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BIOREMEDIATION OF CONTAMINATED SOIL

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ABSTRACT: The aim of this project is to investigating the treatment of contaminated soil treatment by using pseudomonas fluorescens, the application of bio treatment is growing rapidly. The bioremediation process is cost-efficient, safe and nature-based technology which leads to degradation of contaminants. Pseudomonas fluorescens is used for the treatment of sewage sludge. During the recent era of environmental protection, the use of microorganism for the degradation of contaminants from soil. The food and water we consume are often contaminated with a range of chemicals and heavy metals, such as arsenic, cadmium, copper, chromium, gold, lead, nickel, mercury and zinc that are associated with numerous diseases. Many study have established that microbes have the capacity to remove heavy metals from contaminated soils. Among others some of the microbes that play great responsibility in bioremediation of heavy metals involve using pseudomonas fluorescens. This encourageis indication as to the helpfulness of microorganisms and their component for the remediation of the contaminated soil is review in this article.

Keywords: Contaminated soil, heavy metals, bioremediation, pseudomonas fluorescens.

1. INTRODUCTION

The environmental contamination by heavy metals through the industrial waste is one of the main health problems in industrial countries. Metal contaminants can enter to food chain if contaminated water, soils and plants are used for food production. The heavy metals are refer to any metallic component that has beencomparatively high density and toxic or poisonouseven concentration.

As is understandable from the word itself 'bioremediation' (bio + remediation) should involve two components the bio i.e. the live element and remediation i.e. the treatment of the pollutant. The terms denote the existence of some pollutant in the matrix which is to be remediated. Therefore, before going into the typical definitions of bioremediation let us discuss what act we need to remediate and why. The react we will soon find once we start to analyze the present state of the environment. Pseudomonas fluorescens encompass a group of common, nonpathogenic saprophytes that colonize soil, water and plant surface environment. It is a common gram harmful, rod-shaped bacterium. It is motile by means of various polar flagella. Pseudomonas fluorescens has simple nutritional requirements and grow well in mineral salts media supplemented with any of a huge number of carbon sources. Because they are well modified in soil, Pseudomonas fluorescens strain have being investigate extensively for use in applications that require the discharge and survival of microorganisms in the soil. Chief among these are bio control of pathogens in agriculture and bioremediation of various organic compounds.We have selected the sewage sludge sample for treating andremove the contaminants from the sewage sludge and it can be used for agriculture purposes in the environment. The sewage sludge is collected from the sewage treatment plant in which it consists of contaminants like copper, zinc, lead, arsenic, chromium, cadmium, nitrogen, and other harmful compounds. The sewages are collected around the surrounding areas and then the soil and wastewater are separated on sewage treatment plant.This paper provide summary information on a wide variety of

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technologies for the treatment of polluted soil. The in situ technology is presented in involvingthe applying biological process for subsurface to degrade, reduce, remove, or immobilize. The treatment technology presented include common practices as well as innovative alternatives for treating polluted soil. The pseudomonas fluorescens is used for mimic properties in the soil.

2.Bioremediation:

Microorganism and plant are regularly used for the elimination of heavy metals. Process of involvement of microorganisms to reduce contaminant concentration is known as bioremediation which is a natural development and its importance of biodiversity (above or below the ground) is increasingly considered for clean-up of metal contaminated and polluted ecosystem. All the metals are toxic,but some of these are helpful in low concentration. These metal toxicity cause serious morbidity and mortality.

3.Definition for contaminated soil:

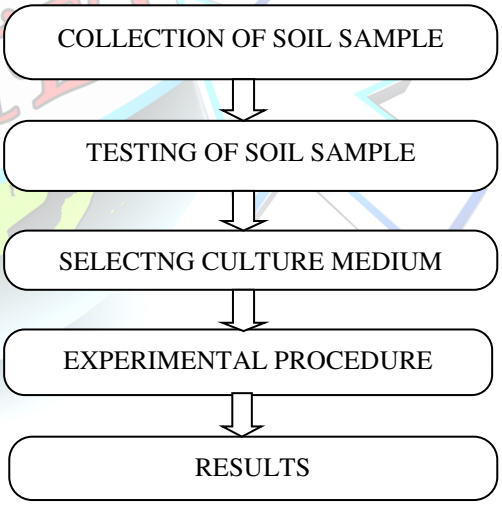
Soil contamination

Soil contamination or soil pollution as part of land degradation is cause by the presence of xenon biotic (human–made) chemicals or other alteration in the normal soil environment. It is typically caused by industrial activity,agricultural chemical or improper disposal of waste. Soil contamination refers to the presence of un-natural (human-made) chemicals or other substances in the soil. The pollutants presented in the soil are that can be in solid or liquid state. This is also true if they enter the ground, as they can then be transported as considerable distance away from the original site of the contamination. Definition of Pseudomonas fluorescensPseudomonas fluorescens encompass a group of ordinary, nonpathogenic saprophytes that take over soil, water and plant surface environment. It is a common gram negative, rod-shaped bacterium. It secretes a soluble greenish fluorescent called fluorescens, particularly under condition and low iron availability.

4.Objectives

- To remove the pollution from the contaminated soil to regain normal condition
- Treating the contaminated soil the pollution is reduced in the soil and heavy metals are also reduced.
- To reduce the health problem and diseases causes in the environment.
- The contaminants removed area can be used for agriculture
- Increases the land availability

5.Methodology



6.Contaminants presented on the soil

Heavy Metals:

These heavy metals are natural constituentof the environment, but changemake use for thehuman

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intention has altered their geochemical procedure and biochemical stability. This results in excess discharge of heavy metals such as cadmium, copper, lead, zinc etc. into natural resources like the earth and aquatic environment. Prolonged exposure and higher accumulation of such heavy metals can have deleterious health personal property on human life and aquatic biota. The role of microorganisms and plants in biotransformation of heavy metals into nontoxic forms is well-documented, and understanding the molecular mechanism of metal accumulation has numerous biotechnological implications for bioremediation of metal contaminated soil.

7.Bioremediation process:

7.1. Bioremediation: Introducing Microbe Based Clean Up System

Remediation of the surroundingsplace such as soil, sediments and water adjust with heavy metals can be achieved throughoutorganicallydeterminedalteration in the oxidation condition. Bioremediation is the microbe-mediated process for clearance or immobilization of the contaminants, including all achievable toxins like hydrocarbons, agrochemicals and other organic toxicants. Non-living toxic compound such as heavy metals, microbes are unable to simplify them into undamaging compounds, and they should be used according to their specialization for the large quantity of contaminants. Thus, the bioremediation approach for heavy metals depends on the active metabolizing capabilities of microorganisms. Several bacteria are known to require varying amounts of heavy metals as important micronutrients for development and development.

7.2. Factors of Bioremediation:

The control and optimization of bioremediation processes is a complex system of severalfactor. These factors include: the subsistence of a microbial population capable of degrading the contaminant; the availability of contaminants to the microbial population; the surroundingsfactor (type of soil, temperature, pH, the presence of oxygen or other electron acceptors, and nutrients).Biodegradation - the use of live organisms such as microorganisms, fungi, and plants to degrade chemical compounds. Bioremediation – procedure of cleaning up environmental sites contaminated with chemical pollutants by using living organisms to degrade hazardous materials into less toxic substance. Most approaches change harmful pollutants into relatively undamaging materials such as carbon dioxide, chloride, water, and simple organic molecules. Processes are in general cleaner.

7.3. Isolation of Bacteria:

Kings'B medium was used for the isolation of hydrocarbon degrading microorganisms. Kings'B medium was prepared by adding pepton 20 gram, glycerol 10 ml, dipottasium hydrogen phosphate 1.5 gram, magnesium sulphate 1.5 gram and finally 15g/L of agar were added; 1000ml of water was added to the flask contain Bushnell-Haas medium. Since the media had a total volume of more than 1000ml hence 700ml of this mixture was transferred to a separate 1 liter flask to avoid spill over during autoclaving. The kings'B medium was then autoclaved at 121˚C for 15 minutes. The sub samples of 1g soil were accurately weighed before being transferred aseptically to a bottle containing 99ml of sterile dilute saline solution to attain a dilution of 10-2.The mixture was then taken aback vigorously and was allowable for the soil to settle at the bottom of the bottle and 0.1ml of the water was transferred to a test tube containing 9ml of sterile dilute saline explanation to attain a dilution of 10-3. A series dilution method subsequently yielded 4 additional test tubes with dilutions of 10-4, 10-5, 10-6. Using a pipette, 1ml of the solution from each oneof the test tubes was inoculated in Kings'B medium containing Petri dishes by means of platting and spreading technique. This process are usedselected for organisms that exhibit hydrocarbon

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degrading capability. The Petridishes were incubated at 37˚C for 24-48 hours.

