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# EFFECT OF HEAVY METALS ON LEAFY VEGETABLE (*TRIGONELLA FOENUM-GRAECUM* L.) AND ITS REMEDIATION

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## Abstract

The present research study is to evaluate and to know the toxicity nature of heavy metals in plant parts of *Trigonella foenum-graecum* L. and its remediation. Pot culture experiments were conducted with three treatments till productivity levels at Greenhouse of Botanical Garden, Department of Botany, Osmania University, Hyderabad. The three treatments consist of control without any addition of heavy metals to the soil, Treatment I - heavy metals spiked into the soil, and Treatment. II, 1% of Calcium Hydroxide added along with heavy metals to the soil. The results showed when compared to control and $Ca(OH)_2$ in treatment-I the high concentrations of heavy metals (Ni, Cd, and Cr) are found in leaf and root and stem of *Trigonella foenum-graecum* L., when compared with 1% Calcium hydroxide, treated soil (Treatment-II). In addition, the plants grown in 1% Calcium hydroxide treated soil, reversed the growth suppression, and inhibited the heavy metal toxicity in plant parts. The study concludes that leafy vegetable *Trigonella foenum-graecum* L. affected with heavy metals can be treated by using calcium hydroxide.

***Keywords*** **:** Fenugreek, heavy metals, Calcium hydroxide, atomic absorption spectroscopy, remediation, leafy vegetable.

## Introduction

*Trigonella foenum-graecum* L. is a leguminous annual plant that grows to around 60 cm tall. It is cultivated worldwide as semi-arid crop. The young leaves are often used in Asian dishes and can be included in salad mixes. Traditionally, fenugreek tea made from the crushed seed was used medicinally for a range of ailments. The plant was fed to animals both tonic and as a valuable food source. The problem of heavy metal contamination is getting serious all over the world especially in developing countries. Consumption of food crops contaminated by heavy metals is a major food chain route for human exposure (Talukdar *et al.*, 2011). Heavy metal pollution is of considerable importance and relevant to the present scenario due to the increasing levels of pollution and its obvious impact on human health through the food chain (Ali *et al.*, 2015). Heavy metal finds its entry through food from natural sources like soil, air and water wastewater, irrigation, solid waste disposal, mining, smelting, sludge applications, vehicular exhaust, fertilizers, fungicides and industrial activities (Shankar *et al.*, 2005, Bakar *et al.*, 2016). Heavy metal pollution is one of the current and most troublesome environmental problems due to mismanagement of effluents coming out from industries that contribute significantly to soil contamination. At elevated levels; these toxic substances may pose considerable loss to human and animal health as well as enormous loss in yield. Most metal toxicity occurs as a result of anthropogenic disturbance, such as mining, where unnaturally high amounts of metals are released during various processes (Lajayer *et al.*, 2017). Heavy metals are of great interest for research purposes with respect to toxicological importance to human health, plants and animals by APHA standards (Ugulur *et al.*, 2019; Aggarwal *et al.*, 2007). A significant reduction in root and shoot of mung bean /green gram was recorded due to heavy metals treatments; the reduction in root and shoot is due to the accumulation of heavy metals in plant tissues & its interactions with the minerals (Fahimirad *et al.*, 2017). Higher uptake of heavy metals by roots resulted in a reduction of the fresh dry weight of seedling (Bashari *et al.*, 2015, Bakar *et al.*, 2015). The Ni concentration increases the inhibition seed of germination and seedling of different plant SPS (Ghori et al 2019). The effect of ionic strength and pH variation on the absorption of Cd, Cr and Ni metals are variable for different soils (Tanwar *et al.*, 2013). Until now, methods used for their remediation such as excavation and landfill, thermal treatment, acid leaching and electro acclamation are not suitable for practical applications, because of their high cost, low efficiency, large destruction of soil structure and fertility. Thus, the development of bioremediation strategies for heavy metals contamination is necessary (Fatima *et al.*, 2020, Halim et al, 2015).

## Materials and Methods

### Plant Material

*Trigonella foenum greacum* L. seeds were obtained from National Seed Corporation Hyderabad, Telangana state, India. Plants were grown in earthen pots, at Greenhouse of Botanical Garden, Department of Botany, Osmania University Hyderabad. The plants were grown without pesticides, fertilizers and any type of manure. The crop was harvested after the productivity level.

**Soil sample preparation:** 20 Kgs of black soil was used for the pot experiments, the soil consists of 15.4% of clay, 3.5% of total carbon was maintained at pH 6.5.

