ROLE OF PLANT-BASED IRON NANOPARTICLES FOR THE ADSORPTION OF HEAVY METALS

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I dedicate this thesis to my loving parents who continuously stood by my side and fully supported me.

ABSTRACT

The research synthesized iron oxide nanoparticles by employing neem leaf extract and subsequently examined the properties of the synthesized nanoparticles using UV-visible spectrophotometer, SEM/EDX, and FTIR. The analysis found that the synthesized iron oxide nanoparticles exhibited UV-visible absorption peaks at 220 nm, indicating their photosensitivity. The SEM results for iron oxide nanoparticles reveal spherical particles with a rough, irregular surface, observed at magnifications of X27,000, 5500, and 10,000, with an average size of approximately 100 nm. The production of iron oxide nanoparticles was effectively carried out, and their effectiveness in removing heavy metal-contaminated soil was assessed. The synthesized nanoparticles were analyzed using FTIR to identify the various functional groups present. The existence of multiple functional groups such as OH, carboxylic, aromatic rings, methyl, alkyl halide, and aldehyde in iron oxide nanoparticles were detected by peaks seen at 3853.9 cm-1, 3344.68 cm-1, 2922.25 cm-1, 2850.88 cm-1, 2364.81 cm-1, 1627.97 cm-1, 1193.98 cm-1, and 825.56 cm-1.. The FTIR spectra exhibited absorption peaks that were indicative of O-H, C=C, and C-O bonds at wavenumbers of 3117.37 cm-1, 1459.24 cm-1, 1577 cm-1, and 1017.96 cm-1, respectively. Furthermore, iron oxide nanoparticles have demonstrated potential in remedying soil sprayed with lead and cadmium in site I-9 and Gujar Khan. Following a significant amount of nanoparticle exposure, the levels of lead and cadmium in the soil dropped. Nanoparticle treatment for one week on lead and cadmium in soil samples from location I-9 produced various the reduction capacities. The control samples showed reduction potential of 12.8% and 15.5% for lead and 21.7% and 23.9% for cadmium from both the soils. At 2% nanoparticle dosage, declines improved over 14 days. Iron Oxide nanoparticles at 5% were consistently effective, with noticeable increases after 14 days. The soil samples from Gujar Khan site 2 had 91.3% less Cd and 93.1% less Pb than the control samples after one week. The decrease lasted two weeks. Samples with 2% nanoparticles decreased Cd and Pb potentials by 92.3% and 93%, respectively. This study demonstrates iron oxide nanoparticles' practicality for soil pollution cleanup. The results indicated that Sector I-9 soil was more successful in removing cadmium than Gujar Khan. The success of lead removal varied between the two soils based on the treatment concentration.

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ABBREVIATIONS

ASS Atomic Absorption Spectroscopy

CNTs Carbon Nanotubes

EDX Energy Dispersive X-ray

FTIR Fourier Transform Infrared Spectroscopy

GSMNPs Green Synthesis Magnetic Nanoparticles

HMs Heavy Metals

I**ONs** Iron Oxide Nanoparticles

LDHs Layered Double Hydroxides

NPs Nanoparticles

PGPR Plant Growth Promoting Rhizobacteria

SEM Scanning Electron Microscope

UV Ultraviolet–Visible Spectroscopy

XRD X-Ray Diffraction Analysis

ZVI Zero Valent Iron

CHAPTER 1

INTRODUCTION

Contamination of soil with heavy metals proves to be a huge environmental problem in Pakistan that threatens the entire ecosystem and human health. Studies have observed that traditional remediation methods can be costly and harmful to the atmosphere. Therefore, the usage of iron oxide nanoparticles is becoming more popular for remediating soil, as this method is more environment-friendly and efficient Sarim et al. (2022). According to numerous studies, iron oxide nanoparticles can remove heavy metals from the soil via different mechanisms, like precipitation, reduction, and adsorption. For instance, Huang et al. (2022) analysed how carboxymethyl chitosan-coated iron oxide nanoparticles remove lead (Pb) from polluted soil. The outcomes showed that the adsorption capacity of iron oxide nanoparticles was high for Pb.

The contamination of heavy metals poses a significant risk to the overall safety of ecosystems and the human population. The global prevalence of these perilous pollutants stems from anthropogenic activities such as industrial manufacturing and mining. Consequently, the need for sustainable and effective approaches to mitigate the adverse effects stemming from heavy metal contamination is increasing rapidly (Yadav et al., 2021). Furthermore, there has been a remarkable interest in using nanotechnology for the goal of environmental cleanup in recent years. The utilization of iron nanoparticles sourced from plants has recently garnered attention as a feasible solution for heavy metals' adsorption, owing to their unique features and diverse array of potential applications. Using plant-based nanoparticles presents a more environmentally friendly and economically viable substitute for conventional adsorbents (Eid et al., 2023). Hence, the primary objective of this chapter is to present an exhaustive assessment of the existing body of knowledge concerning the role of iron nanoparticles obtained from plants in the process of adsorbing heavy metals. The purpose of this analysis is to determine gaps to evaluate the effectiveness of plant-based nanoparticles and contemplate potential avenues for further investigation by analysing the current state of research and comprehension in this field.

1.1 Soil contamination and heavy metals

The existence of pollutants or harmful substances in the soil due to foreign or natural factors is called soil contamination. These pollutants include industrial chemicals, pesticides, heavy metals, and other dangerous materials released into the environment via different sources, such as agricultural practices, improper waste disposal, and industrial activities (Brandl et al., 2015). Heavy metals are the most common kind of pollutants that contaminate the soil. These toxic materials are naturally present in a high amount within the soil. Different human activities lead to soil contamination by heavy metals, such as smelting, usage of agrochemicals i.e. pesticides and fertilizers, industrial activities, and mining.

Heavy metals are introduced into soil through various natural and anthropogenic activities. Natural sources include weathering of rocks and volcanic eruptions, while anthropogenic sources include industrial activities, mining, agriculture, and urbanization. These metals, such as lead, cadmium, mercury, and arsenic, persist in the environment due to their non-biodegradable nature and can accumulate in soil over time. Contamination occurs when heavy metals leach from industrial sites, waste disposal areas, or agricultural lands into the soil, either through direct deposition or runoff from contaminated water. Once in the soil, these metals bind to soil particles and can persist for long periods, posing risks to environmental and human health through exposure pathways such as ingestion, inhalation, or absorption by plants. Lead concentrations in urban soils near industrial sites or highways may exceed 1000 ppm, while agricultural soils may contain less than 100 ppm. Similarly, cadmium concentrations in contaminated soils can range from 0.1 to 10 ppm, with higher levels often found near industrial facilities or waste disposal sites.

Due to these activities, heavy metals are released, which can keep piling up to harmful levels in the soil. High concentrations of heavy metals in the soil can disrupt the physiological processes of plants, such as photosynthesis, nutrient uptake, and water balance, leading to stunted growth, reduced crop yields, and even plant death. This not only impacts agricultural productivity but also has ecological consequences, as plants play a crucial role in maintaining soil fertility and providing habitat for other organisms (Huang et al., 2019). Additionally, heavy metals can adversely influence animals, humans, and plants. Heavy metals tend to penetrate the food chain, thus causing neurological damage, developmental abnormalities, and cancer (Mathur et al., 2022). For instance, according to Mahurpawar (2015), lead (Pb) exposure is known to cause neurological damage, particularly in children, leading to cognitive impairments, developmental abnormalities, and behavioral disorders. Cadmium (Cd) exposure has been linked to kidney damage, respiratory problems, and various forms of cancer (Duruibe et al., 2007). Arsenic (As) is a potent carcinogen, and chronic exposure can lead to skin lesions, cardiovascular diseases, and adverse effects on the respiratory and nervous systems (Rehman et al., 2018).

According to Kumar et al. (2023), the issue of the contamination of heavy metals in global soil has emerged as a significant ecological concern. Heavy metals including chromium (Cr), arsenic (As), cadmium (Cd), lead (Pb), mercury (Hg), and other elements characterized by significant atomic weights and densities can be heavy metals. The natural concentrations of heavy metals in soil have experienced an elevation, primarily because of anthropogenic projects like industrial operations, inadequate waste management, mining, and agriculture. The deleterious pollutants possess the capacity to persist in the environment for prolonged durations, thereby posing risks to both ecological systems and human well-being. Various sources may release heavy metals into the soil, including atmospheric deposition, industrial discharges, agricultural practices, and the utilization of contaminated water for irrigation (Priya et al., 2023). Moreover, heavy metals possess the capacity to accumulate and disperse within the soil, resulting in the contamination of crops, groundwater, and subsequently, the food chain. Luo et al. (2020) also asserted that the presence of heavy metal contamination in soil and its associated ecosystems gives rise to a multitude of adverse consequences. The presence of these substances could potentially exert a deleterious effect on the development and growth of plants, as well as on soil fertility and nutrient cycling. Moreover, the potential for heavy metals to infiltrate into groundwater poses a significant threat to both potable water sources and adjacent ecological systems (Hu et al., 2020). Thus, effective soil remediation procedures are vital for minimizing the impacts of heavy metal pollution, which may have devastating consequences for the environment and human health if left unchecked.

1.2 Nanotechnology for soil remediation

With its novel techniques to addressing soil contamination and improving soil quality, nanotechnology has come out as a promising alternative for soil remediation. Sustainable farming methods have been aided by the use of various nanomaterials, including engineered nanomaterials (ENMs) and nanoparticles (NPs), for the removal of organic pollutants and heavy metals from soil (Niroumand, et al., 2023). In addition to helping to clean up contaminated places, nanotechnology has the ability to increase soil fertility and return contaminated areas to their original state. The rigidity and strength of soil can also be improved by the use of nanotechnology, demonstrating notable advancements in geotechnical qualities for soil enhancement (Sharma, et al., 2022). Since nanotechnology is multidisciplinary, it may be used to build remediation solutions that are both focused and efficient. This highlights the need of ongoing research and development in this area for the purpose of sustainable soil management. Sustainable soil remediation makes use of nanotechnology, which provides environmentally acceptable methods for identifying and eliminating pollutants, replenishing soil fertility, and improving the health of soil ecosystems (Vu & Mulligan, 2023).

1.2.1 Iron oxide nanoparticles for soil remediation

Iron nanoparticles, with a range of 1-100 nanometers, are called iron nanoparticles. Iron nanoparticles are greatly reactive because of their large surface area. They exhibit distinctive physicochemical properties, which makes them popular in the fields of biomedicine, catalysis, and environmental remediation (Thilakan et al., 2022). Hans et al. (2022) maintain that iron nanoparticles are of two types: iron oxide nanoparticles and zero-valent iron nanoparticles (ZVI). ZVI nanoparticles are purely metallic particles of iron. Their zero-oxidation states make them well-suited for remediating inorganic and organic pollutants in groundwater and soil. On the other side, iron oxide nanoparticles are composed of oxygen and iron atoms and comprise distinctive magnetic properties.

Iron oxide nanoparticles have gained recognition for their high surface area and exceptional reactivity, which enables them to interact with heavy metal ions present in contaminated soil. Because of their large surface area-to-volume ratio and small size, iron oxide nanoparticles offer enhanced accessibility to heavy metal pollutants, facilitating their efficient removal (Sawan et al., 2020).

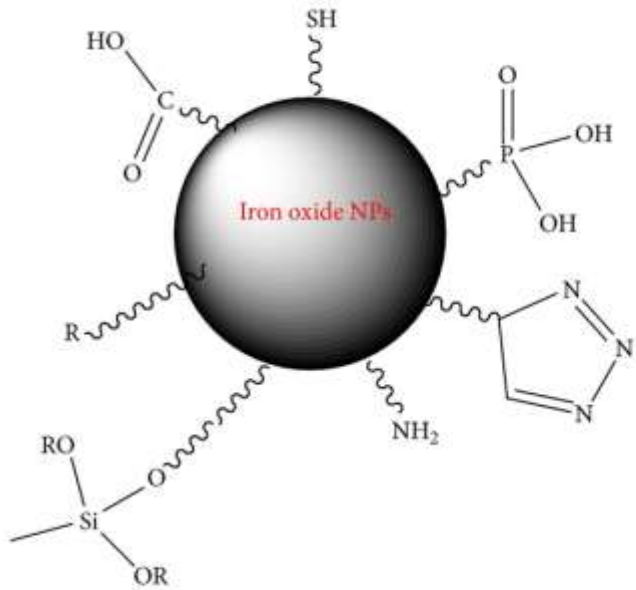


Figure 1.1: Schematic representation of surface modification of iron oxide nanoparticles.

Iron oxide nanoparticles are mostly utilized for removing organic pollutants and heavy metals present in water and soil. Han et al. (2022) demonstrated the effective adsorption of the pesticide chlorpyrifos by iron oxide nanoparticles, resulting in a significant decrease in its concentration in the contaminated soil. This highlights the versatility of iron oxide nanoparticles in addressing multiple types of soil pollution.

1.3 Soil contamination in Pakistan and application of nanotechnology

Because of practices like coal mining and wastewater irrigation, which result in substantial amounts of harmful metals, soil contamination in Pakistan is a serious concern. Studies conducted in a number of locations, including Makarwal, Drosh-Shishi valley, Vehari, and Lahore, have shown that heavy metals like Cd, Co, Cr, Cu, Mn, Ni, Pb, and Zn is present in excess of permitted levels (Ali, et al., 2023). Geological sources as well as human activities like development and agricultural compounds have been cited for the contamination. Toxic materials built up in soil and crops can have serious health effects on humans as well as ecological repercussions. To reduce health risks and guarantee food safety in Pakistan's impacted areas, it is imperative to monitor and manage the amount of heavy metals in the soil (Sarim, et al., 2022). In this context, where heavy metal contamination in soil poses a significant threat to the environment, the use of iron oxide nanoparticles for remediation purposes holds great promise. Traditional remediation methods can be expensive and environmentally harmful, making the adoption of eco-friendly and efficient approaches crucial. Iron oxide nanoparticles offer a sustainable solution for reducing heavy metal concentrations in soil, mitigating the risks to human health and the environment (Lingamdinne et al., 2017).

