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Review on phytoremediation: Environment friendly technique to clean environment

Brilisha Joddha[1*], Dhruv Pandya[2], Himanshu Pandya[1], Archana Mankad[1]
[1] Department of Botany, Bioinformatics and Climate Change and Impacts Management, School of Science, Gujarat University, Ahmedabad, Gujarat, India
[2] Department of Botany, President Science College, Ghatlodiya, Ahmedabad, Gujarat, India

Abstract
This review study is based on the overall Phytoremediation technique which is considered as an ecofriendly, cost –effective method to clean the environment. This is the technique in which we can reduce amount of pollutants from soil, water and air with the help of certain plants. Nowadays, environmental pollution by heavy metals like Pb, Cd, As, Hg, Cr, Cu, Zn, and Sn has become a serious issue in the world. To degrade these heavy metals or to lower their concentrations beyond their threshold limits, phytoremediation is found to be very promising solution. Phytoremediation implies various techniques to degrade, metabolize or immobilize the heavy metals. The technique includes Rhizofiltration, Phytostabilization, Phytoextraction, Phytovolatilization and Phytodegradation. This review was focused on different phytoremediation experiments studied by different scientists. Review specially focused on heavy metals and their remediation techniques.

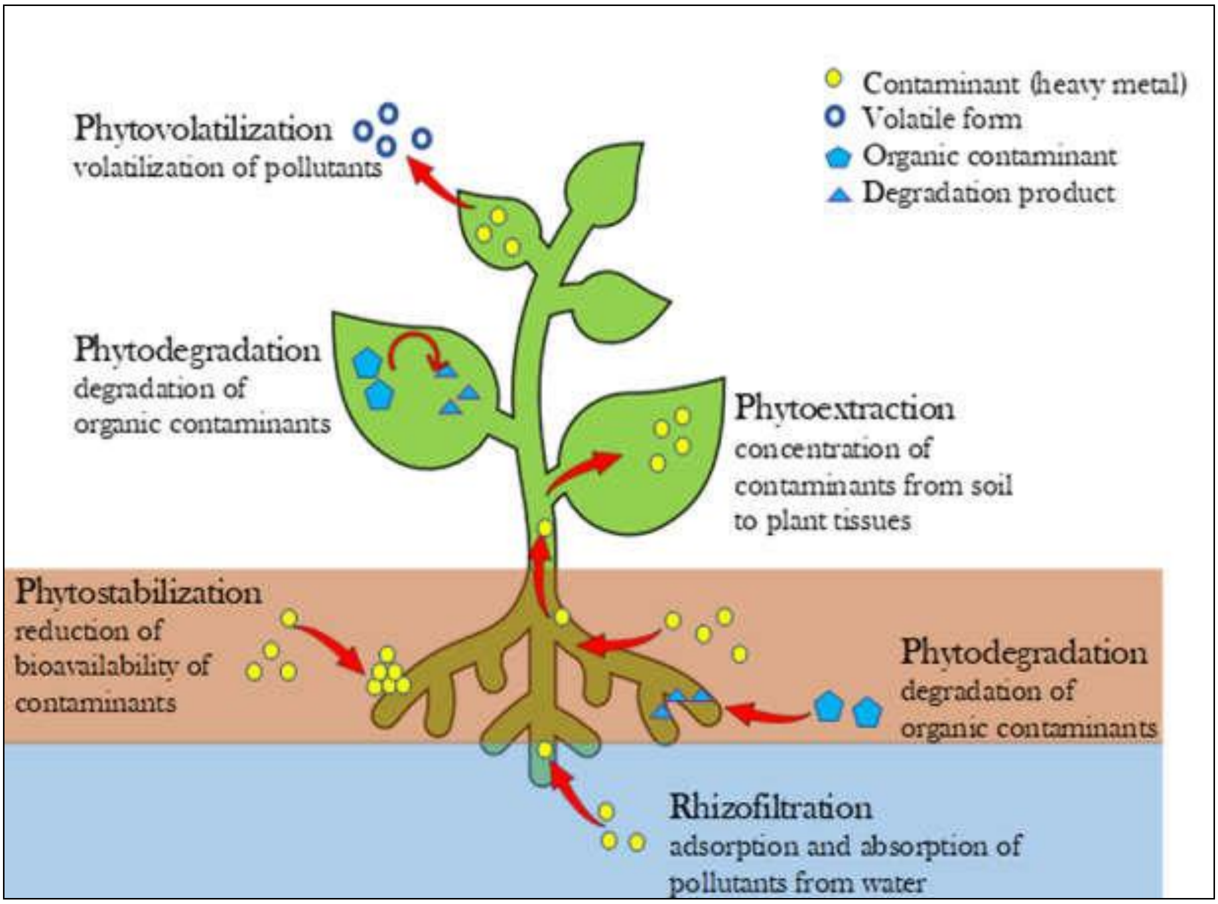
Keywords: phytoremediation, heavy metals, hyperaccumulators