**Heavy metal solution and soil preparation:** The heavy metal solution was prepared in the laboratory by following the guidelines. The different concentrations of heavy metals prepared are Cadmium (10ppm), Chromium (20ppm), and Nickel (16ppm). These heavy metals were dissolved in 150 liters of distilled water and sprayed on 600kg of black soil and dried in shade for 10 days for proper mixing of heavy metals in soil.

**$Ca(OH)_2$ solution preparation:** 1.5 kg of 1% $Ca(OH)_2$ was added to the 300 kgs of soil spiked with heavy metals.

**Pot preparation Treatment:** 20 Kg of black soil was filled in 15 clay pots as control, Treatment-I: 20 Kg of black soil spiked with heavy metals was filled in 15 clay pots, Treatment-II: 20 Kg of black soil spiked with heavy metals and 1% Ca $(OH)_2$ was filled in 15 clay pots.

**Acid digestion of plant samples:** The plant material of *Trigonella foenum-graecum* L. such as root, leaf, and stem were taken after harvesting and the plant material were dried, grind into a fine powder. The 10mg of powder (root, leaf, and stem) was digested in triple acid ($HCLO_4$: HCL: $HNO_3$ (5:1:1) and heated at 80-1000C for 3 hours on the hot plate. After digestion, samples were diluted with 20ml Milli Q water (Millipore instrument) and incubated for about 24 hours and analyzed directly by using Atomic Absorption Spectrophotometer (Model: Perkin Elmer Analyst 100) for detection of Ni, Cd and Cr.

## Results and Discussion

### Nickel:

Nickel, one of the important heavy metal pollutants is of considerable concern because its concentration is rapidly increasing in soils of different parts of the world. It was observed that the plants are grown in control soil, the concentration of nickel in root the plants are grown in control soil we 0.335 ± 0.013 mg/gm, plants grown in heavy metal treated soil the Ni concentration in root was 1.973 ± 0.040 mg/gm and in plants grown with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil Ni concentration in root was 0.421 ± 0.026 mg/gm. the stem was 0.232 ± 0.035 mg /gm, plants grown in heavy metal treated soil the Ni concentration in the stem was 1.572 ± 0.026 mg/gm and in plants grown with heavy metal+%(OH), 2 treated as barrier soil the Ni concentration in the stem was 0.396 ± 0.024 mg /gm. Plants grown in control soil leaf was 0.346 ± 0.041 mg /gm, plants grown in heavy metal treated soil the Ni concentration in leaf was 1.647 ± 0.023 mg /gm and in plants grown with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil the Ni concentration leaf was 0.397± 0.031 mg/ gm. The excess of Ni in the soil causes various physiological alternations and diverse toxicity symptoms such as chlorosis and necrosis in different plant SPS 18-20. The set permissible limit for Ni is 1.683 pp21. Thus, in *Trigonella foenum-graecum* L. concentrations of nickel in root, leaf, the stem is within the permissible limit. Hence, it is recommended that the application of 1% Ca $(OH)_2$ is a good barrier to stop heavy metal entry into *Trigonella foenum-graecum* L. Fig-1, 2, 3, and 4.

### Cadmium

Among the top, 20 highly toxic metals Cadmium (Cd) has been ranked No.7.The concentration of Cadmium in root the plant grown in Control soil was 0.129 ± 0.025 mg/gm, Plants grown in heavy metal treated soil. The Cd concentration in root was 1.001 ±0.027 mg/gm and plants grown in with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil, Cd concentration in root was 0.325 ± 0.024 mg/gm. The plant is grown in Control soil, Cadmium in the stem was 0.228 ± 0.036 mg /gm, Plants were grown in heavy metal treated soil, the Cd concentration in the stem was 1.254 ± 0.024 mg/gm and in plants grown with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil the Cd concentration in the stem was 0.325 ± 0.038 mg /gm. Cadmium in leaf was 0.332 ± 0.039 mg /gm, Plants were grown in heavy metal treated soil the Cd concentration in leaf was 0.930 ± 0.031 mg/gm and in plants grown in soil with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil the Cd concentration in leaf was 0.389 ± 0.026 mg/ gm. .Regulatory limit of Cadmium (Cd) in agricultural soils is 100mg/Kg soil 0.16ppm to 0.253 ppm, 0.041 ppm to 1.50 ppm which is depicted in Fig-1, 2, 3, 4. The permissible limit set by WHO is 0.2 to 0.81 ppm. Thus, in *Trigonella foenum-graecum* L. concentrations of Cadmium in root, leaf, the stem is within the permissible limit. Hence, it is recommended that the application of 1% Ca $(OH)_2$ is a good barrier to stop heavy metal entry into *Trigonella foenum-graecum* L. Fig-1, 2,3,4.

