ABSTRACT

Pesticides are a vital form of biocide that plays a crucial role in modern agriculture. Numerous chemical pesticides have been developed to safeguard crops against pests and boost agricultural productivity. However, the advantages of pesticides are outweighed by their limitations, including their accumulation and persistence in the environment, ultimately leading to severe adverse impacts on the environment, non-target organisms, and human beings.

Imidacloprid is a widely used neonicotinoid insecticide of systemic nature. Imidacloprid works by interfering with the nervous system of insects, specifically by binding to nicotinic acetylcholine receptors, resulting in the disruption of nerve impulses and ultimately leading to the insect's death. Imidacloprid has been found to act against non-target organisms such as honey bees and also affect the aquatic ecosystem. Several physical, chemical, and biological approaches have been adopted for the degradation of imidacloprid. Bioremediation is the preferred method for the removal of such compounds since it is an economically viable and environmentally sustainable approach for the degradation of xenobiotics, including pesticides.

The main objective of this study is to investigate the efficacy of bioremediation in removing imidacloprid from different environmental matrices, namely water, slurry, and soil. The study aims to assess the impact of varying environmental conditions on the bioremediation process and to determine the effectiveness of this approach in degrading imidacloprid in each matrix. The research will provide valuable insights into the potential application of bioremediation as a sustainable solution for imidacloprid contamination in diverse environmental conditions.

The thesis consists of nine main chapters, followed by the references. Chapter 1 provides an overview of pesticides, including their classification, production, usage, and their impact on the environment and human health. It also discusses various treatment technologies for pesticide abatement. Chapter 2 presents a comprehensive review of bioremediation using different microorganisms and the effects of environmental parameters on the biodegradation process. It also includes a brief description of imidacloprid bioremediation and its toxicity assessment and highlights a few prominent studies related to metagenomics.

In Chapter 3, gives a detailed description of the materials and methods used in the experimental studies. The chapter covers information about the various chemicals and media used in the study, as well as the analytical techniques employed. Additionally, the chapter discusses the methods used to isolate and identify imidacloprid-degrading bacteria, conduct batch studies for imidacloprid degradation, optimize environmental parameters for biodegradation, study the process within soil microcosms, assess phytotoxicity, evaluate acute and chronic toxicity in luminescent bacteria, conduct cytotoxicity assessments, and perform metagenomic analysis. Furthermore, the chapter also outlines the methodology for lyophilization and encapsulation of bacteria, along with their applications in soil microcosms.

In Chapter 4, several studies on the bioremediation of imidacloprid are described in detail. A soil sample was collected from an agricultural field and prepared to isolate efficient imidacloprid-degrading bacteria. The study investigated the influence of environmental factors such as pH, temperature, imidacloprid concentration, and shaking speed. Bioremediation was conducted in a batch bioreactor and stirred tank batch bioreactor under optimized conditions. Key engineering parameters such as BOD, COD, and TOC were continuously monitored throughout the degradation process. Kinetic analysis was performed using different models, and mass balance and stoichiometric analysis were conducted.

In Chapter 5, various studies on the assessment of ecotoxicity were conducted. These included evaluating the reduction in toxicity of imidacloprid after bioremediation, assessing phytotoxicity using *Cicer arietinum* seeds, and examining the acute and chronic toxicity of imidacloprid and its metabolites using luminescent bacteria. Cytotoxicity assays were also performed using mammalian cell lines.

Chapter 6 encompasses a description of investigations into imidacloprid degradation in slurry and soil microcosms to provide some understanding of its degradation under environmental conditions. The biodegradation of imidacloprid was evaluated through experiments in slurry and soil microcosms. The biodegradation rates, formation of intermediate products and thus, transformation of imidacloprid were monitored over time. During the biodegradation process, a number of experimental studies were carried out to understand the roles of indigenous bacteria existing within soil microcosms and those of isolated bacteria. In addition, experiments were also

performed to study the amount of imidacloprid absorbed by *Cicer arietinum* plants. Further, the amount of imidacloprid lost through volatilization was also noted.

In Chapter 7, the metagenomic analysis was utilized to investigate the dynamics of the microbial community and the metabolic pathways involved in the degradation of imidacloprid. This analysis facilitated a comprehensive understanding of the broader ecological implications and potential interactions within the microbial community during the degradation process. A soil sample was collected for the study, and a fraction of the sample underwent bioremediation for a duration of 30 days. Subsequently, the post-bioremediation sample was collected and subjected to a comparative analysis of bacterial and functional diversity in relation to the initial sample.

In Chapter 8 of the thesis, an investigation was undertaken to explore the lyophilization and encapsulation of bacteria for potential application in imidacloprid biodegradation. The study assessed the viability of lyophilized bacterial cells after storage periods of 30, 60, 90, 120, 180, and 360 days. Soil microcosms were prepared for evaluating imidacloprid degradation, followed by biostimulation with urea, bioaugmentation with lyophilized and encapsulated bacteria, and combined biostimulation and bioaugmentation studies within the microcosms.

Chapter 9 summarizes the thesis and draws major conclusions from the research. In addition, potential areas for future research on such topics have been suggested.