UTILIZATION OF RHIZOBACTERIA FROM WATER HYACINTH (*EICHHORNIA CRASSIPES*) AND ITS BIOMASS AS A SUSTAINABLE CARRIER IN BIOFERTILIZER DEVELOPMENT

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Introduction:

Agricultural sustainability is a growing global concern due to the overuse of chemical fertilizers that degrade soil health and pollute ecosystems. Biofertilizers, particularly those developed using plant growth-promoting Rhizobacteria (PGPR), offer an eco-friendly alternative that can enhance crop productivity while preserving environmental balance. Water hyacinth (*Eichhornia crassipes*), an invasive aquatic weed, is often regarded as a nuisance due to its rapid growth and tendency to clog water bodies. However, its prolific root system harbors diverse Rhizobacteria with significant biofertilizer potential. This project aims to explore the dual-purpose utility of water hyacinth by isolating beneficial rhizobacteria from its roots and using its biomass as a sustainable carrier material. By transforming an environmental problem into an agricultural solution, the study promotes resource efficiency and waste valorization. The rhizobacteria will be screened for key PGP traits such as nitrogen fixation, phosphate solubilization, and hormone production. Simultaneously, the water hyacinth biomass will be processed into a carrier medium to support microbial viability. Through the formulation and testing of this biofertilizer, the project seeks to provide a cost-effective and sustainable solution for enhancing crop growth. It also aims to contribute to weed management strategies and circular economy practices in agriculture.

Objectives:

1. To isolate and identify Rhizobacteria from the roots of water hyacinth collected from freshwater ecosystems.
2. To assess the plant growth-promoting properties of the isolated Rhizobacteria and simultaneously process water hyacinth biomass into a viable biofertilizer carrier material.
3. To formulate a biofertilizer using the selected bacterial strains and prepared biomass and evaluate its effectiveness in enhancing plant growth under both laboratory and field conditions.

Methodology:

1. Isolation and Characterization of Bacteria from Water Hyacinth Rhizosphere:

Sample Collection: Roots of *Eichhornia crassipes* is collected from freshwater bodies and transported in sterile containers.

Isolation of Bacteria: The roots will be washed with sterile distilled water, homogenized, serially diluted, and plated on Nutrient Agar and PDA. Plates are incubated at 28°C for 24–72 hours. Distinct colonies are sub-cultured to obtain pure isolates.

Characterization of Bacteria: Isolates will be characterized based on colony morphology, Gram staining, and biochemical tests, including catalase, oxidase, indole production, citrate utilization, and sugar fermentation.

2. Evaluation of Plant Growth-Promoting Traits and Biomass Preparation:

The isolated Rhizobacteria will be evaluated for key plant growth-promoting traits, including nitrogen fixation (Jensen’s medium), phosphate solubilisation (Pikovskaya’s agar), IAA production (Salkowski’s reagent), siderophore production (CAS assay), and antifungal activity through dual culture methods against pathogens like *Fusarium* and *Rhizoctonia*.

Simultaneously, fresh water hyacinth biomass is collected, washed, chopped, and dried at 50–60°C until the moisture content is below 10%. The dried material is ground into a fine powder and sterilised by autoclaving at 121°C for 15 minutes to serve as a carrier medium for biofertilizer formulation.

3. Biofertilizer Formulation and Efficacy Evaluation:

Selected bacterial isolates will be cultured in nutrient broth to obtain a high cell density, then mixed with sterilised water hyacinth powder in appropriate ratios to formulate the biofertilizer. The viability of the microbial inoculum will be monitored periodically by CFU counts on nutrient agar.

The formulated biofertilizer will be tested on Maize as a model plant under controlled conditions through pot experiments. Growth parameters such as shoot and root length, biomass, and chlorophyll content will be recorded. Field trials will be conducted using a Randomised Block Design (RBD) to evaluate the impact on plant growth, yield, and soil health (Figure 1).

Data Analysis:

Statistical tools like ANOVA will be used to determine the effects of the biofertilizer on plant growth and soil health. Correlation and regression analyses will evaluate relationships between biofertilizer concentration and plant performance.

Figure 1. This flowchart summarizes the key steps involved in the isolation formulation and evaluation process of the biofertilizer.

Result and Conclusion:

The isolation of Rhizobacteria from water hyacinth roots was successfully carried out under sterile conditions. The obtained cultures were subcultured multiple times to ensure purity, and representative isolates have been submitted to a certified laboratory for molecular identification through 16S rRNA gene sequencing. While species-level identification is currently underway, initial observation based on morphological characteristics under microscopy suggests the presence of diverse bacterial forms.

According to existing literature and preliminary visual analysis, water hyacinth rhizospheres are known to harbor beneficial plant growth-promoting rhizobacteria such as *Azospirillum*, *Pseudomonas,* and *Bacillus* species. These microbes are recognized for their roles in nitrogen fixation. Phosphate solubilization and hormone production. The results of molecular analysis will further confirm the identity and potential of the isolated strains for biofertilizer development. The project is anticipated to provide a sustainable and low-cost biofertilizer option while offering an effective use for invasive water hyacinth biomass.