Moreover, iron oxide nanoparticles have been established as a promising solution for the adsorption of heavy metals in contaminated soil. Their unique properties, like high reactivity, large surface area, and magnetic behavior, make them effective in removing heavy metal pollutants (Zia-Ur-Rehman et al., 2018). The mechanisms by which iron oxide nanoparticles adsorb heavy metals include precipitation, reduction, and adsorption onto their surface (Sawan et al., 2020). Additionally, adsorption is the primary mechanism through which iron oxide nanoparticles interact with heavy metal ions in the soil. When iron oxide nanoparticles come into contact with heavy metal pollutants, electrostatic forces, surface charges, and chemical interactions drive the adsorption process. The surface functional groups of iron oxide nanoparticles, including carboxyl (-COOH) groups and hydroxyl (-OH), serve an important part in capturing heavy metal ions (Sosun et al., 2022). The nanoparticles' large surface area provides more adsorption sites, increasing their capacity to detoxify soil of toxic metals.

Furthermore, iron oxide nanoparticles also exhibit magnetic properties, which enable their separation from the soil after adsorbing heavy metals. This magnetic behavior facilitates their recovery and reuse, making them a sustainable and cost-effective remediation method (Lingamdinne et al., 2017). Moreover, the soil matrix and iron oxide nanoparticles could be readily separated using an external magnetic field, resulting in mainly purified soil with lower levels of heavy metals. Iron oxide nanoparticles have been shown in several experiments to be efficient in removing particular heavy metals from polluted soil. For instance, iron oxide nanoparticles coated with carboxymethyl chitosan have shown a significant ability to bind lead (Pb) (Bhateria and Singh, 2019). The process of adsorption includes the interaction of the positively charged heavy metal ions with the negatively charged surface of the nanoparticles, which results in their immobilization and eventual removal from the soil matrix.

According to Li et al. (2023), plant-based iron nanoparticles provide a sustainable and environmentally beneficial option for heavy metal destruction. Nanoparticle production using plant materials has the dual benefits of reducing the usage of synthetic chemicals and increasing the utilization of renewable resources. Furthermore, plant materials including biomass, agricultural waste, and plant extracts could be used as low-cost building blocks in the production of nanoparticles. Plants are a promising option for massive environmental clean-up activities due to their wealth of resources and capacity for nanoparticle manufacturing. The availability of iron in plants also makes iron nanoparticles easy to synthesize, which reduces manufacturing costs and opens up a broad range of potential environmental uses (Vu & Mulligan, 2023).

Because of their unique features and possible benefits (can concentrate on particular pollutants, are more reactive, and have an elevated surface area-to-volume ratio) nanoparticles have become known as a viable technique for soil remediation (Yadav et al., 2021). According to Sikiru et al. (2022), in soil structures, nanoparticles could be developed and manufactured to have improved motility, catalytic function, and adsorption capabilities. Furthermore, nanoparticles like zero-valent iron (nZVI) and metal oxide nanoparticles have demonstrated positive outcomes in heavy metal-contaminated soil restoration by adsorbing, decreasing, or immobilizing heavy metals. However, careful consideration must be given to the long-term destiny, possible toxicity, and environmental dangers involved with the usage of nanoparticles (Eid et al., 2023).

Nanotechnology research has shown significant growth in recent years. Nanomaterials composed of nanoparticles smaller than 100 nanometers are becoming more significant due to their wide range of uses in various fields such as biomedicine, the food industry, environmental biological remediation, storing electricity, fish farming, and even more (Lin, et al., 2019). The growing number of applications has shifted the focus of research towards creating innovations that are both economical and beneficial to the environment. Nanoparticles are suggested as a creative solution for specific societal demands. The approaches for acquiring these nanoparticles primarily revolve around three strategies: physical, chemical, and biological (Bhateria & Singh, 2019). The constraints in the advancement of physical and chemical techniques include high costs, low efficiency, and excessive energy usage. Additionally, the application of substances and surfactants, known for their toxic, acidic, and flammable properties, can have adverse effects on the natural world and the health of humans. Chemical approaches for synthesizing nanoparticles in biomedical applications were limited because of the harmful effects, unpredictability, and reduced biological compatibility of the compounds. Various procedures and strategies, such as the sol-gel method, hydrothermal synthesis, laser ablation, thermal annealing, and "powered detonations," have been associated with reducing basic materials utilizing extrinsic chemical substances (Dave & Chopda, 2014). The techniques have successfully mastered comprehensive reduction synthesizing operations, but they are complicated, costly, and environmentally unsustainable due to high toxicity. Green nanotechnologies, also known as the biological technique, offer a green alternative to traditional technologies by enhancing the generation of nanomaterials through effective and long-lasting operations that respect the natural environment. Iron oxide nanoparticles (IONPs) possess versatile features that make them highly suitable for various applications, which may result in their introduction into soil habitats. IONPs have varying adsorption characteristics towards harmful substances such as heavy metals and organic compounds, affecting their efficiency of adsorption for various toxins and the chemical reactions that occur at the IONPs-pollutants interface (Tao, et al., 2023).

1.4 Research Objectives

The goal of the research study is to assess the effectiveness of iron oxide nanoparticles in removing heavy metals from soil to prevent contamination. Keeping in view the research, the following research objectives are developed for the proposed research study,

1. To synthesize and characterize iron oxide nanoparticles
2. To investigate the reduction potential of prepared iron oxide nanoparticles in removing heavy metals from soil.

1.5 Literature Review

Heavy metal contamination is a significant worldwide problem resulting from diverse human activities. Exposure to heavy metals poses a threat to both natural habitats and human health due to its persistent, bio-accumulative, and non-biodegradable nature. Hence, the imperative task of eliminating heavy metals from the soil environment is essential to establish a stable, sustainable, and waste-free environment (Goutam, et al., 2018). There are multiple methodologies for eradicating heavy metals from the environment, however, each approach possesses its advantages and disadvantages. The utilization of biological substances to break down contaminants is referred to as bioremediation. Nano-phytoremediation is a novel bioremediation strategy that utilizes nanoparticles created by living organisms and plants to eliminate harmful heavy metals from the environment. This approach is characterized by its efficiency, cost-effectiveness, and environmental friendliness (Abisharani, et al., 2019). Engineered nanoparticles have been found to significantly mitigate the detrimental effects of metal exposure on various plant species. Due to their small size and large surface area, nanomaterials are strongly attracted to metals and can rapidly infiltrate environments that are polluted with metals. This review offers an in-depth analysis of many facets of nano-phytoremediation for heavy metal cleanup. Further investigation is required to ascertain the potentially toxic impact of nanoparticles on plants and the natural world. Additionally, it is imperative to examine the basic process by which nanoparticles are introduced into the environment. To ensure the effectiveness of the excellent quality remediation procedure, it is imperative to carefully select the appropriate plant species and nanoparticles that can effectively absorb toxins (Prakash, 2023).

The environment has seen significant changes due to increased globalization over the past two decades. As a result, there has been a growing need to find environmentally friendly remediation methods. The several origins of soil contamination encompass the utilization of pesticides and fertilizers with chemical industrial effluents and the altered byproducts of these piled chemical remnants. These events can impede the formation and soil biosphere. Various strategies encompassing physical, chemical, and biological methods have been utilized to address this difficulty (Abebe, et al., 2018). In the past ten years, there has been a notable increase in the use of nanotechnology to treat and eliminate pollutants. Nanomaterial research has provided a novel approach to address the issue of contaminated soils, offering a new perspective for remediation. Engineered nanomaterials (NMs) have been employed not only for remediating polluted places but also for effectively addressing the discharge of soil contaminants. They have pioneered environmentally conscious methods for detecting pollutants and restoring contaminated areas to their original state, hence promoting soil health. A comprehensive study that is particular to the subject and goal is necessary to achieve the desired results in nano-enabled remedial methods. The study examined current advances, highlighted areas that need advancement, and aimed to comprehend the necessity of a multifaceted approach in utilizing nanotechnology for multifunctional cleanup techniques involving several toxins (Sharma, et al., Sustainable Use of Nano-Assisted Remediation for Mitigation of Heavy Metals and Mine Spills., 2022). Comparably, Liu et al. (2020) documented the process by which iron oxide nanoparticles can effectively eliminate the herbicide chlorpyrifos from the soil. The results indicated that iron oxide nanoparticles efficiently adsorbed the pesticide, resulting in a significant reduction of pesticide levels in the polluted soil.

Several Researchers have extensively investigated the use of improved plant-based adsorbents to filter sewage for both home and commercial water treatment, with a focus on sequestering pollutants such as heavy metals and chemicals. The adsorption process is regarded as an optimal solution to costly ways of removing contaminants from sewage, despite the existence of several other viable strategies. A study by Yadav, et al., (2021) investigated the application of altered plant-derived adsorbents in effectively eliminating heavy metals and dyes. The study provided a detailed discussion of different techniques for stimulation and alterations, including physical, chemical, and composite production. It explores the use of different elements such as polymers, carbon nanotubes (CNT), graphene, layered double hydroxides (LDHs), and metal oxides. Overall, these alterations of plant-based adsorbents led to a substantial increase in the adsorption capacity. In addition, the impact of several factors, such as the amount of adsorbent used, the initial amount of contaminants, the pH level, the temperature, and the duration of proximity, on the effectiveness of adsorption was also investigated. The reactivation of the applied adsorbent averted the production of solid waste. Researchers engaged in the application of enhanced plant-based adsorbents for sewage treatment would find this assessment particularly advantageous (Yadav, et al., 2021).

In the past few years, nanoparticles have come to prominence as a novel technique for treating wastewater. Extensive investigations have been conducted to study the production of metal and metal oxide nanoparticles. Plant extracts containing polyphenols serve as effective sources of reducing chemicals for synthesizing metal and metal oxide nanoparticles with diverse shapes and sizes. This process can be carried out under varied reaction circumstances, using an environmentally friendly approach. Researchers have used bioinspired methods of green synthesis to create iron oxide nanoparticles (IONs). These IONs are highly suitable for sewage treatment due to their exceptional surface energy and optimal magnetic characteristics. Specifically, IONs can efficiently bind to heavy metal ions present in wastewater. Nevertheless, there is a scarcity of research on the production of IONs using environmentally friendly methods, as opposed to chemically synthesized IONs, to eliminate heavy metals. This study conducted by Jawed, et al., (2021) focused on the production of IONs utilizing bioinspiration, including plant polyphenols. It also explored the potential use of these IONs for removing heavy metals. This study offered useful information into this exciting area of research (Jawed, et al., 2021).

Heavy metals (HMs) are non-destroyable or non-degradable and have a long-lasting presence in the environment. Therefore, it is imperative to address and mitigate the contamination of soil ecosystems caused by HMs to establish a secure and environmentally sustainable ecosystem. Phytoremediation is a method by which plants, through their intimate proximity to the soil, can effectively contribute to soil cleaning up (Memon & Schroder, 2009). Nevertheless, when exposed to environments contaminated with heavy metals (HM), plants experience different issues such as deficits in nutrients and minerals, as well as disruptions in physical and biological functions, resulting in a decrease in the development of the plant rate. Conversely, the biological absorption of heavy metals (HMs) is an additional issue that hinders phytoremediation, as the majority of HMs are not readily accessible to plants, hence impeding effective phytoremediation (Rao, et al., 2015).

To improve the effectiveness of phytoremediation, the hindered plant development and limited accessibility of heavy metals (HMs) can be addressed by integrating nanotechnology, namely nanoparticles (NPs), or plant growth encouraging rhizobacteria (PGPR) with the phytoremediation process. Combining NPs with PGPR in a single integration can enhance plant growth by improving accessibility and absorption of nutrients, as well as controlling plant growth regulators, especially in settings where there is contamination by heavy metals. Nevertheless, there exist certain constraints, such as a substantial concentration of nanoparticles (NPs) that could potentially induce hazardous repercussions on plants (Hall, 2002). Therefore, the integration of PGPR and NPs-based remediation can overcome the constraints of each approach and therefore improve the effectiveness of phytoremediation. This review focuses on the idea of phytoremediation, which involves using plants to address the harmful effects of heavy metals (HMs) on the environment and living organisms. Specifically, it explores the use of nanoparticles (NPs) and plant growth-promoting rhizobacteria (PGPR) in combination to enhance the ability of plants to manage HMs. Furthermore, this study examines the challenges associated with the utilization of Nanoparticles and PGPR in the phytoremediation process, to identify potential areas for future studies. This analysis will help to encourage further investigation in this sector and enhance the practical application of phytoremediation (Gulzar & Mazumder, 2022).

The extensive utilization of traditional herbicides and fertilizers has exerted significant strain on both agriculture and the environment. Recently, nanoparticles (NPs) have gained attention in various industries because of their cost-effectiveness, eco-friendliness, and exceptional performance, particularly in organic farming and the management of soils. Conventional NP manufacturing processes are characterized by high energy use and have detrimental effects on the environment. Developing nanoparticles based on metals using plants is similar to chemical reactions, but biological materials are utilized rather than biochemical-reducing chemicals. This reduces the use of traditional chemicals and produces nanoparticles that are more cost-effective, efficient, less toxic, and less damaging to the environment. Green synthesized metal nanoparticles (GS-MNPs) are widely used in land cultivation to improve crop quality and efficiency. A study by Jiang, et al., (2022) presented a thorough and complete examination of green synthesis magnetic nanoparticles (GS-MNPs) in the context of soil agriculture. The study highlighted the importance of green synthesis, compared the efficacy of conventional nanoparticles (NPs) with GS-MNPs, and emphasized the advantages of GS-MNPs in land farming. The numerous applications of these nanoparticles in agriculture, including promoting plant development, controlling plant diseases, and mitigating the negative impacts of heavy metals through different methods, are described. To offer direction for sustainable agriculture, this study highlighted the limitations and potential of GS-MNPs in agricultural applications (Jiang, et al., 2022).

