

Integration of Science and Engineering: *Pathways to Global Sustainability*



Dr. S. PURUSHOTHAMAN

Dr. P. RAJESWARI

Dr. T. SOMANATHAN

Dr. D.R.P. RAJARATHNAM



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Dr. S. PURUSHOTHAMAN

Principal
School of Engineering and Technology
Jaipur National University, Jaipur, Rajasthan-302017

Dr. P. RAJESWARI

Professor
School of Computer and System Sciences
Jaipur National University, Jaipur, Rajasthan-302017

Dr. T. SOMANATHAN

Professor of Chemistry at the School of Basic Sciences
Vels Institute of Science, Technology & Advanced Studies
Chennai, India

Dr. D.R.P. RAJARATHNAM

Professor and Head, Department of Mechatronics Engineering
Paavai Engineering College, Namakkal, India

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editors@srrbooks.in, contact@srrbooks.in
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PREFACE

The rapid advancements in science and engineering have ushered in a new era of innovation aimed at addressing the world's most pressing sustainability challenges. The integration of diverse disciplines—from artificial intelligence and biotechnology to materials science and mechanical engineering—has created unprecedented opportunities to design solutions that promote global health, environmental resilience, and sustainable industrial growth. This book, *Integration of Science and Engineering: Pathways to Global Sustainability*, brings together contemporary research and forward-thinking perspectives that highlight how scientific ingenuity and engineering precision can collectively contribute to a sustainable future.

In the field of healthcare and medicine, technological innovations are redefining traditional paradigms. Artificial intelligence, for example, is revolutionizing drug discovery and enhancing the resilience of healthcare systems. Through predictive modeling and data-driven insights, AI enables faster, safer, and more efficient identification of therapeutic compounds—bridging the gap between laboratory research and real-world treatment outcomes. Such advancements illustrate how computational science and human biology can merge to create transformative pathways for global well-being.

Parallel to the medical domain, manufacturing and materials engineering are also experiencing a renaissance through the integration of additive manufacturing, hybrid processes, and biomaterials. These technologies not only improve precision and production efficiency but also minimize waste and environmental impact. Emerging studies on sustainable materials—such as mycelium-based composites and seaweed-derived nanoparticles—demonstrate

how natural resources can inspire next-generation innovations in packaging, construction, and nanotechnology, fostering a circular economy driven by ecological responsibility.

The environmental dimension of sustainability is further explored through biotechnological interventions that harness microorganisms for ecological restoration. Endophytic bacteria capable of bioremediating heavy metals exemplify nature-inspired engineering at its finest—using living systems to detoxify pollutants and restore balance to fragile ecosystems. In parallel, the integration of enzymatic and plasma-based technologies for domestic waste management showcases the potential of hybrid biotechnological-mechanical systems to convert household waste into usable energy and materials, bridging environmental protection with urban sustainability.

Beyond the realm of physical and biological sciences, sustainability also encompasses social and organizational dimensions. The evolving mindset of Generation Z towards employment reflects a shift in global values—prioritizing purpose, flexibility, and ethical responsibility. Likewise, digital transformation is reshaping business ecosystems, influencing not only consumer behavior in sectors like FMCG but also organizational citizenship in workplaces increasingly guided by artificial intelligence and data-driven decision-making. These insights emphasize that sustainability must also be human-centered, integrating technological progress with empathy, equity, and collective growth.

Ultimately, the chapters compiled in this volume embody the convergence of scientific discovery and engineering innovation as vital forces in shaping a sustainable world. From sustainable chemistry and green pharmaceuticals to smart materials and digital ecosystems, each contribution demonstrates the transformative potential of

interdisciplinary collaboration. By weaving together these diverse threads, *Integration of Science and Engineering: Pathways to Global Sustainability* underscores that the pursuit of sustainability is not a singular endeavor but a collective commitment—one that demands creativity, cooperation, and a shared vision for the future of humanity and the planet. We would like to extend our sincere thanks to our publisher, **Scientific Research Reports, Chennai, India**, for their dedicated efforts in preparing this book, which provides enriched content.