Introduction

"Phytoremediation", is the use of plants to remediate contamination of soil with organic or inorganic wastes. Plants will then directly or indirectly absorb, sequester, or degrade the contaminant. To maximize this remedial effect plants and fertilization, irrigation, and cropping schemes will be managed. By growing plants over a years, their aim is to either remove the pollutant metal from the contaminated matrix or to alter the nature of the contaminant present within the soil so that it no longer causes a risk to human health and the environment. (Cunningham and Ow, 1996) [1] (1) Environmental pollution by heavy metal has become a serious problem in today's world. The accumulation of heavy metals in the soils and waters poses a risk to the environmental and human health. Heavy metals enter the environment or soil from natural and anthropogenic sources which cause adverse effects on human health. Regarding their toxicities, the most dangerous heavy metals are Hg, Cd, Pb, As, Cu, Zn, Sn and Cr. Out of which Hg, Cd, Pb and as are non –essential while Cu and Zn are essential heavy metals. Heavy metal concentrations beyond threshold limits causes adverse health effects because they interfere with the normal functioning of living systems. (Ali *et al*, 2013) (2).
Phytoremediation basically refers to the use of plants and associated soil microorganisms to reduce the concentrations or toxic effects of contaminants or pollutants from the environments. It can be used for removal of heavy metals and radionuclides as well as for organic pollutants like polynuclear aromatic hydrocarbons, polychlorinated biphenyls, and pesticides. It is a novel, cost- effective, efficient, and eco-friendly. The term Phytoremediation is a combination of two words: Greek *phyto* (means plant) and Latin *remedium* (meaning to correct or remove an evil). The establishment of vegetation on contaminated soils also helps to prevent erosion and leaching of metal. From an economic point of view, the purpose of Phytoremediation of polluted land can be three-sided: (1) risk containment (phytostabilization); (2) phytoextraction of metals having market value such as Ni, Tl, and Au; (3) durable land management where phytoextraction gradually improves quality of soil for subsequent cultivation of crops with higher market value (Ali *et al*,2013) (2).
Phytoremediation is considered to be a 'Green Revolution' in this field which is considered to be an innovative cleanup technologies. It is the use of plants to clean –up contaminated hazardous wastes sites. The idea of using metal-accumulating plants to remove heavy metals and other pollutant compounds was first introduced in 1983,but the concept was actually been implemented for the past 300 years on wastewater discharges (Henry, 2000) (6). Phytoremediation can be defined as the use of plants to remove, metabolize, degrade and immobilize transfer or detoxifying pollutants such as metals, hydrocarbons, dyes, and toxic substances from the soil, water, or air through their natural metabolic pathways and functions (Dhanwal *et al*, 2017) [7].

Different Processes/ Techniques Involved in Phytoremediation

Phytoremediation consists of five main processes, they are Rhizofiltration, Phytostabilization, Phytoextraction, Phytovolatilization, and Phytodegradation/Phytotransformation.