Iron nanoparticles (FeNPs) have attracted significant attention from experts due to their exceptional physical-chemical properties, making them highly suitable for diverse applications in various sectors. Due to their high cost and potential harm to living organisms and the environment, physical and chemical processes are not ideal for nanotechnology. Instead, biosynthetic strategies are both viable and crucial in this field. Microorganisms, fungi, and plant cells can be utilized to convert metal ions into neutral nanoparticles of metal. Biosynthesized nanoparticles possess optimal characteristics in terms of their form, size, production, and strength, making them very appropriate for environmental cleanup purposes. Industrial wastewater, which contains a diverse range of pollutants, poses a significant risk to both living organisms and the environment. Contaminants must be either eliminated or broken down before entering the natural environment and soils. The FeNPs exhibit high efficacy in the remediation of both organic and inorganic contaminants. FeNPs have demonstrated enhanced efficacy against multiple chemicals such as dyes, phenols, nitrates, phosphates, and chromium. A study by Pattanayak, et al., (2021) has specifically examined the bio-synthesis and utilization of FeNPs for remediating environment-polluted soils. The existing studies on the topic have been thoroughly examined in the literature, and the systematic presentation of findings from many researchers worldwide has been conducted. The study investigated various environmentally friendly pathways, specifically focusing on the use of plants to synthesize iron nanoparticles (FeNPs) and zero-valent iron nanoparticles (NZVI). The biosynthesis pathway was straightforward, secure, non-hazardous, and not harmful to the environment. The synthesis of nanoparticles is achieved using renewable sources. Therefore, it presented a favorable substitute for the physical-chemical synthesis method of nanoparticles. Biosynthesis offers many benefits, including lower energy requirements and a decrease in the production of hazardous byproducts during the synthesis process (Pattanayak, et al., 2021).

A study by Afzal & Singh, (2022) evaluated the effects of zinc oxide (ZnO) and iron oxide (FeO) nanoparticles (NPs) with a size of less than 36 nm, as well as their sulfate salt (bulk) a component, on growth rate and seed state of rice. Additionally, the study investigated the effect of these nanoparticles on the community of microorganisms in the rhizosphere environment of rice. Different concentrations of 0, 25, and 100 mg/kg were used for the nanoparticles and their bulk equivalent. Throughout the rice cultivation period of 2021-22, all trials were carried out under a controlled greenhouse environment. The greenhouse maintained a temperature of 30 °C during the day and 20 °C at night, with a relative humidity of 70%. The light period consisted of 16 hours of daylight followed by 8 hours of darkness. The tests were carried out using rice field soil. The findings indicated that the presence of FeO and ZnO nanoparticles at low levels (25 mg/kg) had a positive impact on the growth of grain, as evidenced by increases in height (29%, 16%) and pigment level (2%, 3%). Additionally, these nanoparticles enhanced grain quality metrics, including grains per spike (8%, 9%) and dry volume of grains (12%, 14%). The Zn (2%) and Fe (5%) deposits in the NP solutions at low levels exhibited activation when contrasted with the control group. Our findings revealed that the soil microorganisms exhibited greater variety and richness at low levels of both nanoparticles compared to the bulk-handled and control soil groups. The addition of NPs had an impact on several phyla; however, the most significant modifications were seen in abundance changes of three specific phyla: Proteobacteria, Actinobacteria, and Planctomycetes. The rhizosphere ecosystem exhibited a significant increase in microbes belonging to functional categories such as possible streptomycin makers, carbon and nitrogen fixers, and lignin degraders. However, the populations of microorganisms responsible for chitin degradation, ammonia oxidation, and nitrite lowering showed a decrease. The study findings emphasize notable alterations in many plant-related outcomes, as well as the microorganisms present in the soil around the plant roots (rhizosphere). This study provided additional insights into the unique effects of micronutrient compounds on the nanoscale level, focusing on their effect on rice plants and the microorganisms in the surrounding soil (Afzal & Singh, 2022).

Lead (Pb), chromium (Cr), cadmium (Cd), arsenic (As), and mercury (Hg) are heavy metals that are commonly found in the environment, soil, and water streams, making them significant pollutants. The pollution caused by these metals is attributed to multiple processes, such as mine waste, automobile emissions, and industrial activities related to paper, plastic, fossil fuels, and paint production. Even in small amounts, metals including Pb, Cd, Cr, and Hg can exhibit possible toxic effects. The toxicity potential is contingent upon multiple parameters, including the dosage of heavy metal beginning, soil organic matter content, soil pH levels, levels of competing ions, excrement from roots, the presence of plant species in the soil, and the age of the plants. Hence, it is crucial to accurately detect and analyze these toxic metals in both living organisms and water habitats to effectively identify and create possible solutions for their removal. Traditional practices that rely on experimental methods typically offer excellent sensitivity for multi element assessment. Nevertheless, these investigations need substantial resources, are difficult, time-intensive, and lack adaptability. Due to their advantageous optical qualities and the ability to adjust their size based on separation, metallic nanoparticles are employed to assess the level of heavy metal contamination in soil and water systems. Employing metal nanoparticles (NPs) as sensors for detecting heavy metal ions has several advantages, such as cost-effectiveness, simplicity, and high sensitivity, even at concentrations below one part per million (sub-ppm). A recent study by Dubey, et al., (2022) discovered that silver nanoparticles (AgNPs) generated from several plant-based compositions can be used as colorimetric instruments to accurately measure the level of heavy metal ions such as Cd2 +, Cr3 +, Hg2 +, Ca2 +, and Zn2 + in water. Similarly, silver NPs produced with onion extract showed specific detection capabilities for Hg2+, whilst L-tyrosine-stabilized silver NPs and AuNPs synthesized under sunlight revealed specific detection features for Pb2+, Hg2+, and Mn2+. Moreover, silver nanoparticles synthesized through exposure to sunlight demonstrated an extremely successful colorimetric sensing ability for both Co2+ and Ni2+ ions. Experts are actively attempting to produce novel materials with greater properties by harnessing the immense potential of green nanoparticles (GNPs). The study discussed the existing techniques for synthesizing GNP and explored its ability to detect and treat heavy metal waste streams produced by mining activities. The study discussed the protection, environmental effects, and future possibilities of GNP, providing valuable insights for research on this emerging topic (Dubey, et al., 2022).

Iron oxide nanoparticles (IONPs) are tiny particles composed of iron oxide, with sizes that vary from 1 to 100 nm. These particles can be both magnetite (Fe3O4) or maghemite (y-Fe2O3), and they have diverse applications in different sectors. IONPs can be produced utilizing extracts derived from various plant components, including leaves, stems, bark, flowers, seed fruits, or waste materials. The plant component consists of several metabolites, including phenols, terpenoids, flavonoids, alcohols, alkaloids, proteins, sugars, and phenols. These metabolites can serve as sealing and minimizing agents during the creation of nanoparticles. A study carried out by Adamu, et al., (2023) utilized nanoparticles commonly used in the cleanup of environmental contaminants, among other diverse uses. The primary emphasis was put on utilizing plants for nanoparticle production lies in plant species that exhibit vibrant color and a pleasant scent in their fruits, leaves, or roots, as these are recognized to hold the necessary compounds for decreasing metal ions and serving as decontaminating agents. The form and size of nanoparticles are influenced by factors such as the plant extracts used, the percentage by volume of plant extract to metallic brine solution, and the reaction circumstances including the level of plant extract, pH, period for incubation, and the metal salt. This study primarily examined the process for producing iron oxide nanoparticles using various plant extracts. It also explores the analysis of these nanoparticles and their potential use in eliminating developing pollutants, including medicines, pigments, pesticides, and heavy metals, from the natural world (Adamu, et al., 2023).

The application of nanotechnology in the cleanup and disposal of different waste streams has been progressively investigated. Nanoscale zerovalent iron (nZVI) has been thoroughly studied because of its exceptional volatility and potent reduction capabilities. Nevertheless, traditional techniques for producing nZVI particles have various restrictions, prompting the development of an environmentally friendly approach using plant-derived components. Plant extracts include diverse substances that are suitable for nZVI production, obviating the necessity for hazardous chemicals and minimizing the use of energy. Furthermore, the utilization of different plant species for the production of nZVI leads to distinct physicochemical characteristics of the nanoparticles. This review study by Kheskwani & Ahammed, (2023) presents a comprehensive examination of the production, properties, and utilization of plant-based nZVI particles for the elimination of various types of contaminants including coloring agents, heavy metals, nutrients, and trace organic contaminants from water. Further investigation into plant-based nZVI particles is required to comprehend their application in sewage treatment comprehensively. The study is particularly important for effectively eliminating a broader range of pollutants, enhancing long-term viability, and minimizing both the financial and environmental consequences of the entire procedure. Plant-based nanoscale zero-valent iron (nZVI) has demonstrated a significant ability to adsorb various heavy metals (Kheskwani & Ahammed, 2023).

Du et al. (2023) demonstrated that nanoscale zero-valent iron (nZVI) produced from green tea leaves exhibited a significantly greater ability to adsorb for Pb(II) in comparison with nZVI generated using chemical means, with a sevenfold increase. The researchers observed higher effectiveness in removal when Fe2(SO4)3 was utilized as the precursor iron in plant-based nZVI, in comparison to FeSO4. When Fe2(SO4)3 was utilized, the adsorption capacity reached 377.3 mg/g, surpassing the 327.6 mg/g achieved with nZVI created with plant extract and FeSO4. This difference can be attributed to the FeSO4 particles' tendency to agglomerate more easily (Du, et al., 2023). Research has been carried out to investigate the impact of many parameters, such as pH, contact time, temperature, and the amounts of absorbent and adsorbate, on the procedure of adsorption of heavy metals by plant-based nZVI. The optimal pH is determined by factors such as the functional groups found in nZVI, which depends on the specific plant chosen, the charge of the contaminant to be eliminated, and the electrostatic attraction.

In their study, Rana et al. (2018) examined the process of oxidizing As(III) utilizing nZVI that was generated from guava plants. The experiment was conducted at various pH levels. At a pH of 3, within the initial 2 minutes, 70% of As(III) underwent oxidation to As(V) due to a Fenton-like reaction occurring at an acidic pH. This process prevented the creation of an oxide layer covering, which might potentially impede adsorption (Rana, et al., 2018). Under conditions of neutral pH, a 30% oxidation occurred over 2 minutes, whereas a 74% oxidation occurred within 10 minutes. In the initial 2-minute period, there was no notable occurrence of oxidation due to the simultaneous presence of As(III) and As(V), which resulted in competition for the active binding regions. At a pH of 9, the efficiency of elimination via oxidation significantly fell to 25% due to the formation of ferrous and ferric hydroxides on the surface. This resulted in a reduction of active sites available for adsorption. Additionally, they proposed that when the pH is high, the polyphenols present in the plant extract become ionized, resulting in the unavailability of the sealing agents. As a result, ZVI undergoes a reaction with oxygen in the atmosphere, leading to the formation of hydroxides. Therefore, a decrease in pH results in improved efficiency (Arshadi, et al., 2017).

Increased pH levels have been found to result in greater elimination of heavy metals. Arshadi et al. (2017) discovered that there was a greater elimination of heavy metals, such as Pb(II) and Hg(II), within the pH range of 5–9. At an elevated pH level, the adsorbent's surface acquired a negative charge, resulting in the formation of Pb(OH)2 precipitates and the protonation and deprotonation of hydroxyl species on the surface of nZVI. While the absorption was significant under alkaline conditions, the presence of iron hydroxides impeded the uptake process (Yan, et al., 2012). Pollutant elimination exhibits a positive correlation with the duration of contact, reaching a state of equilibrium at a certain point. Subsequently, the absorption becomes insignificant because all available active sites on the adsorbent are occupied. With a rise in the amount of adsorbent, the absorption also increases due to the greater number of active sites for adsorption. However, exceeding a specific threshold may lead to the aggregation of nZVI particles, resulting in a reduction in efficiency. Higher pollutant concentrations necessitate longer removal times and result in reduced removal efficiency, as the contaminants quickly fill the available sites on the adsorbent. The processes that comprise the elimination of heavy metals using nZVI encompass adsorption, absorption, precipitation, co-precipitation, oxidation, and reduction. The primary process is contingent upon the redox properties of the contaminant and the pH of the solution, along with the ionic capacity and concentration of the adsorbent (Weng, et al., 2016).

CHAPTER 2

MATERIALS AND METHODS

2.1 Research methodology

The goal of the present research was to characterize and synthesize iron oxide nanoparticles to treat the contaminated soils with heavy metals such as Cd, Cr, Pb, and As. The research first collected the raw material of *Azadirachta indica* leaves (neem plant leaves). The two soil samples were obtained from agricultural sites, one from the sector I-9 Islamabad and one from Gujjar Khan.

The research has several reasons for choosing soil samples from Gujar Khan and Sector I-9 in Islamabad. It makes it possible for the study evaluate the degree of regional contamination, analyzing the effectiveness of nanoparticles in a range of settings, improving the generalizability of the results, and carrying out comparative analysis to comprehend location-specific factors influencing nanoparticle remediation processes. This method offers a thorough grasp of the interactions between iron oxide nanoparticles and different types of soil, providing information on local environmental issues and the practicality of using nanoparticle-based remediation techniques in actual situations.

