial, and agricultural sectors as well as in nanotechnology for the fabrication of various nanoparticles incorporated into different applications. In this paper we will discuss Bacterial Endophytes: Recent Developments and Applications.

Keywords:

Bacterial Endophytes, Developments, Applications, Plant–Microbe, Plant-Bacteria, Biotechnological, Biocontrol, Host Plant, Volatile Organic Compounds, Rhizosphere Bacteria.

4.1 Introduction:

An endophyte is an endosymbiont that inhabits a plant for a portion of its life cycle but does not appear to be pathogenic. These endosymbionts are frequently bacteria or fungi. Although endophytes are common and have been identified in every plant species that has been investigated thus far, little is known about the majority of the connections between endophytes and plants. Certain endophytes have the potential to improve a plant's capacity to withstand abiotic stressors like drought and reduce biotic stressors like insect, disease, and herbivore attacks by fortifying the plant's defenses against these entities. While endophytic fungi and bacteria are often researched, endophytic archaea—which are a part of a plant's core microbiome—are also being explored more and more for their potential to promote plant growth.

Because of their broad range of metabolic potential, a great number of endophytic bacteria have been thoroughly described from a variety of medicinal plants. Plants can withstand a variety of stressful situations thanks to the mutualistic relationship that endophytes have with them. These bacteria' capacity for biocontrol gives them the potential to shield plants from a variety of harmful threats.

Although crop diseases are managed with chemical pesticides and fertilizers, their toxicity and environmental contamination necessitate the development of environmentally friendly methods to increase agricultural productivity in a way that is both safe and economical.

As a result, endophytic microbiome has been recognized as a noteworthy substitute to be utilized in the field of agriculture. Despite the fact that the endophytic bacterial association with medicinal plants has been the subject of numerous research to date, little is known about their adaptation mechanisms and metabolite potential. [1]

Different physical, molecular, and biochemical methods are employed by endophytic bacteria to exhibit and carry out a range of growth and biocontrol characteristics. The specific endophytic bacteria can be further studied and cultivated for commercial applications since they possess the majority of the features that promote plant development and biological control, making them a dependable agent in plant growth, reproduction, and protection.

Copper, iron, zinc, and magnesium are among the crucial organic acid-metal complexes that are absorbed by the majority of beneficial endophytic bacteria. Plants are able to remove these metals from the bacteria through the entry of endophytic bacteria into the roots of the plants.

Conversely, introducing endophytic bacteria into plants can prevent the development of illness symptoms that are brought on by pathogens such viruses, nematodes, fungus, insects, and bacteria.

As a more appealing source of nutrients than chemical fertilizers, endophytic bacteria can be employed. Nonetheless, there are still a number of research and application gaps. For instance, numerous studies have demonstrated that endophytic bacteria fix atmospheric nitrogen gas and transform it into a form that plants can use. On the other hand, not as much research has been done on the other two important nutrients, potassium and phosphorus. [2]

Zinc and iron are among the trace elements that endophytic bacteria solubilize. Nevertheless, it is unclear if they can completely utilize their capacity to solubilize and make other essential minor elements (such manganese and molybdenum) available for plant use.

There's no denying that a large portion of research has focused on endophytes' capacity to synthesize and produce indole-3-acetic acid; however, there is a dearth of information regarding other hormones, like zeatin, abscisic acids, and gibberellic acids, and how these vital plant growth regulators are produced by endophytic bacteria or how they affect plant growth and development.

Researching certain endophytic bacterial features will therefore contribute to the production of more bioproducts than growers now have access to.

According to reports from other studies, bioformulations are characterized by their ease of delivery, ability to improve plant development and stress resistance, boost biomass and yield, and facilitate technological exploitation and marketing.