(**Source:** Bioremediation Methods for the Recovery of Lead-Contaminated Soils: A Review - Scientific Figure on Research Gate. Available from: https://www.researchgate.net/figure/Schematic-representation-of-different-phytoremediation-approaches-by-plants-under-study_fig1_ 341531208 [accessed 18 Dec, 2021]

Fig 1: Processes Involved in Phytoremediation

Rhizofiltration

It can be defined as the use of plants, both terrestrial and aquatic ; to absorb, concentrate, and precipitate contaminants from polluted aqueous sources with low concentration of contaminants in their roots(Ghosh and Singh, 2005) (3). Phytofiltration encompasses the use of plants roots and other plant parts like seedlings and expurgated shoots to adsorb or absorb heavy metals ions and organic pollutants from polluted surfaces of waters, and wastewaters for cleaning numerous aquatic environments. It is the most common utilized technique for contaminant elimination (Ahmed *et al*, 2018) (8). Rhizofiltration can be used for Pb, Cd, Cu, Ni, Zn, and Cr, which are commonly retained within the roots. The advantages associated with this technique are the ability to use both terrestrial and aquatic plants for either in situ or ex situ applications. Another advantage is that contaminants or pollutants do not have to be translocated to the shoots. So, species other than hyperaccumulaters may be used (Jadia and Fulekar, 2009) (4).

Phytostabilization

It is mostly used for the remediation of soil, sediment and sludges and it depends on ability of root to limit contaminant mobility and bioavailability in the soil. It can occur through the absorption, precipitation, complex action, or metal valence reduction. A dense root system stabilizes and prevents erosion of soil. It is very effective when rapid immobilization is needed to conserve ground and surface water and disposal of biomass is not required in it. However the major disadvantage is that, the pollutant remains in soil as it is, and thus requires regular monitoring (Ghosh and Singh, 2005) (3). This Phytoremediation technique utilizes the use of plants to lower the mobility of contaminants by immobilizing or precipitating them from the polluted or contaminated sites, therefore by lessening or avert their availability and relocation to ground water or ultimate entrance into the bionetwork (Ahmed *et al*, 2018) (8).

Phytoextraction

Phytoextraction, also known as Phytoabsorption, phytosequestration, or phytoaccumulation, is one of the best techniques of Phytoremediation which involves majorly the use of contaminant-accumulating plants to confiscate perilous compounds or elements, particularly heavy metals that may be harmful to microorganisms even at very low concentrations, from polluted environments(Ahmed *et al*,2018) (8). It is the best method to remove the contamination primarily from the soil and isolating it, without destroying the soil structure and fertility. It is best suited for the phytoremediation of diffusely polluted areas, where pollutants occur only at relatively very low concentrations and superficially. There are two basic techniques involved in phytoextraction, which have finally developed are 1) Chelate assisted phytoextraction or induced phytoextraction, in which artificial chelates are added to increase the mobility and uptake of metal contaminant. 2) Continuos phytoextraction in this the removal of metal depends on the natural ability of the plant to remediate; only the number of plant growth repetitions are controlled (Ghosh and Singh, 2005) (3).

Pytovolatilization
The technique involves the use of plants to take up contaminants from the soil, transforming them into the volatile form and transpiring them into the atmosphere. It occurs as growing trees and other plants take up water and the organic and inorganic pollutants. Some of these pollutants can pass through the plants to the leaves and volatilize into the atmosphere at relatively low concentrations (Gosh and Singh, 2005) (3). Phytovolatilization is among the techniques of Phytoremediation which involves majorly the use of plants to remove volatile organic carbons (VOC) and some few inorganic carbons that could exist in volatile forms such as arsenic, mercury and selenium ions(Ahmed *et al*, 2018) (8).

Phytodegradation/ Phytotransformation
In phytoremediation of organic compounds, plant metabolisms contributes to the contaminant reduction by transformation, break-down, stabilization or volatilizing pollutant compounds from the soil and groundwater. Phytodegradation is the breakdown of organic compounds taken up by the plant to simpler molecules that are incorporated into the plant tissues (Ghosh and Singh, 2005) (3). Generally, plants contain enzymes, such as reductases,dehalogenases, and oxygenases which are responsible in breaking down and converting contaminants including chlorinated solvents like trichloroethylene and some herbicides(Ahmed *et al*,2018) (8).