2.2 Preparation of nanoparticles

2.2.1 Synthesis of Iron oxide (FeO_2) nanoparticles (Green synthesis)

Iron oxide nanoparticles were produced using Neem leaf extracts through an environmentally friendly method. The leaf extracts were prepared by taking 100 grams of green Neem leaves (Iqbal, et al., 2021). The leaves were washed with double distilled water twice and then cut into pieces and boiled in 200 ml of distilled water at 60˚C for 20 mins to avoid any inactivation of biochemical compounds present in the neem leaf which may help in the synthesis of iron oxide nanoparticles. The resulting solution was filtered through Whatman filter paper, and the filtrate was retained and stored at 4℃ in the refrigerator. The 100 ml filtrate was then mixed with $FeSO_4$ solution and stirred on a magnetic stirrer for 2 hours. The black suspension obtained was then filtered, washed with distilled water and ethanol several times, and finally dried at 100℃ overnight. The obtained powder was further characterized to know more about its material properties. The Neem leaf extract and iron chloride were taken at 2:1 concentration. The solution was heated and continuously stirred with a magnetic stirrer. A large amount of black precipitates ere formed following 24 hours. Filtration process took place for up to two to three hours (Kolekar et al., 2011). In the next step, it was centrifuged at 1300 revolutions per minute (RPM) for 40 minutes. Further, it was oven-dried and crushed using a pestle and mortar. The resulting nanoparticles were obtained in a plastic container. Nanoparticle characterization was performed through FTIR and SEM analysis to determine the physical and chemical properties of iron oxide nanoparticles. After the filtration process, the precipitates were washed with distilled water and kept in an oven at 105.7 C temperature. The main purpose of drying the nanoparticles was that there should be no humidity in the particles. This process took place for up to two to three hours. After this step, nanoparticles were then grounded to a very fine powder. In the end, calcination of nanoparticles is done at different temperatures such as 50, 700, and 900 C. The color of nanoparticles was changed at different temperatures. The main purpose of calcination was to know the effect of calcination on iron oxide NPs as an increase in calcination temperature leads to an increase in crystallized size. Band gap values increase with the increase in calcination temperature (Mohan et al., 2016).

Figure 2.1(a): Preparation for nanoparticles

Figure 2.1(b) Neem leaf extracts for the green synthesis of nanoparticles

2.2.2 Characterization of Synthesized nanoparticles

The characterization approach was used to study the spectral properties of iron oxide nanoparticles. Characterization of nanoparticles defines size, morphology, surface chemistry, elemental composition, and crystallinity. SEM/EDX, UV, and FTIR were used here for analysis of iron oxide nanoparticles (Thakral et al., 2021). The characterization of the surface of nanoparticles means which functional group is present in samples that will be responsible for the reduction of metal nanoparticles (Hossain et al., 2010).

2.3 Scanning Electron Spectroscopy

SEM is a technique used to analyze the external morphology, topography, chemical composition, crystalline structure, and arrangement of materials making up a sample. It is an efficient approach for the analysis of organic and inorganic materials on a nanoscale to micrometer scale. The system employs a variety of electron guns, and the electron beams they produce have a wide range of properties (Berthomieu et al., 2009)..

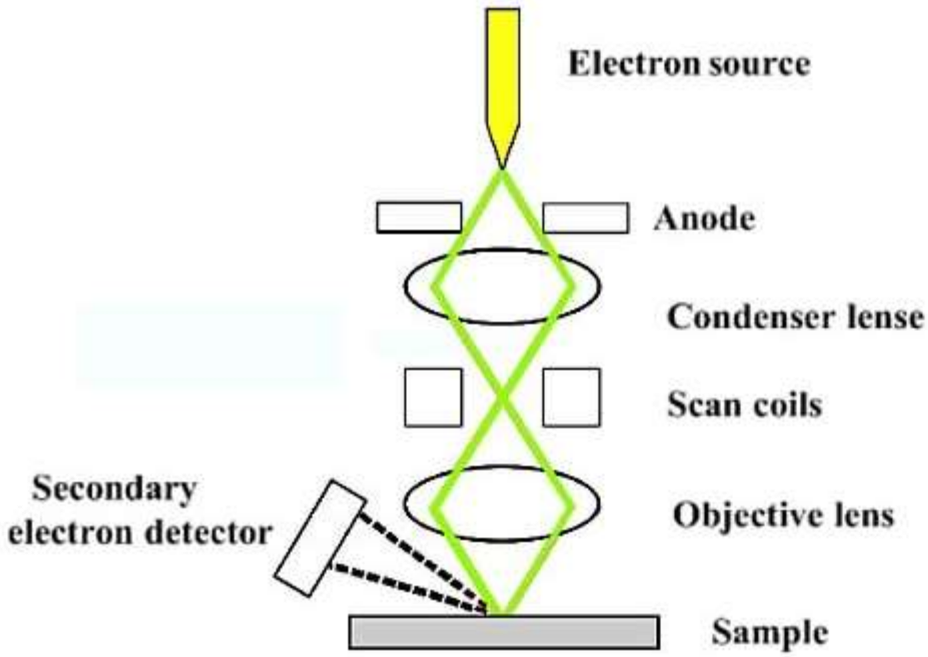


Figure 2.2: Working of SEM (Scanning electron microscopy)

2.4 FTIR (Fourier Transform infrared spectroscopy)

FTIR spectroscopy investigates vibrational properties of amino acids and cofactors to identify chemical groups in reactions. It's used to characterize nanoparticles by identifying different materials present on the absorption spectrum.(Gaffney et al., 2002). (Eid et al., 2022).

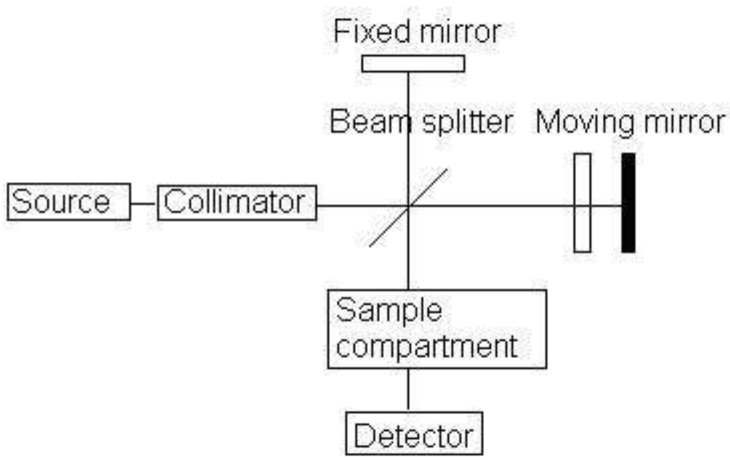


Figure 2.3: Working principle of FTIR Spectroscopy

2.5 Ultra-visible Spectroscopy

UV-visible spectroscopy is a method to determine the optical absorption of particles in analytical chemistry. It measures the absorption and electronic transitions of a sample when it absorbs UV or visible light. The amount of light absorbed by the sample at different wavelengths is quantified. In the first step, 0.1% ethanol is taken, and the absorbance peak is given at 370 nm. Scanning is done to identify the peaks of iron oxide nanoparticles.(Kumar et al., 2013).

2.6 Soil sampling and primary analysis

Two soil samples were collected from agriculture sites, one in Islamabad and the other from Gujjar Khan agricultural site. Samples were obtained at a depth of 0 to 50 cm. Five soil samples from each site were collected into plastic bags and were transported, and refrigerated in the Environmental Sciences Lab, Bahria University for further analysis. Soil samples were primarily analyzed for basic soil parameters (pH, EC, Temp). At the initial stage presence of heavy metals was assessed in the soil sample through ICP-OES.

Figure 2.4: Collected soil samples from agricultural sites, Gujjar Khan and Islamabad

2.6.1 Sample preparation of soil samples

To evaluate the effectiveness of nanoparticle treatment for immobilizing metal and compounds, samples of 20 g of polluted soil were treated with different NP doses. For the preparation of soil samples, the soil samples were irrigated with distilled water until the water-holding capacity of the soil was reached. After this, different doses of nanoparticles 0%, 2%, and 5% (w:w) in plastic bottles were added and in the soil respectively. The experiment was performed in triplicates. Bottles were placed on the shaker for 72 hours at 60 rpm. After incubation soil was dried and sampled for analysis. For the digestion of soil samples, perchloric acid ($HClO_4$) was utilized. 5 grams of soil was put into the digestion flask, and 10ml of acid was added to it. The flask was heated on the hot plate for 20 minutes at 100℃. 30 ml of distilled water was also added into the digestion flask before putting it onto the hot plate. After 20 minutes the flask was removed from the hot plate and was placed to cool down. After this, the solution was filtered using the Whatman filter paper. The filtrate was collected and properly labeled to make it ready for testing through ICP-OES for analysis of heavy metals in soil.

Figure 2.5(a): Prepared nanoparticles for soil treatment

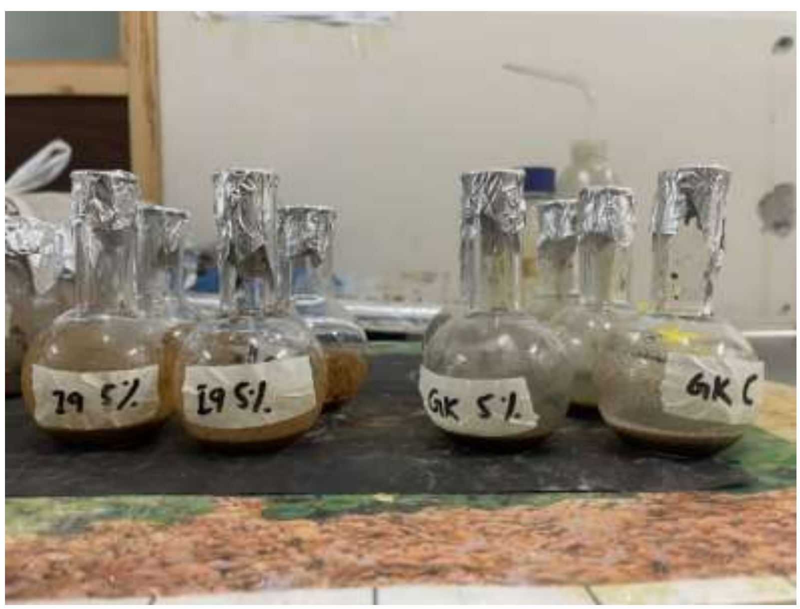


Figure 2.5(b): Acid digestion of soil samples with perchloric acid

2.7 Heavy Metal Analysis

For the quantification analysis of heavy metals by atomic absorption, 2.5 gm of each of the samples was acid digested with aqua-regia (HNO_3: HCl; 1:3), 2.5 grams of soil sample was digested with 15 ml of aqua-regia (HNO3: HCl;1:3). Sample were left overnight at room temperature, followed by heating on a hotplate to 100˚C for 3 hours until almost dried. After this, they were allowed to cool at room temperature and diluted with 20 ml of 2% nitric acid (HNO3) followed by filtration using Whatman qualitative filter paper No. 1. The filtrate was marked and labeled properly before being sent for analysis. Heavy metals were analyzed through ICP-OES.

The results of analysis of both soil samples showed values of Lead (Pb) and cadmium (Cd) below detection limit (BDL). For this reason, both the soils were sprayed with solutions of Lead and Cadmium nitrate to see removal efficiency of prepared nanoparticles. A solution consisting of 200 parts per million (ppm) of lead nitrate and cadmium nitrate, including a total of 16 milligrams (mg) of lead and cadmium nitrate dissolved in 80 milliliters (ml) of distilled water, was applied as a spray over soil samples obtained from I-9 and Gujar Khan. The soil that underwent treatment was allowed to remain in the laboratory for the duration of one night. Afterwards, 0.5 grammes of nanoparticles were introduced into the soil that had been treated with lead and cadmium. Following a week, the initial five samples from both I-9 and Gujar Khan soils were examined using atomic absorption spectroscopy. After two weeks, an additional set of five samples from every site with 1g of nanoparticles were examined to evaluate the effectiveness of nanoparticle elimination.

Figure 2.6: Prepared twenty samples of the soil for heavy metal analysis

2.7.1 Inductively Coupled Plasma Optical Emission Spectroscopy ICP-OES Principle

Analysis techniques like inductively coupled plasma optical emission spectroscopy (ICP-OES) can determine how much of a specific element is present in a sample. In order to move electrons from the ground state to an excited state, the ICP-OES concept relies on the idea that atoms and ions may absorb energy. The energy source in ICP-OES is heat from an argon plasma working at 10,000 kelvins. Excited atoms must emit light at specific wavelengths upon deceleration to a lower energy level in order for the ICP-OES principle to operate. The amount of light produced at each wavelength depends on how many atoms or ions makes the transition. The Beer Lambert law outlines the connection between light intensity and element concentration (Morrison, et al., 2020).

Inductively coupled plasma has been utilized extensively to analyze trace metals in a system. Inductively coupled plasma-optical emission spectroscopy (ICP-OES) and inductively coupled plasma-mass spectrometry are two techniques that can be utilized to perform ICP (ICP-MS). One of the main benefits of employing ICP-MS from a lab perspective is its multi-element capacity, which allows many parameters to be measured simultaneously in a single analysis. When used in the lab, ICP has the ability to process samples at an incredibly high capacity owing to its quick analytical times and simple sample preparation. (Bulska & Wagner, 2016)

In a wide range of research fields, including forensic, earth, environmental, and biological sciences as well as the food, material, chemical, integrated circuit, and nuclear sectors, it is common practice to use inductively coupled plasma (ICP) mass spectrometry (MS) and Emission Optical Spectroscopy (OES). Due to their high ion density and high temperature, plasmas may efficiently atomize and elementally ionize all types of samples and matrices given via a range of specialized equipment. High sensitivity (ppt-ppq), relative salt tolerance, compound-independent element response, and optimal quantitation accuracy are just a few of the unique characteristics that contribute to ICP's unmatched performance in accurately detecting, classifying, and characterizing trace elements. (Ammann, 2007)**.** ICP-OES is currently considered as the most powerful technique due to its accuracy, sensitivity, excellent detection properties, least detection time, negligible chemical interferences, and no need for multiple dilutions as it can detect multiple elements from an analysis. It can detect up to two to seventy elements (Khan, 2022).