A. Isolation and Biodiversity of Bacterial Endophytes:

For bacteria that are able to colonize and establish in planta, the endophytic niche provides protection from the environment. These bacteria have been identified from every plant compartment, including the seeds, and they typically inhabit the intercellular gaps. Both monocotyledonous and dicotyledonous plants, including woody tree species like oak and pear as well as herbaceous crop plants like sugar beet and maize, have been found to harbor endophytic bacteria. Traditionally, research on the variety of bacterial endophytes has concentrated on describing isolates that were taken from interior tissues after plant surfaces were cleaned with sodium hypochlorite or other comparable chemicals.

B. Application of Endophytes via their Metabolic Capacity:

It is becoming more and more obvious that many endophytes have special metabolic abilities, which most likely arose as a response to living in the endosphere of plants. Based on these special metabolic abilities, endophytes have a number of potential uses in a range of industries. [3]

4.2 Review of Literature:

Many studies have been conducted on the beneficial interactions between microbes and plants that support plant growth and health. Additionally, recent research has looked into their ability to improve the biodegradation of contaminants in soil.

The majority of these investigations have concentrated on microorganisms found in plants' rhizospheres. Of the approximately 300,000 plant species that exist on Earth, every single plant is home to one or more endophytes. Endophytes are defined as bacteria that colonize the internal tissue of the plant while displaying no outward signs of infection or detrimental effect on their host (Strobel et al., 2004). [4]

Bacterial endophytes are found in a variety of agricultural, horticultural, and forest species' stem, root, leaf, tuber, and fruit tissues. They are occasionally regarded as a component of the rhizospheric microbe population. Compared to rhizospheric bacteria found in plants, endophytic bacteria have a better advantage since they come into close touch with plant tissues.

They also provide more benefits to the plant than bacteria found in the rhizospheric region or outside of plants. Such endophytic bacterial populations are widely distributed and frequently occur, suggesting that healthy plants attract a greater number of endophytic populations and offer these organisms a vast and relatively unexplored biological niche (Hardoim et al., 2015). [5]

Many endophytes have demonstrated a natural ability for xenobiotic degradation or may function as vectors to impart degradative features in addition to producing new compounds. Certain endophytes' capacity to break down organic compounds and exhibit resistance to heavy metals and antibiotics is likely a result of their exposure to a variety of substances found in the plant/soil niche. To improve phytoremediation, this inherent capacity to break down these xenobiotics is being studied (Siciliano et al., 2001). [6]

Studies on the relationships between plants and bacteria have been conducted for many years. The results indicate that bacteria have a beneficial effect on plant growth and health, and that plants can "select" their microbiome or core microbiome to obtain beneficial bacterial colonizers, such as endophytes, which are bacteria that live inside plant tissues (De Souza et al., 2016).

According to a functional definition of endophytic behavior put out by some authors, a bacterium is considered an endophyte if it can be isolated from the inside of a plant or from the disinfected surface of a plant tissue without evidently harming the plant. [7]

4.3 Objectives:

- The study of endophytes from bacteria: current advancements and uses.
- Categorization of endophytes in bacteria.

- Endophytes actively inhibit diseases and increase crop yields through biocontrol.
- As they coexist peacefully inside their hosts, beneficial endophytes generate a variety of chemicals that are helpful for safeguarding plants from the elements and promoting plant development and sustainability.

4.4 Research Methodology:

This study's overall design was exploratory. The research paper is an endeavor that is founded on secondary data that was obtained from reliable online resources, newspapers, textbooks, journals, and publications. The research design of the study is mostly descriptive in nature.

4.5 Result and Discussion:

Fungal and bacterial species known as endophytes live inside the plant endosphere without causing harm to their hosts.

They exist in the cellular environment of plants without causing any symptoms and carry out specialized tasks related to symbiosis, like producing secondary metabolites or signaling molecules that operate as both internal and exterior stimuli during mutualistic interactions.

The biochemical and pharmaceutical sectors can obtain new biomolecules from endophytic microorganisms. With great potential for use in medicine, the pharmaceutical industry, or agriculture, they generate physiologically active metabolites such as immune-suppressive substances, anticancer drugs, plant growth promoters, antimicrobial volatiles, insecticides, antioxidants, and antibiotics (Figure 4.1).

