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ORIGINAL ARTICLE **OPEN ACCESS**

Phytoaccumulation of Cd and Cr in selected leafy vegetables crops grown in sewage irrigated soils of Prayagraj

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ABSTRACT
The present study was conducted to observe the phytoaccumulation of Cd and Cr in Spinach and Fenugreek grown in sewage irrigated sites of Prayagraj. Four different sewage-discharge points (situated at Naini, Balua Ghat, bakxibandh, and BeliGaonsites of Prayagraj regions) were selected for collecting soil and plant samples. The study revealed that different sewage-discharge points showed enrichment and phytoaccumulation of heavy metals (Cd and Cr) in the soils and plants, respectively, in proportion with the degree of pollution or levels of heavy metals through the sewage-irrigated soils. Depth soilsshowed a detectable amount of Cd 1.27-3.28 mgkg^{-1} and a higher amount of Cr 2.87-7.30 mgkg^{-1}, particularly in sewage-irrigated pots. The Cd content was found 0.63-1.34 mgkg^{-1} and Cr ranged from 1.60-3.50 mgkg^{-1} in the plants grown in sewage-irrigated soil. The study concluded that spinach and fenugreek grown in sewage-irrigated soils accumulated Cd and Cr which pose a health problem to the consumers of these vegetables. The highest phytoaccumulation of heavy metals was observed in the order of Cr > Cd by the selected plant in the order of Spinach (Spinacia oleracea L.)>Fenugreek (Trigonella foenumgraecum L.), resulting in the phytoremediation of the metals in the sewage-irrigated soils.
***Keywords**: Phytoaccumulation, Cadmium, Chromium, Sewage water, Phytoremediation*

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INTRODUCTION

Water contamination by heavy metals has become a great concern around the world. Global stock and flow of Cr, Cd, Ni, Pb, Cu, Fe, Zn, and Ag represents over 98% of the total mass of metals mobilized by human activity at the turn of the 20th century [1]. Currently, in heavy metal-polluted waters both non-essential (e.g. Cr, Cd, Hg, Ag, and Pb) and essential (e.g. Cu, Ni, Fe, and Zn) metals are present[2]. Among heavy metals occurring in polluted waters, both Cr and Zn appear as most relevant, is included in the list of priority pollutants [3, 4]Increasing release of Cr and Zn from industrial activities has strongly increased their levels in the environment producing a great concern in many countries [5].

Pollution is one of the most serious factors that influence the vulnerability of water. The main sources of pollution are untreated industrial effluents, municipal wastewater, and runoff from chemical fertilizers and pesticides from the agriculture sector[6]. Inorganic chemicals are considered a serious threat to aquatic ecosystems and, in consequence, to human health. Usually, the agricultural drainage water receives several contaminants that originate from industrial, agricultural, and municipal activities. All these activities contaminate the water bodies and sediments with huge quantities of inorganic anions and heavy metals [7]. The municipal sewage sludge, as an end product of domestic wastewater, contains organic pollutants as well as heavy metals, which integrated into the agriculture sector and thereby into the food chain, which affects human health[8, 9].Wastewater contains a large number of heavy metals; these include Ni, Cr, Cu, Cd, As, Ag, Hg, and Pb. Heavy metals present in wastewater may enter into the food chain very easily after absorption by living organisms. When they enter the food chain, they may accumulate in large quantities in the bodies of human beings that may cause serious health risks. Therefore, it is important to treat the wastewater contaminated with heavy metals before its discharge into the environment [10].

Metal mining and milling processes, industrial wastes from tanning, textile, electroplating, gas exhausts, energy, and fuel production, petrochemical from all over the world, etc., also contribute to heavy metals pollution of soil, water, and air and causes one of the most severe environmental problem today [11].

Metals occur in a variety of forms as ions, compounds, and complexes in the environment and resulted in heavy metals pollution in the biosphere. As a consequence of accelerated human intervention, soil accumulates one or more of HMs in greater values causing risks to human health[12, 13, 14, 15]. These heavy metals in the biosphere not only cause toxic and deleterious effects on human health causing chronic diseases, but also on plants by reducing their productivity due to their effects on the metabolic processes of plants and soil-plant interactions[14].