Wishes and Regards,

Dr. S. PURUSHOTHAMAN

*School of Engineering and Technology, Jaipur National University,
Jaipur, Rajasthan.*

Dr. P. RAJESWARI

*School of Computer and System Sciences, Jaipur National
University, Jaipur, Rajasthan.*

Dr. T. SOMANATHAN

*Department of Chemistry, Vels Institute of Science, Technology and
Advanced Studies (VISTAS), Chennai.*

Dr. D.R.P. RAJARATHNAM

*Department of Mechatronics Engineering, Paavai Engineering
College, Namakkal.*

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Chapter 5

Endophytic Bacteria as Sustainable Bioagents for Heavy Metal Bioremediation: Isolation, Molecular Characterization, and Application Prospects

Vijay Balaji S A^{a*}, Sai Ramesh A^{a*}

^{a} Department of Bio-Engineering, School of Engineering, Vels Institute of Science, Technology and Advanced Studies (VISTAS), Chennai*

** Corresponding Author: drsairamesh@gmail.com,
vijaybalaji1685@gmail.com*

Abstract

Plants often team up with bacteria living inside them - a partnership where both benefits. This collaboration can help plants withstand harsh conditions, like soil contaminated by metals. Here, we collected these inner bacterial allies from a remarkably resilient medicinal plant, figured out exactly who they are genetically, then tested their ability to cope with stress. Plant samples underwent careful cleaning, then scientists retrieved microbes living inside. These cultures went through detailed study - appearance, chemical makeup, also genetic testing - to pinpoint exactly what they are. This work allowed precise grouping of strains seemingly able to withstand harsh metals. Researchers grew these strains in labs containing significant amounts of chromium, lead, or cadmium to measure how well they cope with, even absorb, those substances. We found some robust strains that readily gather metals via lab tests. Then, to make them more durable and useful, we crafted them into stable forms. Testing showed these chosen strains successfully lowered metal levels in mock polluted water within a controlled system. Despite everything, findings indicate certain bacteria living within plants could be great

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at safely collecting pollutants - a greener option than current cleanup methods. Essentially, this research shows how vital these hidden plant partners are for cleaning up contaminated environments, especially when dealing with heavy metals.

Keywords: Endophytic bacteria, Heavy metal tolerance, Bioaccumulation, 16S rRNA sequencing, Bioremediation, Environmental biotechnology.

1. Introduction

Worldwide, ecosystems alongside people face increasing danger from heavy metal contamination. These metals - cadmium, lead, chromium, mercury, and arsenic - don't break down; instead they build up in the environment, within soil, waterways, even plants and animals (WHO, 2006; Engwa et al., 2019). This buildup stems from factory releases, mining, plating processes, agricultural drainage via fertilizers/pesticides, also careless trash handling (MN Pollution Control Agency, n.d.; Engwa et al., 2019). Once released, metals enter ecosystems - moving through food webs where they build up to dangerous amounts. Health impacts differ depending on the specific metal, potentially causing damage to kidneys or lungs, harming child development, weakening bones, contributing to heart problems, or even sparking cancer (Chakraborty, Mukhopadhyay, & Roy, 2013; Engwa et al., 2019; WHO, 2006). Since these metals linger and amass, typical cleanup approaches frequently prove inadequate, so we require eco-friendly ways using living organisms to break down or eliminate them.

1.1 Heavy metals (Cd, Pb, Cr) are toxic and non-biodegradable

Cadmium, lead, likewise chromium are seriously dangerous contaminants because they stick around - they don't break down -

and are poisonous. As a result, these metals build up in both land and water, moving into our food supply. This presents genuine risks to people, potentially causing harm to the brain, kidneys, even cancer. Specifically, cadmium messes with how cells work; lead impacts nerves plus blood production. Hexavalent chromium, meanwhile, creates damaging internal stress alongside genetic harm. Heavy metals stay in the environment - they don't break down like many pollutants - so cleaning them up is tough, costly work (Muhammad Ali et al., 2019). Because these metals show up everywhere - from factory waste to mining byproducts and farm drainage - we urgently require eco-friendly cleanup solutions, perhaps using microbes to help (Muhammad Ali et al., 2019).