### Chromium

Chromium is one important pollutant among other pollutants. Due to its wide industrial use, chromium is considered a serious environmental pollutant. Contamination of soil and water by chromium is a recent concern. Contamination of soil and groundwater due to the use of various activities has become a serious source of concern to plant and animal scientists over the past decade. the concentration of Chromium in root was 0.229 ± 0.028 mg/gm, plants are grown in heavy metal treated soil, the Cr concentration in root was 1.648 ±0.031 mg/gm and in plants grown with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil Cr concentration in root was 0.321± 0.027 mg/gm. Chromium in the stem was 0.328 ± 0.032 mg /gm, plants are grown in heavy metal treated soil the Cr concentration in the stem was 1.646 ± 0.025 mg/gm and in plants grown with heavy metal + 1% Ca (OH) 2 treated as barrier soil the Cr concentration in the stem was 0.279 ± 0.024 mg /gm. Chromium in leaf was 0.321 ± 0.051 mg /gm, plants are grown in heavy metal treated soil the Cr concentration in leaf was 1.913 ± 0.032 mg /g and in plants grown with heavy metal + 1% Ca $(OH)_2$ treated as barrier soil the Cr concentration leaf was 0.394 ± 0.024 mg/ gm. shown in Fig-1, 2,3,4. Chromium is a heavy metal that causes serious environmental contamination in soil, sediments and ground water23, Chromium in the plants 0.0006 ppm to 0.018 ppm modified from crop plants; the WHO (2005) limits for Chromium have not yet been established. However, permissible limits for Chromium set by Canada were 2 ppm in food crop material and 0.02mg/day in oil yielding crop WHO. Comparison of metal levels in the crop plants investigated with those proposed by FAO/WHO showed that the herbs have Chromium concentrations equivalent to the limits permissible in edible plants. Thus, in *Trigonella foenum-graecum* L. concentrations of Chromium in root, leaf, stem is within the permissible limit. Hence, it is recommended that application1 of % Ca $(OH)_2$ is a good barrier to stop heavy metal entry into *Trigonella foenum-graecum* L. Fig-1, 2, 3.

**Table 1:** Heavy metal content and treatment of *Trigonella foenum-graecum* L. mg/gm of dry weight).

| **Parameter** | **Heavy metal** | **Control Soil** | **Treatment-I. (Soil+Heavy metal)** | **Treatment-II (Soil+Heavy metal+ 1% Ca $(OH)_2$** |
|---|---|---|---|---|
| | | Mean ± S.D | Mean ± S. D | Mean ± S.D |
| Root | Ni | 0.335 ± 0.013 | 1.973 ± 0.040 | 0.421 ± 0.026 |
| | Cd | 0.129 ± 0.025 | 1.001 ±0.027 | 0.325 ± 0.024 |
| | Cr | 0.229 ± 0.028 | 1.648 ±0.031 | 0.321± 0.027 |
| Stem | Ni | 0.232 ± 0.035 | 1.572 ± 0.026 | 0.396 ± 0.024 |
| | Cd | 0.228 ± 0.036 | 1.254 ± 0.024 | 0.325 ± 0.038 |
| | Cr | 0.328 ± 0.032 | 1.646 ± 0.025 | 0.279 ± 0.024 |
| Leaves | Ni | 0.346 ± 0.041 | 1.647 ± 0.023 | 0.397± 0.031 |
| | Cd | 0.332 ± 0.039 | 0.930 ± 0.031 | 0.389 ± 0.026 |
| | Cr | 0.321 ± 0.051 | 1.913 ± 0.032 | 0.394 ± 0.024 |

