CHETANA
INTERNATIONAL JOURNAL OF EDUCATION (CIJE)
Peer Reviewed/Refereed Journal
(ISSN: 2455-8729 (E) / 2231-3613 (P))
Impact Factor
SJIF 2023 - 7.286

Prof. A.P. Sharma
Founder Editor, CIJE
(25.12.1932 - 09.01.2019)

Impact of Heavy Metal (Cu) on the Growth and Physiology of Calotropis Procera in Rajasthan

Omendra Singh Shekhawat
Research Scholar
Dr R B Sharma
Professor
Dept. of Life Science, University of technology, Jaipur, Rajasthan
Email: omendrasingh442@gmail.com, Mobile-8949998153

First draft received: 02.03.2024, Reviewed: 05.03.2024, Final proof received: 08.03.2024, Accepted: 15.03.2024

Abstract

Heavy metals (HMs) are widely recognized for their toxicity and have serious environmental implications as technology advances and public pressure mounts to guarantee the safest and healthiest environment. This study evaluates the phyto-remediation potential of HMs i.e., Copper (Cu), Zinc (Zn), Lead (Pb), and Cadmium (Cd) by Calotropis procera (Aiton) W.T. Aiton, also known as Sodom apple, along an urban–rural gradient and its effect on communities' diversity, forage and medicinal quality in semi-arid region of Jamdoli Jaipur Rajasthan. The HM concentration was investigated along with the urban–rural gradients by sampling C. procera and soil samples. Acid-digested samples were tested for metal concentration using an atomic absorption spectrophotometer (AAS). We used principal component analysis and cluster analysis to identify the pattern of metal distribution in plants and soil. To comprehend the species' diversity of plant communities in polluted sites, the species' composition of C. procera communities was explored. Our results showed that the concentration of HMs in the soil and plant decreased from Zn to Cd (Zn > Cu > Pb > Cd). Likewise, more than half of the soil metal accumulated in the roots and aerial part of the plant, indicating the bioaccumulation potential of the plant species for these metals. Zn, Cu, Pb, and Cd translocation ratio varied from root > stem > leaf > flower. Root to stem transfer of metal was poor, but strongly mobilized to the leaves when available in the stems. Carthamus lanatus, Sonchus asper, Cynodon dactylon, Xanthium strumarium, and Silybum marianum were the leading species in three groups of 36 plant species. Pearson's correlation revealed a significant relationship between HM concentrations and diversity indices. Zn and Cu content in the soil influenced plant species richness, Shannon–Wiener index (H'), and evenness index (Eh). Given the environmental toxicity of HMs, Cd concentrations in soil exceeded the permissible level, suggesting residents should be warned about potential health risks. As a result, the species chosen for this study can be employed as a bio-monitor and phyto--remediator of soil contaminated by these HMs, as it can accumulate HMs to a toxic level.

Key-words*: heavy metals; phyto-remediation; Calotropis procera; species diversity etc*.

Introduction

Heavy metal (HM) pollution has spread across the biosphere, making it a serious worldwide concern because of its risk to humans and animals. Various anthropogenic activities are mostly related to urban–rural gradients and industrialization, such as mining, application of fertilizers, smelting and industrial manufacturing processes, which can lead to the release of the HMs into the environment. HMs are a serious matter of concern as these elements are non-biodegradable and persist in the soil for a much longer time than in other parts of the environment. High concentrations of toxic metals, including Zn, Cu, and Cd are mostly due to human activities. Metal contamination in the environment would affect the diversity, microbial properties, and functional diversity of soils. Moreover, increased concentration of metal in the soils can be transferred and deposited into plants and likewise to human beings, producing numerous serious health disorders.

Numerous tools for remediation of contaminated soils have been undertaken, such as chemical/physical treatment and excavation of contaminated material. However, significant efforts have been made lately to identify cost-effective solutions for remediation of HMs-contaminated soil. Recently, these cost-effective approaches, such as phyto-remediation, have become a subject of public and scientific interest to alleviate HM pollutants from the soil, water, and air. Phyto-remediation is the use of plants to remove contamination from the soil, sediments, and water. Metal hyper accumulator plant species have been reported, but they are slow-growing, less abundant, and produce little biomass. Phyto-remediation treatments may be more successful if native plant species can overcome these hurdles. Native plants are frequently efficient in growth, reproduction, and survival when faced with challenging circumstances. Therefore, the search for native plants with phyto-remediation potential is important and continues.