Cadmium is ingested into multiple organs within the human body *i.e.,* kidney, liver, lungs, thymus testes, heart, epididymis, prostate, and salivary glands, leading to malfunctioning of multi-organ and ultimately death[16, 17]. The Itai-Itai epidemic with 184 patients and 388 possible victims was a well-known environmental hazard associated with Cd infection. Faulty farming practices and the use of hazardous plant agro-chemicals allow Cd to invade the food chain of humans. Commonly, trace elements level is typically higher in the roots, however in certain leafy vegetables (*e.g.,* lettuce and spinach), Cd is accumulated in plant leaves owing to its fast absorption and mobility within the plant system[18].

On the opposite hand, vegetables irrigated with sewage water are a crucial source of dietary uptake of heavy metals over an in depth range of concentrations[19, 20]. The heavy metals mainly enter through roots and may translocate to the edible parts of the vegetables[21, 22]. The excess of heavy metals decreased the essential nutrient uptake by vegetables and as a result, decreased the quality of the vegetables[23]. However, metal uptake and its effects on vegetables considerably varied with vegetable species[24, 19].To our knowledge, so far, no prediction models for the uptake of heavy metals by spinach plants grown on soil amended with sewage sludge can be found in the literature. Spinach is a common leafy vegetable that belongs to the family Chenopodiaceae and is cultivated worldwide because it is highly nutritious [25]. It is an annual edible dioecious plant that grows rapidly and has the ability to survive over a moderate winter. It is versatile and has been used as a salad, a stewed vegetable, or an ingredient in numerous other cooked meat and vegetable dishes due to its health benefits[26].

The study was conducted to evaluate the depth-wise distribution, of Cd and Cr phytoaccumulated of heavy metals (Cd and Cr) by some spinach and fenugreek (*Spinacia oleracea* L. and *Trigonella foenumgraecum* L.) leafy vegetables crops, the objectives (1) to remove Cd and Cr toxicity related problems and sewage-irrigated soils and phytoaccumulated of heavy metals (Cd and Cr) by growing spinach and fenugreek (*Spinaciaoleracea* L. and *Trigonella foenumgraecum* L.) leafy vegetables crops (2) To review the accumulation of heavy metals (Cd and Cr) in the sewage-irrigated soils and plants, at different sites of the Prayagraj region of Utter Pradesh. (3) To ensure capacity of *Spinacia oleracea* and *Trigonella foenumgraecum* crop for phytoremediation of Cd and Cr in the sewage-irrigated soils.

MATERIAL AND METHODS

The investigational site is situated in northern India at 24°58′N latitude and 80°56′E longitude on the south-east, facing slopes of similar inclination at altitudes between 170 and 85 m above sea level. A sand clay loam soil, derived from sewage-irrigated Indo-Gangetic alluvial soils of Sheila Dhar Institute of Soil Science farm situated on the coming in concert of Ganga and Yamuna alluvial deposit was sampled from Prayagraj city, (U.P.) India. The properties of the soil were:

Table-1 Physicochemical properties of the investigation

Parameters	Soil
pH	7.8 ± 0.2
EC(dSm^{-1}) at ^0C	0.26 ± 0.03
Organic carbon (%)	0.56 ± 0.15
CEC (cmol(P^+)kg^{-1})	20±2.16
Total nitrogen (%)	0.07 ± 0.02
Total phosphorus (%)	0.04 ± 0.01
Texture- Sand (>0.2mm), Silt (0.002-0.2mm), Clay (<0.002) (%)	55.0±5.2, 21.0±4.5, 24.0±4.5

Extract for Cd and Cr content in soil

Soil samples collected from different discharge points of Naini, BaluaGhat, bakxibandh, and BeliGaon sites of Prayagraj regions. In each sampling unit, soil samples were drawn from several spots in a zigzag pattern leaving about 2 m area along the field margins. Silt and clay were separated by the Pipette method and fine sand by decantation. For total Cd and Cr content, 2 grams of soil was mixed in 5 ml of HNO_3 (16M, 71%) and 5ml of $HClO_4$ (11M, 71%). The composite was heated up to dryness. The volume was made up to 50 ml with hot distilled water. The clean filtrate was used for the estimation of the heavy metals (Cd and Cr) by Atomic Absorption Spectrophotometer (AAS) (AAnalyst600, Perkin Elmer Inc., MA, USA).

