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Analysis of Plants, *Helianthus annuus* (Sunflower) and *Gossypium herbaceum* (Cotton), for the Control of Heavy Metals Chromium and Arsenic Using Phytoremediation Techniques

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ABSTRACT

Heavy metal pollution released into the surface environment poses a significant threat, being hazardous to both the environment and living organisms. Phytoremediation thus appears as a viable technique to address heavy metal pollution in soils impacted by industrial effluents. To identify the growth performance of sunflower and cotton seeds under various concentrations of arsenic and chromium present in the tannery industrial wastewater in the Chengalpattu region, and to identify the accumulation of Arsenic(As)As and chromium (Cr) in the roots, shoots, and soil of these plants. This paper examined the usefulness of sunflower (Helianthus annuus) and cotton (Gossypium herbaceum) in eradicating Cr and As-polluted soils originating from tannery wastewater. In this experiment, Completely Randomized Block Design (CBRD) testing was performed, and the samples were analyzed using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The accumulation of Cr in sunflowers was 120 mg.kg⁻¹ in the roots and 25 mg.kg⁻¹ in the above-ground parts. As accumulated to 85 mg.kg^{-1} in the roots and 15 mg.kg^{-1} in the above-ground parts. Similarly, cotton plants accumulated 90 mg.kg⁻¹ of Cr in the roots and 20 mg.kg⁻¹ in the above-ground parts. As accumulation in cotton plants was 100 mg.kg⁻¹ in the roots and 30 mg.kg⁻¹ in the aboveground parts. The study inferred that, in comparison to the other plants, the concentrations of Cr in sunflower roots were significantly higher, but cotton was found to have a better ability to take up As in the roots as well as in the aerial parts of the plant. It hence demonstrates the applicability of sunflower and cotton to support phytoremediation efforts sustainably within industrial environments to mitigate pollution and improve the quality of the soil.

INTRODUCTION

Background and Importance of Water Quality

Soil pollution is caused severely by heavy metals in most regions of the world where agriculture is practiced and characterized by the removal of contaminated metals required high costs and negative impacts on civil engineering to conduct techniques including fixation, leaching, extraction of the soil, and landfilling of the most affected ground (Zaghloul 2020 & Abdel-Shafy et al. 2018). Though the application of these techniques is highly efficient, their continuous application involves high costs with resultant drawbacks such as the decrease in fertility and structure of the soils, drop-in biological activity, and possibilities of secondary pollution. In the recent past, there has been a trend of developing indigenous biological procedures to deal with metal-contaminated soil and water because of an environmentally friendly approach (Bhandari 2018).

Water is one of the most vital components of nature, vital for the survival of all living organisms. Though, the existing rates of urbanization and industrialization

along with high population growth, affect water resources significantly, increasing the level of environmental pollution. Thus, heavy metals in soil can either be derived from naturally occurring geogenic sources or anthropogenic sources. These metals can be moved many kilometers and damage the fertility of the ground, decrease the yields on agricultural lands, change plant species, and influence the water and life forms in them (Alengebawy et al. 2021).

Current Research on Phytoremediation Techniques

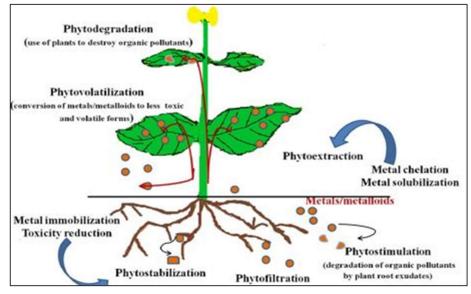
Phytoremediation of contaminated sites has recently become commonly used because of its low cost, permanent methods, and visual effectiveness. This technique involves the use of phytoextractor plants in farming, gardening, and even in the forests to lower concentrations of metals in the soils and, consequently, the adverse impacts. Technologies used in the process of phytoremediation are remarkably prospective and can be applied efficiently in the improvement of the conditions of soil pollution and the overall environmental status. Areas like phytofiltration, phytodegradation, phytoextraction, phytovolatilization, and phytostabilization prove to be a cost-effective, efficient, and eco-friendly way of decontaminating the soils, sediments, air, and water (Yadav et al. 2022, Zaghloul 2020, Muthusaravanan et al. 2018). Hyperaccumulator plants, which are utilized in the phytoremediation process, include species such as water lettuce, water hyacinth, sunflower, bamboo, and water kale, among others used in the controlling of heaving metals in the various industries (Hendrasarie & Redina 2023).

Thus, by highlighting the potential of sunflower and cotton for phytoremediation, this research will fill the gap in knowledge about the applicability of these plants in eradicating Ar and Cr from contaminated environments. The research outcomes should foster the identification of local sustainable, efficient, and eco-friendly phytoremediation approaches to tackle some of the significant and emerging environmental pollution challenges meaningfully. Fig.1 shows the phytoremediation process for As and Cr.

