

Biomonitoring of cadmium accumulation in plants treated with cattle manure to enhance morph-physiological attributes of Spinach (*Spinacia oleracea* L.) and fenugreek (*Trigonella foenum-gracum* L.)

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ABSTRACT

Toxicology of cadmium stress was evaluated in two different varieties of (*Spinacia oleracea* and *Trigonella foenum-gracum* L.) named as Dasi palak, Lahori palak and Kasuri methi, Guj, 1 methi respectively with the application of cattle manure the current study investigates the impact of cattle manure on the morpho-physiological traits of cadmium-stressed spinach and fenugreek. Seeds of both species were taken from Ayyub Agricultural Research Institute, Faisalabad (AARI).

Introduction

As a member of the order Caryophyllales, spinach (*Spinacia oleracea* L.) exhibits the ability to absorb heavy metals due to its enormous surface area. Single seed and less place even it can be grown in a balcony is enough for the cultivation of 3-4 plants which is very beneficial for the farmer. Due to having a short duration, 30-35 days are enough for its harvesting after sowing. Nutritionist recommended the nutritional value of spinach due to presence of iron, vitamins, B9 also known as folate (which prevent from anemia and is helpful for pregnant women approved by WHO minerals and mineral salts). The best sowing periods for spinach are the late winter or early spring. Dark green leaves are the edible portion of spinach which contains large amount of nitrogen in it but it absorbs less nitrogen from soil [6]. Fenugreek (*Trigonella foenum-gracum* L.) is very valuable, annual, having less volume herbaceous plant belong to the family leguminosae. The leaves of fenugreek are used as a vegetable because mineral and vitamins present in it but its seed are rich source of protein used as a spice and medicine for the menstrual pain, diabetes and heart disease. Latin word *Trigonella* which is the genus due to triangular shape of white-yellowish flowers and "Greek hay" refer to its specie name. Presence of N-compounds, volatile constituents, amino acid and polyphenolic compound in fenugreek show its medicinal value [3].

Environmental pollutants like heavy metals

Industrial expansion and anthropogenic activities produce large number of environmental pollutants like heavy metals which is the global issue of the world. Heavy metals belong to the 4th periods of the periodic table having density and atomic number greater than 5g/cm³ and 20g/cm³. Major source of cadmium are phosphate fertilizers, industrial wastes and sewage sludge. Uses of the phosphate fertilizer is to enhance the soil quality and crop productivity of the plants but due to presence of more than 300 Cd mg/kg it is key source of heavy metal like cadmium [1].

Excessive amount of heavy metal in plant causes many physiological disturbances

Excessive amount of heavy metal in plant causes many physiological disturbances like water uptake, enzymatic activities, seed germination, photosynthesis, stomatal movement, membrane stability, protein synthesis, hormonal balance and transpiration. Cadmium directly affects the plant nutrients like potassium, phosphorus, magnesium, calcium and enzymes which led the nutrition imbalance that ultimately decrease the photosynthesis and respiration rate. Cadmium also has indirect effects on plants by producing reactive oxygen species like hydrogen peroxide (H₂O₂), hydroxyl radicle (OH[·]) and super oxide dismutase (SOD) which damages the metabolic processes of the plants. Toxicity of heavy metal in plants reduced by the using of organic and inorganic approaches [8].

Nutrient present in animal manure

Micronutrients, phosphorus, organic matter, carbon, nitrogen and potassium are involved in the composition of animal manure which is the best source for plants to grow and produce as much as possible. Animal manure containing (SOM) that accelerate the soil porosity, moisture and nutrient retention which ultimately increase the soil fertility. Nutrient present in animal manure stored for a long time in soil but it released very slowly that is the best for root development and crop yield [2].

Application of charcoal and animal manure

The plants to grow and flourish, normal conditions are needed. However, a lot of Cd in the soil throws off the plants' overall system. The current study showed that when there was a high concentration of Cd in the soil, the morphological and biomass characteristics of spinach and fenugreek decreased. The soil's highest level of Cd content caused the hypertonic condition in the soils and ultimately less availability of water for uptake, in results decreased the plants morphological attributes. The effects of Cd metal on spinach positively correlate with the different absorption of heavy metal (Cd) on spinach decreased the biomass and morphological parameters (plant length, number of leaves and fresh and dry weight of plants). Utilization of animal manure enhances the morphological attributes of spinach and fenugreek under stress condition and the similar results by application of charcoal and animal manure. Treatment of animal manure stops the mobilization of Cd metal or make. It less available or convert the metal into less soluble form [4].