Soil Physico-chemical analysis

Soil pH was measured with a 1:2.5 soil-water ratio using an electrical digital pH meter. Organic carbon was determined by chromic acid digestion method and CEC using neutral ammonium acetate solution. For nitrogen, a known weight of soil (1 g) was taken in a 150 ml conical flask and treated with 10 ml of digestion mixture containing sulphuric acid and selenium dioxide. Double distilled water was used for the preparation by chromic acid digestion method, cation exchange capacity (CEC) by neutral 1 N ammonium acetate solution, total nitrogen by digestion mixture (containing sulphuric acid) selenium dioxide and salicylic acid) using the micro-Kjeldahl method, Glass Agencies, Ambala, India [27], and total phosphorus by hot plate digestion with HNO_3(16M, 71%) and extraction by standard ammonium molybdate solution[28, 27].

Plant Samples

Plants were harvested after 60 days having higher phytochemicals at their maturity stage as suggested by[29]. Plant samples were carefully rinsed with tap water followed by 0.2 % detergent solution, 0.1 N HCl, deionized water, and double-distilled water. Samples were then soaked with tissue paper, air-dried for 2–3 days in a dust- and contaminant-free environment, placed in clean paper envelopes, dried in a hot-air oven at a temperature of 45◦C, and ground to a fine powder. Plant biomass dry weights were recorded. Roots and shoots were separated and analysed.

Analysis of Cd and Cr in plant extract

One gram of ground plant material was digested with 15 ml of a tri-acid mixture containing conc. HNO_3 (16 M, 71 %), H_2SO_4 (18 M, 96 %) and $HClO_4$ (11 M, 71 %) in5:1:2). The composite was heated on a hot plate at low heat(60◦C) for 30 min, and the volume was reduced to about5 ml until a transparent solution was obtained. After cooling, 20 ml distilled water was added and the content was filtered through Whatman filter paper No. 42[27]. The Total content of Cd and Cr in the extracts of the tri-acid mixture was determined by the Atomic Absorption Spectrophotometer (AAnalyst600, PerkinElmer Inc., MA, USA).

Data Analysis

A bioaccumulation factor (BF) of Cd and Cr was calculated to determine the efficiency of the plants to accumulate a heavy metal from the soil [30]: BF = concentration of a metalin the roots (mg kg^{-1}) / concentration of the same metal in soil (mg kg^{-1}). A translocation factor(TF) of Cd and Cr was calculated to determine the ability of the plants to translocate a heavy metal from the roots to the shoot: TF = concentration of a metal in the shoot (mg kg^{-1}) / concentration of the same metal in the roots (mg kg^{-1}).
Results were expressed as the mean ± standard error of the mean (SEM) of three replicates. GraphPad Prism 9 software was used for drawing Figures.

RESULT AND DISCUSSION

The adsorptions of Cd and Cr in sewage-irrigated soils in the depth distribution of sewage-irrigated soil concentration of Cd and Cr was sewage irrigated soil at all the three sites depth (in cm.) 0-15, 15-30, and 30-60. In sites Naini are highest 3.28-1.86 mg kg^{-1} and Cr 7.30-3.36 mg kg^{-1}, BaluaGhat Cd 3.18-1.72 mg kg^{-1}, Cr 6.89-3.28 mg kg^{-1}, Bakxibandh in Cd 3.0-1.66 mg kg^{-1}, Cr 6.66-3.17 mg kg^{-1} and lowest BeliGaon in Cd 2.88-1.27 mg kg^{-1} , Cr 5.73-2.87 mg kg^{-1}. The among study was Naini profile having to fund comparatively higher sewage-sludge is characterized by the higher heavy metal contents as compared to the other sewage sites of Prayagraj region because which resulted in the available Cd and Cr of the sewage irrigated soils, the higher build-up of Cr > Cd was because the Cr and Cd content increased due to the discharge of industrial waste i.e. dye industries, tanneries, batteries (Fig. 1, 2). Along with the studies depth-wise, Naini profile having received comparatively higher sewage-sludge are characterized by the higher heavy metal contents as compared to the other soil types of the study area, because of higher metal adsorbent phases. It has a substantial role in determining the availability and mobility of heavy metals in soils because it could reduce the bioavailability of heavy metals in soils by adsorption or creating stable complexes with humic substances[31]. In addition, the organic matter content contributes to providing organic chemicals to the soil solution that may work as chelates and raise heavy metal availability to plants[32].