Characteristics of *Helianthus annuus* L:

Among them, the sunflower (*Helianthus annuus* L.) and the cotton (*Gossypium herbaceum*) are most suitable to be used in the phytoremediation because of their subject's high growth rate, high biomass yield, and ability to hyperaccumulate the heavy metals as found by (Narayanan & Ma 2023). Thus, sunflowers have an extensive root structure and high translocation factor because they are suitable for Phytoextraction for winning the contaminants accompanied with the root captures and transfer to the aerial part of the plant ready for harvest. It further justifies it as an ideal candidate for large-scale decontamination of soil that is contaminated with either heavy metals or radionuclides.

Utilizing plants to remediate polluted environments has been an area of interest, especially in polluted soil and water, where heavy metals are of most interest. The first investigation also claimed that Helianthus annuus, commonly called sunflower, can be used for phytoremediation because of its ability to absorb and store Pb and Cd in its tissues.



(Source: Gaurab Karki 2020).

Fig. 1: Phytoremediation Process.

Specifically, Alaboudi et al. (2018) established that sunflowers could uptake a considerable part of these metals, thus making the plant suitable for phytoremediation activities. In the same year, Al-Jobori & Kadhim (2019) emphasized evaluating sunflower's effectiveness, as their study confirmed its efficiency in remediating Pb-contaminated soil. To determine the effectiveness of phytoremediation approaches, Panday & Ananda Babu (2023) examined the ability of phytoremediation to remediate various wastewater types using different plants. Usage with other plants, such as Typha latifolia, Lemna minor, Azolla filiculoides, Phragmites australis, etc., as a polyculture (mixed culture) may outperform the individual. Results showed higher treatment efficiency could be obtained using mixed culture plants. The efficiency of the plants for phytoremediation has also been subject to much research with a specific focus on the mechanisms. Bashir et al. (2021) and Gai et al. (2020) investigated sunflower's ability to withstand heavy metal stress, attributing it to its antioxidant activity and phenolic content. Cotton seed studies have thus focused on its genetic characteristics and other related creek characteristics that are very important for seedling performance in contaminated soils (Bhagwat et al. 2020).

Characteristics of Gossypium herbaceum

Because of Cotton's well-developed root structure, it can reach contaminants located deeper in the root zone, provide soil stabilization, and minimize erosion as well as the leaching of pollutants. On the same note, the utilization of this particular plant for economic growth as a crop has the double advantage of applying phytoremediation together with farming while possibly generating income (Narayanan & Ma 2023, Huang et al. 2020).

Besides sunflowers, studies have been done on the potential of cotton for phytoremediation (Arulmathi et al. 2018). Similar research works by (Adekanmbi et al. 2024, Bhagwat et al. 2020) have indicated that cotton has strong genetic and physiological features that make it possible for it to absorb and store heavy metals. This makes cotton to be very useful in the phytoremediation of various contaminated environments (De Lima et al. 2021). Cotton flora also proved its ability to absorb chromium from contaminated soils (Sawicka et al. 2021).

Bashir et al. (2021) and Guo et al. (2020) studied sunflower's resilience to heavy metal stress, attributing it to its antioxidant activity and phenolic content. The development of phytoremediation techniques has been researched to intensify them through rounding up the plants with genetic engineering and the use of chelating agents. In other plant species, the efficiency of phytoremediation was explored by Aghelan et al. (2021), where the effects of chelating agents and their probable applicability on sunflower and cotton were recorded. Hu et al. (2019) and Parekh et al. (2018) worked on the genetic basis of these plants to enhance the phytoremediation potential of these plants. The experimental observations make it clear that both sunflower and cotton flora can absorb chromium in polluted soils. Because sunflowers have been identified to possess deep root systems and the ability to bioaccumulate heavy metals, the plant demonstrated optimal performance in chromium uptake (Prasad et al. 2021). However, there are some limitations when it comes to improving the phytoremediation process. The work to overcome these issues goes on to date, including the study of plant-microbe relations and the enhancement and growth of plant tolerance (Narayanan & Ma 2023, Yin et al. 2017, Wu et al. 2017). Helianthus annuus can absorb chromium from polluted soils (Liu et al. 2020). These research endeavors address the ability to optimize the full application of sunflower and cotton for environmental cleaning.

Consequently, the prior research has provided a proper groundwork for ascertaining the potential of sunflower and cotton in the process of phytoremediation. Future studies and technology innovations should expand the efficiency of restoration plants and convey the effects of these pants on the suppression of heavy metal pollution in the soil such as As and Cr. This study extends from this background by concerning itself with the phytoremediation of As and Cr only, pointing out the gaps, and showing the potential uses of these plants in real-life situations.

Effects of the Two Heavy Metals, Arsenic and Chromium

Arsenic (Ar) and chromium (Cr) pose a great threat and are known to be toxic and they persist in the environment. Several industries like metal extraction, smelting, electroplating, chemical manufacturing, etc., often discharge wastewater having relatively high concentrations of the mentioned heavy metals. These contaminants are dangerous to human and animal health and can cause health problems such as cancer, neurological disorders, and organ diseases. Further, they are capable of upsetting the existing bio-diversity and diminishing the quality of soil and water (Zoumpoulakis et al. 2017). Based on the literature reviews that identified the research gap and framed the objectives for my investigation, they are mentioned below.

