Research Article

Enhancement effect of Hydroxyl Benzoic Acid (HBA) foliar application on growth performance of *Triticum aestivum* L. (Wheat) under induced chromium toxicity

Sami Ullah^{1*}, Musawar Khan², Muhammad Idress², Sajjad Ali²,
Muhammad Nauman Khan², Muhammad Adnan³ and Fethi Ahmet Ozdemir⁴

1. Department of Botany, UOP, KP, 25120-Pakistan

2. Department of Botany, BKUC, KP, 24420-Pakistan

3. Department of Chemistry, BKUC, KP, 24420-Pakistan

4. Department of Molecular Biology and Genetics, Faculty of Science and Art, Bingol University, 12000, Bingol-Turkey

*Corresponding author's email: sami_jan69@yahoo.com

Citation

Sami Ullah, Musawar Khan, Muhammad Idress, Sajjad Ali, Muhammad Nauman Khan, Muhammad Adnan and Fethi Ahmet Ozdemir. Enhancement effect of Hydroxyl Benzoic Acid (HBA) foliar application on growth performance of *Triticum aestivum* L. (Wheat) under induced chromium toxicity. Pure and Applied Biology. Vol. 9, Issue 4, pp2529-2542. <http://dx.doi.org/10.19045/bspab.2020.90269>

Received: 16/04/2020

Revised: 01/07/2020

Accepted: 09/07/2020

Online First: 06/08/2020

Abstract

Heavy metals are toxic to plants even at low concentration, resulting in growth and total biomass reduction. Present research was intended to assess the effect of induced chromium toxicity on growth and performance of wheat varieties (Ta-Habib and Sahar) extensively growing in Peshawar region, Khyber Pakhtunkhwa surrounded by industrial area, adding 2-4 ppm Cr^{+3} in the form of effluents to irrigation water. The study also focused on the enhancement effect of Hydroxyl benzoic acid (HBA) foliar spray under chromium toxicity wheat plant. Highest amplitude of agronomic characteristics, germination percentage and vigor index was observed in Ta-Habib indicated tolerant variety as compared to variety Sahar which has been proved by the hyper accumulation of Cr^{+3} in the mentioned variety. It was reported that treatments T1 (2ppm Cr^{+3}) and T3 (4ppm Cr^{+3}) showed adverse effects which were mitigated by the foliar application of Hydroxyl benzoic acid (HBA).

Keywords: Chromium toxicity; Foliar application; Hydroxyl benzoic acid; Wheat

Introduction

Metals being essential component of the environment chiefly deposited in the soil due the over use of chemical fertilizers, pesticides and poor municipal waste disposal [1, 2]. The term “heavy metals” refers to any metal element having high density and toxic in nature even at little

concentration. Increasing heavy metal concentration in soil also increases the uptake of heavy metals by plants depending upon the type of soil, plant growth stages and species [3]. Heavy metals reduce nutrient uptake in plants and causing chaos in plants metabolic activities such as lowering chlorophyll content, mineral

nutrients deficiency and finally chlorosis [4]. Chromium is amongst one of the prominent heavy metals whose concentration in the atmosphere is increasing at an alarming rate. Furthermore, degradation of chromium is not possible into harmless products, hence it retains in the environment for longer period of time [5]. Higher concentrations of Cr^{+3} is potentially harmful at germination and early seedling growth stages [6]. High levels of chromium cause inhibition of plant growth, chlorosis in young leaves, nutrient imbalance, wilting of tops, and root injury [7]. Heavy metals entering into plant tissues inhibit most of the physiological processes of metabolism including respiration, photosynthesis and nitrate assimilation [8]. In Pakistan the toxic heavy metals from industries are entering in food chain and ground water supplies; leading to serious ailments in local population [9]. The industrial effluents (untreated) are the main sources of heavy metal pollution [10]. Besides this, leather and tanning industries are the major sources of chromium and other potentially detrimental heavy metals in both water and soil (through leaching) thus polluting both to a great extent. Chromium releases in waste water from different industries such as food processing and match producing industries, due to which the vegetable fields irrigated with sewage around Peshawar and Faisalabad contained significantly higher concentration of chromium [11, 12]. According to Ali *et al.*, [13] chromium stress causes ultra-structural disorders in leaves, root ultra-structural modification, disruption and disappearance of nucleus. Higher concentration of Cr^{+6} in the plant root zone affects many physiological processes and inhibits plant growth [14]. Hexavalent chromium (Cr^{+6}) causes a decrease in shoot length of wheat [15]. Common agricultural crops such as wheat, Indian mustard, sorghum and maize are affected by chromium, higher chromium concentration inhibited germination, plumule and radicle length [16].