The twenty samples of soil were evaluated and analyzed using Inductively Coupled Plasma Optical Emission Spectroscopy ICP-OES (Model-5110), for two selected heavy metals naming Lead (Pb), and Cadmium, to evaluate the effectiveness and reduction potential of iron oxide nanoparticles.

CHAPTER 3

RESULTS AND DISCUSSION

The chapter discusses the results of the synthesis of iron oxide nanoparticles for the removal and treatment of contaminated soil with heavy metals. The chapter will also discuss the removal efficiency of iron oxide nanoparticles in the treatment of soil contaminated with heavy metals. All the results are discussed hereafter.

The research work consisted of synthesis of iron oxide nanoparticles and their characterization through FTIR, SEM, and UV spectrophotometer. The results for the removal of heavy metals, Cadmium, and Lead from soil carried out through ICP-OES, evaluating the reduction potential and removal efficiency of iron oxide nanoparticles.

3.1 Characterization through FTIR

FT-IR was done to investigate the functionalized chemical group and chemical constitution of iron oxide nanoparticles. There is an interpretation of results by the help of graphs before and after the calcination of iron oxide nanoparticles by using and without using surfactant at 600C. Calcination of synthesized nanoparticles is done to determine the crystalline structure of iron nanoparticles. Calcinated samples of iron FTIR show that by increasing calcination temperature, the sharpness of the characteristic peaks for metal oxide increases. By increase in particle size due to calcination stability of iron oxide nanoparticles increases. In the FTIR spectrum on X –the axis there is wavenumber cm-3 and on the y-axis there is transmission %. FTIR spectrum shows three types of peaks weak, strong and broad peaks. Broad and strong signals have polar regions that have hydrogen bonding and sharp signals 28 have nonpolar regions. Wave number decreases when the mass of functional groups increases, and wavenumber increases when bond length decreases. The presence of functional groups in Iron oxide nanoparticles was investigated by FTIR. To differentiate the bonding of the compounds before and after calcination the FTIR spectra were studied to analyze chemical bonding of structure. Fig 3.1 illustrates the FTIR spectrum that explains that synthesized functional groups are exhibited at 3853.9 cm^{-1}, 3344.68 cm^{-1}, 2922.25cm^{-1}, 2850.88 cm^{-1}, 2364.81 cm^{-1}, 1627.97 cm^{-1}, 1193.98 cm^{-1}, and 825.56 cm^{-1}. The band at 3124.99cm-1 was due to the OH band of the carboxylic group.

The absorption peaks at 1577cm^{-1} indicate the presence of aromatic rings in the molecule and 1460-3 cm^{-1} shows the presence of methyl group. Small absorption peaks at 1018cm^{-1} showed alkyl halide functional groups and at 1125.97 cm^{-1} showed methyl group. Fig illustrates the absorption peaks at 3115.77cm-1 that show stretching vibrations of aldehyde groups. When iron oxide nanoparticles are calcinated at 600 C then the peaks at 1159.cm^{-1} disappeared and the Fe-O stretching band was formed at peak 426cm^{-1} (Yang et al., 2016). Fig 3.1 illustrates the chemical bonding between polyvinyl alcohol and Fe_2O_3 nanoparticles that were observed by FTIR spectroscopy. The crystallinity of iron oxide nanoparticles increases with the increase of calcination temperature and the results show absorption peaks of O-H, C=C, and C-O at wave numbers 3117.37 cm^{-1}, 1459.24cm^{-1}, 1577cm^{-1}, and 1017.96cm^{-1} before calcination (Kim et al., 2008). According to existing literature, Fig 3.4 illustrates the same absorption peaks with minor differences in their intensity and the existence of Fe_2O_3 vibration modes confirming the existence of iron oxide nanoparticles at 400cm-1 (Norouzi et al., 2021). Therefore, it may be inferred that the metabolites involved in the reduction process belong to the aromatic compound's family. The Neem extract in both solvents contains simple phenols and variants of phenolic chemicals, which may contribute to reducing metal ions and forming FeO NPs. The reducing process of the parent nanoparticle forms a Fe0-phenolate combination through chelation, leading to the formation and development of the nanoparticle. The varied details of the samples helped consolidate crucial information. The FeO nanoparticles produced using the extract in 96% Neem leaf extract exhibited distinct properties compared to the colloid formed using the extract in 100% solvent.

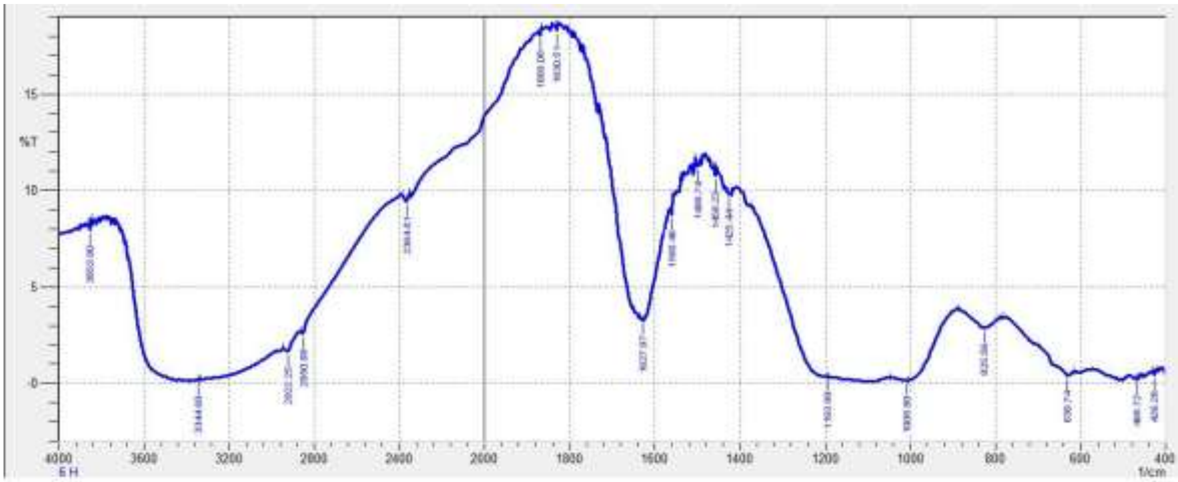


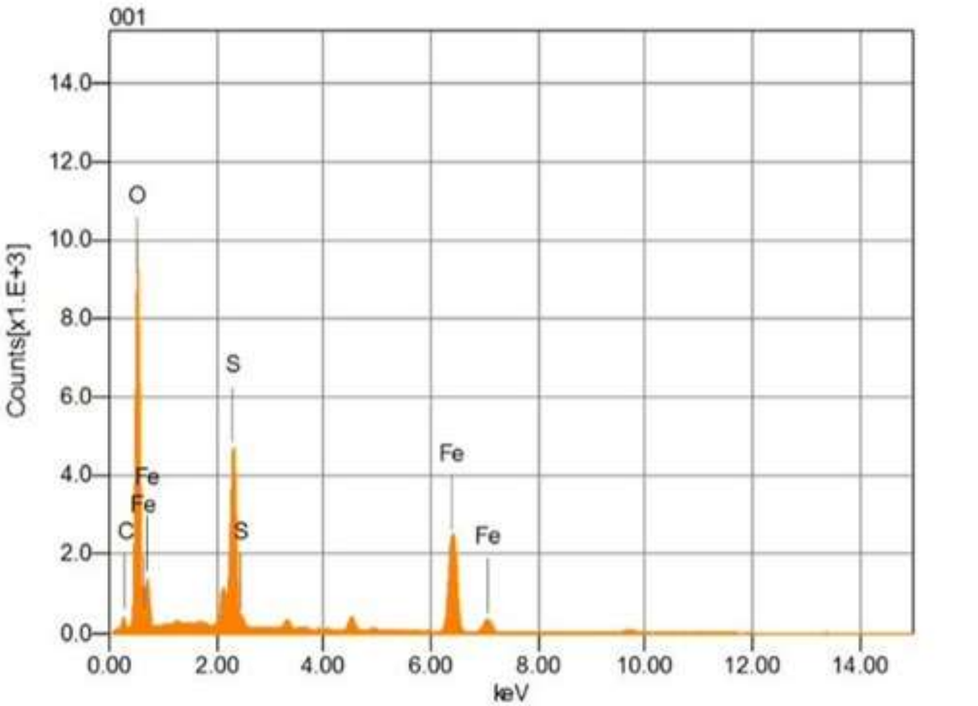
Figure 3.1: Results of characterization of nanoparticles (Iron oxide) through FTIR spectroscopy

Table 3.1: FTIR results of Iron oxide nanoparticles

Wavenumber (cm-1)	**Functional groups**
3124.977 cm-1	O-H bands
1577cm -1	Aromatic C=C
1460cm-1	CH2 bending vibration
1125.97	Methyl group
426	Iron oxide
3115.77	Stretching bands of aldehyde groups
3117.37	O-H group
1459.24	C=C
1577	C-O

3.2 SEM analysis

Figure 3.2 and 3.3 show the SEM results, particle size and images of iron oxide nanoparticles. The SEM image was taken at X27,000, 5500, and 10,000 magnifications. The image shows iron oxide particles are spherical in shape with rough and irregular surface and the size of the particles was around 100 nm.



Formula	mass%	Atom%	Sigma	Net	K ratio	Line
C	6.80	11.90	0.02	9278	0.0027502	K
O	53.18	69.84	0.07	256378	0.3472987	K
S	11.50	7.53	0.04	189494	0.0914461	K
Fe	28.52	10.73	0.08	173470	0.2423252	K
Total	100.00	100.00				

Figure 3.2: Results of SEM analysis

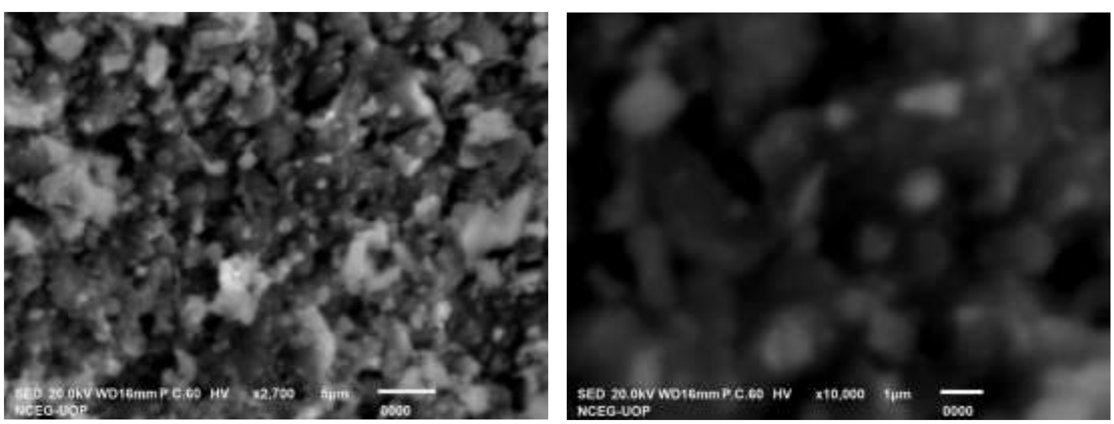


Figure 3.3(a): SEM Images

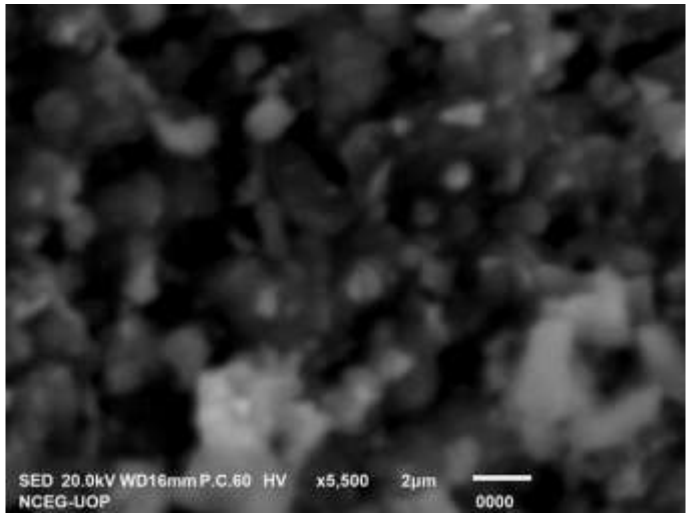


Figure 3.3 (b): SEM Images

3.3 UV Spectroscopy

The results of UV spectrophotometry showed that on wavelength of 220nm the absorbance was 1.909, and the wavelength of highest peak was 237m. At different wavelength different absorbance was noted. 2.22 on 700nm, 1.50 on 476nm, 2.46 on 237nm, 2.28 on 531nm, 1.49 on 468nm, 1.48 on 494nm and 2.18 on 371 nm. However, upon change in particle size or particle shape, a slight shift in the absorption was observed. The average wavelength exhibited by nanoparticles was 220nm indicating their photosensitivity.

3.4 Iron oxide nanoparticles for the removal of heavy metals from soil (Cd and Pb)

Lead is employed in diverse industries such as battery production, construction, munitions, electronics, and radiation shielding due to its distinctive characteristics, such as high density and malleability. Nevertheless, the extensive utilization of it has led to substantial environmental contamination. Industrial activities and the use of lead petrol, along with its inclusion in paints and soldering materials, are sources of lead emissions that contaminate the air, land, and water (Rahimi, et al., 2015). The pollution presents significant health hazards leading to delays in growth, neurological impairments, and several other health problems. Attempts to limit lead contamination encompass legislative measures, such as the prohibition of leaded petrol and paints, with remediation endeavors to cleanse polluted locations and avert more environmental harm. (Andrade-Zavaleta, et al., 2022). Fe_3O_4 nanoparticles were used to remove Pb(II) ions from water employing sequential adsorption. An in-depth study has been conducted on how temperature, pH, and the presence of other ions impact the adsorption of Pb(II). Adsorption equilibrium was reached in 30 minutes. There is a spike in the rate of adsorption of Pb(II) with rising temperature, suggesting endothermic adsorption. Additionally, the presence of coexisting cations does not affect the adsorption. The compact dimensions and extensive surface area of magnetite nanoparticles render them well-suited for adsorption (Recillas, et al., 2011). Heavy metals efficiently spread throughout the active layer of Fe_3O_4 nanoparticles. The outermost area of nanoparticles of magnetic material plays an essential role in the adsorption step. High-surface nanoparticles are prone to agglomeration, reducing their efficiency. Therefore, it is crucial to change the structure of IONPs to enhance their ability to function. The surface of iron oxide nanoparticles can be altered by bonding them with appropriate functional groups such carboxylic acids, phosphoric acid, silanol, thio, and amine, as well as tiny organic compounds, biological molecules polymers, and other metallic nanoparticles (Li, et al., 2014).