A few natural, well-adapted plants have been utilized extensively for HM phyto-remediation applications, including lemongrass (vetiver), Siam weed, wild grasses, Crotalaria, Avena, and Sesbania species. Calotropis procera, a traditional medicinal plant that may accumulate considerable amounts of HMs, is a dominating species in Jamdoli Jaipur Rajasthan, and has been evaluated for its phyto-remediation potential in this study. Furthermore, Calotropis procera was chosen as a HMs pollution bio-monitor for a variety of reasons: it may be found on the roadside, in urban and rural regions; it has a wide geographical range and ecological distribution; and sampling, identification, and cultivation are simple and inexpensive.

The World Health Organization (WHO) and the United States Environmental Protection Agency (USEPA) have recognized strict criteria for maximum acceptable limits for accumulation of HMs in soil. Toxicant levels in urban soils have been closely monitored and extensively recorded in developed nations, such as North America and Europe, Still, HM pollution and remediation data are critically deficient in underdeveloped countries. The lack of funds for other treatments makes phyto-remediation especially well-suited to these developing nations, where labour, knowledge, and cost-cutting measures are crucial. This study was carried out to fill the gap by presenting a thorough picture of HM contamination in the semi-arid region of Jamdoli Jaipur, as well as identifying prospective candidate species for effective, practical phyto-remediation. The present study aims to investigate the toxic levels of Zn, Pb, Cu, and Cd using Calotropis procera L. as a bio-indicator; and to evaluate the extent of metal accumulation potential of Calotropis procera in Jamdoli Jaipur Rajasthan.

Review of Literature

Water from tanning industry has increased level of chromium. Concentration of chromium through anthropogenic discharge in water bodies is 3550mt (Nriagu et al. 1990)[1].

A study conducted at Faislabad by (Qadir et al.1998)[2] indicated accumulation of lead above the recommended level in plants and soil being irrigated by city effluents.

Furthermore the industrial zones of Lahore, Sheikhopura and Kala Shah Kaku indicated soil and water contamination of salts and heavy metals in their respective areas (Lone et al. 2000)[3].

Industrial revolution has resulted in polluting the biosphere (Swaminathan, 2003)[4] In soils, heavy metals are accumulated through various sources and practices. Industrial wastes aid in accumulation of toxic contaminants into the soil and are affecting the food quality and also posing adverse effects on human life as well

Toxic heavy metals affecting microorganisms are involved in impairment of biochemical systems of plants and animals. As the contaminants are released into water channels that are used for the irrigation purpose are observed to contaminate the plants as well as their products. Occupational workers and consumers both are at risk of exposure to such activities (Adachi and Tainosho, 2004) [5]. Certain metals like cadmium, arsenic, and zinc are harmful contaminants once they enter the soil and water.

Heavy metals like chromium contaminate soil, sediments, and ground water (Shanker et al. 2005) [6].

Microorganisms, plants and animals all are affected by the presence of heavy metals (Igwe et al. 2005) [7].

(Onder S, Dursun S, Gezgin S, Demirbas 2007)[8] There are limited studies on environmental pollution by heavy metals in Turkey. The investigation of showed that Pb accumulation in the grass growing near a main road junction in ankara increased during a 6 month experimental period. Analyzed samples collected from seven different sampling points in Sivas city during the months of June and July to show the environmental pollution effect of heavy metal levels in plant materials indicated that heavy metal content was as follows: zn, 12.4, Pb 6.8, ni 6.8, Cu 5.5 and Cd 0.2 µg g-1 in plant material. The results showed that all metal values may be found in industrial regions. A similar investigation in Erzurum [19] showed that Pb and Cu are important environmental problems in the winter period but that there was no problem with zn.