Furthermore, in adverse circumstances like nutrient stress, temperature stress, salinity, trace metal stress, or drought, endophytic bacteria might enhance plant growth.

They can also aid in the growth of plants in polluted areas by breaking down potentially harmful substances. We outline the key ideas behind the use of endophytes in biotechnology and agriculture.

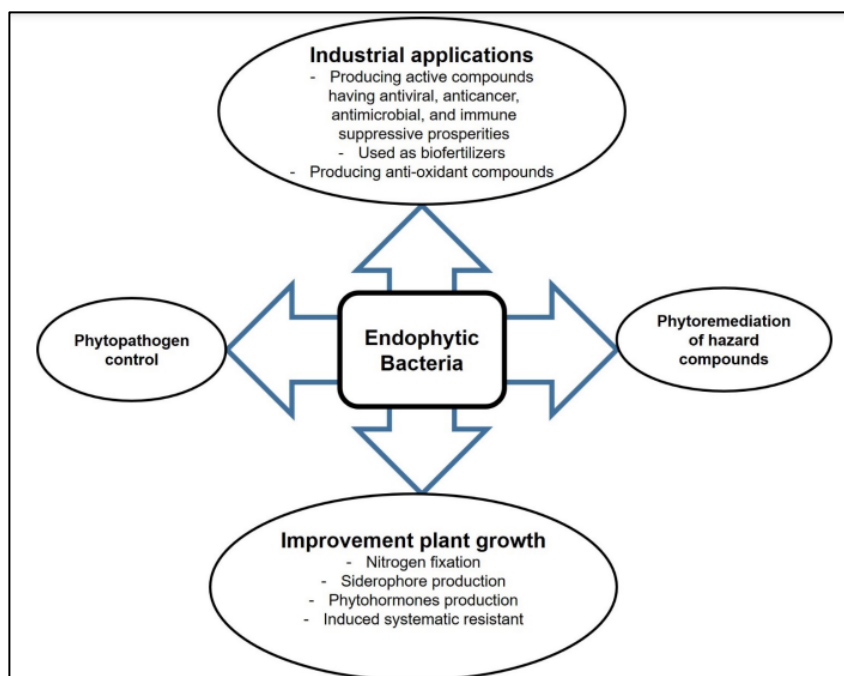


Figure 4.1: Prospective biotechnological applications of endophytic bacteria [8]

4.5.1 Biocontrol of Plant Pathogens:

Due to their capacity to be passed down sustainably to the following generation, endophytic bacteria are reported to be appropriate biocontrol agents. Another benefit of endophytic bacteria in biocontrol is that they enhance and support the health of their host plant rather than competing with it for resources and space. In a previous review, some of the endophytic bacteria with biocontrol abilities were thoroughly documented.

The most effective endophytic bacteria for biocontrol applications against plant diseases and pathogens are those belonging to the genera *Arthrobacter*, *Pseudomonas*, *Serratia*, *Bacillus*, and *Curt* bacterium. Endophytes are typically examined using genetic screening methods and dual plate assays following their separation from the host plant. Because *Bacillus* species are capable of producing a large variety of physiologically active compounds that are strong plant pathogen inhibitors, they have been suggested to be effective biocontrol agents. Tomato bacterial wilt was treated with a few seed-associated endophytic bacteria, including *Bacillus subtilis*, *Bacillus velezensis*, *Leuconostoc*

mesenteroides, *Lactococcus lactis*, and *Bacillus amyloliquefaciens*. All of the isolates were able to demonstrate biocontrol qualities. It has been observed that additional related bacterial endophytes generate secondary metabolites, which may contribute to the biocontrol of plant diseases. Furthermore, in both in vitro and field studies, *Bacillus velezensis* 8-4 was shown to inhibit potato fungal infections, including *S. galilaeus*, *Phoma foveat*, *Rhizoctonia solani*, *Fusarium avenaceum*, and *Colletotrichum coccodes*. These are but a few instances of the numerous ways that endophytic bacteria have been employed to regulate the spread and development of important plant diseases.