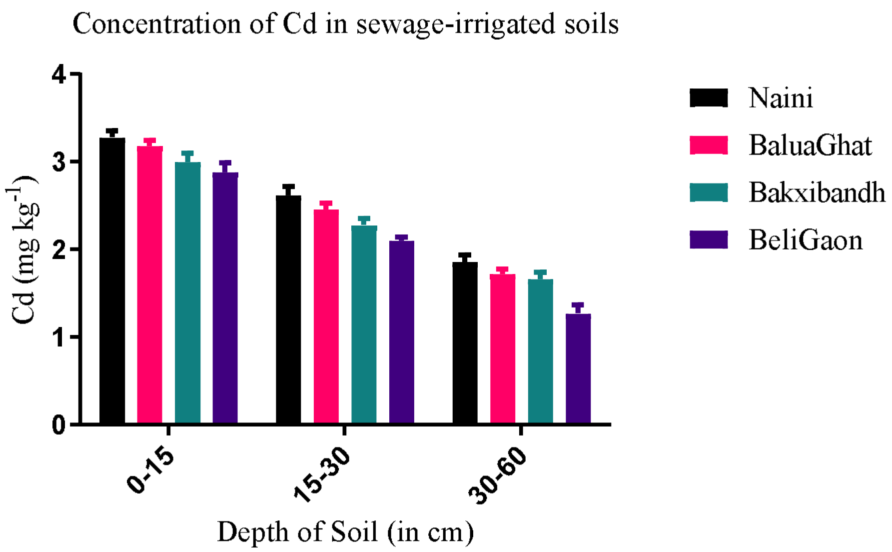

Fig.1: Adsorption of Cd in sewage irrigated soil

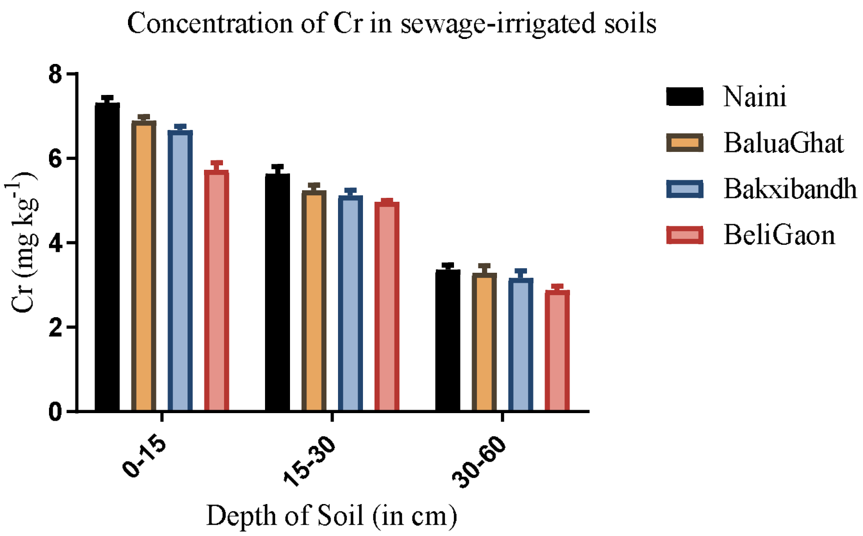

Fig.2: Adsorption of Cr in sewage irrigated soil

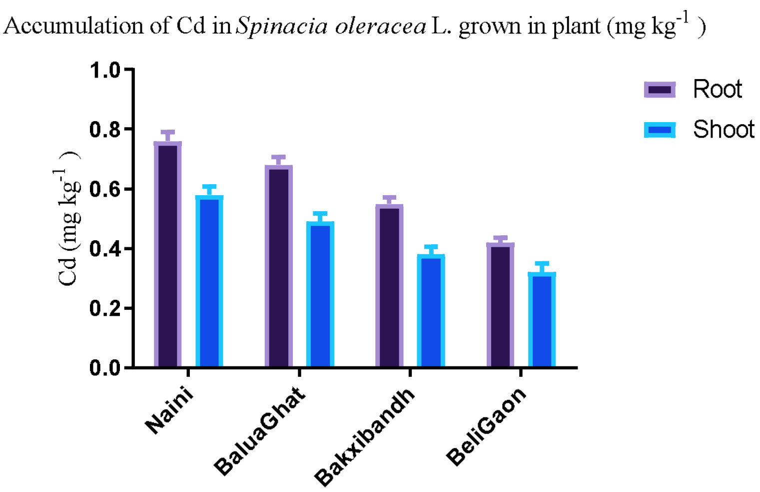

Fig.3: Phytoaccumulation of Cd in spinach grown in plant (mg kg^{-1})