***Trigonella foenum graecum* L.-Root**

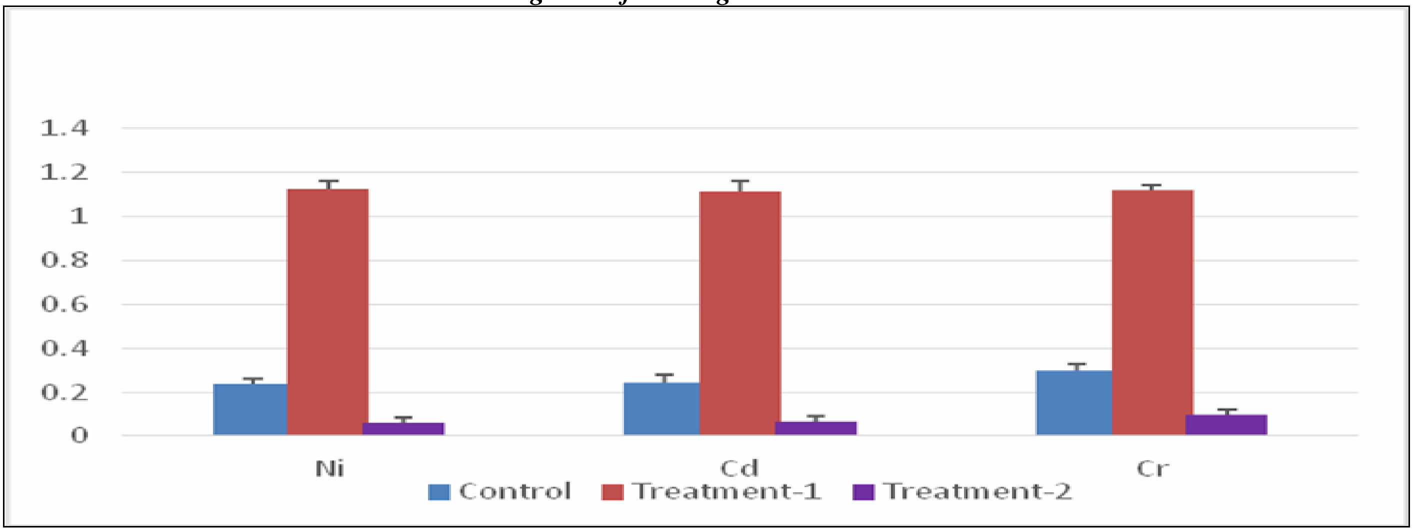


**Fig. 1 :** Heavy Metal contents of Root of *Trigonella foenum graecum* L. in three different treatments (mg/gm of dry weight)