Some of the mechanisms have been described as researched in the past few years.

- Genes related to host defense that are overexpressed.
- rivalry for food and resources.
- volatile organic compounds.
- synthesis of antibiotics.
- synthesis of lytic enzymes

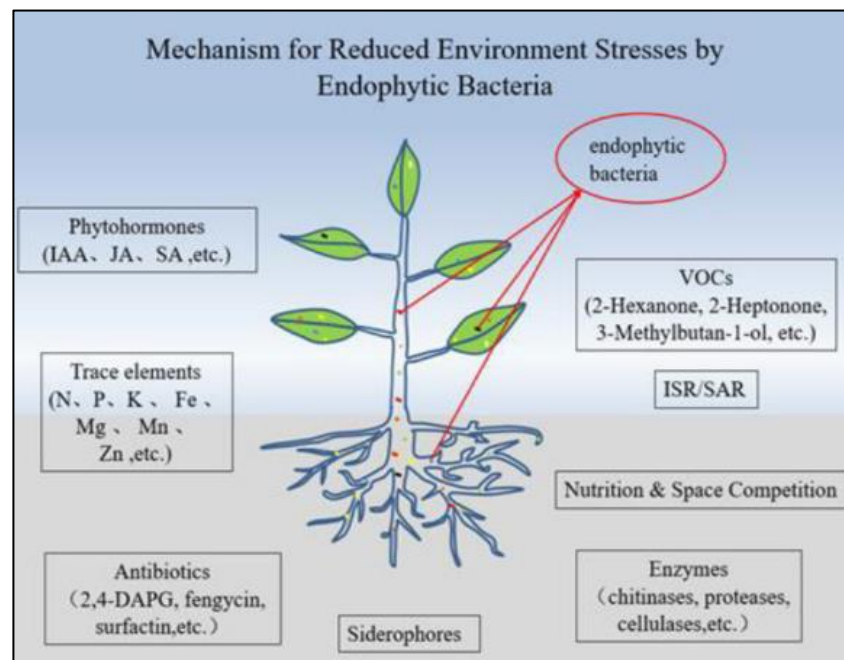


Figure 4.2: Summary of roles of endophytic bacteria in promoting plant growth and the biological control of plant pathogens. [9]

4.5.2 Applications of Bacterial Endophytes:

A. Agricultural Applications:

- **Plant Growth Promotion:**

Endophytic bacteria enhance plant growth in agriculture, horticulture, silviculture, and phytoremediation by employing the same methods as rhizosphere bacteria. It is possible for the growth-promoting effect to be direct or indirect (Figure 4.3).