Plant Response to Heavy Metals
Plants have three basic processes for growth on metal contaminated soil, they are;

Metal Excluders
They prevent metal from entering the aerial parts of plants or maintain low and constant metal concentration over a wide range of metal concentration in soil, they mainly restrict metal in their roots. The plant may alter its cell membrane permeability, change metal binding capacity of cell walls or exude more chelating substances (Ghosh and Singh, 2005)(3).

Metal Indicator
Plant species which actively accumulate metal in their aerial tissues and generally shows up metal level in the soil. Plant indicator species tolerate the existing concentration level of metals by producing intracellular metal binding compounds chelators, or alter metal compartmentalization pattern by storing metals in non-sensitive parts(Ghosh and Singh, 2005) (3).

Metal Accumulator Plant Species
Plant species can concentrate or accumulate metal in their aerial parts, to levels far exceeding than soil. Hyperaccumulaters are plant species which can absorb high levels of contaminants concentrated either in roots, shoots or leaves (Ghosh and Singh, 2005) (3).

Advantages of Phytoremediation
It is easily applicable to a variety of organic and inorganic compounds, *in situ/ ex situ* application, *In situ* application of the technique decrease the amount of soil disturbance compared to conventional methods, It also reduces the concentration of waste to be landfilled upto 95%, It does not require expensive instruments or highly specialized personnel, and it is environmental- friendly and aesthetically pleasing to the people (Henry, 2000) [6]. It has low investment cost and minimum instrument requirement, prevents erosion of soils, especially thinner inorganic soils, It reduces leaching of particulate substance and spreading of toxicants. Contaminants can be recovered from the plant tissues and offer a good opportunity for commercialization. It can be used for phytoremediation of soils that are non-productive for agricultural land (Dhanwal *et al*, 2017) [7].

Disadvantages of Phytoremediation
It shows incomplete removal of pollutants with long-term low performance, it has limited applicability to different types of wastes, especially with high level toxicity wastes. It is mainly applicable for the upper layer of the soil and mine tailings. It’s effectiveness is affected by seasonal factors. It poses risk of bioaccumulation in the food chain. Proper disposal of plant matter or substance is required with proper risk assessment and which has possibility of introduction and spreading of undesirable invasive species of plants (Dhanwal *et al*, 2017) [7]. It is restricted to sites with shallow or low contamination within rooting zone of remediative species. It may take upto several years to remediate a polluted site. Harvested plant biomass by phytoextraction method may be classified as a RCRA hazardous waste (Henry, 2000) [6]. Accumulation of pollutant metal or contaminant in fruit and other edible parts of crop and vegetables (Farraji *et al*, 2016) [5].

Some of the important reviewed research articles:

Sekar *et al*,in 2013 investigated the physiological capacity of *Golden doranda, Balsam,*and *Erwa* plants in tolerating heavy metals like chromium by pot culturing and the analysis of heavy metals in the whole plant was done by using inductively coupled plasma optical emission spectroscopy. Pandya *et al*, 2021 assessed the heavy metal extraction capacity of *Impatiens balsamia* by two approaches *in-vitro* and *in-vivo* with two metals lead and cadmium using different concentrations was assessed by Atomic Absorption Spectroscopy. They concluded that as compare to *in-vivo* produced plants *in-vitro* produced plants has more capacity to accumulate lead and cadmium. Patil and Jadhav in 2013 studied the *Tagetes patula* for remediating textile reactive blue dye 160 by *in-vitro*. There was a considerable (90%) decolorization of dye within 4 days of incubation as confirmed by UV-vis, HPLC and FTIR analysis.