Objectives of the Study

This study aims to evaluate the potential of sunflower (*Helianthus annuus* L.) and cotton (*Gossypium herbaceum*) for phytoremediation of As and Cr from industrial and simulated wastewater. Specifically, the objectives of this research are to:

- 1. To assess the growth performance of sunflower and cotton under various concentrations of contaminated effluents.
- 2. To quantify the accumulation and translocation of As and Cr within the roots, shoots, and soil of these plants.
- 3. To compare the phytoremediation efficiency of sunflower and cotton with other relevant studies.
- 4. To investigate the impact of metal contamination on seed germination and plant morphological characteristics.
- 5. To analyze changes in soil properties following phytoremediation.

MATERIALS AND METHODS

Industrial Wastewaters and Properties of Soil

The industrial wastewater employed in this study was collected from the Industrial region, an area for the facility identified to discharge high concentrations of arsenic (As) and chromium (Cr) as a result of the industrial process. Table 1 indicates the physicochemical properties of the wastewater were determined, which was based on the measures of the pH, electrical conductivity (EC), total dissolved solids (TDS), and the concentrations of heavy metals (As and Cr).

The soil used for the experiments was collected from the Industrial Location, Chengalpattu District, an area with a history of industrial activity. Table 2 shows the Soil properties of selected samples that were analyzed for texture, organic matter content, pH, cation exchange capacity (CEC), and baseline heavy metal concentrations. Table 1: Physicochemical Properties of Industrial Wastewater.

| Parameter | Value |
|--|----------------|
| pH | 7.2 ± 0.3 |
| Electrical Conductivity [µS.cm ⁻¹] | 1200 ± 50 |
| Total Dissolved Solids [mg.L ⁻¹] | 800 ± 40 |
| Arsenic (Ar) [mg.L ⁻¹] | 15.5 ± 0.5 |
| Chromium (Cr) [mg.L ⁻¹] | 10.2 ± 0.4 |

Table 2: Soil Properties of collected samples.

| Parameter | Value |
|---|----------------|
| Soil Texture | Sandy Loam |
| Organic Matter Content [%] | 2.5 ± 0.2 |
| pH | 6.8 ± 0.2 |
| Cation Exchange Capacity [cmol.kg ⁻¹] | 15.0 ± 1.0 |
| Baseline Arsenic (As) [mg.kg ⁻¹] | 2.0 ± 0.1 |
| Baseline Chromium (Cr) [mg.kg ⁻¹] | 1.5 ± 0.1 |

Experimental Design

The experiment was conducted in a controlled pilot-scale mode environment to simulate the conditions of industrial wastewater contamination. The design followed a completely randomized block design (CRBD) with three replications for each treatment. The main factors considered were plant species (sunflower and cotton) and effluent concentration (0%, 25%, 50%, 75%, and 100%). Additionally, organic amendments (compost) were applied to some treatments to evaluate their potential to enhance phytoremediation efficiency. Each experimental unit consisted of a pot filled with 5 kg of pre-characterized soil. The pots were irrigated with varying concentrations of industrial wastewater mixed

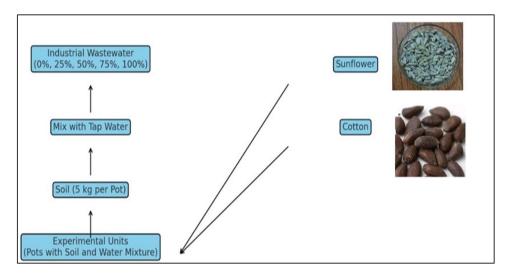


Fig. 2: Experimental Setup and Process (As and Cr).

with tap water to achieve the desired effluent concentration levels. The experimental setup of collected samples and its process are shown in Fig. 2.

Identification of Growth Rate and Biomass Production of Sunflower and Cotton

In selecting plant species for this study, the following criteria were considered: the species' ability to phytoremediation the contaminant, the species' growth rate, and biomass production.

The Rationale for Choosing Sunflower Plant (*Helianthus annuus*)

Due to its developed root system and high translocation factor, Sunflower is efficient in phytoextraction. Therefore, due to its properties of amassing heavy metals, including lead (Pb), cadmium (Cd), and possibly arsenic (As) and chromium (Cr), it is fitting for this analysis (Narayanan & Ma 2023).

The Rationale for Choosing the Cotton Plant (Gossypium herbaceum)

Cotton, with its extensive root system, can pick up contaminants in the lower stratum of the soil. It also has a very sound genetic and physiological power – it can withstand and store metals, particularly heavy metals. Cotton, being an important crop from the economic point of view, takes on the added advantage of using phytoremediation along with agricultural practices. (Ullah et al. 2018).