2. Role of Endophytic Bacteria in Plant-Microbe Interactions

In the interior of every plant there are good-natured bacteria - endophytes - that not only help the plants survive but also increase their resilience during tough times. These bacteria establish themselves within the plant tissues, causing no harm to the host, but rather, they are turning the host's life into wellness. Their activities include nitrogen fixation, IAA-like hormone production, nutrient unlocking (e.g. phosphorus), and resource capture via siderophores. Consequently, the presence of these bacteria during the exposure to toxic metals will result in less severe effects; they will encapsulate or transform the metals, produce antioxidants, alter (e.g. reduce) the level of ethylene hormone in the plant and activate the natural defenses of the plant. The interaction between these microbes and plants is of a double nature, they not only help but also intensify the plants' natural resistance to diseases and at the same time regulating how the plant deals with the harmful molecules. This eventually results in less damage from harsh chemicals or metal pollution

(Beneficial role...; Plant-microbe interactions influence plant performance..., 2025). Thus, these bacteria become the heroes of a plant's struggle to survive and even thrive under unfavorable conditions—bringing in the mix both the means of development and resilience.

2.1 Endophytes live inside plant tissues without causing harm

One of the main reasons for this is that a lot of the times, plants have tiny creatures - bacteria primarily but also fungi - living inside them without being noticed doing no harm (Stone et al., 2018). Both the microbes and plants are getting the most out of the collaboration; the former are not only helping the latter in growing bigger but also in overcoming stresses, developing their defense against diseases through hormone production, iron acquisition, and the synthesis of protecting compounds (Santoyo et al., 2016). Endophytes enjoy the safety of the plant's interior and nourishment from its metabolic activities; in return, plants have their nutrients obtained, nitrogen fixed, and disease resistance enhanced. Since the endophytes are residing in the tissue of the plant, they are able to survive and thrive even when the environment changes - a capability that is useful in such areas as cleaning up of the earth by pollution or plant growth enhancement.

2.2 Improve plant tolerance to stress, including metals

Bacteria living inside plants help them withstand hardships like high concentrations of toxic metals. They do this in several ways - releasing stress-reducing chemicals, boosting antioxidant defenses, or creating substances that bind to the metals (Mammy et al., 2016). Moreover, endophytes lessen metal absorption alongside its harmful effects via secretion of sticky polymers and proteins called

metallothioneins; these trap and contain the dangerous metals near roots or inside the plant itself (Rajkumar et al., 2012). Microbes tweak how plants deal with stress - they control plant hormones such as abscisic acid alongside salicylic acid, boosting photosynthesis, nutrients, then defenses against oxidation (Compant et al., 2021). Consequently, owing to these team-ups, endophytes shield plants from harm caused by metals, simultaneously improving growth plus output even where the ground is polluted (Kumar et al., 2022).

3. Heavy Metal Tolerance Mechanisms in Endophytic Bacteria

Inside plants, certain bacteria handle poisonous metals in diverse ways, letting them flourish where others can't. They deal with these metals through collecting them, sticking them to their surfaces, breaking them down using enzymes, or pumping them out. Cells gather metal ions, storing them inside while making proteins like metallothioneins alongside phytochelatins - this lowers how much metal is available to cause harm (Liu et al., 2024). However, systems exist that pump out too many metal ions, keeping things balanced (Nnaji, 2024). Moreover, bacteria use chemical changes, like redox reactions, to turn dangerous metals into safer ones, shielding themselves (Bhardwaj et al., 2025). Endophytes also shift how plants absorb metals - they tweak plant genes involved in transport while releasing substances changing metal availability near roots (Bhardwaj et al., 2025). This complex approach demonstrates how useful these bacteria could be for cleaning up contaminated areas.

3.1 Bioaccumulation and biosorption reduce metal toxicity

Endophytic bacteria help the environment in polluted areas with heavy metals by two principal mechanisms: sequestration and biosorption. Sequestration refers to the active processes where metals

are taken up and stored within; while biosorption is a simpler attachment using materials like fats, proteins, and sugars (Volesky et al., 2003). Thus, the total toxic metal present gets reduced, and plants and other organisms are shielded by these (Gadd, 2010). Endophytes through chemical processes convert metals into less toxic forms or by capturing them which contributes to environment purification (Rajkumar et al., 2012). Thus, both methods of metal trapping and surface adhesion by these bacteria provide eco-friendly and permanent solutions for contamination cleanup, as opposed to the routine chemical treatments (Liu et al., 2024).