***Trigonella foenum greacum* L.-Stem**

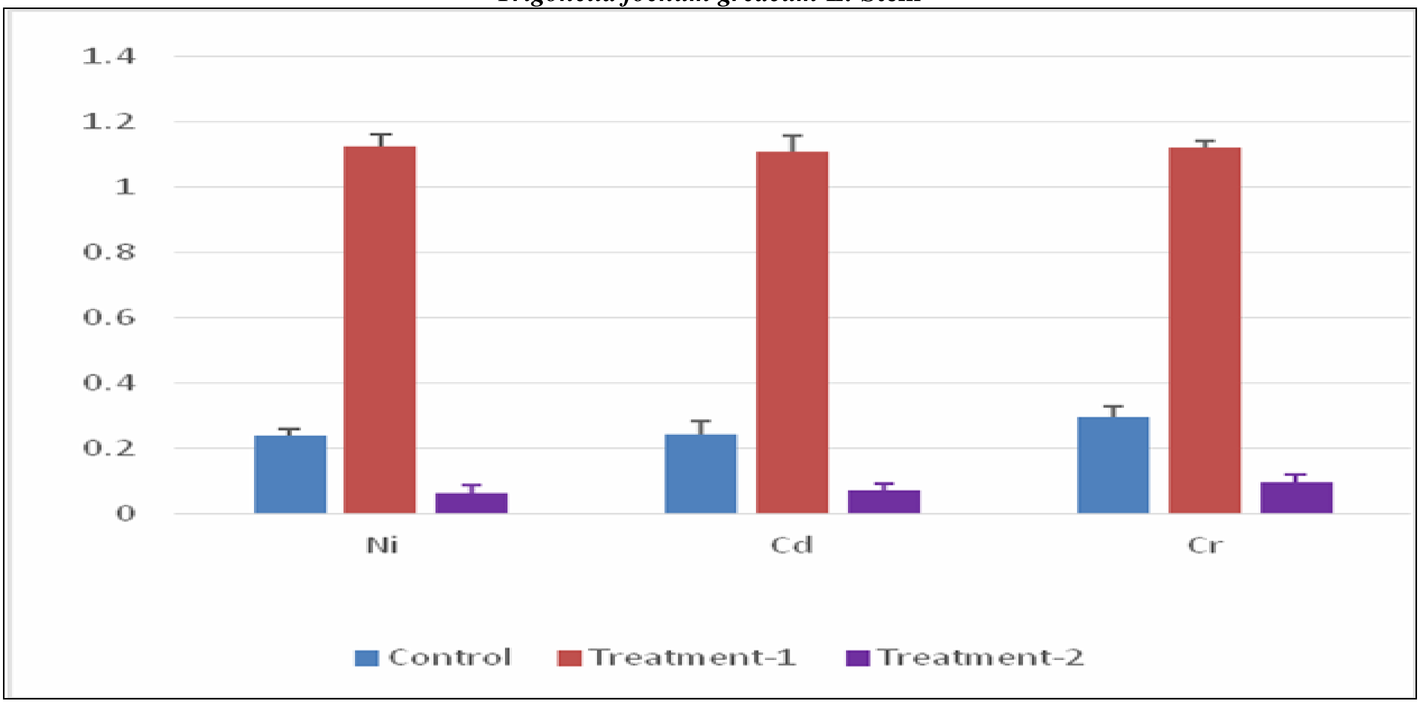


**Fig. 2 :** Heavy Metal contents of Stem of *Trigonella foenum graecum* L. in three different treatments (mg/gm of dry weight).

***Trigonella foenum graecum* L. - Leaves**

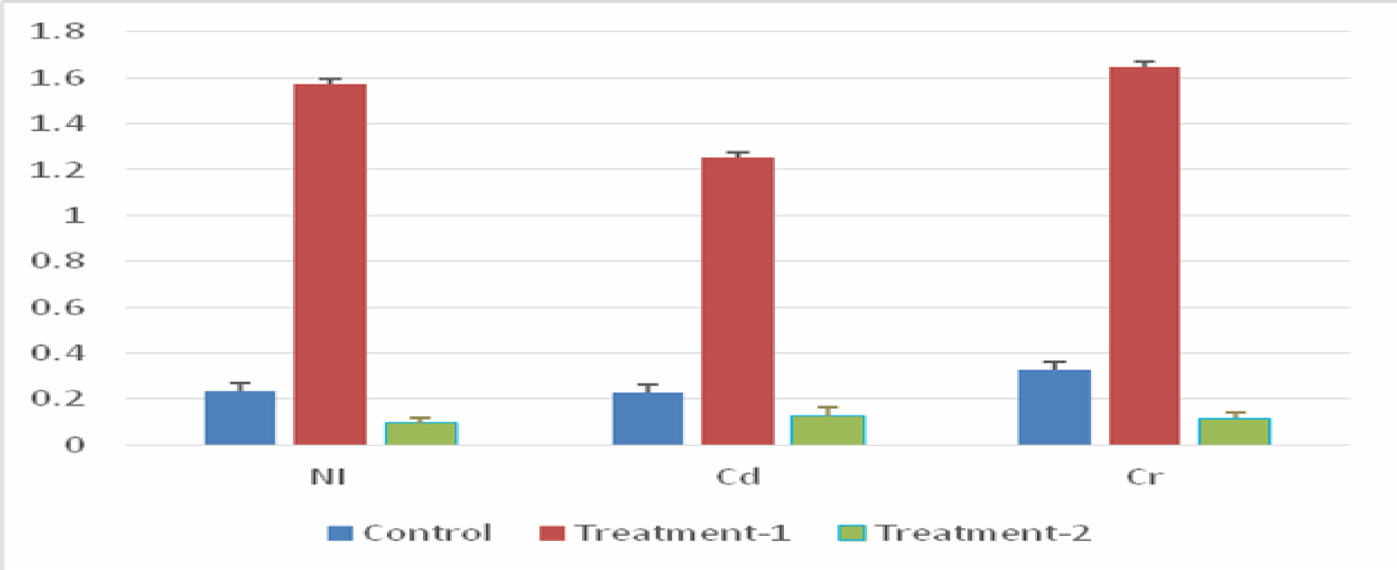


**Fig. 3 :** Heavy Metal contents of Leaf of *Trigonella foenum graecum* L. in three different treatments (mg/gm of dry weight).

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## Conclusion

The results highlights that leafy vegetable crop plants used for human consumption and for preparation of curry in daily life and used for fodder animal husbandry should be collected from an unpolluted natural habitat. Heavy metal content in the plants, growing in polluted areas can be reduced when the soil is treated with of 1% Ca $(OH)_2$. By this method, we can achieve heavy metal free *Trigonella foenum graecum* L. which is a leafy vegetable plant.

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