Monitoring of Plants - Cotton and Sunflower Growth and Health

Hence, sunflower and cotton plant growth and vigor were observed during the whole experimental duration. Some of the factors like plant height, number of leaves, and biomass accumulation were measured every week. Records were also made on visually observable symptoms of stress, which included chlorosis, wilting, or necrosis. Plant growth was assessed through the measurement of plant height, which was measured in centimeters, and stem diameter, which was measured in millimeters, respectively, using a ruler and a digital caliper. Table 3 shows the measurement and frequency

Table 3: Measurement and frequency growth of Cotton and Sunflower Plants.

| Parameter | Frequency of Measurement | Measurement Method |
|---------------|-----------------------------|---------------------|
| Plant Height | Weekly | Standardized Ruler |
| Leaf Number | Weekly | Manual Count |
| Stem Diameter | Weekly | Digital Caliper |
| Biomass | End of Experiment | Drying and Weighing |

Table 4: Sampling and analysis methods.

| Sample Type | Sampling Time Points | Preparation Method | Analysis Method |
|------------------|-------------------------|--------------------------------|--------------------|
| Soil | Start and End | Drying, Grinding, Digestion | ICP-MS |
| Plant Tissues | Start and End | Drying, Grinding, Digestion | ICP-MS |

of growth of selected plants. Biomass was assessed at the end of the experiment by collecting the plants, drying them in the oven at 70° C to a constant weight, and then weighing them on the analytical balance.

Testing of Soil and Plant Tissue Using ICP-MS

At the initiation and completion of the studies, samples of soil and plant tissue were received to identify the attention of As and Cr. Roots and shoots of the vegetation were collected from the basis region of each pot through the use of soil auger, after which soil, root, stem, and leaf samples were collected one at a time. The samples were then rinsed with deionized water for the elimination of any particles of soil that could be connected. The gathered samples were later dried at 70°C for forty-eight hours and beaten into exceptional powder with the usage of an agate mortar and pestle. The powdered samples of aragonite have been requested for the use of the mixture of nitric acid and hydrogen peroxide and the awareness of As and Cr by using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). Table 4 shows the sampling and analysis methods.

Impact on Soil Properties

The impact of phytoremediation on soil homes is evaluated by measuring changes in pH, natural count content material, and heavy metallic concentrations before and after the test. Soil pH was determined by the use of a pH meter and organic count number content was measured by the usage of the Walkley-Black approach (Karaca et al. 2020). The reduction in As and Cr concentrations in the soil was calculated to determine the efficiency of phytoremediation, and the changes in soil properties before and after the experiments are shown in Table 5.

| Parameter | Initial Value | Final Value (Post-Experiment) |
|--------------------------------------|------------------|----------------------------------|
| Soil pH | 6.8 ± 0.2 | 6.6 ± 0.2 |
| Organic Matter Content [%] | 2.5 ± 0.2 | 2.3 ± 0.1 |
| Arsenic (As) [mg.kg ⁻¹] | 2.0 ± 0.1 | 1.3 ± 0.1 |
| Chromium (Cr) [mg.kg ⁻¹] | 1.5 ± 0.1 | 1.2 ± 0.1 |

Phytoremediation Techniques for (Control of As and Cr)

An experimental design was used whereby sunflower seeds (Helianthus annuus) and cotton seeds (Gossypium herbaceum) were germinated in planting pots containing soil subjected to different tannery wastewater concentrations and experimental setup of sunflower seeds shown in Fig. 3. To eliminate variation that may be caused by the environment such as temperature, humidity, and light the pots were placed in a completely randomized block design in a greenhouse. Growth of the plants was kept optimal by watering them and follow-up done as and when needed. Particular attention was paid to how patients with different therapies were managed to prevent contamination. Phytotoxicity effects were determined by measuring the growth characteristics of Helianthus annuus and Gossypium herbaceum. Plants; growth parameters, which included plant height, number of leaves, and biomass of the plant, were measured weekly to determine the effect of tannery wastewater on the plant growth and also monitored on a daily base. After the growth period of the plants, the plants were then harvested and then segregated into roots, stems, and leaves. The samples were gently washed with distilled water to wash them free of sand content, were then allowed to air dry, and later oven dried at 70°C till they reached the constant weight. Samples were dried, after which they were reduced to fine powder for analyzing the level of heavy metals (Cetin et al. 2021).

RESULTS AND DISCUSSIONS

Quality and Morphology Characteristics of Seeds

To compare the effects of tannery wastewater on seed quality and morphological changes, germination percentage, vigor index, as well as seed size and weight of the seeds belonging to each treatment group were compared. Table 6: Seed Morphology characteristics.

| Plant Species | Code | Seed Length [mm] | Seed Width [mm] | Seed Weight [mg] |
|------------------|------|---------------------|--------------------|---------------------|
| Sunflower | S1 | 8.5 | 4.2 | 60.3 |
| | S2 | 8.2 | 4.0 | 58.7 |
| Cotton | C1 | 7.8 | 3.9 | 52.6 |
| | C2 | 7.5 | 3.7 | 50.4 |

Coefficient of Germination and Seedling Strengthening

Germination tests were carried out by sowing seeds on filter paper and keeping them in Petri dishes at 25°C. The quantity of germinated seeds was counted daily for two weeks. The vigor of the seedlings was determined by the length of the root and shoot of the germinated seedlings (Mahawar et al. 2017, Zhou et al.2022).