Cadmium, and Lead are significant metals in industry used in numerous goods and operations with consequences for the environment. Remediating these metals in polluted areas poses practical and financial challenges due to the high cost and complexity of existing approaches. Iron oxide (Fe_2O_3) nanoparticles were synthesized, characterized, and investigated for removing Cr, Pb, Cd, and As from soil contaminated with these metals in the current study. Fe_2O_3 nanoparticles were produced through the Neem leaf extracts and were analyzed using UV–Vis and FTIR spectroscopy. Group studies were conducted with varying amounts of Fe_2O_3 nanoparticles to reduce soil contaminated with Cr, Cd, As, and Pb. The nanoparticles of iron oxide at an average concentration of 0.5 and 1g exhibited nearly 95% reduction capability within two weeks.

3.5 Heavy metals concentrations in soil with nanoparticles

Heavy metals are naturally occurring elements that have high atomic weight and number. These metals are considered serious harmful pollutants in the environment because they have toxic effects and persistent bioaccumulation problems. Soils samples were obtained from two sites, sector I-9 agricultural site and Gujar Khan agricultural site. The resultant values obtained after the treatment of heavy metals were compared on the basis of time period for soil samples with the duration of one and two weeks. Heavy metals are the most detrimental contaminants in the environment. Environmental mediums such as soil, water, and air are contaminated by human activities, farming operations, and the discharge of waste from factories and heavy metals. They tend to form chemical bonds with proteins and enzymes, leading to disruptions in human biological systems. Nanotechnology is the most effective approach for the elimination and management of heavy metals due to its capacity to promote sustainability. In the study, soil samples from Islamabad and Gujar Khan were collected to investigate the reduction potential of nanoparticles (iron oxide) to remove lead and cadmium from the soil with the addition of iron oxide nanoparticles at varying doses of control, 2% and 5%. The soil samples were tested though ICP-OES.

Table 3.2: Results of heavy metals Cd, and Pb analyzed in soil samples through ICP-OES

S. No	**Sample Code**	**Cd (ppm)**	**Pb (ppm)**
1	I-9 2% (1-8)	0.135	28.562
2	I-9 5% (1-8)	0.179	26.214
3	I-9C (8-16)	0.138	41.297
4	I-9 5% (8-16	0.171	51.111
5	I-9 2% (8-16)	0.173	53.079
6	1-9-8%	0.00019	0.00331
7	1-9-2%	0.00015	0.00011
8	(1-8) 1-9 5%	0.00013	0.00402
9	1-9-2%	0.00018	0.0011
10	GK 2% (1-8)	0.177	33.512
11	GK 5% (1-8)	0.128	18.361
12	GK 2% (8-16)	0.122	39.670
13	GK 5% (8-16)	0.158	44.979
14	GKC (8-16)	0.151	53.365
15	GK-2%	0.00015	0.00189
16	GK-5%	0.00019	0.00289
17	GK-5% 8-16	0.00016	0.00325
18	1-8 GK-2%	0.00017	0.00289
19	I-9 Control	25.6	43.55
20	GK Control	31.1	47.88

Soil samples collected from locations in I-9, Islamabad, and Gujar Khan were analyzed for the presence of cadmium (Cd) and lead (Pb) concentrations. During the I-9 experiment, various procedures were applied, involving the use of 2% and 5% nanoparticle solutions. The findings revealed varying levels of Cd and Pb among the treatment and control groups. The control samples owing to high metal concentration had shown reduced reduction potentials. In I-9, the samples that were treated with 2% nanoparticles showed Cd values that varied from 0.135 ppm to 0.173 ppm and the amount of Pb varied from 28.562 ppm to 53.079 ppm. Furthermore, in Gujar Khan, similar treatments were administered, with the amounts of Cd and Pb fluctuating correspondingly. The results of this study offer valuable information about the effectiveness of nanoparticle treatments in decreasing the levels of heavy metals in soil samples collected from both I-9, Islamabad, and Gujar Khan.

Table 3.3: Results of nanoparticles treatment for lead and cadmium for Site 1 sector I-9 soil

Sample Codes	**Percentage of reduction potential (Cd)**	**Percentage of reduction potential (Pb)**
After one week		
I-9 Control	12.8	21.7
I-9 2% (0.5g)	85.6	85.7
I-9 2% (0.5g)	89.7	86.1
After two weeks		
I-9 Control	12.8	21.7
I-9 2% (0.5g)	90.2	90.3
I-9 2% (0.5g)	91.9	93.3
After one week		
I-9 Control	12.8	21.7
I-9 5% (1g np)	91.2	86.8
I-9 5% (1g np)	90.7	92
After two weeks		
I-9 Control	12.8	21.7
I-9 5%(1g np)	91.1	94
I-9 5%(1g np)	93	99.1

For the soil samples from site I-9 treated with nanoparticles for lead and cadmium, the percentage reduction potential after one week varied across different treatments. The control samples had no nanoparticles, however in relation to reduction potential and metal concentration they showed percentages of 12.8% and 21.7% for lead and cadmium. When treated with a 2% concentration of nanoparticles (0.5g), reduction percentages after one week were recorded at 85.6% for cadmium and 85.7% for lead. However, with the same nanoparticle concentration, after two weeks, improvements were observed, with reduction percentages increasing to 90.2% for cadmium and 90.3% for lead. Additional testing with a greater quantity of nanoparticles (5%, 1g NPs) showed an additional pronounced decrease in possibilities for reduction. The following week, the levels of cadmium decreased by 91.2% and lead decreased by 86.8%. After a period of two weeks, the reduction improved to 91.1% for cadmium and 94% for lead in the initially conducted experiment, and 93% for cadmium and 99.1% for lead in the following experiment. The results indicate that the effectiveness of Iron Oxide nanoparticles remediation improves over time, and larger quantities lead to greater decreases in the amount of heavy metals in the soil.

Table 3.4: Results of nanoparticles treatment for lead and cadmium for Site 2 Gujar Khan soil

Sample Codes	Percentage reduction potential (Cd)	Percentage reduction potential (Pb)
After one week		
GK Control	15.5	23.9
GK 2% (0.5gnp)	89.1	84.2
GK 2% (0.5gnp)	88.7	85.1
After two weeks		
GK Control	15.5	23.9
Gk2% (0.5g np)	91.1	89.3
GK 2% (0.5g np)	92.3	93
After one week		
GK Control	15.5	23.9
GK 5% (1g np)	89.3	88.1

GK 5% (1g np)	90.1	91.1
After two weeks		
GK Control	15.5	23.9
GK 5%(1g np)	92.1	90.2
GK 5%(1g np)	91.7	92

The soil samples collected from location 2 in Gujar Khan were analyzed to determine the possibility of reducing the levels of cadmium (Cd) and lead (Pb) after treatments with nanoparticles, lead, and cadmium nitrate. The examination was conducted using ICP-OES. The control samples had no nanoparticles, however in relation to reduction potential and metal concentration they showed percentages of 15.5% and 23.9% for lead and cadmium. The samples exposed to a nanoparticle concentration of 2% (0.5g np) exhibited somewhat lower reducing possibilities, with Cd levels varying from 88.7% to 89.1% and Pb levels varying from 84.2% to 85.1%. Significantly, after a duration of two weeks, substantial enhancements were detected in all treatment options, while the control samples exhibited comparable reduction capacities. Conversely, the samples that were exposed to a concentration of 2% nanoparticles exhibited improved reduction possibilities, with the ability to reduce Cd by up to 92.3% and Pb by up to 93%. In a similar way samples exposed to a 5% concentration of nanoparticles (1g np) showed different levels of potential for reduction after both the first and second weeks. The decrease in Cd ranged from 89.3% to 92.1%, while the corresponding decrease in Pb ranged from 88.1% to 92%.

Applying iron oxide nanoparticles to soil samples from both sites shows potential for effectively addressing heavy metal pollution through restoration. The enormous size and physical properties of iron oxide nanoparticles make them highly effective in adsorbing heavy metals such as cadmium and lead. By incorporating iron oxide nanoparticles into the soil, they can efficiently combine with the heavy metal ions, thereby decreasing their movement and accessibility for absorption by vegetation or seepage into groundwater. Furthermore, the magnetic characteristics of iron oxide nanoparticles provide effortless extraction from the soil following examination, hence reducing the potential environmental consequences. By applying and utilizing iron oxide nanoparticles, we can effectively reduce the concentration of heavy metals in soil, leading to restoration of the environment and protection of human well-being. The results showed a starting removal effectiveness of 80-90% within the first week. Further examination after a period of two weeks revealed removal efficiencies ranging from 95 to 99%, successfully eliminating the soil of heavy metal pollutants.

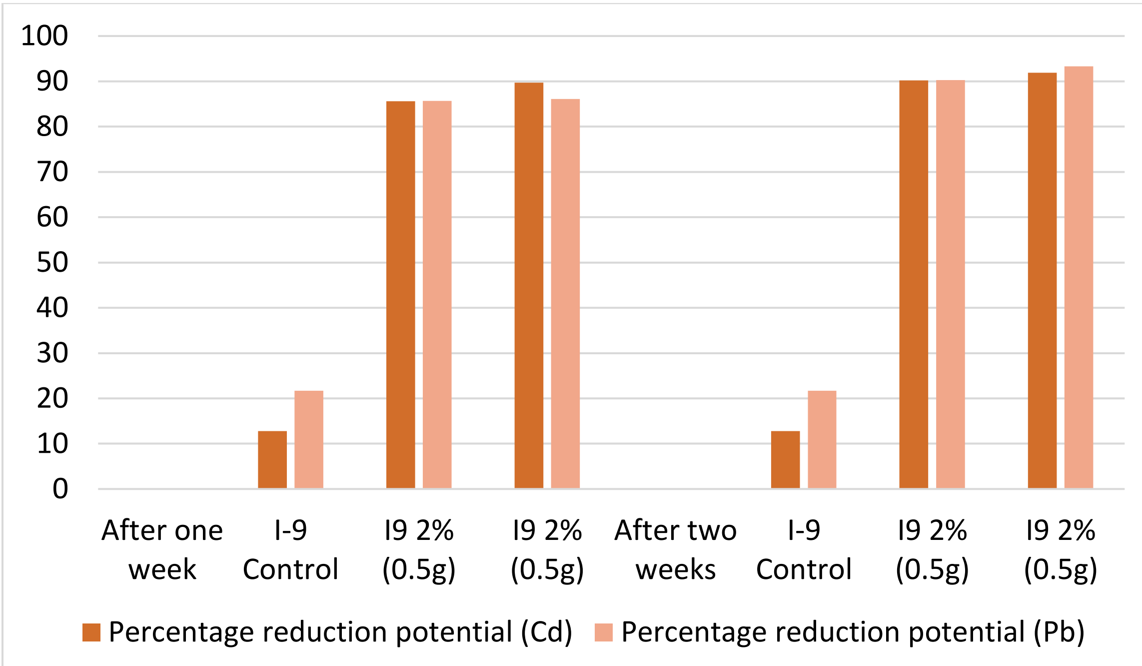


Figure 3.4: Reduction potential of nanoparticles for soil obtained from I-9 with 2% NPs

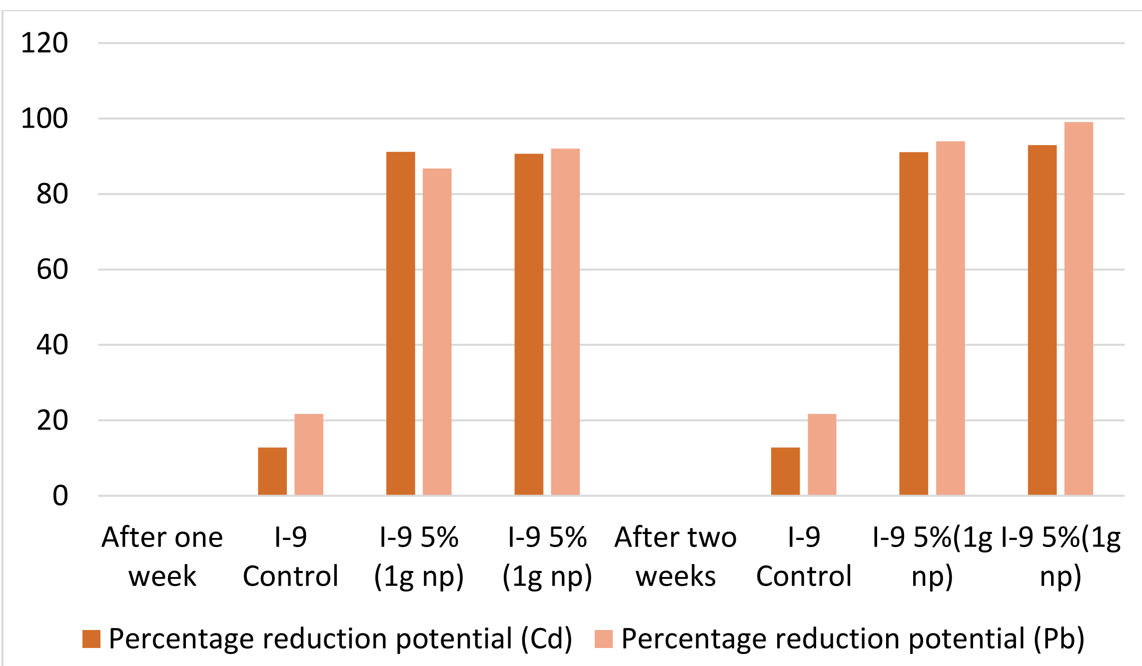


Figure 3.5: Reduction potential of nanoparticles for soil obtained from I-9 with 5% NPs