(Agrawal Jagrati, Gupta Nitin, Bharadwaj Nilima and Kalpana S. 2011)[9] Time to time several studies have been made by number of researchers for determining heavy metal levels in spices1, vegetables, weeds 6 as well as in medicinal plants to identify and indicate their dangerous effects. Present studies have been initiated to investigate the levels and limits of uptake of heavy metals i.e. Pb, Cd, Zn, Fe and Cu to the various usable/edible parts of three chosen medicinal plants

(Singh P, Mishra G, Sangeeta, Srivastav S, Jha KK, Khosa RL. 2011)[10] Comprehensive review on the Phyto chemical and pharmacological aspects of Capparis decidua. Capparis decidua, climbing, thorny shrub, densely branched, spinous shrub or tree, up to 6 meters in height, is widely used in traditional medicinal system of India, has been reported to possess carminative, tonic, emmenagogue, aphrodisiac, alexipharmic; improves the appetite, antirheumatic, lumbago, hiccough, cough and asthma. It is known as a rich source of alkaloids, phenols, sterols and glycosides. The innumerable medicinal properties and therapeutic uses of Capparis decidua as well as its phytochemical investigations prove its importance as a valuable medicinal plant.

(Ghosh, A.; Bhatt, M.; Agrawal, H. 2012)[11] Treated sewage water (TSW), groundwater (GW), soil and plant samples were collected from peri urban vegetable growing areas of Northern India (Varanasi) and analysed to assess the long-term effect of irrigation with TSW on Cd, Cr, Ni and Pb build-up in soils and its subsequent transfer into commonly grown vegetable crops. Results indicate that TSW was richer in essential plant nutrients but contained Cd, Cr and Ni in amounts well above the permissible limits for its use as irrigation water.

(Bhat R.S., Aldaihan S. 2013)[12] The present study investigates antibacterial activity of Garcinia mangostana in different solvents by disc diffusion method. Seven different extracts were prepared, using different solvents viz., methanol, ethanol, acetone, chloroform, ethyl ether, petroleum sprit and water. The result of antibacterial screening showed that all the extracts show moderate inhibition against pathogenic bacteria, both gram positive including Staphylococcus aureus, Streptococcus pyogenes, and Bacilllus subtillis and gram negative bacteria including Escherichia coli and Pseudomonas aeruginosa.

(Patil S B, Naikwade N S. 2018)[13] Heavy metal content in plant material is dependent on extrinsic factors like the soil and treatments etc. They are important for the safety profile and risk analysis of the drug thus should be studied as safety profile parameter. High levels of heavy metals in the herb are hazardous as they are non biodegradable. These toxins accumulate in different organs and may disturb the normal functions of central nervous system, liver, lungs, heart, kidney, brain and produce serious health hazards such as injury to the kidneys, symptoms of chronic toxicity, renal failure, and liver damage [18] and sometimes different types of cancers, skin eruptions, intestinal ulcers, etc

Table: 1.	Effect of different types of heavy metals Calotropis Procera Need of the Study
Copper (Cu)	Decrease in antioxidant activities. Impairment of photosynthetic parameters. Decrease in chlorophyll content. Reduced length and weights of embryonic axis of germinating seeds.

(Casciaro B., Calcaterra A., Cappiello F., Mori M., et al. 2019)[14] A class of natural compounds with a variety of biological activities is represented by alkaloids, important secondary metabolites produced by a large number of organisms including bacteria, fungi, plants, and animals. In this work, starting from the screening of 39 alkaloids retrieved from a unique in-house library, we identified a heterodimer β-carboline alkaloid, nigritanine, with a potent anti-Staphylococcus action. Nigritanine, isolated from Strychnos nigritana, was characterized for its antimicrobial activity against a reference and three clinical isolates of S. aureus. Its potential cytotoxicity was also evaluated at short and long term against mammalian red blood cells and human keratinocytes, respectively. Nigritanine showed a remarkable antimicrobial activity (minimum inhibitory concentration of 128 µM) without being toxic in vitro to both tested cells

(Akhtar et al. 2021)[15] Demonstrated that combining ZnO NPs and bacteria plays a significant role in plant tolerance and HM removal from water. Rice seeds primed with bacteria grown in HM-polluted water containing ZnO NPs (5 mg/L) demonstrated decreased HM uptake in roots, shoots, and leaves, resulting in increased plant growth. Furthermore, their combined effects reduced the bio-accumulation index and metallothionine (MTs) content while increasing plant tolerance. This study found that treating bacteria with lower concentrations of ZnO NPs synergistically helped plants reduce heavy metal toxicity, particularly Pb and Cu, and improved plant growth.