Shah *et al,* in 2017 assessed the cadmium accumulation and it's effects on growth and biochemical parameters in *Tagetes erecta* L which was raised in pots containing different concentration of cadmium. Results of quantitative estimation of cadmium in plant parts showed that roots accumulated highest amount of Cd followed by stems and leaves whereas inflorescence contained least amount of cadmium. Satashiya in 2017 conducted a survey work on balsam, Gomphrena, Marigold, Gaillardia in pot culture for remediating Cd, Ni and Pb contaminated soil. He concluded that for phytoremediation of cadmium, lead and nickel marigold and Gomphrena can be used as its biomass yield, heavy metal uptake as well as bio-concentration factor and mobility of heavy metals in the plants was higher.

Shah *et al*, in 2017 assessed the lead accumulation and it's effect on growth and biochemical parameters in *Tagetes erecta* L which was raised in pots containing different concentration of lead. Biochemical analysis showed that amount of protein continuously decreased whereas amount of free amino acid continuously increased with increasing concentration of lead. Ekta and Modi, in 2020 assessed the bioaccumulation study of *Zinnia elegans*. L with different concentration of lead from contaminated soils by AOAC method 1990 to find out plant species suitable for uptaking heavy metal (Pb). Pandya and Mankad, in 2019 assessed the effect of metal toxicity on plant growth and metabolism of *Datura stramoinum* L. After 105 days leaf weight was higher in lead treated plants. Shoot weight and root weight was higher in cadmium treated plants.

Bilek *et al*, in 2020 conducted to assess the stress tolerance in *Populus balsamifera* and *Salix eriocephela* and one hybrid willow to salinity and hydraulic fracturing wastewater. He concluded that *P.balsimifera* was relatively found salt intolerant compared to two other species. Salido *et al* in 2003 assessed the performance of arsenic and lead using Chinese brake fern (*Pteris*) and *Brassica juncea*. Results indicate that increasing pH increase the arsenic removal by *Pteris* and *Brassica* can extract upto 32 mg of lead from the field. Warthakar and Jadhav in 2014 growned *in-vitro consortium Petunia grandiflora* and *Gaillardia grandiflora* to degrade a dye mixture. Consortium was found to be more usefull than individual plants. BOD and COD of the dye mixture was reduced upto 69% and 73% respectively.

Ehsan et *al*, in 2016 assessed the phytoremediation potential of vinca by pot culturing in chromium contaminated soil by using different concentrations. The results indicated that plants height, fresh and dry weight increased at low concentration, but decreased at high contamination levels of chromium. Gaur *et al*, in 2017 conducted to assess phytoremediation potential of coriander for lead and arsenic by pot experiment and filter paper. They concluded that coriander seeds grown in pot was more effective in removing lead and arsenic than filter paper.

Ghazaryan et *al*, in 2017 determine the contents of copper in agricultural soils containing Coriander, Garlic. Results showed that in coriander (in aerial parts) and in garlic (aerial and underground parts) as well as in the leaves of horseradish the exceeding maximum permissible concentration of copper was observed. Swain *et al*,in 2014 demonstrated the phytoremediation potential of water hyacinth for the removal of copper and cadmium. The experiment showed that at all the levels the plants accumulated the highest concentration of cadmium in roots while the highest concentration of copper was accumulated in stems.

Dheri *et al*, in 2007 carried out glasshouse investigation to evaluate natural potential of fenugreek, spinanch and raya for cleanup of chromium contaminated silty loam and sandy soils. The findings indicated that family cruciferae (raya) was most tolerant to Cr toxicity followed by chenopodiaceae (spinach) and leguminosae (Fenugreek). Kaur in 2016 study the responses of fenugreek along with remediation potential was tested against lead (Pb). The result showed that fenugreek accumulated Pb and translocated in the harvestable parts of plants. Ehsan *et al*, in 2014 carried out citric acid assisted phytoremediation of cadmium in *Brassica napus*. They noted that Cd stress significantly reduced plant growth and biomass production and addition of citric acid significantly reduced Cd toxicity in *Brassica napus* plants. Thus citric acid treatment caused enhanced phytoextraction of cadmium in *Brassica* plants.