Biochemical aspects like photosynthetic pigments (Chlorophyll a and Chlorophyll b), total protein and amino acids content decreased with increasing chromium concentration [17]. Different applied treatments of foliar spray increased all studied growth parameters as number of branches and leaves per plant, leaf area per plant and leaves dry weight as well [18]. Under the influence of 4-HBA superoxide dismutase and apoplastic forms of peroxidase in wheat coleoptiles were increased [19]. Chromium effluent is highly toxic to plant and is harmful to their growth and development [20]. Toxicity of chromium in plants also results in inhibition of enzymatic activities, impairment of photosynthesis, nutrient and oxidative imbalances, and mutagenesis [21]. High levels of chromium interfere with important metabolic processes causing toxicity and ultimately reduction in growth and yield. It leads to stunted growth decrease of root-shoot growth and total biomass, chlorosis and eventually plant death [22]. However many reports have focused on the toxic effects of Cr^{+3} and Cr^{+6} on plants, our knowledge about its toxicity is shallow, and the detailed mechanisms are not richly understood. Although due to our poor knowledge, we understood up to some extent that heavy metal chromium have negative effects on crop plants. To achieve the objective of studies, little efforts have been made to determine:

- The effect of hydroxyl benzoic acid foliar application on growth performance of wheat.
- The effect of hydroxyl benzoic acid foliar application on morphological characters of *Triticum aestivum* L.
- The effect of hydroxyl benzoic acid application on agronomic characters of wheat and
- To determine the Chromium toxicity of rhizospheric soil on growth of wheat.

Materials and methods

Plant material and growing conditions

Seeds of two selected varieties of *Triticum aestivum* L. (wheat) including Ta-Habib and

Saher were obtained from Cereal Crop Research Institute Persabaq Nowshehra (CCRI). The seeds were sterilized in 5% chlorox for 2 minutes then washed with 50% ethanol for 3 minutes. Seeds were washed with distilled water and sown in plastic pots filled with soil and sand in ratio of 3:1. Proper ventilation was maintained, weeds were eradicated periodically and pots were protected from harsh weather regimes. For each treatment 3 pots (replicates) were taken. The numbers of total pots was 30. Numbers of seeds per pot were 12. Ta-Habib was selected as variety-1 (V1) and Saher was variety-2 (V2).

Nutrient culture experiment

The seedling was grown in 2ppm and 4ppm of Cr^{+3} concentrations. These various concentrations of chromium were dilute to 100ml (used water) in volumetric flask.

Application of heavy metal (Cr^{+3}) concentrations to plants

Required concentrations of heavy metal chromium are 2pp and 4ppm were applied to required replicates of plants in amount of 5ml was imposed withholding water supply for a period of 10 days. The first heavy metal treatment was started at 2ppm and 4ppm, the second and third was same as first. First treatment of heavy metal (Cr^{+3}) concentrations (2ppm and 4ppm) were given to plants after 24 days of sowing. Second treatment of heavy metal (Cr^{+3}) concentrations (2ppm and 4ppm) were given to require replicates of plants after 10 days of first treatment. Third treatment of heavy metal (Cr^{+3}) concentrations (2ppm and 4ppm) were also given to plants after 10 days of the second treatment.

Application of HBA foliar spray

During experimental work HBA (Hydroxyl Benzoic Acid) foliar spray were prepared. 1ml of HBA taken by the help of puppet and were poured in 100ml of water (1ml/1liter). Then the prepared HBA spray was sprayed on required replicates by the help of spray pressure pump. First treatment of HBA spray was given to required replicates of plants on 24 days of sowing. Second treatment of HBA spray was given to

required replicates of plants after 10 days of the first treatment. Third treatment of HBA spray was also given to required replicates of plants after 10 days of the second treatment.

Sampling

Sampling was done three times with intervals of 7, 14 and 21 days after the onset of chromium treatment. For each treatment 3 replicates were taken. The samples were stored and weighed till further analysis.