Project Outcome & Industry Relevance:

The project is expected to result in the development of a sustainable biofertilizer using rhizobacteria isolated from water hyacinth and its processed biomass as a carrier. This offers an innovative solution to two major concerns: the need for eco-friendly agricultural inputs and the management of invasive aquatic weeds.

The biofertilizer, once developed and tested, can enhance plant growth, soil health, and reduce dependence on chemical fertilizers, making it suitable for organic farming. The approach also adds value to water hyacinth, often considered a nuisance, by converting it into a useful agricultural product (Figure 2).

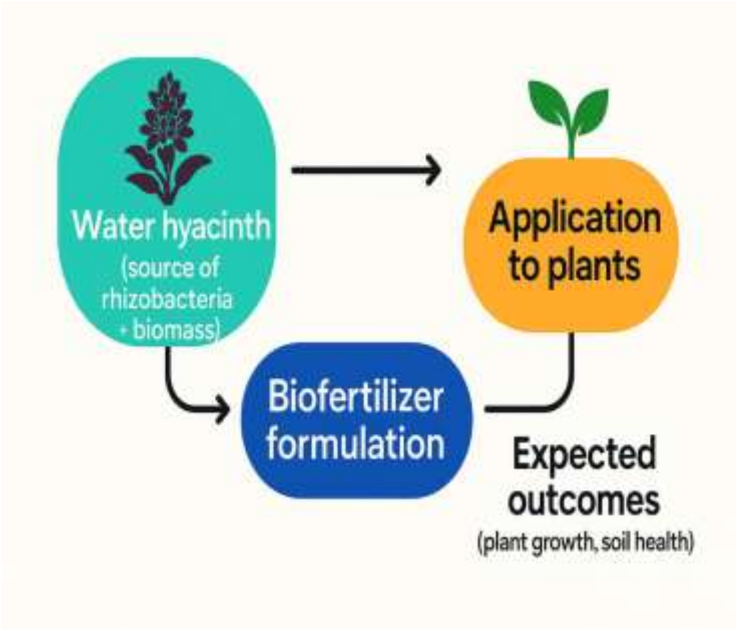

Figure 2. Conceptual framework illustrating the integration of water hyacinth as a source of Rhizobacteria and biomass for biofertilizer formulation, its application to plants, and expected outcomes in plant growth and soil health.

This concept has strong industry relevance, particularly for agro-based startups, rural entrepreneurship, and environmental restoration initiatives. It can be scaled for commercial production and promoted through sustainable agriculture programs and government-supported bio-input schemes.

Working Model vs. Simulation/Study:

The project is primarily a theoretical and experimental study focused on the isolation and characterization of rhizobacteria from water hyacinth and their potential application in biofertilizer development. It also explores the use of water hyacinth biomass as a sustainable carrier material. The study involves a literature review, laboratory-based experiments, and a planned pot experiment to evaluate the biofertilizer's effectiveness. No physical working model is being developed; the focus remains on scientific analysis and empirical validation.

Project Outcomes and Learnings:

The expected outcomes of this project include the successful isolation and identification of rhizobacteria from water hyacinth, assessment of their plant growth-promoting traits, and evaluation of water hyacinth biomass as a sustainable carrier for biofertilizer formulation. The planned pot experiments are expected to provide insights into the effectiveness of the formulated biofertilizer on plant growth under controlled conditions.

Through the process of conducting this project, key learnings include:

- Gaining hands-on experience in microbiological techniques such as bacterial isolation, culture, and characterization.
- Developing a deeper understanding of plant-microbe interactions and their role in sustainable agriculture.
- Enhancing skills in scientific literature review, experimental design, and data analysis.
- Recognizing the potential of invasive plant species like water hyacinth in eco-friendly agricultural applications.

This project fosters critical thinking, scientific inquiry, and a multidisciplinary approach toward solving environmental and agricultural challenges.

Future Scope:

The future scope of the project includes:

1. Conduct field trials to test the effectiveness of the formulated biofertilizer under various soil and climatic conditions.
2. Optimize carrier material composition to improve stability, shelf-life, and nutrient content.
3. Develop standardized protocols for mass production and quality control of the biofertilizer.
4. Perform molecular and genetic studies to identify key plant growth-promoting genes in the rhizobacteria.
5. Explore the combined effect of rhizobacteria with other beneficial microbes (e.g., mycorrhizae, nitrogen-fixers).
6. Study the interaction of formulated biofertilizer with different crop species.
7. Investigate the long-term impact on soil health and microbial diversity.
8. Assess the role of the biofertilizer in improving crop yield and plant resilience under stress conditions.

9. Evaluate the environmental benefits of converting invasive water hyacinth into a useful agricultural product.
10. Explore potential for commercialization through collaboration with agro-industries.