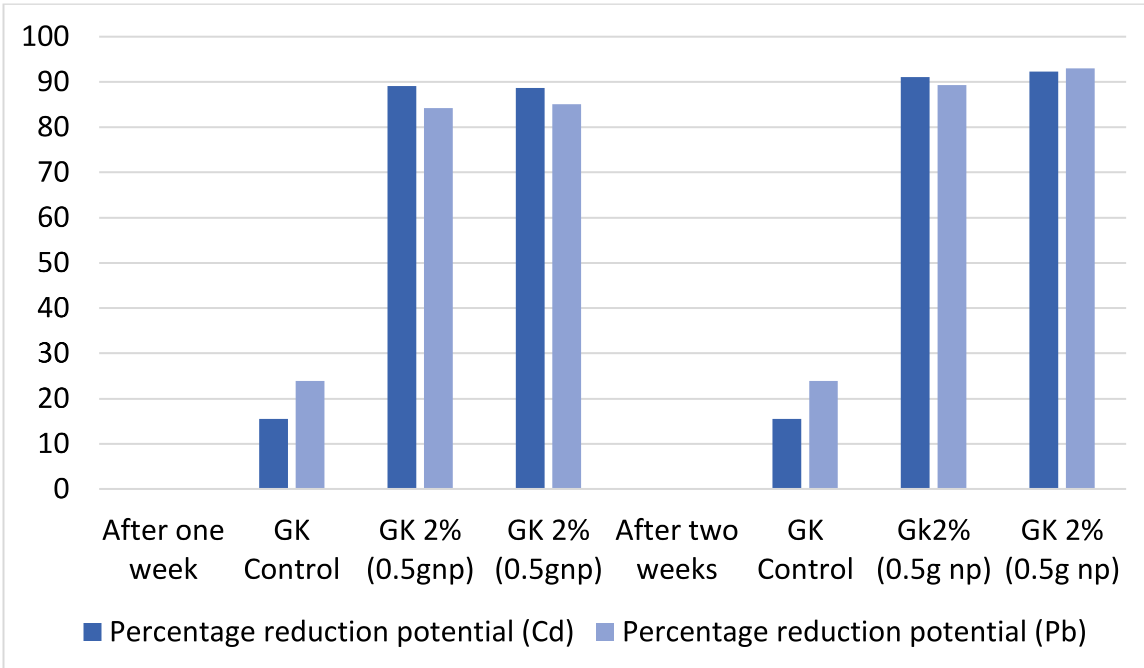


Figure 3.6: Reduction potential of nanoparticles for soil obtained from Gujar Khan with 2% NPs

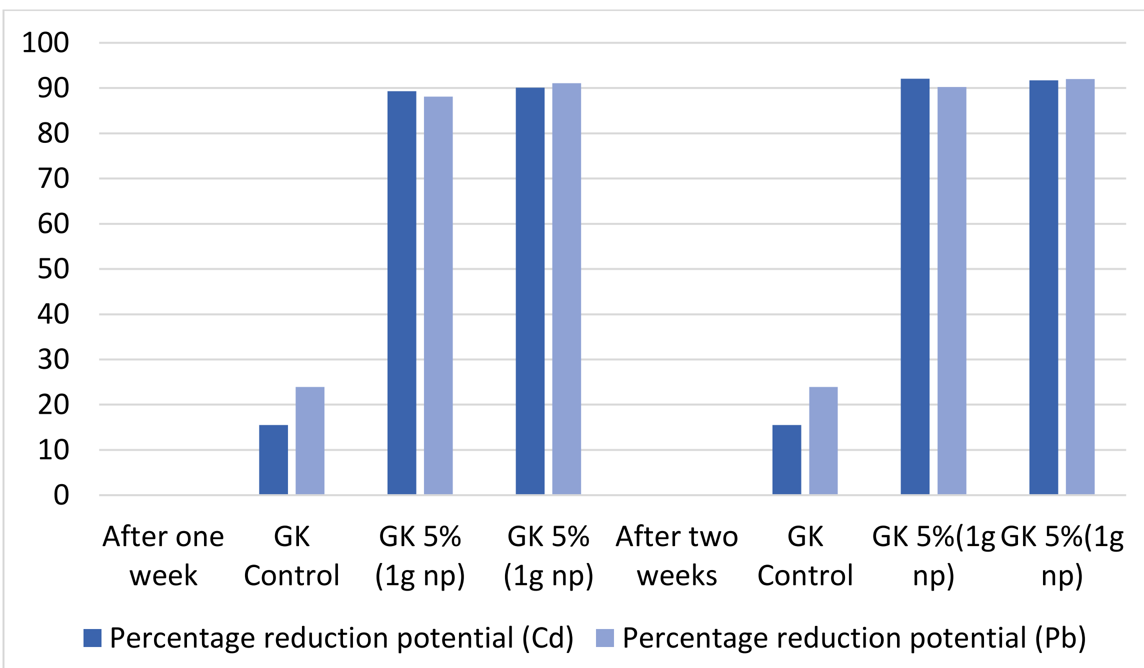


Figure 3.7: Reduction potential of nanoparticles for soil obtained from Gujar Khan with 5% NPs

3.6 Comparative analysis of removal efficiency for both the soils

A comparison was made regarding the removal efficiency of heavy metals in the two soils based on the findings presented in Table 3.2 from the study, which examined the levels of heavy metals Cd and Pb in soil samples from Sector I-9 and Gujar Khan following treatment with iron oxide nanoparticles at different doses. Sector I-9 soil had a better Cd removal efficiency than Gujar Khan soil in both the 2% and 5% iron oxide nanoparticle treatments. Sector I-9's reduced Cd contents following treatment suggest that the cadmium in that soil was removed more successfully. Sector I-9 soil had a greater Pb removal effectiveness than Gujar Khan soil for the 2% iron oxide nanoparticle treatment. Gujar Khan soil had a better Pb removal effectiveness with the 5% treatment, nevertheless. Sector I-9 soil showed higher efficacy in the removal of cadmium (Cd) in both 2% and 5% treatments. The lead (Pb) removal effectiveness of the two soils differed depending on the treatment concentration. The results indicated that Sector I-9 soil was more successful in removing cadmium than Gujar Khan. The success of lead removal varied between the two soils based on the treatment concentration.

DISCUSSION

The research synthesized iron oxide nanoparticles by employing Neem leaf extract and subsequently examined the properties of the synthesized nanoparticles using UV-visible spectrophotometer, SEM, and FTIR. The analysis found that the synthesized iron oxide nanoparticles exhibited UV-visible absorption peaks at 220 nm, indicating their photosensitivity. The synthesized nanoparticles were analyzed using FTIR to identify the various functional groups present. The goal of the study was to utilize synthesized nanoparticles for remediation of heavy metal-contaminated soil. Heavy metals, including cadmium (Cd), and lead (Pb), are adverse contaminants in soil. The production of iron oxide nanoparticles was effectively carried out, and their effectiveness in remedying heavy metal-contaminated soil was assessed. The synthesized nanoparticles demonstrated suitable characteristics for efficient metal adsorption through the use of thorough evaluation methods such as FTIR and UV analysis (Abdullah, et al., 2020). In addition, the effectiveness of iron oxide nanoparticles in remedying heavy metal-contaminated soil was evaluated using Atomic Absorption Spectroscopy (ASS), which showed substantial decreases in heavy metal levels after treatment. The results emphasize the capability of iron oxide nanoparticles to be a successful solution for reducing soil pollution caused by heavy metals. This contributes to the progress of environmentally friendly administration methods. The SEM results for iron oxide nanoparticles reveal spherical particles with a rough, irregular surface, observed at magnifications of X27,000, 5500, and 10,000, with an average size of approximately 100 nm.

The Fourier Transform Infrared (FTIR) study of iron oxide nanoparticles exhibited discernible absorption peaks that corresponded to certain functional groups and chemical bonds. The presence of several functional groups such as OH, carboxylic, aromatic rings, methyl, alkyl halide, and aldehyde in iron oxide nanoparticles was suggested by peaks observed at 3853.9 cm-1, 3344.68 cm-1, 2922.25 cm-1, 2850.88 cm-1, 2364.81 cm-1, 1627.97 cm-1, 1193.98 cm-1, and 825.56 cm-1. Upon calcination at a temperature of 600°C, specific peaks vanished, and Fe-O stretching bands appeared at a frequency of 405 cm-1. The FTIR spectra exhibited absorption peaks that were indicative of O-H, C=C, and C-O bonds at wavenumbers of 3117.37 cm^{-1}, 1459.24 cm^{-1}, 1577 cm^{-1}, and 1017.96 cm^{-1}, respectively. The magnitude of these peaks exhibited variations in accordance with the temperature at which calcination occurred, suggesting alterations in the degree of crystallinity. Furthermore, the identification of Fe_2O_3 patterns of vibration at a frequency of 400 cm^{-1} provided conclusive evidence for the occurrence of iron oxide nanoparticles (Karami, 2018).

Iron oxide (Fe_2O_3) nanoparticles were synthesized and examined for their potential use in removing cadmium (Cd) and lead (Pb) from contaminated soil. The nanoparticles were synthesized by using Neem leaf extracts using a standardized procedure, and were thoroughly examined using several spectroscopic and imaging techniques. An experiment was conducted to assess the ability of nanoparticles, lead, and cadmium nitrate to reduce the levels of Cd and Pb in soil samples taken from site 2 in Gujar Khan. The control samples exhibited a reduction potential of 15.5% for Cd and 23.9% for Pb after one week, which remained steady over the course of two weeks. The samples that were exposed to a dose of 2% nanoparticles exhibited an increase in reduction capacities over time, with Cd achieving 92.3% and Pb achieving 93%. On the other hand, the samples subjected to 5%

nanoparticles displayed varying but usually higher reduction abilities. This demonstrates the effectiveness of using nanoparticles for treatment purposes. FTIR spectroscopy revealed the existence of a ferric oxyhydroxide (FeOOH) coating on Fe_2O_3 nanoparticles, which was characterized by characteristic peaks at 3,421 cm^{-1} and 1,641 cm^{-1} corresponding to OH bending and stretched energies. The findings suggest that Fe_2O_3 nanoparticles have promising potential as catalysts for remedying soil affected with heavy metals, offering an alternative solution to the technological and financial difficulties associated with conventional cleanup methods. (Rajendran & Sengodan, 2017).

Location I-9 soil samples treated with nanoparticles for lead and cadmium showed varying potential for decrease after a week of treatment. Control samples reduced cadmium and lead 12.8% and 21.7% initially and remained same for the course of two weeks. However, at the 2% nanoparticles concentration, reductions increased over a period of two weeks. After a period of two weeks, Iron Oxide nanoparticles application at 5% reduced soil heavy metal values significantly (Venkatraman & Priya, 2022). The results from characterizing the FeO NPs were crucial in determining the schematic model associated with metal removal. Oxide presents in nanoparticle stages facilitate the adsorption of metals by providing active sites. The quantity of active sites on the surface is determined by the organic functional groups obtained from eucalyptus leaf extract, as shown in prior studies. The finding was confirmed via the FT-IR study used to characterize the nanoparticles produced through the green method. Prior studies demonstrated the correlation between the number of active sites on the adsorbent and the overall quantity of adsorbates (Kamath, et al., 2020).

CONCLUSION

Green synthesis of iron oxide nanoparticles is proposed as an environmentally friendly option for creating nanomaterials with several possible applications. Promising results have been obtained from the production of iron oxide nanoparticles for the purpose of remediating soil polluted with heavy metals. The synthesis of iron oxide nanoparticles was effectively carried out, and their effectiveness in cleaning heavy metal-contaminated soil was assessed. The existence of multiple functional groups such as OH, carboxylic, aromatic rings, methyl, alkyl halide, and aldehyde in iron oxide nanoparticles were detected by peaks seen at 3853.9 cm-1, 3344.68 cm-1, 2922.25 cm-1, 2850.88 cm-1, 2364.81 cm-1, 1627.97 cm-1, 1193.98 cm-1, and 825.56 cm-1 through the FTIR spectroscopy. UV–visible spectroscopy detected peak levels of absorption at a wavelength of 230 nm indicating their photosensitivity. The FTIR spectra exhibited absorption peaks that were indicative of O-H, C=C, and C-O bonds at wavenumbers of 3117.37 cm^{-1}, 1459.24 cm^{-1}, 1577 cm^{-1}, and 1017.96 cm^{-1}, respectively. The SEM results for iron oxide nanoparticles reveal spherical particles with a rough, irregular surface, observed at magnifications of X27,000, 5500, and 10,000, with an average size of approximately 100 nm. The goal was to determine the efficacy of iron oxide nanoparticles in cleaning soil that was sprayed with Pb and Cd. The results revealed a substantial potential for reducing heavy metal contamination. Ultimately, nanoparticles have demonstrated potential in remedying soil sprayed with lead and cadmium in site I-9 and Gujar Khan. Following a significant amount of nanoparticle exposure, the levels of lead and cadmium in the soil dropped. Nanoparticle treatment for one week on lead and cadmium in soil samples from location I-9 produced various the reduction capacities. The control samples showed reduction potential of 12.8% and 15.5% for lead and 21.7% and 23.9% for cadmium from both the soils. At 2% nanoparticle dosage, declines improved over 14 days. Iron Oxide nanoparticles at 5% were consistently effective, with noticeable increases after 14 days. The soil samples from Gujar Khan site 2 had 91.3% less Cd and 93.1% less Pb than the control samples after one week. This decreases lasted two weeks. Samples with 2% nanoparticles decreased Cd and Pb potentials by 92.3% and 93%, respectively. These studies demonstrate iron oxide nanoparticles' practicality for soil pollution cleanup.