Assessment and measurement of agronomic characters

Leaf, shoot, root and soil fresh and dry weight in grams (g) were measured for all the replicates with the aid of electric balance.

Plant powder preparation

For further analysis plant samples were kept and dried at room temperature after drying the samples, grinded to fine powders then stored in moisture proof plastic bags for further analysis.

Chemical analysis of rhizospheric soil **Soil moisture content**

Soil (10 gram) was taken from uniform depth i.e. 6 inches from the surfaces of pots. Dry weight was determined after drying the soil in oven for 72 hrs at 70 °C till constant weight. The %age moisture content of soil is calculated by following formula:

$$\text{MC (\%)} = \frac{\text{Fresh weight of soil (g)} - \text{Dry weight of soil (g)}}{\text{Weight of dry soil (g)}} \times 100$$

Percent field capacity of rhizospheric soil

The field capacity (%) of rhizospheric soil can be calculated by following mathematical equation:

$$\text{FC (\%)} = \frac{\text{Weight of wet soil (g)} - \text{Weight of dry soil (g)}}{\text{Weight of dry soil (g)}} \times 100$$

Chromium analysis

The rhizospheric soil and plant powder was analyzed for chromium (Cr^{+3}). Methodologies for the preparation of different reagents, stock solutions, working solution and standards solutions are given in appendix.

Chromium analysis of rhizospheric soil

0.25gm of rhizospheric soil extract and 10-20ml of distilled water was taken in a test tube and analyzed for Cr^{+3} on atomic absorption Spectrophotometer. For the determination of chromium, stock solutions were made. 100 ppm stock solution of the Cr^{+3} were prepared by dissolving required amount of salts in distilled water. The availability of Cr^{+3} elements in the selected varieties of plants from Charsadda district was determined by Perchloric-acid digestion method [23].

Chromium analysis of selected plants of Charsadda district

For the determination of the above-mentioned element, stock solutions were made. 100 ppm stock solution of the Cr^{+3} were prepared by dissolving required amount of salts in distilled water. The availability of Cr elements in the selected varieties of plants from Charsadda district was determined by Perchloric-acid digestion method [23]. For this whole plants powder were also 0.25 gm taken from each treatment. Dried samples (0.25g) were taken in 100 ml beaker and add 5 ml of mixed acid solution i.e., Perchloric acid, sulfuric acid and nitric acid having ratios of 1:0.1:5 respectively. Afterward the mixture was boiled in fume hood on hot plate till the digestion has been completed; indicated by white fumes coming out from the flasks. Moreover, few drops of distilled water were mixed and allowed to cool. The digested samples were transferred in 100 ml volumetric flasks and the volume was made up to 100ml by adding distilled water. Then filtered the extract with Whitman filter paper and filtrate were collected in labeled plastic bottles. Concentration of these elements in the entire samples was determined by Shimadzu AA-670 Atomic Absorption Spectrophotometer.

Results and discussion

Results in (Table 1) indicated that maximum percent moisture content, field capacity,

percent germination has been reported in Ta-Habib and Saher has been recorded in treatment T5 (control) with 16.05% and 15.75% followed by treatment T2 (2ppm Cr^{+3} +HBA) with 13.40% and 12.70% after 7, 14 and 21 days respectively, indicating that Hydroxyl benzoic acid play a positive role in increasing of moisture content and field capacity, while chromium exhibiting negative effect on these parameters. Maximum shoot fresh weight (Table 2) has been recorded in treatment T2 (2ppm Cr^{+3} +HBA) after 7 days with 0.238g followed by treatment T5 (control) with 0.238g in cultivar Ta-Habib while that of Saher has been recorded in treatment T5 (control) with shoot fresh weight 0.20g followed by T2 (2ppm Cr^{+3} +HBA) with 0.19g, while minimum shoot fresh has been recorded in T3 (4ppm Cr^{+3}) with 0.181g and 0.16g respectively, result after 14 days of treatments shows that maximum shoot fresh weight has been recorded in the treatment T5 (control) with 0.40g and 0.42g followed by treatment T2 (2ppm Cr^{+3} +HBA) with 0.34g and 0.40g, while minimum shoot fresh weight has been recorded in the treatment T3 (4ppm Cr^{+3}) with 0.18g and 0.35g respectively in Ta-Habib and Saher. Maximum shoot dry weight (Table 2) has been recorded in treatment T5 (control) after 7 days in both wheat varieties (Ta-Habib and Saher) with 0.07g and 0.05g, while lowest shoot dry weight in both varieties has been recorded in treatment T3 (4ppm Cr^{+3}) with 0.03 and 0.04g followed by T1 (2ppm Cr^{+3}) with 0.061g and 0.04g respectively. Result after 14 days shows that maximum shoot dry weight in Ta-Habib and Saher has also been recorded in treatment T5 (control) with 0.13g and 0.13g followed by treatment T2 (2ppm Cr^{+3}) with 0.12g and 0.13g, whereas lowest shoot dry weight after 14 days has been recorded in treatment T3 (4ppm Cr^{+3}) with 0.06g and 0.09g respectively.