Animal manure enhances the photosynthetic activity of plants

Current investigation clearly indicated that maximum quantity of the Cd stress in the soil decreased the chlorophyll contents of the plants. Increased Cd uptake by plants caused amplification of cation production in leaves, high energy cation production in plants inhibit the synthesis of chlorophyll contents calculated that the Cd toxicity effects on spinach and mustard plants and concluded similar results. Previous study on spinach showed that presence of Cd in the soils decreased the chlorophyll and carotenoids contents in different plants such as wheat spinach. Application of animal manure enhances the photosynthetic activity of plants due to synthesis of chlorophyll contents. Similar results investigated that biochar increased the activity of photosynthetic pigment by increasing the adsorption of nutrient by the plant from soil. Current investigation is that cadmium stress mitigates the concentration of free amino acids and contents of total soluble proteins in both varieties of spinach and fenugreek (Desi, Lahori palak, Gujh-1 and Kasuri methi). Cadmium stress inhibited the uptake of nitrogen content from soil by plant protease enzymes resultant lessen amount of protein in plant observed by the researcher. The structural and functional component of amino acids that are also the constituents of plant proteins is nitrogen. Our result was like the previous work done by the revealed that cadmium stress decreased the uptake of protein content in pea plant. In the present work, cadmium stress showed various outcomes on biochemical parameters in both cultivars of spinach and fenugreek plants. Cadmium stress decreases the effect of soluble protein content in both varieties of spinach and fenugreek plant. Our result correlates with the uptake and translocation of different level of Cd metal on rape (*Brassica napus* L.) Soluble protein in plants shows the heavy metal presence in plants. Total soluble proteins maintain the osmotic potential of cell. Continuously uptake of cadmium metal reduced the effect of soluble proteins in rape plants to disturbed the transportation pathway of sugar molecules that helpful in photosynthetic system [9].

Application of animal manure

Application of animal manure improved the biochemical parameters like total soluble proteins and amino acid content in both varieties of spinach and fenugreek plant. Animal manure reduced the de novo enzymes production

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that lowered the production of proteins in sunflower, maize and soybean. Our result correlate with the biochar impact on physiological and biochemical attributes of spinach (*Spinacia oleracea* L.) in nickel contaminated soil. Biochar application increase the total soluble proteins and amino acid content in *Spinacia oleracea* (L.) that are helpful in carbon and nitrogen metabolism. It was because the promotion of de novo synthesis of enzymes. The promotion of these Enzymes raised the levels of metal-binding complexes containing heavy metals (metallothioneins and phytochelatins) determined. Present study showed that secondary metabolites like flavonoids, and phenolics decreased by increasing cadmium stress in both varieties of spinach and fenugreek plant. The metabolites present in plant tissue or organs decline the metal toxicity and increase the efficiency of signals transduction that helpful to prevent the membrane alteration. Our findings were consistent with earlier research described how the phenolic compounds responded with heavy metal stress in *zea mays*. Secondary metabolites like phenolics and flavonoids decreased by the heavy metal in tea leaves (*Camellia sinensis* L.) that affected the membrane permeability or water stressed like conditions [5].

Animal manure improved the biochemical parameters

Utilization of animal manure improved the biochemical parameters like flavonoids, and phenolics in both varieties of spinach and fenugreek plant. Animal manure decline the effect of oxidant (ROS) and improve the uptake of nutrients produced by the Cd metal examined the influence of biochar and ascorbic acid applied topically on the physio-biochemical the characteristics of barley (*Hordeum vulgare* L.) grown in cadmium-contaminated soil. Treatment of biochar improve the plant vigor by reducing the toxic effect of cadmium metal because minerals and other micronutrients found in biochar act as osmoprotectants [7].

Conclusion

Based on the study's findings, it was determined that metal (Cd) stress reduced fenugreek (*Trigonella foenum-graecum* L.) and spinach (*Spinacia oleracea* L.) growth and yield by reduction in the photosynthetic pigments and increasing the activity of ROS. Morphological and biochemical parameter reduced in 70 mg/L cadmium concentration than 35 mg/L and control. Because animal manure decreased the mobilization of cadmium metal and increased the surface charge that decreased the cadmium metals into the soil that available to the plant. Cadmium mobility and accumulation from soil to root and leaves depend on cation channels or calcium transporters but no visible chlorotic or necrotic symptoms were found because leafy vegetables also have tolerance against metal toxicity. Treatment of animal manure was good sorbent to alleviate the harmful effect of cadmium on morpho-physiological attributes by increasing antioxidant enzyme activities which ultimately stable the chlorophyll pigments and mobility of mineral nutrients. Animal manure is inexpensive and more research is needed to discover the added values of this natural sorbent in industrial applications.

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