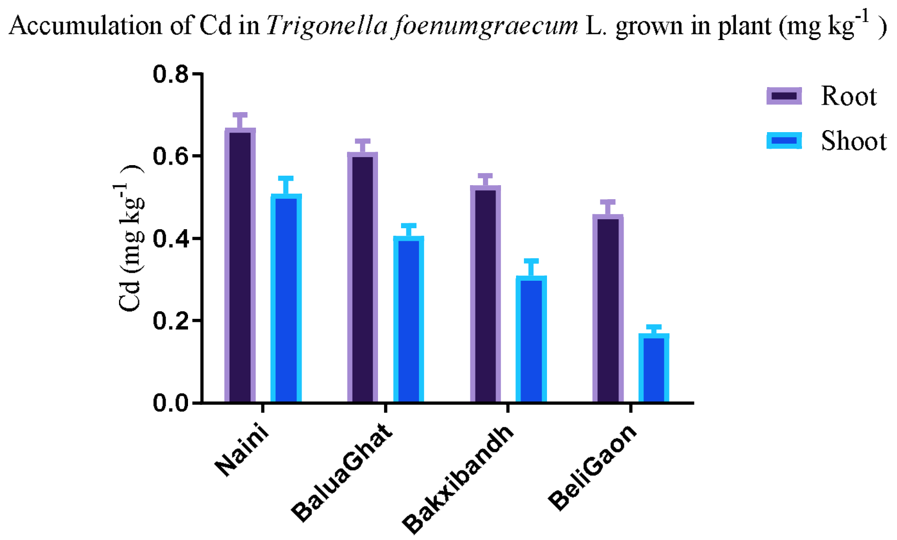

Fig.4: Phytoaccumulation of Cd in fenugreek grown in plant (mg kg^{-1})

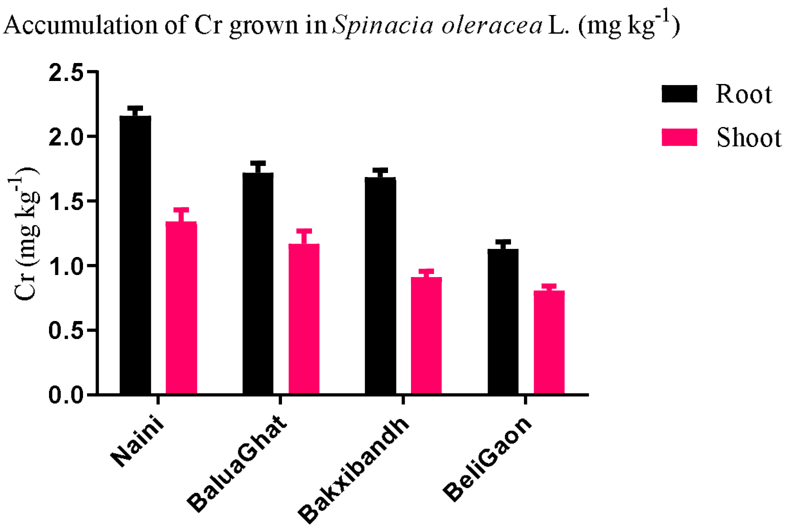

Fig.5: Phytoaccumulation of Cr in spinach grown in plant (mg kg^{-1})