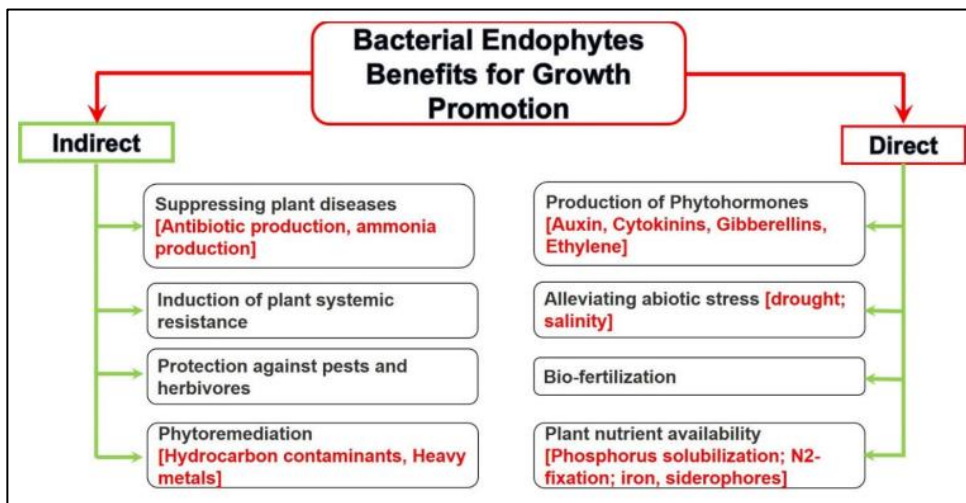


Figure 4.3: Direct and indirect growth-promoting attributes by endophytic bacteria.

Plants that can adjust their hormone levels in response to environmental stressors grow and develop more quickly thanks to phytohormones. Low levels of hormone release in plants result in less responsiveness, despite the fact that plant hormones regulate the growth and response of plants. Then, endophytic bacteria's phytohormones can control the amounts of endogenous plant hormones and ultimately affect various aspects of plant growth and development. Auxin is a substance released by endophytic bacteria in plants that promotes root growth and causes lateral roots, adventitious roots, and root hairs. Auxin is a multifunctional protein in plant cells that helps with cell elongation by controlling cell wall composition and stiffness. Additionally, it fortifies the plant's defenses against outside stressors.

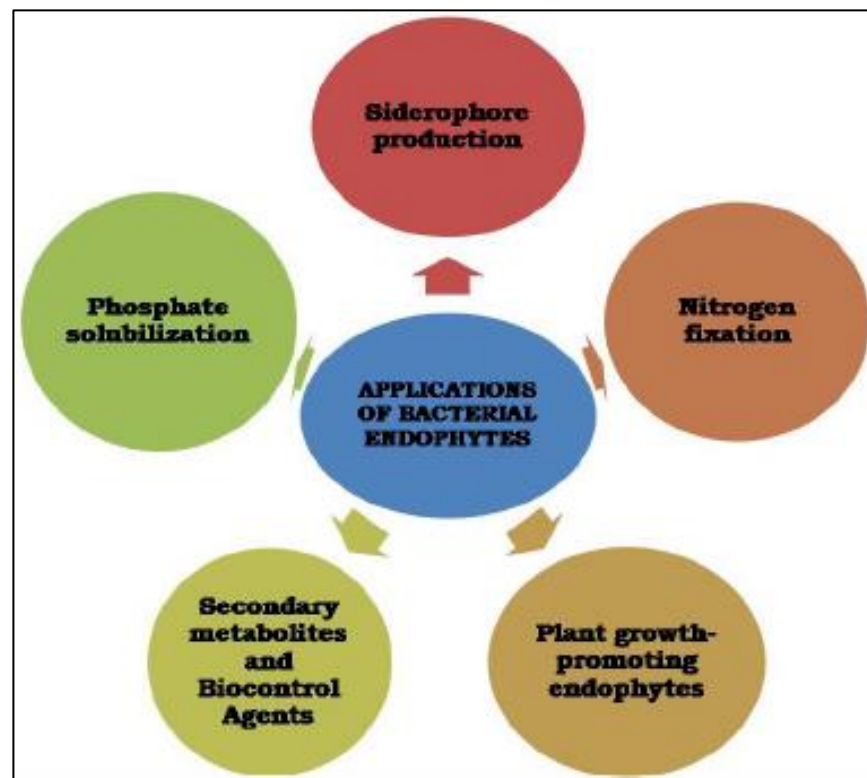


Figure 4.4: Potential applications of endophytes.

- **Nitrogen Fixation and Phosphate Solubilization:**

For plants, nitrogen serves as a nutritional component. According to a report, endophytic bacterial inoculants use biological nitrogen fixation to provide nitrogen for growth and development.

A handful of promising endophytic bacteria, including *Bacillus*, *Fusarium*, *Pseudomonas*, *Rhizobium*, and *Klebsiella*, provide the nitrogen needs of both leguminous and non-leguminous plants. Endophytic bacteria fix nitrogen and act as a substitute for chemical fertilizers in the agriculture industry.