Study Area

Rajasthan is a state in northern India. It covers 342,239 square kilometres (132,139 sq mi) or 10.4 per cent of India's total geographical area. It is the largest Indian state by area and the seventh largest by population. It is on India's north western side, where it comprises most of the wide and inhospitable Thar Desert (also known as the Great Indian Desert) and shares a border with the Pakistani provinces of Punjab to the northwest and Sindh to the west, along the Sutlej-Indus River valley. It is bordered by five other Indian states: Punjab to the north; Haryana and Uttar Pradesh to the northeast; Madhya Pradesh to the southeast; and Gujarat to the southwest. Its geographical location is 230.3' to 300.12' North latitude and 690.30' to 78°.17' East longitude, with the Tropic of Cancer passing through its southernmost tip.

Need of the Study

"Toxicity" refers to the quality of being extremely harmful or poisonous, or containing an unwanted or excessive amount of any element. Heavy metals such as Hg, Cu, and Zn are toxic to plants. Heavy metals are primarily found in industrial waste. Which have a significant impact on soil quality as well as biological, chemical, and physical properties. Because industrial water is used on agricultural land, heavy metals are transferred and eventually concentrated in plant cells, causing toxicity in plant cells? These have potentially harmful effects on plant growth and development. These metals have an atomic weight greater than 4g/cm3 and are heavier than water. The toxicity of heavy metal elements varies by species, specific metal concentration, chemical form, Ph, and so on. These must be in small quantities or none at all.

Data Collection

We collect data for a study on the physiology of a few plant genus' resistance to heavy metals. There are two kinds of data collections. There are two types of data collection: primary data and secondary data. We visit kanota, jamdoli, vatica in Jaipur Rajasthan for primary data collection. In secondary data collection, we conduct research on various research papers and thesis related to the physiology of a few plant genus and heavy metal toxicity.

Samples Collection

Plant parts as well as soil samples were collected from three different areas of jaipur i.e. populated (Spot 1), less populated (Spot 2) and non-populated (Spot 3) areas. The spot 3 was selected in hilly and rocky area. Soil samples were taken from the upper 8-10 cm of soil. Plants parts particularly roots were washed in tape water to remove dust and dirt, and then rinsed properly with de-ionized water. The rinsed plant parts were then dried under shed. The dried samples were grinded by using pistol and mortar and then stored in clean, dried plastic bottles. Heavy metals like Fe, Zn, Cu, Pb, Ni, Cr, Mn and Co were determined in roots, root bark, stem bark, flower and fruit as well as in soil of this plant.

Fig.1 & 2 Sample collected at various spot in jamdoli area of jaipur.

Tools and Techniques

The microbes were discovered biochemically, and their ability to resist heavy metals such as zinc and copper will be assessed. Recent advances in bioremediation techniques have occurred in the last two decades, with the ultimate goal of effectively restoring polluted environments in an eco-friendly and low-cost manner. Different bioremediation techniques have been developed and modelled; however, due to the nature and/or type of pollutant, there is no single bioremediation technique that serves as a "silver bullet" to restore polluted environments.

Analysis of Samples

(a) Acid Digestion of Soil Samples

For heavy metals determination, all the soil samples were crushed lightly and were sieved to pass through 2-mm mesh. One gram of soil sample was treated with 10 ml of HNO_3 for 24h in china dish at room temperature, then 5 ml of $HClO_4$ (70%) was added. The contents of the china dish were heated on a hot plate until the volume of contents was reducing to 2 mL. The contents of the china dish were cooled and filtered through what man filter paper into 25 mL volumetric flask and diluted to the mark with distilled water. Heavy metals like Fe, Zn, Cu, Pb, Ni, Cr, Mn and Co were analyzed with Flame Atomic Absorption Spectro-photometer [FAAS] (Perkin Elmer 400).