Table 1. Effect of Hydroxyl benzoic acid (HBA) foliar spray on physicochemical characteristics of rhizospheric soil and on %germination of *Triticum aestivum* L. (Wheat) under various concentration of chromium

Treatments	%Moisture Content						%Field Capacity						% Germination					
	7 days		14 days		21 days		7 days		14 days		21 days		7 days		14 days		21 days	
	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2
T1	12.90±1.83	11.10±2.12	8.85±2.60	9.00±1.27	11.83±6.83	7.45±0.63	14.83±2.42	12.51±2.68	10.13±0.68	9.90±1.54	11.60±0.52	7.87±0.49	91.66±11.7	91.99±0.0	91.6±11.70	91.99±0.00	91.6±11.70	91.99±0.0
T2	13.40±3.11	12.70±0.35	9.20±0.56	14.15±0.64	12.55±2.19	8.25±1.48	16.14±1.33	14.48±0.46	10.63±1.38	17.63±3.42	12.14±6.03	9.39±3.29	100.0±0.0	100.±0	100.00±0.00	100.00±0.00	100.0±0.0	100.0±0.0
T3	12.87±2.36	11.00±1.41	5.96±0.49	5.31±5.37	10.40±0.42	5.66±1.57	14.78±3.07	12.500±1.78	9.05±3.61	7.52±4.89	6.64±9.29	7.81±2.49	83.33±0.00	83.33±0.0	83.33±0.0	83.33±0.0	83.33±0.0	83.33±0.0
T4	13.90±0.98	11.11±4.80	8.30±3.04	6.90±4.25	11.15±0.07	6.80±1.5	15.54±4.15	12.651±6.0	9.71±0.94	10.27±0.36	12.54±0.08	8.04±0.72	91.66±11.7	100.0±0	91.66±11.7	100.0±0	91.66±11.7	100.0±0.0
T5	16.05±1.20	15.75±1.62	9.60±1.132	14.95±2.47	13.00±2.82	8.55±2.76	18.71±2.29	18.71±2.29	12.68±0.45	17.98±1.28	15.00±3.74	11.24±1.40	100.0±0.0	100.0±0.0	100.0±0.0	100.0±0.0	100.0±0.0	100.0±0.0

V1= (Ta-Habib), V2= (Saher), T1= (2ppm Cr⁺³), T2= (2ppm Cr⁺³+HBA), T3= (4ppm Cr⁺³), T4= (4ppm Cr⁺³+HBA), T5= (Control)

Result after 21 days of germination shows that maximum shoot dry weight in cultivar Ta-Habib and Saher has been recorded in treatment T2 (2ppm Cr⁺³+HBA) with 0.58g and 0.44g followed by treatment T5 (control) with 0.2g and 0.25g, while lowest shoot dry weight in both varieties has been recorded in treatment T3 (4ppm Cr⁺³) with 0.14g and 0.17g respectively, showing that Hydroxyl benzoic acid (HBA) can increase shoot dry weight of the selected varieties under the effect of heavy metal chromium. Result regarding to root length shows in (Table 3), result after 7 days shows that the maximum root length in both varieties (Ta-Habib and Saher) has been recorded in treatment T5 (control) with 5.60cm and 6.65cm, whereas the smallest values of root length has been reported in treatment T3 (4ppm Cr⁺³) with 2.75cm and 4.15cm respectively. Result showing greatest root