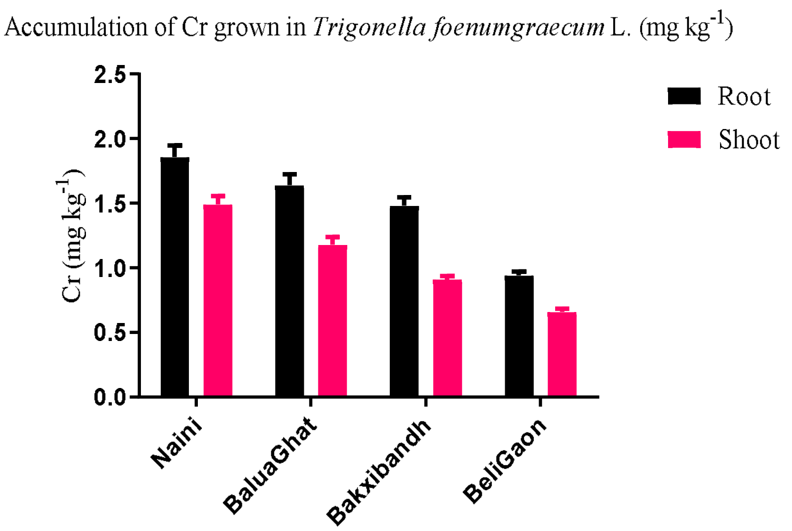

Fig.6: Phytoaccumulation of Cr in fenugreek grown in plant (mg kg^{-1})

Phytoaccumulation of Cd and Cr in Spinach (*Spinacia oleracea* L.) and Fenugreek (*Trigonella foenumgraecum* L.) grown in sewage-irrigated soils

The available heavy metals in four sites of the Prayagraj region in the sewage-irrigated soils were Cd and Cr (Fig. 1, 2). The two vegetables (spinach and fenugreek) grown in heavy metal-enriched sewage irrigated soil took up metal ions in varying degrees. The plant biological system was largely influenced by the availability of Cd and Cr (heavy metals) in terms of metal uptake. The uptake of Cd and Cr in root and shoot of *Spinaciaoleracea* grown in Naini sites was observed as Cd 42% and Cr 48%, resulted in the highest accumulation of Cd and Cr in spinach, and the uptake of Cd and Cr in root and shoot of *Trigonella foenumgraecum* grown in Naini sites was observed as Cd 36% and Cr 46% vegetables plants grown in sewage-irrigated soils. Because, the sewage-irrigated soils of Naini sites have a higher concentration of heavy metals in comparison to other sites, therefore, the contents of heavy metals in sewage irrigated soils were found more. Whereas, in the BaluaGhat sites, the uptake of heavy metals in root and shoot of *Spinacia oleracea* L. and *Trigonella foenumgraecum* L. was observed as Cd 37% and Cr 42%, and in *Trigonellafoenumgraecum* in Cd 32% and Cr 41%, which resulted in second-highest phytoaccumulation of heavy metals. However, the Bakxibandh sites, the uptake of heavy metals in root and shoot of *Spinaciaoleracea* L. and *Trigonella foenumgraecum* L. was observed as Cd 31% and Cr 39%, and in *Trigonella foenumgraecum* in Cd 28% and Cr 33%, which resulted in third phytoaccumulation of heavy metals. Whereas, BeliGaon sites, the uptake of heavy metals in root and shoot of *Spinacia oleracea* L. and *Trigonella foenumgraecum* L. grown was observed as follow: Cd 26% and Cr 34%, and in *Trigonella foenumgraecum* in Cd 22% and Cr 28%, resulted in the lowest phytoaccumulation of Cadmium and chromium in vegetables plants grown in sewage-irrigated soils. However, the accumulation of heavy metals was found higher in root in comparison to shoot. Which all sites of the Prayagraj region have grown in *Spinacia oleracea* highest and lowest *Trigonella foenumgraecum* phytoaccumulation of Cd and Cr in sewage-irrigated soils? The order of phytoaccumulation of the heavy metals was Cr > Cd in the sewage-irrigated sites (Fig. 3, 4, 5, 6). Different vegetables accumulate different metals depending on environmental conditions, type, and available form of metals. Leafy vegetables and root crops are known to accumulate higher amounts of heavy metals than grain crops [33]. The use of sewage water for irrigation may result in the accumulation of toxic metals in the soil up to hazardous levels[34].Studies have shown the increased accumulation of toxic metals in different plant parts when grown in sewage and industrial water or sludge-amended soils[35, 36].