By using the acetylene reduction assay, endophytic bacteria's capacity to fix nitrogen is verified. In non-legume plants, endophytic bacteria such as *Herbaspirillum*, *Azospirillum*, and *Azoarcus* spp. fix nitrogen.

- **Siderophore Production:**

Low molecular weight iron-binding siderophores are secreted by endophytes to aid in plant growth. Assays using Chrome azurol S agar plates are commonly used to identify siderophore synthesis in bacteria.

An alternate method for identifying and analyzing siderophores produced by both wild-type and mutant bacteria is the cross-feeding assay.

- **Secondary Metabolites and Biocontrol Agents:**

Secondary metabolites with anticancer, antibacterial, anti-insect, and other qualities are stored in endophytes. Secondary metabolites, which have antifungal, antibacterial, and insecticidal qualities and inhibit plant diseases, are produced by a variety of bacterial and fungal endophytes. Endophytic bacteria secrete biocontrol chemicals, which inhibit or prevent the entry of pathogens and infections.

- **Bacterial Endophytes:**

Plants must establish mutualistic ties with other living things in the ecological system for them to successfully flourish in their ecological niche. The relationship that exists between microbes and plants is one example of this positive correlation.

Certain tissue-colonizing bacteria develop a tight relationship with their host plant and even benefit it in favorable and unfavorable circumstances.

The benefits that these endophytic bacteria provide to their host plants include assisting them in reducing the effects of biotic and abiotic factors on growth. Endophytic bacteria give their host plant the ability to withstand stress.

They both enhance the growth of their host plant and cause allelopathic consequences. The ability of endophytic bacteria emphasized functional qualities in Table 4.1 to help their host plants flourish and survive in their specific ecological niches has been linked to these traits.

Table 4.1: Plant productivity improvement using bacterial endophytes.

Bacteria Endophyte	Host Plant	Bioactive Influence
<i>Bradyrhizobium</i> sp SUTNa-2	<i>Oryza sativa</i>	Plant-growth-promoting
<i>Pantoea dispersa</i> IAC-BECa-132; <i>Pseudomonas</i> sp; <i>Enterobacter</i> sp	<i>Saccharum officinarum</i>	Plant-growth-promoting
<i>Enterobacter cloacae</i> RCA25; <i>Herbaspirillum huttiense</i> RCA24	<i>Oryza sativa</i>	Plant-growth-promoting
<i>Pseudomonas granadensis</i> T6; <i>Rhizobium larrymoorei</i> E2	<i>Oryza sativa</i>	Plant-growth-promoting and pesticide tolerance
<i>Bacillus amyloliquefaciens</i> EPP90; <i>Bacillus subtilis</i> ; <i>Bacillus pumilus</i>	<i>Pennisetum glaucum</i>	PGP and abiotic stress tolerance
<i>Gordonea terrae</i>	<i>Avicena marina</i>	Plant-growth-promoting
<i>Pantoea</i> , <i>Pseudomonas</i> , <i>Enterobacter</i>	<i>Eleusine coracana</i>	Plant-growth-promoting
<i>Bacillus subtilis</i> LE24, <i>Bacillus amyloliquefaciens</i> LE109, <i>Bacillus tequilensis</i> PO80	<i>Citrus spp</i>	Biocontrol of pathogens
<i>Curtobacterium</i> sp SAK 1	<i>Glycine max</i>	PGP and salinity stress tolerance
<i>Bacillus tequilensis</i> (PBE1)	<i>Solanum lycopersicum</i>	PGP and biocontrol of pathogens

Numerous plant hosts, habitats, and sections of the plant, such as the root tissues, stems, leaves, seeds, fruits, tubers, ovules, and nodules, have been found to harbor bacterial endophytes. On the other hand, bacterial endophytes are found in root tissues more often than in aerial plant tissues. Several studies have documented the ability of bacterial endophytes to promote crop growth in a variety of crops, such as tomato, canola, rice, wheat, and potatoes. Numerous exploratory research projects have hinted at the enormous agrobiotechnological potential of using endophytic bacteria as bio-inoculants to create a long-lasting, environmentally friendly, and sustainable agricultural production system.