length in Ta-Habib and Saher after 14 days in treatment T5 (control) with 6.30cm and 5.25cm followed by T2 (2ppm Cr⁺³+HBA) with 6.0cm and 4.65cm, after 21 days greatest root length been recorded in T5 (control) with 5.80cm and 8.20cm followed by T2 (2ppm Cr⁺³+HBA) with 5.65cm and 5.05cm respectively, while the smallest root length has been reported in both wheat varieties (Ta-Habib and Saher) in T3 (4ppm Cr⁺³) after 14 days with 3.70cm and 0.07cm, and after 21 days with 4.20cm and 3.80cm. Result shows in (Table 3) that the maximum root fresh weight in wheat (*Triticum aestivum* L.) varieties (Ta-Habib and Saher) has been recorded in treatment T5 (control) after 7 days with similar value 0.08g and 0.08g, after 14 days with root fresh weight, 0.44g and 0.10g and after 21 days with maximum root fresh weight 0.46g and 0.18g respectively. While minimum root fresh

weight has been reported in both wheat varieties (Ta-Habib and Saher) in treatment T3 (4ppm Cr^{+3}) after 7 days with 0.05g and 0.04g, after 14 days with 0.07 and 0.05g followed by treatment T1 (2ppm Cr^{+3}) with 0.09 and 0.06g respectively, whereas after 21 days minimum root fresh weight has also been reported in both wheat varieties (Ta-

Habib and Saher) in treatment T3 (4ppm Cr^{+3}) with 0.14g and 0.09g, showing that Hydroxyl benzoic acid has constructive role in augmentation of root length and fresh weight under the effect heavy metal chromium, while without spray treatments showing that Cr^{+3} has negative effect on root.

Table 2. Effect of Hydroxyl benzoic acid (HBA) foliar spray on shoot length, shoot fresh weight and shoot dry weight of *Triticum aestivum* L. (Wheat) under various concentrations of chromium

Treatments							Shoot fresh weight						Shoot dry weight					
	7 days		14 days		21 days		7 days		14 days		21 days		7 days		14 days		21 days	
	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2
T1	14.90±0.70	12.15±3.18	16.80±0.70	18.75±3.33	24.0±2.54	25.15±1.06	0.19±0.0	0.18±0.0	0.26±0.05	0.34±0.02	0.45±0.05	0.69±0.22	0.06±0.0	0.04±0.03	0.08±0.04	0.11±0.02	0.16±0.02	0.21±0.0
T2	15.05±2.05	13.55±1.06	17.05±2.75	19.40±2.12	24.25±3.32	28.30±9.61	0.23±0.0	0.19±0.0	0.34±0.02	0.40±0.0	0.86±0.21	0.81±0.0	0.04±0.0	0.04±0.0	0.12±0.0	0.13±0.03	0.58±0.06	0.44±0.29
T3	12.91±2.47	10.80±0.70	14.05±1.13	15.60±1.69	21.30±2.12	24.60±15.9	0.18±0.0	0.16±0.0	0.18±0.00	0.35±0.0	0.46±0.21	0.53±0.00	0.03±0.0	0.04±0.01	0.06±0.0	0.09±0.01	0.14±0.04	0.17±0.02
T4	14.95±2.61	11.85±4.31	14.90±1.55	18.50±2.54	23.20±3.67	27.75±2.82	0.20±0.0	0.17±0.0	0.34±0.07	0.37±0.04	0.57±0.40	0.74±0.22	0.04±0.0	0.04±0.02	0.08±0.01	0.11±0.01	0.19±0.0	0.17±0.05
T5	15.90±2.26	14.75±0.49	17.70±0.28	19.55±1.76	25.60±5.37	28.40±4.66	0.20±0.0	0.20±0.0	0.40±0.18	0.42±0.01	0.83±0.17	1.08±0.65	0.07±0.0	0.05±0.01	0.13±0.05	0.13±0.0	0.21±0.07	0.26±0.05

V1= (Ta-Habib), V2= (Saher), T1= (2ppm Cr^{+3}), T2= (2ppm Cr^{+3} +HBA), T3= (4ppm Cr^{+3}), T4= (4ppm Cr^{+3} +HBA), T5= (Control)

Result given in (Table 3) shows that maximum root dry weight in Ta-Habib and Saher has been recorded in treatment T5 (control) after 7 days with similar values 0.04g and 0.04g, after 14 days the maximum root dry weight in Ta-Habib and Saher were 0.05g and 0.03g, whereas after 21 days the maximum root dry weight has been recorded in treatment T2 (2ppm Cr^{+3} +HBA) with 0.05g followed by treatment T5