7.4. Mechanisms of Bioremediation:

Microorganisms are omnipresent that dominate in heavy metal-contaminated soil and can simply convert heavy metals into non-toxic forms. In bioremediation process, bacteria mineralize the organic contaminants to end-products such as carbon dioxide and water, or to metabolic intermediates which are use as the major substrates for cell growth process. Microorganisms are capable of two-way defense viz. Production of degradation enzymes for the target contamination as well as resistance to relevant heavy metals.

8. Sewage Sludge:

Due to the nature of the physicochemical process involved in waste water treatment, sludge tends to concentrate heavy metals and other harmful constituents in attendance in waste water. The heavy metals content of sludge is restricted by governmental regulations such as the Sludge Directive 86/278/EEC of the European Union and EPA Part 503 of the United States. Concentrations of up to 10 heavy metals (As, Cr, Cd, Cu, Pb, Hg, Mo, Ni, Se and Zn) are typically monitored in sewage sludge. When maximum permissible concentrations

S. No	Lead	chromium	cadmium	arsenic
Before testing	2.61±1.47	7.67±2.59	0.38±0.05	1.32±0.23
After testing	5.50±1.79	11.73±6.93	0.75±0.13	5.05±1.18

are not met, sludge is mainly disposed of by incineration.

8.1.Reaction of Pseudomonas Fluorescence with Soil:Pseudomonas fluorescens 1.50% liquid formation is an eco- friendly bio fungicide containing pseudomonas fluorescens and useful against contaminants present in the soil.

Properties of soil samples used in this study

Soil Property	Lead	chromium	cadmium	Arsenic
% Gravel	NA	<5	35	20
% Sand	20	36	54	18
% Silt	34	19	11	32
% Clay	46	40	10	25
% Carbon	21	18	18	23
% Moisture (before drying)	0.84	4.2	7.2	4.5
% Moisture (after drying)	4.21	2.51	1.73	1.43
% Iron	4.9	8.5	5.2	4.4
pH	6.44	6.57	6.3	6.33

8.2.Results:

Reaction of pseudomonas fluorescens with the soil

Concentration of the heavy metal

Chart 1. Contaminants reduced %

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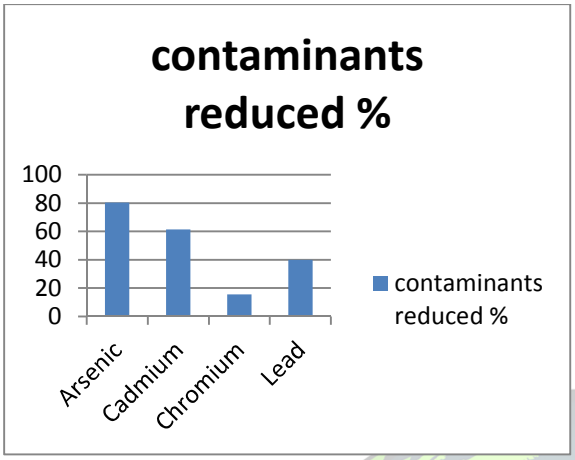

9. Conclusion:

Bioremediation provides a technique for cleaning up pollution by attractive the natural biodegradation processes. So by developing an understanding of microbial communities and their response to the natural environment and pollutants, expanding the knowledge of the genetics of the microorganisms to increase capabilities to degrade the pollutants, conducting field trials of new bioremediation techniques which are cost effective, and dedicating sites which are set aside for long term research purpose, these opportunities offer potential for important advances. The results from the pseudo-total analysis indicated that the majority of the heavy metals had exceeded MPL while the rest approached the limit. These heavy metals contaminationvarious spatially, across the entire area, and vertically down the profile with some areas and/or depths having greater concentrations than the others. Most of these heavy metals leach down to ground water particularly Zn and As. More of these heavy metals occur in their available forms, and accumulate in plants, especially the amaranths. Accumulations of these metals pose risks of ground and surface water contamination, andhuman health of those community consuming vegetables growing on this land.Pseudomonas fluorescens is one such proven biological control agent.

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