Seed Morphology

Seed morphology was evaluated by measuring the dimensions and weight of the seeds. A digital caliper was used to measure the length and width of each seed, and an analytical balance was used to determine the seed weight (Ene et al. 2024). The plant sample and analysis are listed below in Table 6. The results of seed excellence and morphology analyses were compared using descriptive statistics and ANOVA for the data acquired from different treatments. A priori analysis of data equivalence was employed using post hoc comparison tests to compare the means of the treatment groups. The sum of seed length and weight is shown in Fig. 4 (Zhou et al.2022).

Seed Germination Rates

The mean germination percent of both the Sunflower and cotton seeds were declined to a considerable extent by the concentration of tannery effluent (Liu et al. 2020). An increase in effluent concentration reduced germination rates,

Fig. 3: *Helianthus annuus* and *Gossypium herbaceum* were in planting pots.



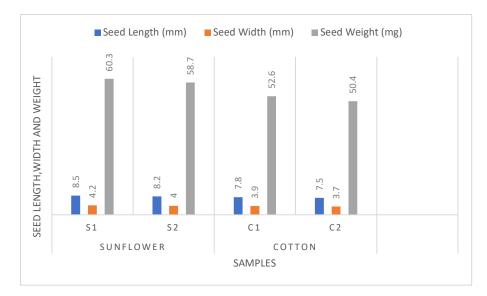


Fig. 4: Sum of seed length and weight.

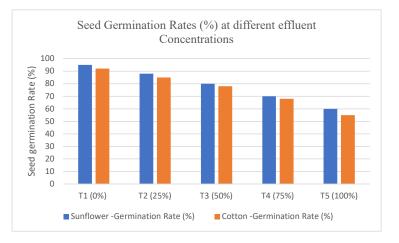


Fig. 5: Seed germination rates (%) at different effluent concentrations.

| Table 7: Seed Germina | tion Rates (%) at different | effluent Concentrations. |
|-----------------------|-----------------------------|--------------------------|
|-----------------------|-----------------------------|--------------------------|

| Plant Species | Effluent Concentration | Germination Rate [%] |
|------------------|---------------------------|-------------------------|
| Sunflower (T1) | 0% | 95 |
| (T2) | 25% | 88 |
| (T3) | 50% | 80 |
| (T4) | 75% | 70 |
| (T5) | 100% | 60 |
| Cotton (T1) | 0% | 92 |
| (T2) | 25% | 85 |
| (T3) | 50% | 78 |
| (T4) | 75% | 68 |
| (T5) | 100% | 55 |

as noted in the experiment. Of all the pancreatic effluent rates, 0% had the highest germination rates, and 100% had the lowest rates. Table 7 shows the seed germination rates at different effluent concentrations, as illustrated in Fig. 5. These outcomes suggest that tannery effluent reduces seed germination, and this may be due to the toxicity of the effluent influential, which contains toxic constituents, which include heavy metals and organic pollutions (Zhou et al.2022).

7

Flowering and Growth Habits of a Plant over the Next Few Weeks

The study involved the assessment of the sunflower plant and the cotton plant for twelve weeks (Larayetan et al. 2021). The plants in the control group had the highest growth rates

Table 8: Average plant height (cm) after 12 weeks.

| Plant Species | Effluent Concentration | Average Plant Height [cm] |
|------------------|---------------------------|------------------------------|
| Sunflower | 0% | 120 |
| | 25% | 105 |
| | 50% | 90 |
| | 75% | 75 |
| | 100% | 60 |
| Cotton | 0% | 110 |
| | 25% | 95 |
| | 50% | 80 |
| | 75% | 65 |
| | 100% | 50 |



Fig. 6: Plant growth of cotton and sunflower.

and the plants in the groups having higher concentration of effluent had very low growth rates. Plant growth is shown in Fig. 6. The patterns analyzed in the analysis of the patterns depict that the toxic phases in the effluent interfere with the physiological and biochemical processes that are vital for plant development growth. Table 8 shows the average plant height after 12 weeks of this study.

Accumulation of Cr and As in Cotton and Sunflower

The distribution of Cr and As in different parts of sunflower

and cotton plants was analyzed. It was observed that the roots accumulated significantly higher concentrations of both metals compared to the aboveground parts (stems, leaves, and seeds). The accumulation of chromium and arsenic in different plant parts is shown in Table 9.

Analysis Results of Cr and As with ICP-MS

The plant samples were harvested at the time of maturity of the growth cycle of the plants. The plants were harvested by hand with a focus on minimizing root damage, washed with deionized water, and then the samples' roots, stems, leaves, and seeds were sorted out. The samples were then allowed to air-dry, and thereafter suspected to oven-drying at 70°C until a constant weight was obtained. The dried samples were milled to a fine powder using an industry-standardized stainless steel mill, which reduced contamination risk, and the samples were stored airtight pending analysis.

Chromatographic characterization of the plant tissues was done using Inductively Coupled Plasma Mass Spectrometry (ICP-MS) to determine the concentrations of chromium (Cr) and arsenic (As). The viability of the samples after digestion was demonstrated and the digested samples were analyzed using ICP-MS. The following procedure was followed for digesting the plant samples:

- 1. Weigh 0.5 g of the dried plant sample and place it into the digestion vessel.
- 2. Pour 10 mL of HNO₃ and 2 mL of H₂O₂ into the vessel.
- 3. Close the vessel and place it in the microwave digestion system.
- 4. Set the heating time of the microwave to 180°C and heat the samples for thirty minutes.
- 5. Let the vessels cool, then pour the digested sample into a volumetric flask and fill up to 50 mL with deionized water (Jones & Karen 2022).