Bioaccumulation Factor (BF) and Translocation Factor (TF)

The BF and TF values have been illustrated under the (Fig.7-10). The capability to move or be moved freely and easily of Cd and Cr from soil solution to plant tissues was enhanced by the found in four sites of sewage-irrigated soils in the Prayagraj regions of Cd from 1.27- 3.28 mg kg^{-1} (Fig. 1), and Cr 2.87-7.30 mg kg^{-1} (Fig. 2).

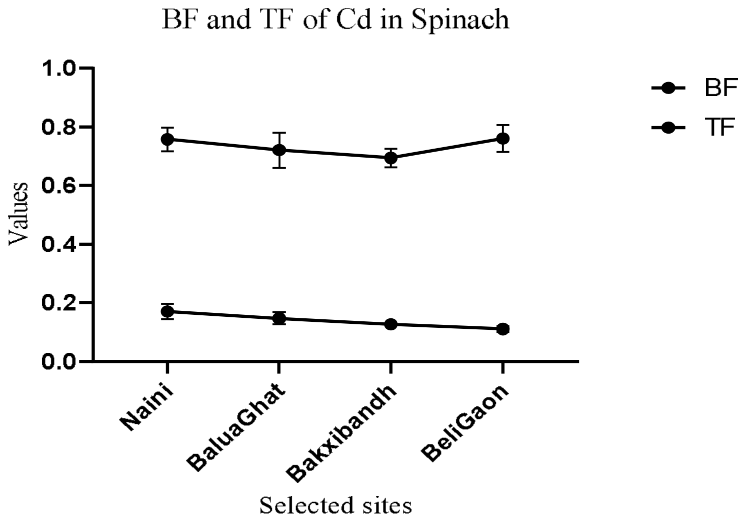

Fig.7: Bioaccumulation factor (BF) and Translocation factor (TF) of Cd in Spinach

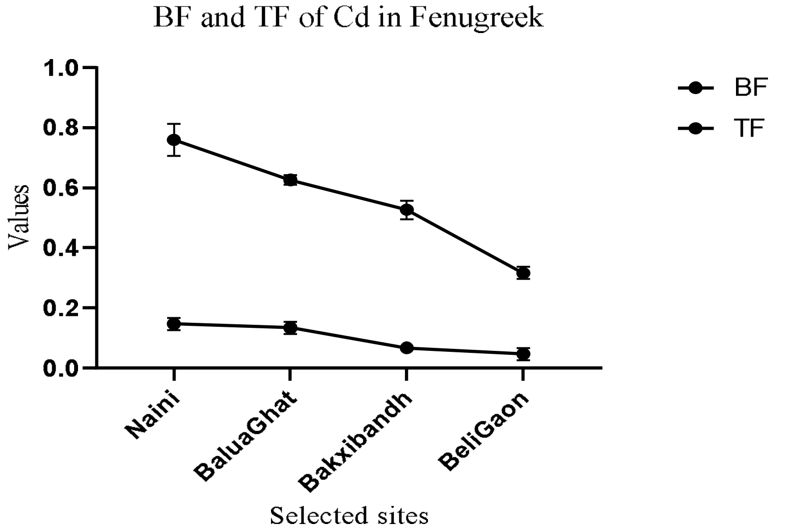

Fig. 8: Bioaccumulation factor (BF) and Translocation factor (TF) of Cd in Fenugreek

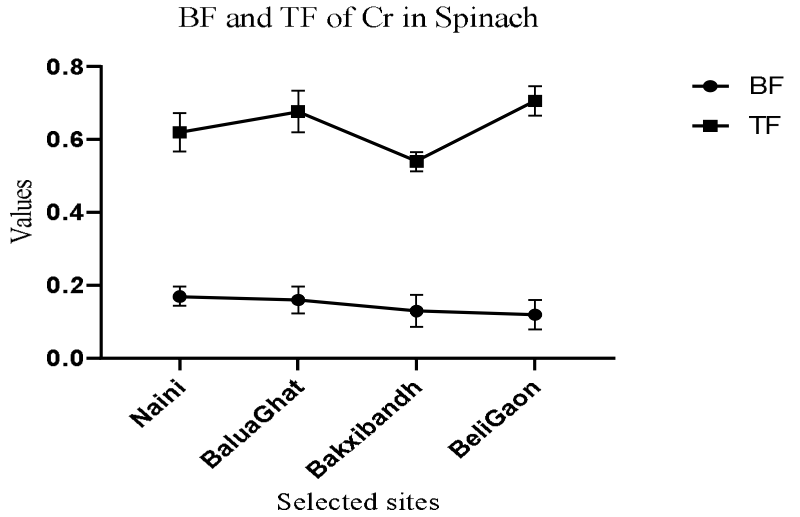

Fig. 9: Bioaccumulation factor (BF) and Translocation factor (TF) of Cr in Spinach