(control) with 0.04g in Ta-Habib, while that of Saher has been found in T5 (control) with 0.05g. While minimum root dry weight in both wheat varieties (Ta-Habib and Saher) has been recorded in treatment T3 (4ppm Cr^{+3}) after 7 days with similar values 0.01 and 0.01g, after 14 days with 0.02 and 0.01g and after 21 days with 0.03g and 0.02g, followed by T1 (2ppm Cr^{+3}) with 0.02g and 0.01g respectively.

Table 3. Effect of Hydroxyl benzoic acid (HBA) foliar spray on root length, root fresh weight and root dry weight of *Triticum aestivum* L. (Wheat) under various concentration of chromium

Treatments	Root length						Root fresh weight						Root dry weight					
	7 days		14 days		21 days		7 days		14 days		21 days		7 days		14 days		21 days	
	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2
T1	3.250±0.91 _a	4.30±0.35	4.40±0.14	4.10±1.41	4.80±0.5	5.40±1.69	0.06±0.01	0.06±0.01	0.09±0.02	0.06±0.0	0.09±0.01	0.10±0.05	0.02±0.0	0.02±0.0	0.03±0.0	0.02±0.0	0.02±0.01	0.01±0.0
T2	5.150±0.21 _c	5.600±0.28	6.00±1.55	4.65±2.19	5.65±1.48	5.05±1.62	0.07±0.0	0.06±0.0	0.15±0.01	0.07±0.01	0.21±0.03	0.14±0.11	0.03±0.0	0.02±0.0	0.03±0.0	0.01±0.0	0.05±0.02	0.04±0.02
T3	2.750±0.91 _a	4.15±0.35	4.35±0.70	3.70±1.55	4.20±1.41	3.80±1.55	0.05±0.0	0.04±0.03	0.07±0.01	0.05±0.0	0.14±0.03	0.09±0.03	0.01±0.01	0.01±0.0	0.02±0.0	0.01±0.0	0.03±0.0	0.02±0.01
T4	4.000±0.70 _b	4.40±2.12	5.80±0.84	4.12±1.23	5.20±0.70	4.25±1.62	0.06±0.03	0.04±0.02	0.11±0.06	0.09±0.0	0.20±0.14	0.13±0.0	0.03±0.03	0.02±0.0	0.038±0.01	0.01±0.12	0.04±0.0	0.03±0.02
T5	5.600±2.40 _d	6.65±1.06	6.30±1.69	5.25±0.91	5.80±0.98	8.20±0.56	0.08±0.01	0.08±0.0	0.44±0.20	0.10±0.03	0.46±0.32	0.18±0.14	0.04±0.0	0.04±0.0	0.05±0.03	0.03±0.0	0.04±0.0	0.05±0.0

V1= (Ta-Habib), V2= (Saher), T1= (2ppm Cr⁺³), T2= (2ppm Cr⁺³+HBA), T3= (4ppm Cr⁺³), T4= (4ppm Cr⁺³+HBA), T5= (Control)

Result shows in (Table 4) after 7, 14 and 21 days of treatments that the largest leaf length in cultivar Ta-Habib has been found in treatment T2 (2ppm Cr⁺³+HBA) ranges from 10.75cm, 11.15cm and 14.35cm while that of Saher has also been recorded in treatment T2 (2ppm Cr⁺³+HBA) with 10.0cm, 12.1cm and 15.85cm respectively. While smallest leaf length in Ta-Habib has been recorded with the above intervals in treatment T3 (4ppm Cr⁺³) ranges from 9.15cm, 8.20cm and 11.80cm, while that of cultivar Saher has also been recorded in treatment T3 (4ppm Cr⁺³) ranges from 7.75cm, 9.30cm and 12.55cm respectively. Result shows in (Table 4) that maximum fresh weight of leaf in cultivar Ta-Habib with intervals of 7 and 14 days has been recorded in treatment T2 (2ppm Cr⁺³+HBA) with 0.04g and 0.05g, after 21 days in T5 (control) with 0.14g, whereas in Saher