(b) Determination of Moisture

Five grams of each fresh plant part was taken in a Petri dish and placed in oven at 90^0C for 3hours. It was then removed, cooled in desiccators and weighed. The sample was further heated in an oven for another 2 hours and the process was repeated until a constant weight was achieved. The percent moisture content was determined.

(c) Ashing

One gram of crushed and powder portion from each part of plant like root, root bark, stem bark, flower and fruit was taken in crucible for heating in an oven at 105^0C to remove moisture. Then the dried sample after charring was placed in furnace. The furnace temperature was gradually increased from room temperature to 550^0C in 1 hr. The sample was ashed for about 4 hr until a white or grey ash residue was obtained. The content of crucible were cooled in desiccators and weighed. The percent ash composition was calculated by the formula reported in literature.

(d) Acid digestion of plant samples

After cooling and weighing of samples, 5 mL of HNO_3 (25%, v/v) solution was added into crucible and, when necessary, the mixture was heated slowly to dissolve its contents. The solution was filtered through what man filter paper into 25 mL flask and diluted to the mark.

Estimation of heavy metals was carried out on Flame Atomic Absorption Spectrophotometer [FAAS] (Perkin Elmer 400 was used.

Results and Discussion

The various parts of Calotropis Procera plant collected from three different spots, populated, less populated and non-populated areas were analyzed for moistures, ash and heavy metals. The percent moisture, percent ash composition and concentration of heavy metals are appended in table respectively.

Interestingly, the plant parts including roots, root bark, stem bark, flowers and fruits as well as in the soil of non-populated area were found to have more heavy metals than populated and less populated areas. This may be because of geological strata of the studied area.

COPPER (Cu)

Although copper is an essential enzymatic element for plant growth and development but can be toxic in high concentration. Its high concentration in Plants can cause Phyto toxicity. High levels of copper may cause metals fumes fever wit flue like symptom dermatitis, irritation of the upper respiratory tract, hair and skin De-coloration, metallic taste in the mouth and nausea. Deficiency of copper results an anaemia. The recommended daily dietary intake of copper is 2-3 mg/day.

Copper level was determined in all the studied samples and was in the range of 9.60-17.20 mg/kg. High level of copper was found in the soil of spot 3 followed by the soil of spot 1. The plant samples collected from spot 3 showed high level of copper as compared to the plant samples collected from spot 1 and spot 2. From the table 1, it is clear that root bark of each spot contains the high level of copper. It is also clear from the table 1, that in case of each spot, the aerial parts of plants contain high level of copper then the underground parts.

Table 1: Percent Ash and Moisture composition in various parts of Calotropis procera

S. No.	Plant Parts	% Ash	% Moisture
Spot 1	Roots	4.01	38.48
	Root Bark	8.93	35.71
	Stem Bark	10.81	18.11
	Flower	8.73	67.22
	Fruit	6.6	66.46
Spot 2	Roots	5.34	36.43
	Root Bark	8.6	35.54
	Stem Bark	11.09	18.39
	Flower	7.64	64.01
	Fruit	5.69	65.23
Spot 3	Roots	6.83	31.01
	Root Bark	11.21	37.51
	Stem Bark	14.21	19.67
	Flower	9.63	64.11
	Fruit	8.01	77.65

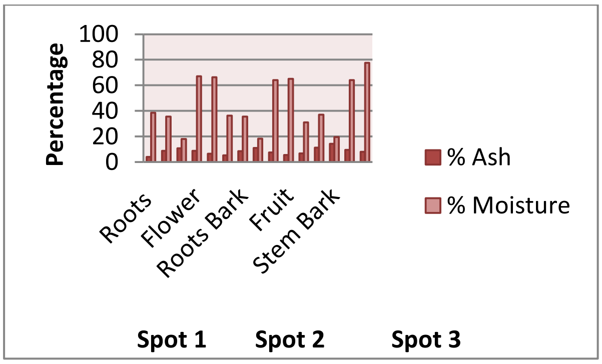

Fig. 3: Showing percentage of Ash and Moisture in Various parts of Calotropis Procera

Conclusions

The study showed that soil collected from non-populated area has high concentrations of heavy metals than populated and less populated areas. It may be due to geological strata of the studied area; as a result the heavy metals uptake by plant in that spot was greater. Medicinal plants are traditionally used by local people for long period of time to achieve desirable results. Accumulation of heavy metals varied from spot to spot. These are lower in residential area than in non-residential area. Thus, it reiterates our belief that every medicinal plant should be analyzed for contaminant load before processing it for further pharmaceutical purposes or for local human consumption.