Bihola et *al*, in 2019 assessed the effect of lead and cadmium on the growth parameters and protein content of *Coleus blumei* Benth and heavy metal extraction capacity. They concluded that *coleus* is one of the heavy metal stress tolerant ornamental species. Yuan *et al*, in 2013 analyzed the effects of selenium (Se) on *Coleus blumei* Benth under lead stress to determine possible mitigating mechanism of Se. Results indicated that Pb is tolerated by *Coleus* plants through allocation plasticity, activation of antioxidants system and improvements in particle size and mineralogical effects. Plant can be usefull in phytoremediation of aquatic system with Pb, especially with low concentration of selenium.

Visoottiviseth *et al*, in 2002 assess the potential of native plant species and soil sample which is contaminated by arsenic. of 36 plant species, only two species of ferns, a herb and a shrub seemed suitable for phytoremediation. The ferns were by far the most proficient plants at accumulating arsenic from soil. Baldwin and Butcher, in 2007 was employed to characterize Phytofiltration for the uptake of arsenic and macronutrients by two arsenic hyperaccumulaters, *Pteris cretica* and *Pteris vittata* in hydroponic environment. Significant differences in the concentration levels of the macronutrients were observed in roots, stems, and leaves between the control and arsenic- exposed plants. Salido *et al*, in 2004 [8] assessed the phytoremediation of lead using Indian mustard *Brassica juncea* with EDTA and electrodics. The accumulation of lead in the shoots using 0.5 mmol/kg EDTA with electric potential increased by two or four fold compared to the use of of EDTA only. The maximum lead accumulation in the shoots was obtained with the application of electric field 1h per day for 9 days with EDTA. Chanu and Gupta, in 2016 assessed the ability of *Ipomoea aquatica* to accumulate lead by exposing it to the graded concentration of metal. Accumulation of lead was highest in root followed by that in stem and leaf with translocation factor (TF) values of less than unity. Ghosh and Manchanda, in 2019 assessed the phytoremediation of heavy metals from water of Yamuna River by *Tagetes patula*, *Brassica scoparia*, *Portulaca grandiflora*. It has been noticed that there was a bioaccumulation of heavy metals in various parts of the plants.

Biswal *et al*, in 2021 conducted pot experiments to explore the phytoremediation potential of two different marigold species grown in heavy metals contaminated red, black, alluvial and recent river clay. The results showed that Tagetes erecta was more efficient in extracting heavy metals from different heavy metals contaminated soils. Karimi, in 2013 carried out the comparative phytoremediation of chromium contaminated soils by Alfalfa (*Medicago sativa*) and *Sorghum bicolor* (*L*) *Moench*. The experiment consisted of five treatments in which chromium concentrations varied from 1 to 10 mg/kg soil. Concentration of chromium in soil in all treatments after phytoremediation of Alfalfa was decreased between 60-74% and after phytoremediation of Sorghum was decreased between 51- 69.5%. Saha *et al*, in 2016 assessed the phytoremediation potential of industrial mines wastewater using water hyacinth. It has been observed that this plant was able to remove 99.5% Cr (VI) of the processed water of mines in 15 days. It is also capable of reducing total dissolved solids (TDS), BOD, COD and other elements of water also.

Alaboudi *et al*, in 2018 assessed the phytoremediation potential of sunflower by using contaminated soils with lead and cadmium. The study concludes that *Helianthus annus* plant was more favorable for Cd uptake compared to Pb, and they suggest its ability for the remediation of lead and cadmium contaminated soils. Shahandeh and Hossner, in 2000 assessed 36 plant species of different agronomic importance, size, dry matter production, and tolerance to heavy metal were evaluated for Cr (III) and Cr (VI). According to results sunflower was the least tolerant to Cr, and Bermudagrass and switchgrass were the most tolerant. Durumin Iya *et al*, in 2019 assessed the phytoremediation potential of *Polyscias fruticose* in the removal of heavy metals from spiked soil. The cuttings were transplanted then grown on 2.00 kg soil spiked with several heavy metals for 300 days. The indices used to show the ease of heavy metals uptake and translocation indicated that Co, Cr, Mn, Ni, and Pb displayed the greatest ease of absorption while Zn, Fe and Cu were accumulated in root but not translocated into the shoot. Fatima *et al*, in 2020 investigated the remediation potential of fenugreek for Cu under the influence of ascorbic acid. It was found that ascorbic acid enhanced Cu concentration maximum upto 42% in leaf, 18% in stem and 45% in roots as compared to Cu treated only plants.