3.2 Resistance genes support adaptation to contaminants

Endophytic bacteria cope with harsh conditions - like those containing heavy metals - thanks to electric resistance genes. These genes create tools like pumps that expel metals, proteins grabbing onto metals, systems detoxifying them chemically, or sequestering them away. All this lessens harm within bacterial cells (Nies et al., 2003). Consequently, genes like *czc*, *mer*, and *ars* help these organisms withstand high levels of cadmium, lead, chromium, alongside various other poisonous ions (Silver & Phung, 2005). Microbes share beneficial genes, quickly spreading abilities to survive harsh conditions - like those found in polluted soil or within plants (Rajkumar et al., 2012). Consequently, bacteria become more robust. Resistance genes give them what they need to handle toxic metals; therefore, cleanup efforts work better.

4. Isolation and Cultivation of Endophytes

To get bacteria from inside plants, researchers follow a careful plan. First, they clean the plant's exterior using things like bleach or alcohol so only inner microbes remain. Then, sterilized pieces of the

plant are broken down, subsequently spread on food-filled dishes to encourage bacterial development. To help endophytic bacteria thrive, they're usually kept between 25–30°C away from light. Then we observe how colonies develop, choosing different forms for more study. Consequently, this process yields bacteria we can grow - bacteria ready for testing and detailed examination (Yu et al., 2022).

4.1 Surface sterilization removes external microbes

Getting rid of surface microbes is key when studying bacteria living inside plants; it cleans the plant without harming those within (Schulz et al., 2015). Usually, this means repeated treatments - like wiping with alcohol, bleach, then rinsing with sterile water - to clear away anything clinging to the outside (Hallmann et al., 1997). Thorough sterilization guarantees any microbes found come only from inside the plant - essential for correctly identifying and studying genuine endophytes (Tao et al., 2021). Poor sterilization, however, risks contamination from outside sources; this could skew research into how plants interact with microbes, likewise affecting assessments of bioremediation possibilities (Santoyo et al., 2016).

4.2 Isolates prepared for further analysis

To get ready for detailed study, researchers isolate pure cultures of bacteria living within plants, then keep them thriving. This preparation generally means growing these bacteria consistently - under controlled settings - so they're healthy and behave similarly during testing (Tao et al., 2021). Getting this right matters because what happens next - like figuring out which microbes are present using DNA, testing how well they survive harmful substances, or gauging if they collect toxins - depends on it. Careful work with these samples guarantees consistent, trustworthy outcomes, allowing us to

properly understand their uses in both technology and nature

5. Molecular Identification

Pinpointing what lives inside plants requires knowing exactly which organisms they are, alongside what those organisms do. Because all bacteria possess a 16S rRNA gene, it serves as a dependable tool for identifying them - its consistent sections make it widely useful. Typically, scientists copy this gene via PCR utilizing broad-reaching starters, then read its sequence. Researchers check product sequences against databases like GenBank via BLAST, figuring out what kind of organism they came from. Afterward, programs - MEGA X for example - help map how different isolates evolved. This approach lets scientists pinpoint bacterial identities alongside understanding where they fit within ecosystems, even suggesting uses in environmental work.

5.1 DNA extracted from pure isolates

To figure out what bacteria live inside plants - specifically, to study their genes - scientists first need to get the bacterial DNA. This involves breaking open the cells, getting rid of unwanted stuff like proteins, subsequently cleaning up the DNA itself. The goal? To have really good DNA ready for things like making copies or reading its code (Tao et al., 2021). DNA reveals where organisms fit on the tree of life, also pinpointing genes linked to tolerating toxic metals, building up substances within tissues, alongside other key abilities (Ambikapathy et al., 2022). Consequently, careful DNA work - both getting it out and treating it right - ensures clean, repeatable results for detailed molecular investigations (Sahu et al., 2022).