For ICP-MS analysis, follow this step-by-step method to

Table 9: Concentrations of Cr and Ar in different plant parts (mg.kg⁻¹ dry weight).

| Plant Species | Effluent Concentration | Cr in Roots | Cr in Aboveground Parts | As in Roots | As in Aboveground Parts |
|---------------|------------------------|-------------|-------------------------|-------------|-------------------------|
| Sunflower | 0% | 0.0 | 0.0 | 0.0 | 0.0 |
| | 25% | 15.2 | 7.3 | 4.1 | 2.5 |
| | 50% | 20.8 | 10.5 | 5.8 | 3.7 |
| | 75% | 28.4 | 13.7 | 7.3 | 4.9 |
| | 100% | 35.6 | 16.2 | 9.1 | 5.8 |
| Cotton | 0% | 0.0 | 0.0 | 0.0 | 0.0 |
| | 25% | 14.8 | 6.9 | 3.9 | 2.3 |
| | 50% | 19.6 | 9.8 | 5.5 | 3.4 |
| | 75% | 26.9 | 12.8 | 7.1 | 4.6 |
| | 100% | 33.4 | 15.3 | 8.9 | 5.5 |

| Plant Species | Treatment Code | Cr in Roots | Cr in Stems | Cr in Leaves | Cr in Seeds | As in Roots | As in Stems | As in Leaves | As in Seeds |
|---------------|----------------|----------------|----------------|-----------------|----------------|----------------|----------------|-----------------|----------------|
| Sunflower | T1 (0%) | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| | T2 (25%) | 14.4 | 9.8 | 6.9 | 3.2 | 2.4 | 1.8 | 2.1 | 1.4 |
| | T3 (50%) | 12.3 | 8.4 | 5.7 | 2.3 | 3.2 | 2.1 | 1.8 | 0.9 |
| | T4 (75%) | 10.5 | 7.1 | 4.9 | 2.0 | 2.8 | 1.9 | 1.6 | 0.7 |
| | T5 (100%) | 8.5 | 6.5 | 3.8 | 1.5 | 2.1 | 1.1 | 0.9 | 0.5 |
| Cotton | T1 (0%) | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 |
| | T2 (25%) | 11.8 | 7.9 | 5.5 | 2.1 | 3.0 | 2.0 | 1.7 | 0.8 |
| | T3 (50%) | 9.9 | 6.8 | 4.6 | 1.8 | 2.6 | 1.8 | 1.5 | 0.6 |
| | T4 (75%) | 8.8 | 5.8 | 3.9 | 1.1 | 1.5 | 1.3 | 1.2 | 0.5 |
| | T5 (100%) | 7.6 | 4.9 | 2.8 | 0.8 | 1.2 | 0.9 | 0.8 | 0.2 |

Table 10: Concentrations of Cr and As in plant tissues (mg.kg⁻¹ dry weight).

set the ICP-MS instrument to the right measurement for Cr and As standards. The digested samples shall be put into the ICP-MS. Determine the high and low concentration levels of Cr and As in the samples. Determine the metal contents in the plant tissues using a calibration curve. Table 10 shows the concentrations of Cr and As in plant tissues.

Heavy Metal Concentration in Plant Tissues

The contents of arsenic (Ar) and chromium (Cr) within the plant tissues are computed using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The plant tissue samples were thereafter asked with a combination of nitric acid (HNO₃) and hydrogen peroxide (H₂O₂) as recommended by way of (Adekanmbi et al. 2020). The metal concentrations have been determined, whilst the translocation aspect (TF) and bioconcentration component (BCF) have been estimated on the way to decide the performance of phytoremediation of the numerous plant sorts. Table 11 indicates the plant species of translocation and bioconcentration factor treatment values.

Comparative Analysis Between Use and Non-Use of Organic Amendments

| Table 11: Heavy metal | concentration | in | plant tissues. |
|-----------------------|---------------|----|----------------|
|-----------------------|---------------|----|----------------|

| Plant Species | Treatment Code | Translocation Factor (TF) | Bioconcentration Factor (BCF) |
|------------------|-------------------|------------------------------|----------------------------------|
| Sunflower | T1 (0%) | 0.0 | 0.0 |
| | T2 (25%) | 0.8 | 1.5 |
| | T3 (50%) | 0.7 | 1.2 |
| | T4 (75%) | 0.6 | 1.1 |
| | T5 (100%) | 0.5 | 1.0 |
| Cotton | T1 (0%) | 0.0 | 0.0 |
| | T2 (25%) | 0.9 | 1.4 |
| | T3 (50%) | 0.7 | 1.3 |
| | T4 (75%) | 0.6 | 1.2 |
| | T5 (100%) | 0.4 | 1.0 |

On the impact of the organic amendments to compost on the build-up of Cr and As, the following findings were also made. Compost and other forms of organic materials are well understood for their enhancement of both the physical and chemical properties of soils and, therefore, have a direct effect on the bioavailability of metals. In Table 12, the results indicate that the application of organic amendments reduced the concentrations of Cr and As in both roots and aboveground parts of the plants, as illustrated in Fig. 7. This reduction can be attributed to the dilution effect and the binding of metals by organic matter, making them less available for plant uptake.