highest fresh weight of leaf has also been recorded after 7 and 14 days in treatment T2 (2ppm Cr⁺³+HBA) with 0.04g and 0.07g, after 21 days in T5 (control) with 0.14g, while minimum fresh weight of leaf has been reported in treatment T3 (4ppm Cr⁺³) in both varieties (Ta-Habib and Saher) after 7 days with 0.03g and 0.02g, after 14 days with 0.03g and 0.05g, after 21 days with 0.07g and 0.09g respectively, indicated that Hydroxyl benzoic acid (HBA) participate a constructive role in enhancement of leaf length and fresh weight under inoculation of heavy metal chromium. Result of leaf dry weight shows in (Table 4) after 7, 14 and 21 days of treatments that the highest dry weight of leaf has been recorded in treatment T3 (4ppm Cr⁺³) after 7 days in wheat variety (Ta-Habib) with 0.05g, while that of Saher were found in treatment T2 and T3 with 0.02g, after 14 days in T2 (2ppm

Cr⁺³+HBA) of Ta-Habib with 0.02g while that of Saher except T3 were found from treatment T1 to T5 with similar high values 0.02g, whereas after 21 days highest leaf dry weight recorded in treatment T2 (2ppm Cr⁺³+HBA) of Ta-Habib 0.04g and that of Saher in T5 (control) with 0.04g. While minimum leaf dry weight has been recorded in treatment T1, T2, and T4 after 7 days with 0.01g in Ta-Habib while that of Saher

in treatment T1, T4 and T5 with 0.01g, whereas after 14 days minimum leaf dry weight in both wheat varieties (Ta-Habib and Saher) founded in T3 (4ppm Cr⁺³) with similar value 0.01g and after 21 days minimum leaf dry weight has also been founded in T3 of Ta-Habib with 0.01g and that of Saher in T3 (4ppm Cr⁺³) and T4(4ppm Cr⁺³+HBA) with similar value 0.02g.

Table 4. Effect of Hydroxyl benzoic acid (HBA) foliar spray on leaf length, leaf fresh weight and leaf dry weight of *Triticum aestivum* L. (Wheat) under various concentrations of chromium

Treatments	Leaf length						Leaf fresh weight						Leaf dry weight					
	7 days		14 days		21 days		7 days		14 days		21 days		7 days		14 days		21 days	
	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2
T1	10.73±1.34	9.70±0.98	8.65±0.21	9.55±0.91	14.30±3.95	14.0±1.13	0.03±0.0	0.03±0.0	0.03±0.0	0.06±0.01	0.09±0.01	0.10±0.03	0.01±0.0	0.01±0.00	0.01±0.0	0.02±0.0	0.02±0.0	0.03±0.0
T2	10.75±4.73	10.0±0.8	11.15±0.21	12.10±3.25	14.35±4.17	15.85±2.05	0.04±0.0	0.04±0.0	0.05±0.02	0.07±0.0	0.10±0.02	0.13±0.1	0.01±0.0	0.02±0.0	0.02±0.0	0.02±0.0	0.04±0.01	0.03±0.01
T3	9.15±1.76	7.75±0.2	8.20±2.40	9.30±0.70	11.80±0.1	12.55±0.07	0.03±0.0	0.02±0.0	0.03±0.0	0.05±0.0	0.07±0.004	0.09±0.0	0.05±0.0	0.02±0.0	0.01±0.0	0.01±0.0	0.02±0.0	0.02±0.0
T4	10.65±1.76	8.25±3.04	8.95±0.21	11.60±0.21	12.50±0.56	12.60±0.28	0.03±0.0	0.03±0.0	0.05±0.02	0.06±0.0	0.10±0.01	0.12±0.02	0.01±0.0	0.01±0.0	0.01±0.0	0.02±0.0	0.03±0.0	0.02±0.0
T5	10.74±0.91	9.0±2.68	10.65±1.76	11.75±1.55	15.25±2.33	15.55±3.74	0.03±0.0	0.031±0.0	0.05±0.01	0.06±0.01	0.14±0.07	0.14±0.07	0.02±0.0	0.01±0.0	0.01±0.0	0.02±0.0	0.03±0.0	0.04±0.02

V1= (Ta-Habib), V2= (Saher), T1= (2ppm Cr⁺³), T2= (2ppm Cr⁺³+HBA), T3= (4ppm Cr⁺³), T4= (4ppm Cr⁺³+HBA), T5= (Control)