Numerous beneficial effects of almost a wide variety of endophytic bacteria include biocontrol activity, as well as acting as an enhancer of N₂ fixation, plant hormone production, phosphate solubilization, and inhibitors of ethylene (C₂H₂) biosynthesis against various biotic and abiotic stresses (Figure 4.5).

In comparison to bacterial pathogens and rhizospheric bacteria, they proliferate at a lower population density and offer superior defense against abiotic stress. When other crops are rotated with different plant hosts, they aid in suppressing the proliferation of nematodes. They typically grow inside plant vascular tissues and cellular spaces.

An estimate states that at least 129 distinct endophytic bacterial species, primarily gram-positive and gram-negative bacteria belonging to over 54 genera, have been identified from various plant species. that while bacterial endophytes aid in plant growth, they haven't been successful in influencing plant growth in Feld circumstances. In addition to promoting plant growth and suppressing diseases and pollutants, endophytes also aid in the solubilization of phosphate and the assembly of nitrogen in plants. [10]

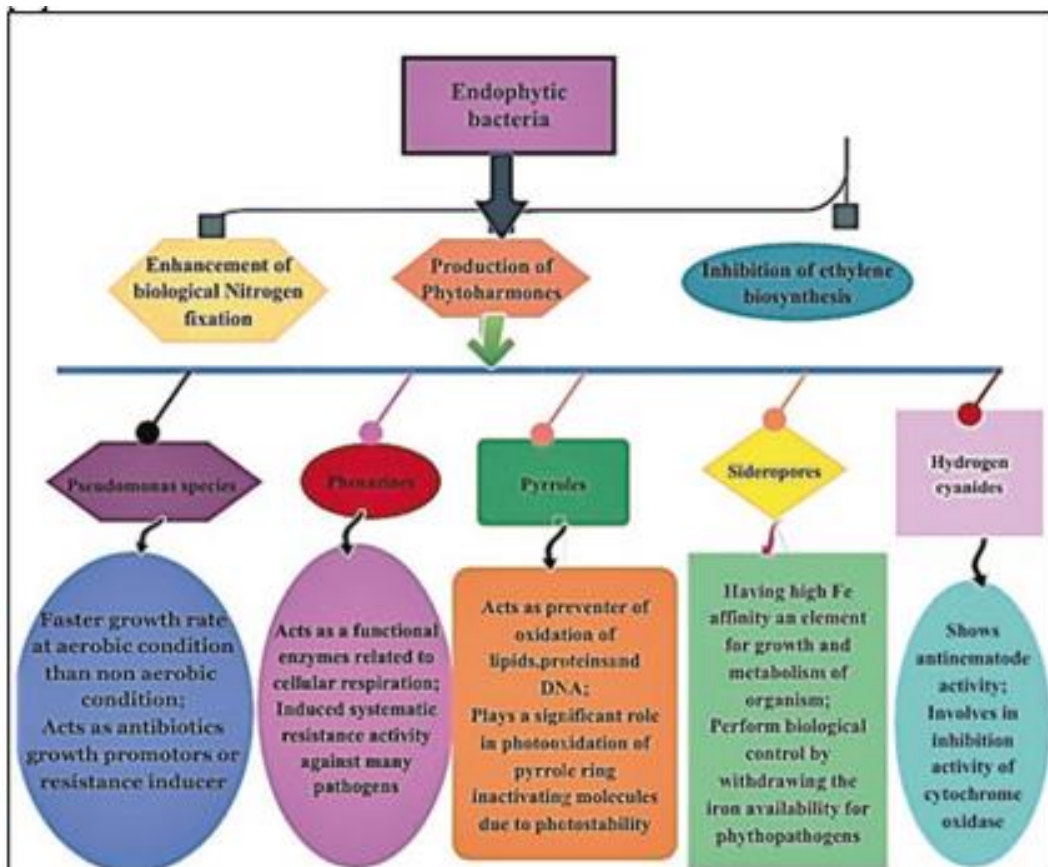


Figure 4.5: Mechanism, classification and significance of endophytic bacteria

4.6 Conclusion:

The improvement of plant health and low-input sustainable agriculture applications for food and nonfood crops can be achieved through the utilization of endophyte–plant interactions. The genes controlling the colonization and establishment of endophytic bacteria in plants can be identified thanks to the availability of entire genome sequences of important endophytic bacteria. The transcriptome and proteome analyses that are now being developed to examine other plant-microbe interactions will be based on this information. Within the interior tissues of their host plant, endophytic bacteria live in a mutualistic or symbiotic relationship. By secreting phytohormones and secondary metabolites, they encourage plant development. Endophytes are thought to fix nitrogen and solubilize phosphate. The majority of endophytes produce siderophores as a biocontrol agent to suppress insects and plant disease pathogens, as well as to supply iron for enhanced growth and output. According to a report, endophytic bacteria use phytoremediation to eliminate pollutants from soil.