6. Vitro Screening for Bioaccumulation

In vitro screening of endophytic bacteria for heavy metal

bioaccumulation is performed to evaluate their tolerance and accumulation capacities under controlled laboratory conditions. Isolates are grown on nutrient rich media enriched with various concentrations of heavy metals including cadmium (Cd), lead (Pb), or nickel (Ni). However, in growing suppression assays, which generally take the form of either measuring the colony diameter or optic denseness of bacterial growth, the period of bacterial growth is measured to assess these bacteria to these different concentrations of metallic elements. In addition, it will be perform quantitative analysis of metals accumulation using atomic absorption spectroscopy (AAS) or inductively coupled plasma mass spectrometry (ICP-MS) to determine the concentration of metals in bacterial cells. Thus, also molecular markers like the presence of metallic element resistance genes can also be distinguished using PCR amplification and sequencing, therefore, indicative of possible genetic metals tolerance mechanisms (January et al., 2019; Liu et al., 2024). This expansive screening strategy can therefore lead to the identification of possible bioaccumulators for use in a bioremediation strategy to reduce heavy metal contamination.

6.1 Isolates grown on metal-supplemented media

To check how well they cope with toxic substances, we grew bacteria taken from inside plants on nutrient mixtures containing metals like cadmium, lead, or chromium. These metal-rich mixtures let us observe – under regulated circumstances – how these bacteria grow, live, and adjust when exposed to metallic challenges (Rajkumar et al., 2012). Tracking how colonies grow alongside metals reveals robust strains ideal for cleaning up pollution – as shown by Kumar and others in 2022. Because this lab testing quickly spots microbes that can gather, expel, or neutralise heavy metals, it lays groundwork for

more detailed research into their real-world performance (Gadd et al., 2010).

6.2 Tolerance levels tested for Cd, Pb, Cr

Researchers checked how well bacteria growing inside plants handled heavy metals – cadmium, lead, and chromium – by giving them different amounts in lab dishes. They also watched how much the bacteria grew, observing colony size alongside total weight, to figure out what level of each metal they could withstand. Tolerant isolates look good for cleaning up pollution because they live well even where metals are a problem (Gadd et al., 2010). Studying them helps us understand how microbes fight off metals - through things like pumping metals out, storing them safely, or breaking them down - knowledge essential for using bugs to fix contaminated sites (Kumar et al., 2022)

7. Applications in Bioremediation

Bacteria living inside plants show real potential for cleaning up pollution, especially when dealing with harmful metals in earth alongside water sources. These tiny helpers boost a plant's natural cleanup abilities via several routes. They might directly grab onto metals, changing them into less dangerous forms, or locking them away inside the plant - lessening harm. Alternatively, these bacteria give plants a lift, encouraging growth by making hormones, helping roots absorb food, then bolstering defenses against difficulties; collectively improving how well plants pull contaminants from the ground plus hold them steady (Liu et al., 2024; Khatoon et al., 2024). Endophytes help break down harmful substances in polluted areas, aiding natural cleanup. Combining these bacteria with plants presents an environmentally sound way to tackle heavy metal

contamination - a method useful for cleaning up water or reviving tainted farmland.

7.1 Endophytes enhance plant-based metal removal

Plants get better at pulling harmful metals from the ground thanks to helpful bacteria living inside them. These bacteria help plants absorb, move around, then neutralize dangerous substances like cadmium, lead, and chromium. They do this by boosting root growth, creating special compounds, also releasing acids – all making these metals easier for plants to take up (Ma et al., 2016). Inside plants, certain microbes grab onto metals, lessening harm while boosting cleanup power (Rajkumar et al., 2012). Consequently, when plants collaborate alongside these microbes, they clear polluted ground faster - also staying healthier despite challenging circumstances. This partnership offers an environmentally sound way to restore land (Compant et al., 2021).

8. Conclusion

Bacteria that live in the interior of the plants can be considered an organic way to get rid of toxic metals. These microorganisms take a position within the plant tissue - without causing any harm - and then conduct gathering, sticking, or pumping out metals and so on as their processes, thus allowing plants to live in and even purify the soil of contaminated areas. By means of genetic markers, scientists identify the useful types of bacteria and consequently those that are the best at resisting or degrading these pollutants get revealed (Gupta et al., 2022).

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