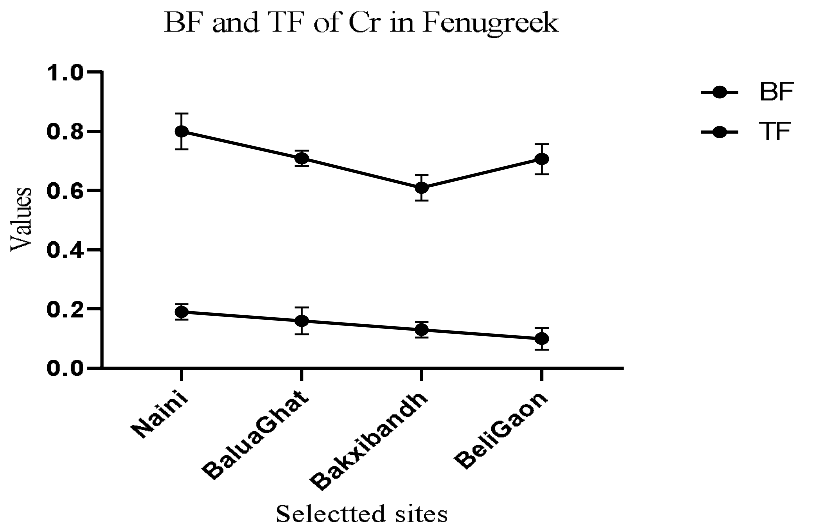

Fig. 10: Bioaccumulation factor (BF) and Translocation factor (TF) of Cr inFenugreek

The capability of the growing spinach and fenugreek plant for Cd and Cr ions accumulated. The value of bioaccumulation factor was observed that high concentration found in sewage soils in the plant with increased concentrations of heavy metals (Cd and Cr) presented higher BFs value Cd 0.17 and Cr 0.17, the obtained result showed that *Spinacia oleracea*, and the *Trigonella foenumgraecum* higher BFs Value Cd 0.15 and Cr 0.19 can be use effectively for the removal of Cd and Cr contaminated soils. The result shows the higher translocation factor (TF) value 0.76, 0.71 of Cd and Cr was observed at the higher concentrations in the root and shoot of *Spinacia oleracea* (in mg kg^{-1}). Whereas, the result shows *Trigonella foenumgraecum* plant the higher TF value 0.76, 0.80 0f Cd and Cr was observed at the higher concentrations in the root and shoot (in mg kg^{-1}).However, this decrease when Cadmium and Chromium accumulation increase from shows that the higher accumulation plant shoots tissues controlled more shoots Cd and Cr bioaccumulation (Fig.7-10). The greatest Translocation Factor (TF) 0.80.The BF generally showed the movement of heavy metals from soil to the plant root indicating the efficiency of uptake of the bio-available metals from the environment and gives an idea whether the plant is an accumulator, excluder or indicator[37].

CONCLUSION

It was observed from the present study that leafy vegetables (Spinach and Fenugreek) can accumulate significant amount of heavy metals (Cd, Cr) in their edible parts when grown in contaminated soil. Edible part of spinach and fenugreek accumulated considerable amount of Cadmium and Chromium. So the consumption of these vegetables may lead to bioaccumulation of Cd and Cr in the consumers of these vegetables and intake of such vegetables on regular basis is detrimental to health. Accumulation of biologically non-essential elements may deplete the transport of essential element into the plant body and long-term consumption of such vegetables can be hazardous to human health and create clinical problems. Therefore, regular monitoring of the presence of heavy metals in soil and vegetables are also necessary to prevent excessive build-up or such metals in human body.

CONFLICT OF INTEREST

The authors declare no conflict of interest to publish this manuscript in the journal.

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