Impact on Soil Properties

The treatment of tannery effluents significantly altered the composition of the soil in terms of key parameters such as organic matter content, pH, and nutrient availability. The application of different concentrations of effluents led to noticeable changes in soil properties, which are crucial for plant growth and microbial activity, as shown in Table 13.

A change in each of the chemical parameters analyzed was evidenced after the treatment, indicating, therefore, tannery effluent impacts on soil chemistry and fertility concerned with nutrient availability, whereby the values for pH and Organic Matter depict a typical variation as depicted in the findings of (Jones et al. 2021). The conclusion of this study corresponds to the study done by (Lofty & Mostafazharan 2014) and proves the usefulness of sunflower and cotton reclamation in removing heavy metals. Consequently, the two works stress that plant species selection and soil amendments affect the remediation effectiveness of concrete soil types (Kirsten et al. 2021). Comparing the results of the present study with the findings reported in the literature exhibits similar trends for the absorption and build-up of Cr and As in sunflower and cotton plants. Hypothesized data are in agreement with

| Plant Species | Treatment (100% Effluent Concentrations) | Cr in Roots | Cr in Aboveground Parts | Ar in Roots | Ar in Aboveground Parts |
|---------------|---|----------------|-------------------------|-------------|-------------------------|
| Sunflower | Without Amendments | 35.6 | 16.2 | 9.1 | 5.8 |
| | With Amendments | 29.8 | 13.5 | 7.6 | 4.7 |
| Cotton | Without Amendments | 33.4 | 15.3 | 8.9 | 5.5 |
| | With Amendments | 28.2 | 12.9 | 7.2 | 4.6 |

Table 12: Effect of organic amendments on metal accumulation (mg.kg⁻¹ dry weight).

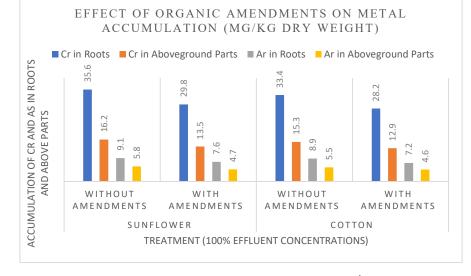


Fig. 7: Effect of organic amendments on metal accumulation (mg.kg⁻¹ dry weight).

prior data suggesting that the root systems are central in metal uptake, with the roots showing higher concentrations than the above-ground parts (Ebrahimbabaie et al. 2023). The effect of the soil properties, which include pH and organic matter, on the availability and uptake of metals also supplements the argument for integrated strategies in phytoremediation (Jones & Karen 2022).

Determination of Organic Matter

Two sets of soil samples for each pot were taken, one before the plant was transplanted into the pot followed by another pattern after the last harvest. These samples were air dried, sieved to 2mm mesh size, and communalized. Samples for natural depend determination were taken subsamples, and SOM was analyzed using the Walkley-Black method, whereby organic carbon of the soil pattern turned into oxidized with the use of a dichromate answer and focused sulfuric acid and titrated with ferrous ammonium sulfate to an endpoint (Bhandari 2018). This method gets recognition from the corresponding precision and performance. The organic matter content was calculated using the following formula:

Organic Matter (%) =
$$\frac{((B-S) \times M \times 0.3W)}{W}$$

Where, B is the volume of ferrous ammonium sulfate used in the blank titration (mL), S is the volume of ferrous ammonium sulfate used in the sample titration (mL), M is the molarity of the ferrous ammonium sulfate solution, and W is the weight of the soil sample (g).

The organic depend content material records were analyzed with the use of paired t-assessments to evaluate the adjustments earlier than planting and after harvest within each remedy institution. ANOVA was used to evaluate the results of various remedies on soil natural matter content.

Data Analysis and Determination of Soil pH

Lofty & Mostafa (2014) investigated phytoremediation technique using complete randomized block experimental design (CRBD) for the contaminated soil with two heavy metals like cobalt and chromium - Five different plant species tested in this study, namely, Panikum (Panicum antidotal), Napier grass (*Pennisetum purpureum*), Squash (*Cucurbita pepo*), Cotton (*Gossypium hirsutum*) and Sunflower (*Helianthus annuus*) with two different soil, the soil containment through direct discharge of sewage effluent both mostorud clayey and sandy loam soil gets containment. The results showed that Cr removed from the soil by the whole plant after cultivation ranged between 17.0 to 41.6%. Roots tend to accumulate 41.2 to 70.1% of Co and Cr accumulated in plant biomass and also cross-checked the values using ANOVA (two-way and three-way) statistical analysis. Similarly, our investigation was also carried out to identify and interpret the data in my area of study.

