Chapter V: Conclusion and Future Prospects

5.1. Isolation and Identification

This study successfully identified nitro-PAH-degrading rhizobacteria and nitro-PAH-accumulating plants. *Bacillus cereus* BG034 (accession numbers PQ394621) and *Bacillus altitudinis* BG05 (accession numbers PQ390296) were isolated from the rhizosphere of nitro-PAH-accumulating plants. The plant species *Cyperus rotundus* L., *Cyperus esculentus* L. (Cyperaceae family), *Axonopus compressus* (Sw.) P. Beauv., and *Imperata cylindrica* (L.) Raeusch. (Poaceae family) were identified as potential accumulators of 1-nitropyrene and 2-nitrofluorene. The isolated bacterial strains exhibited high tolerance to these pollutants and demonstrated significant degradation potential.

5.2. Co-inoculum Development

A bacterial co-inoculum (BGC01) was formulated to enhance nitro-PAH degradation. The co-inoculum showed significantly higher degradation efficiency than individual bacterial strains, achieving 76% and 87.2% degradation of 1-nitropyrene and 2-nitrofluorene, respectively, within 72 hours. Additionally, these bacterial strains exhibited plant growth-promoting properties, such as phosphate solubilization, indole acetic acid synthesis, siderophore production, hydrogen cyanide production, ammonium production, and sucrose fermentation. The application of formulated biostimulant further improved plant growth and development.

5.3. Technology Development

The plant-bacterial co-inoculum (BGCP01) and biostimulant-assisted phytoremediation approach was established and evaluated in soil microcosms under controlled greenhouse conditions. The application of plant-bacterial co-inoculum (BGCP01) and biostimulant enhanced plant growth and mitigated oxidative stress caused by nitro-PAHs, as evidenced by increased activity of superoxide dismutase, ascorbate peroxidase, catalase, and peroxidase. Furthermore, the degradation of nitro-PAHs led to the breakdown of complex aromatic structures into simpler linear aliphatic hydrocarbons, such as decane, while improving the physicochemical properties of contaminated soils.

5.4. Future Prospects

Even though the study demonstrated the significant potential of plant-bacterial co-inoculum (BGCP01) for nitro-PAH degradation under laboratory and greenhouse conditions, several challenges must be addressed for field-scale implementation. Contaminated sites often lack essential nutrients and may contain co-contaminants, including 16 EPA PAHs, heavy metals, microplastics, nanoplastics**,** and other organic emerging contaminants which could limit bacterial survival and functionality. A comprehensive understanding of soil chemistry and climatic conditions will be necessary to optimize in-situ bioremediation techniques.

The success of field applications will depend on the survival of host plants and the effective colonization of pollutant-degrading microbes in the rhizosphere. Future research should focus on identifying the specific enzymes involved in nitro-PAH degradation, as this study did not determine the enzymatic mechanisms underlying the process.