References

1. (Nriagu et al. 1990) [1] Journal of Science of the Total Environment, Volume - 223, Published on - 10 November 1998, Pages 99-117

2. (Qadir et al 1998) [2] Journal of Environment, Development and Sustainability, Volume – 2, Pages -: 11–19, Published on – 8 feb 2000. Cadmium concentration in vegetables Grown on urban soils irrigated with Un-treated municipal sewage

3. (Lone et al. 2000) [3] Journal of Zhejiang University Science B., Volume – 9, published on march, 2008, Pages - 210–220. Phyto remediation of heavy metal polluted soils and water: Progresses and perspectives

4. (Swaminathan, 2003) [4] Magnesium Metabolism and Its Disorders Clinical Biochemistry Review, Volume - 24, Pages: 47-66, Published on – 2003

5. (Adachi, K. and Tainosho, Y. 2004) [5] Journal of Environmental International, Volume - 30, Pages - 1009-1017, Published on- april 2004. Characterization of Heavy Metal Particles Embedded in Tire Dust.

6. (Shanker et al. 2005) [6] Journal of Environmental International, Volume- 31, Published on – july 2005, Chromium toxicity in plants

7. (Igwe et al. 2005) [7] Journal of Land Degradation & Development, Volume 16, Pages - 87-96, published: 04 February 2005. Erodibility in relation to water-dispersible clay for some soils of eastern Nigeria

8. (Onder S, Dursun S, Gezgin S, Demirbas 2007) Polish Journal of Environment Study Published on – 2007, Volume – 16, Pages : 145-154. A Determination of heavy metals pollution in grass and soil of city centre green areas Turkey.

9. (Agrawal Jagrati, Gupta Nitin, Bharadwaj Nilima and Kalpana S. 2011) [9] Determination of heavy metal contents in samples of different medicinal plants. International Journal of Chemical Science, Published on 2011, Volume-9, Pages: 1126-1132.

10. (Singh P, Mishra G, Sangeeta, Srivastav S, Jha KK, Khosa RL. 2011)[10] Traditional uses, phyto chemistry and pharmacological properties of Capparis decidua. Der Pharmacia. Lettre. Published on – 2011, Volume- 3, Pages: 71-82.

11. (Ghosh, A.; Bhatt, M.; Agrawal, H. 2012) [11] Environment Monitoring Assessment, Published on - 2012, Volume - 184, Pages: 1025–1036. Effect of long term application of treated sewage water on heavy metal accumulation in vegetables grown in northern India

12. (Bhat R.S., Aldaihan S. 2013) [12] Anti-microbial activity of Garcinia mangostana using different solvents extracts. International Journal of Bio-science. Published on – 2013, Volume – 3 Pages: 262-272.

13. (Patil S B, Naikwade N S. 2018) [13] International Journal of Current Research and Review. Published on- 2018, Volume- 2, Pages- 16-25. Capparis decidua wild medicinal plant

14. (Casciaro B., Calcaterra A., Cappiello F., Mori M., et al. 2019) [14] Nigritanine as a New Potential Antimicrobial Alkaloid for the Treatment of Staphylococcus aureus-Induced Infections. Journal of Toxicology study, Published on -2019 Volume -11 Pages: 511-519.

15. (Akhtar et al. 2021) [15] Journal of America Chemical Society Omega, Published on – 30 march, 2021, Volume -14, Pages - 9709–9722. Green Synthesis of Zinc Oxide (ZnO) Nano-particles Using Aqueous Fruit Extracts of Myristica fragrans: Their Characterizations and Biological and Environmental Applications