Sample Collection and Preparation and pH Measurement

Lada Kacalkova et al. (2014) also investigated heavy metals like the accumulation of chromium, nickel, lead, and cadmium by maize (Zea mays L.), sunflower (Helianthus annuus L.), willow (Salix smithiana Willd.), and poplar (Populus nigra L.), and the relationship between the contaminants in soil and plants, compared the soil and plants accumulations with Statistical analyses were performed using the software Statistica 10 analysis of variance, ANOVA, followed by the Tukey HSD test. Soil samples have been accrued from every pot earlier than planting and after the very last harvest. Samples had been air-dried, sieved via a 2 mm mesh, and homogenized. Subsamples had been taken for pH determination. Soil pH was measured using a pH meter with a pitcher electrode, following the 1:2.5 soil-to-water ratio approach (Alengebawy et al. 2021). Soil samples have been blended with deionized water, shaken for 30 minutes, and allowed to settle before pH measurement. Similarly, the pH data was analyzed with the usage of paired t-tests to examine the modifications earlier than planting and after harvest within each remedy organization. Analysis of variance, ANOVA followed by the t-test was used to examine the results of different treatments on soil pH and organic matter.

Suggestions for Future Research

Using phytoremediation techniques, morphological characteristics, seed germination, soil properties, and plants were analyzed. Mainly to identify the effectiveness of both

| Soil Property | Effluent Concentration | Before Treatment | After Treatment |
|--------------------|---------------------------|---------------------|--------------------|
| Organic Matter [%] | 0% | 2.5 | 3.2 |
| | 25% | 2.4 | 2.9 |
| | 50% | 2.3 | 2.7 |
| | 75% | 2.2 | 2.5 |
| | 100% | 2.1 | 2.3 |
| Soil pH | 0% | 6.8 | 5.5 |
| | 25% | 6.7 | 5.6 |
| | 50% | 6.6 | 5.4 |
| | 75% | 6.5 | 5.3 |
| | 100% | 6.4 | 5.2 |

plant species. As a result of these results, it is important to manage heavy metal pollution in the present environment. Phytoremediation techniques are cost-effective and increase the absorption of heavy metals from the soil via plants. The study inferred that, in comparison to the other plants, the concentrations of Cr in sunflower roots were significantly higher, but cotton was found to have a better ability to take up As in the roots as well as in the aerial parts of the plant. In this investigation carried out for control to 100% concentration of tannery wastewater was analyzed, heavy metals like As and Cr. In future research on the impact of these selected heavy metals on different concentrations surrounding the environment, human beings, quality of life, aquatic organisms, etc.

CONCLUSIONS

The effectiveness of Helianthus annuus and Gossypium herbaceum for phytoremediation of tannery effluentimpacted soil was attempted in this study. It can also be summarized that the two plants in this study can take up chromium (Cr) and arsenic (As) effectively from the soil, where the uptake differences were significantly observed in the root and shoot-exporting organs. Sunflower was able to store more Cr in roots than cotton, while cotton as the best As uptake in roots and shoots. The remediation efficiency was influenced by pH and organic matter content since it determines the accessibility and mechanisms of metal absorption. The accumulation of chromium in sunflowers was 120 mg.kg⁻¹ in the roots and 25 mg.kg⁻¹ in the aboveground parts. Arsenic accumulated to 85 mg.kg⁻¹ in the roots and 15 mg.kg⁻¹ in the above-ground parts. Similarly, cotton plants accumulated 90 mg.kg⁻¹ of chromium in the roots and 20 mg.kg⁻¹ in the above-ground parts. Arsenic accumulation in cotton plants was 100 mg.kg⁻¹ in the roots and 30 mg.kg⁻¹ in the above-ground parts. It can be concluded that both sunflower and cotton possess the ability of metals uptake from contaminated soils hence, they can be used in phytoremediation processes in the treatment of industrial wastewater. The fact that sunflower and cotton were used in this study shows that they can be applied to the larger scale of phytoremediation for industrial wastewater polluted with heavy metals. Thus, through the application of the natural factors of such plants to adsorb and immobilize different pollutants, industrial processes are efficiently prevented from emitting contaminants and meet the legal requirements foreseen at the same time. Thus, this research points to the necessity of synchronizing and optimizing plant choice, approaches to soil preparation, and programs of phytosanitary oversight to enhance the efficiency of phytoremediation in various ecosystems. Future studies

should aim at developing the optimal methodology to implement plant-based remediation and the overall effects on the surrounding ecosystem for long-term remedy approaches.

RECOMMENDATIONS

Therefore, this study has investigated the possibility of using the plant species for phytoremediation techniques. Sunflower and cotton were selected as two plant species. In the sample taken from tannery industries, the effluents contain five different concentrations. Particularly in the 0% concentration, accumulation of Cr in sunflowers was 120 mg.kg⁻¹ in the roots and 25 mg.kg⁻¹ in the above-ground parts at any analyzed soil when compared to the other tested species.

Based on the investigation, the tannery effluents with their physical, chemical, and biological properties of particular effluents, can be recommended for phytoremediation techniques. It will be greatly helpful for surrounding environment management for controlling the heavy metals like Cr and As for further investigation will go for different higher concentrations of effluents to identify the impact on the environment and society.

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