Liao *et al*, in 2016 assessed the bio-surfactant enhanced phytoremediation by using two bio- surfactant and a synthetic surfactant to facilitate phytoremediation of crude oil contaminated soil by maize. Results showed that these surfactants did not significantly affect the biomass production of maize, but they inhibited the chlorophyll fluorescence of the maize leaf. Aliyu and Adamu, in 2014 done the analysis of some selected heavy metals accumulated in maize planted in contaminated soil for 42 days. Generally, it was observed that the heavy metals accumulation by the plant in soil for both experimental and control is high in maize. Banuelos *et al*, in 2005 done the analysis of three transgenic Indian mustard lines were tested under field conditions for their ability to remove selenium (Se) from Se- and boron contaminated saline sediment. The plants were over-expressed with genes encoding the enzymes APS, ECS and GS respectively. GS plants significantly tolerated the contaminated soil better than wild type. Dhillon and Dhillon, in 2009 conducted field experiments at two locations in the seleniferous region of northwestern India to evaluate the efficiency of four cropping systems. Results show that Brassica- based cropping system lead to significant reductions in Se capital of contaminated soil over 2-3 years.

Salah Elwafa *et al.*, 2019 evaluated heavy metal responsive gene study of *Sorghum*. They did genetic mapping and transcriptional profile of phytoremediation. Nahar *et al.*, 2014 described and *in silico* analysed molecular structures and mechanisms of $AtPCS_1$ Protein involved in the binding with arsenic and cadmium metals in the cell.

Adilovic *et al*., 2019 described *in silico* analysis of signalling genes and proteins from *Brassica oleracea* NCED2/3 are known to be correlated to heavy metal stress including copper, arsenic and cadmium. Gyulai *et al.*, 2012 evaluated remediation capacity of chromium, nickel, zinc, lead through *Cucurbita pepo* and also described NIP, TIP, SIP and Si-TRP sequences which are responsible for the extraction of the heavy metal.

Gavanji *et al.*, 2013 described metal binding sites on the basis of *in silico* analysis in metallothionein proteins. They selected plants of Poaceae family like *Oryza sativa* and *Triticum aestivum*. Bipul Sarkar *et al.*, 2020 evalauated remediation capacity of aluminium through *Amaranthus viridis* weed, they used *in vivo* approach for the study. Kaur Shodhi *et al.*, 2019 evaluated the systematic remediation arsenic contaminat studies through *in silico* aspects with molecular docking studies. G. Thapa, 2010 described heavy metal stress specific phytochelatin synthetase through *in silico* study.

Vinod Kumar *et al.*, 2012 evaluated the stress released protein Phytochelation from *Cynodon dectylon,* they used GOR4, Geno 3D, ProtParam tools for the evaluation. Thapa *et al.*, 2010 described characterization of heavy metal stress specific phytochelatin synthase gene (PCS) from heterologous systems. On the basis of genomics study they tried to sequence the gene and predicted proteins with the help of proteomics where metal can bind.

Conclusion

Phytoremediation is the key technique to clean the environment. Plants has not only capacity to accumulate organic pollutants but it also has capacity to accumulate, extract and degrade inorganic pollutants such as heavy metals in which lead, cadmium and chromium were major. Indian mustard and Maize are major plants which has capacity to extract and degrade organic and inorganic pollutants. In future the researchers has to focus on this thrust area and directly it has to be implemented to reduce pollutants from environment. Molecular approach using different process like docking, simulation between ligand and protein or gene can be fruitful techniques. So, before experiments binding capacity of pollutants with plant proteins can be assessed by which *in vitro* and *in vivo* work and its man power and economy can be reduced.

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