RECOMMENDATIONS

Based on the scope and findings of our study, the following recommendations are made:

1. It is advisable to conduct more investigation and improve the production conditions for iron oxide nanoparticles in order to improve their effectiveness and durability in removing contaminants.
2. Furthermore, it is imperative to prioritize ongoing research endeavors that aim to examine the enduring consequences of nanoparticle utilization on soil well-being and the functioning of ecosystems.
3. The incorporation of cutting-edge technologies and strategies, such as nanotechnology and environmentally friendly production procedures, could provide new remedies for tackling the issue of heavy metal pollution in soil ecosystems.
4. Future research could also examine the possible cumulative benefits of mixing iron oxide nanoparticles with other cleaning strategies, such as phytoremediation or biochar usage, in alongside optimizing manufacturing settings and examining long-term impacts.
5. It is necessary to conduct field-scale research in order to evaluate the sustainability and feasibility of restoration methods using nanoparticles in actual polluted areas. Regular monitoring and evaluation of soil health indicators, such as diversity of bacteria and fertility levels, are crucial for assessing the long-term environmental effects of nanoparticle treatment.
6. The active participation of stakeholders and the local community is crucial for the effective execution of remediation methods that include nanoparticles. This emphasizes the significance of incorporating societal viewpoints and resolving any apprehensions regarding the health and safety and control of nanoparticles.

REFERENCES

Abdullah, J. A., S., Alonso-González, B., Guerrero, A., . . . A. (2020). Green synthesis and characterization of iron oxide nanoparticles by pheonix dactylifera leaf extract and evaluation of their antioxidant activity.

Abebe, B., Murthy, H.C.A., Amare, & E. (2018). Summary on Adsorption and Photocatalysis for Pollutant Remediation: Mini Review. *J.Encapsul. Adsorpt. Sci.* , 08, 225–255.

Abisharani, J., Devikala, S., Kumar, D., . . . P. (2019). Green synthesis of TiO2 Nanoparticles using Cucurbita pepo seeds extract. . *Mater. Today Proc*, 14, 302–307.

Adamu, I., H., Faruruwa, D., M., Adeyemi, M., M., . . . O., A. (2023). Plant Synthesized Iron Oxide Nanoparticles for Removal of Emerging Contaminant. . *Chemistry Africa,* , 1-14.

Afzal, & Singh, S. N. (2022). Effect of zinc and iron oxide nanoparticles on plant physiology, seed quality and microbial community structure in a rice-soil-microbial ecosystem. . *Environmental Pollution*, 314, 120224.

Ahmed F, I. H. (2006). Trace metal concentrations in street dusts of Dhaka city, Bangladesh. *Atmos Environ*, 40:3835–44.

Ahmed, M. M. (2012). *Atomic Absorption Spectroscopy (AAS).* Alexandria: Pharos University .

Ali, L., Ali, S., Khattak, SA., . . . HD. (2023). Distribution, Risk Assessment and Source Identification of Potentially Toxic Elements in Coal Mining Contaminated Soils of Makarwal, Pakistan. *Environmental and Human Health Outcomes. Land,* , 12(4), 821.

Ammann, A. (2007). Inductively coupled plasma mass spectrometry (ICP MS): A versatile tool. . *Journal of mass spectrometry : JMS. , 42*(10.1002/jms.1206.), 419-27. .

Andrade-Zavaleta, K., Chacon-Laiza, Y., Asmat-Campos, D., . . . N. (2022). Green synthesis of superparamagnetic iron oxide nanoparticles with eucalyptus globulus extract and their application in the removal of heavy metals from agricultural soils. *Molecules,* , 27(4), 1367.

Arshadi, M., Abdolmaleki, K., M., Mousavinia, F., . . . A. (2017). Nano modification of NZVI with an aquatic plant Azolla filiculoides to remove Pb(II) and Hg(II) from water: Aging time and mechanism study. . *Journal of Colloid and Interface Science* , 486, 296–308. https://doi.org/10.1016/j.jcis.2016.10.002.

Bhateria, & Singh, R. R. (2019). A review on nanotechnological application of magnetic iron oxides for heavy metal removal. *Journal of Water Process Engineering,*, 31, 100845.

Bulska, & Wagner, E. B. (2016). Quantitative aspects of inductively coupled plasma mass spectrometry. Philosophical Transactions of the Royal Society . *A: Mathematical, Physical and Engineering Sciences,* , 374(2079), 20150369.

Dave, & Chopda, P. L. (2014). Application of iron oxide nanomaterials for the removal of heavy metals. . *Journal of Nanotechnology*.

Du, C., H., C., W., G., W., S., L., P., & N., &. X. (2023). 2023 Green synthesis of nano-zero valence iron with green tea and it's implication in lead removal.. *Bulletin of Environmental Contamination and Toxicology*, 110 (1). https://doi.org/10.1007/s00128-022-03649-6.

Dubey, S., Sheoran, S., H., Kumar, V., . . . G. (2022). Potential of green nanoparticles for sensing and remediation of heavy metals from mining applications. . *In Green Nanomaterials for Industrial Applications*, (pp. 445-476). Elsevier.

Goutam, S.P., Saxena, G., Singh, V., . . . K.B. (2018). Green synthesis of TiO2 nanoparticles using leaf extract of Jatropha curcas L. for photocatalytic degradation of tannery wastewater. . *Chem. Eng. J.*, 336, 386–396.

Gulzar, & Mazumder, A. B. (2022). Helping plants to deal with heavy metal stress: the role of nanotechnology and plant growth promoting rhizobacteria in the process of phytoremediation. *Environmental Science and Pollution Research,*, 29(27), 40319.

Hall, J. (2002). Cellular mechanisms for heavy metal detoxification and tolerance. *J. Exp. Bot.* , 53, 1–11.

Iqbal, J., Abbasi, BA., Yaseen, & T. (2021). Green synthesis of zinc oxide nanoparticles using Elaeagnus angustifolia L. leaf extracts and their multiple in vitro biological applications. *Sci Rep*, 11, 20988 (2021). https://doi.org/10.1038/s41598-021-99839-z.

Jawed, A., Sharma, S., Golder, Ak., . . . M., L. (2021). Plant-polyphenol-mediated synthesis of iron oxide nanomaterials for heavy metal removal: a review. *New Trends in Removal of Heavy Metals from Industrial Wastewater,* , 115-129.

Jiang, Y., Zhou, P., Zhang, P., . . . Y. (2022). Green synthesis of metal-based nanoparticles for sustainable agriculture. . *Environmental Pollution,* , 119755.

Kamath, V., Chandra, P., Jeppu, &., & P., G. (2020). Comparative study of using five different leaf extracts in the green synthesis of iron oxide nanoparticles for removal of arsenic from water. *International journal of phytoremediation*, , 22(12), 1278-1294.

Karami, H. (2018). Synthesis and characterization of iron oxide nanoparticles by solid state chemical reaction method. . *Journal of Cluster Science,*, 21, 11-20.

Khan, S. R. (2022). Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES): a Powerful Analytical Technique for Elemental Analysis. . *Food Analytical Methods, , 15(3),*(https://doi.org/10.1007/s1216), 666–688. .

Kheskwani, & Ahammed, U. M. (2023). Removal of water pollutants using plant-based nanoscale zero-valent iron: A review. *Water Science & Technology,* , 88(5), 1207-1231.

Li, Z., Ma, Z., Kuijp, v. d., J., T., . . . L. (2014). A review of soil heavy metal pollution from mines in China: pollution and health risk assessment. . *Science of the total environment,*, 468, 843-853.

Lin, J., Sun, M., Su, B., . . . Z. (2019). Immobilization of cadmium in polluted soils by phytogenic iron oxide nanoparticles. . *Science of the Total Environment, .*, 659, 491-498.

Memon, & Schroder, A. P. (2009). Implications of metal accumulation mechanisms to phytoremediation. *Environ. Sci. Pollut. Res. Int.*, 16, 162–175.

Morrison, C., Sun, H., Yao, Y., . . . WE. (2020). Methods for the ICP-OES analysis of semiconductor materials. . *Chemistry of Materials,*, 32(5), 1760-1768.

Niroumand, H., Balachowski, L., Parviz, & R. (2023). Nano soil improvement technique using cement. . *Scientific Reports,* , 13(1), 10724.

Pattanayak, DS., Pal, D., Thakur, C., . . . L., G. (2021). Bio-synthesis of iron nanoparticles for environmental remediation: Status till date. . *Materials Today: Proceedings,* , 44, 3150-3155.

Prakash, P. (2023). Nano-phytoremediation of heavy metals from soil: a critical review. *Pollutants*, 3(3), 360-380.

Rahimi, S., Moattari, M., R., Rajabi, L., . . . M. (2015). Iron oxide/hydroxide (α, γ-FeOOH) nanoparticles as high potential adsorbents for lead removal from polluted aquatic media. *Journal of Industrial and Engineering Chemistry,* , 23, 33-43.

Rajendran, & Sengodan, S. K. (2017). (2017). Synthesis and characterization of zinc oxide and iron oxide nanoparticles using Sesbania grandiflora leaf extract as reducing agent. . *Journal of Nanoscience,*.

Rana, A., Kumari, N., Tyagi, M., . . . S. (2018). Leaf-extract mediated zero-valent iron for oxidation of arsenic (III): Preparation, characterization and kinetics. . *Chemical Engineering Journal* , 347, 91–100. https://doi.org/10.1016/j.cej.2018.04.075.

Rao, K.G., Ashok, C.H., Rao, K.V., . . . P. (2015). Green Synthesis of TiO2 Nanoparticles Using Aloe Vera Extract. . *Int. J. Adv. Res. Phys. Sci. (IJARPS)* , 2, 28–34.

Recillas, S., García, A., González, E., . . . X. (2011). Use of CeO2, TiO2 and Fe3O4 nanoparticles for the removal of lead from water: Toxicity of nanoparticles and derived compounds. . *Desalination,*, 277(1-3), 213-220.

Sarim, M., Jan, T., Khattak, SA., . . . E. (2022). Assessment of the Ecological and Health Risks of Potentially Toxic Metals in Agricultural Soils from the Drosh-Shishi Valley, Pakistan. . *Land,* , 11(10), 1663.

Sergio, Ferreira., L., A, M., Bezerra., Adilson, Santos., S., . . . Garcia. (2018). Atomic absorption spectrometry – A multi element technique,. *TrAC Trends in Analytical Chemistry*, Pages 1-6, ISSN 0165-9936, https://doi.org/10.1016/j.trac.2017.12.012.

Sharma, N., Singh, G., Sharma, M., . . . D., V. (2022). Sustainable Use of Nano-Assisted Remediation for Mitigation of Heavy Metals and Mine Spills. *Water,*, 14(23), 3972.

Sharma, N., Singh, G., Sharma, M., . . . VD. (2022). Sustainable Use of Nano-Assisted Remediation for Mitigation of Heavy Metals and Mine Spills. *Water,*, 14(23), 3972.

Sharma, Neetu., Gurpreet, Singh., Monika, Sharma., . . . Rajput. (2022). "Sustainable Use of Nano-Assisted Remediation for Mitigation of Heavy Metals and Mine Spills.". *Water* , 14, no. 23: 3972.

Tao, Z., Zhou, Q., Zheng, T., . . . S. (2023). Iron oxide nanoparticles in the soil environment: Adsorption, transformation, and environmental risk. *Journal of Hazardous Materials*, 132107.

Venkatraman, & Priya, Y. A. (2022). Removal of heavy metal ion concentrations from the wastewater using tobacco leaves coated with iron oxide nanoparticles. . *International Journal of Environmental Science and Technology,* , 19(4), 2721-2736.

Vu, & Mulligan, K. C. (2023). Remediation of organic contaminated soil by Fe-based nanoparticles and surfactants: a review. *Environmental Technology Reviews,* , 12(1), 60-82.

Weng, X., Jin, X., Lin, J., . . . Z. (2016). Removal of mixed contaminants Cr(VI) and Cu(II) by green synthesized iron-based nanoparticles. *Ecological Engineering*, 97, 32–39. https://doi.org/10.1016/j.ecoleng.2016.08.003.

Wilschefski, & Baxter, S. M. (2019). Inductively Coupled Plasma Mass Spectrometry: Introduction to Analytical Aspects. *Clin Biochem Rev.*(doi: 10.33176/AACB-19-00024.).

Wilschefski, & Matthew, R. B. (2019). "Inductively coupled plasma mass spectrometry: introduction to analytical aspects.". *The Clinical Biochemist Reviews* (doi: 10.33176/AACB-19-00024.), 40, no. 3: 115.

Yadav, K., V., Ali, D.., Khan., H., S., . . . S. (2020). Synthesis and characterization of amorphous iron oxide nanoparticles by the sonochemical method and their application for the remediation of heavy metals. *Nanomaterials,* , 10(8), 1551.

Yadav, S., Yadav, A., Bagotia, N., . . . S. (2021). Adsorptive potential of modified plant-based adsorbents for sequestration of dyes and heavy metals from wastewater-A review. *Journal of Water Process Engineering*, 42, 102148.

Yan, W., Ramos, V., M. A., Koel, E., B., . . . X., W. (2012). As(III) sequestration by iron nanoparticles: Study of solid-phase redox transformations with X-ray photoelectron spectroscopy. . *Journal of Physical Chemistry C* , 116 (9), 5303–5311. https://doi.org/10.10.

ROLE OF PLANT-BASED IRON NANOPARTICLES FOR THE ADSORPTION OF HEAVY METALS

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