4.7 References:

1. Aldor LS & Keasling JD (2003) Process design for microbial plastic factories: metabolic engineering of polyhydroxyalkanoates. *Curr Opin Biotechnol* 14: 475–483.
2. Azevedo JL, Maccheroni J Jr, Pereira O & Ara WL (2000) Endophytic microorganisms: a review on insect control and recent advances on tropical plants. *Electr J Biotech* 3: 40–65.
3. Catalán AI, Ferreira F, Gill PR & Batista S (2007) Production of polyhydroxyalkanoates by *Herbaspirillum seropedicae* grown with different sole carbon sources and on lactose when engineered to express the *lacZlacY* genes. *Enzyme Microbial Technol* 40: 1352–1367.
4. Strobel G, Daisy B, Castillo U, Harper J (2004) Natural products from endophytic microorganisms. *J Nat Prod* 67: 257–268.
5. Hardoim, P. R., Van Overbeek, L. S., Berg, G., Pirttilä, A. M., Compant, S., Campisano, A., et al. (2015). The hidden world within plants: ecological and evolutionary considerations for defining functioning of microbial endophytes. *M. M. B. R.* 79 (3), 293–320. doi: 10.1128/MMBR.00050-14

6. Siciliano S Fortin N Himoc N et al. (2001) Selection of specific endophytic bacterial genotypes by plants in response to soil contamination. *Appl Environ Microbiol* 67: 2469–2475.
7. De Souza, V.K. Okura, J.S.L. Armanhi, B. Jorrín, N. Lozano, M.J. Da Silva, et al. Unlocking the bacterial and fungal communities' assemblages of sugarcane microbiome *Sci. Rep.*, 6 (2016), p. 28774.
8. Antifungal, plant growth-promoting and genomic properties of an endophytic actinobacterium *Streptomyces* sp. NEAU-S7GS2 *Front. Microbiol.*, 10 (2019), p. 2077.
9. Monowar, T.; Rahman, M.S.; Bhore, S.J.; Raju, G.; Sathasivam, K.V. Silver Nanoparticles Synthesized by Using the Endophytic Bacterium *Pantoea ananatis* are Promising Antimicrobial Agents against Multidrug Resistant Bacteria. *Molecules* 2018, 23, 3220.
10. Alvin, A.; Miller, K.I.; Neilan, B.A. Exploring the potential of endophytes from medicinal plants as sources of antimycobacterial compounds. *Microbiol. Res.* 2014, 169, 483–495, Doi: 10.1016/j.micres.2013.12.009.