Result indicated in (Table 5) that highest number of leaf in Ta-Habib and Saher has been recorded in treatment T2 (2ppm Cr⁺³+HBA), after 7 days with 3.50 and 4.02, after 14 days with 5.50 and 6.00 and after 21 days maximum number of leaf has been found in treatment T5 (control) of both varieties with similar value i.e. 6.00, while smallest number of leaf in cultivar Ta-Habib and Saher has been recorded in treatment T3

(4ppm Cr⁺³) after 7 days with similar value 3.00, after 14 days 4.00 and 4.50 and after 21 days with similar value, 5.00 respectively in Ta-Habib and Saher, indicating that leaf numbers were reduced by chromium treatments while Hydroxyl benzoic acid (HBA) can play a helpful role in enhancement of leaf dry weight and leaf numbers under inoculation of heavy metal chromium. Result shows in (Table 5) that

the largest leaf width after 7 days of treatment in wheat variety Ta-Habib has been recorded in treatment T2 (2ppm Cr^{+3} +HBA) and T5 (control) with similar values 0.45cm and that of Saher also has been found in T2 (2ppm Cr^{+3} +HBA) followed by T5 (control) with 0.40 and 0.39cm, after 14 days in both varieties (Ta-Habib and Saher) leaf width recorded in T5 (control) with similar value 0.50cm, after 21

days in both varieties (Ta-Habib and Saher) leaf width has also been recorded in T5 (control) with similar value 0.75cm. While minimum leaf width has been recorded in the treatment T3 (4ppm Cr^{+3}) in both wheat varieties (Ta-Habib and Saher) after 7 days with 0.30cm and 0.35cm, after 14 days with similar value 4.00cm and after 21 days with minimum leaf width 0.60cm and 0.64cm respectively.

Table 5. Effect of Hydroxyl benzoic acid (HBA) foliar spray on leaf numbers, leaf width and vigor index of *Triticum aestivum* L. (Wheat) under various concentrations of chromium

Treatments	Leaf numbers						Leaf width						Vigor index					
	7 days		14 days		21 days		7 days		14 days		21 days		7 days		14 days		21 days	
	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2
T1	3.40±0.7	3.0±0.0	5.0±0.0	5.0±0.0	5.0±0.0	5.00±0.7	0.36±0.0	0.36±0.0	4.50±0.7	4.50±0.7	0.55±0.0	0.64±0.0	577.36±81.57	477.95±115.1	975.60±43.27	1149.26±500.04	1408.08±356.04	1496.16±122.18
T2	3.50±0.7	4.02±1.4	5.50±0.0	6.0±0.0	5.50±0.7	5.50±0.7	0.45±0.0	0.40±0.0	4.90±0.0	4.80±0.0	0.65±0.2	0.74±0.0	632.58±263.6	709.56±112.8	1097.55±463.08	1151.46±43.70	1966.68±561.55	1700.76±580.13
T3	3.±0.0	3.0±0.70	4.0±0.0	4.50±0.7	5.00±0.0	5.00±0.7	0.30±0.0	0.35±0.0	4.00±0.0	4.0±0.0	0.54±0.0	0.60±0.0	469.26±72.04	600.28±165.0	807.92±212.87	627.6±151.04	820.80±357.060	1000.14±3365.1
T4	3.40±0.7	4.01±0.0	5.50±1.4	5.00±0.0	5.50±0.7	5.00±0.7	0.35±0.0	0.35±0.0	4.70±0.0	4.51±0.7	0.60±0.1	0.64±0.0	772.70±166.4	603.90±230.6	987.30±205.77	999.18±136.19	1338.10±552.25	16087.86±257.7
T5	3.40±0.7	4.0±0.0	5.0±0.0	4.50±0.7	6.00±0.0	6.00±0.0	0.45±0.0	0.39±0.0	5.00±0.0	5.0±0.0	0.75±0.0	0.75±0.0	885.96±233.	1173.9±148.2	1111.68±456.17	1873.04±713.73	1346.88±191.08	2328.60±392.02

V1= (Ta-Habib), V2= (Saher), T1= (2ppm Cr^{+3}), T2= (2ppm Cr^{+3} +HBA), T3= (4ppm Cr^{+3}), T4= (4ppm Cr^{+3} +HBA), T5= (Control)

Result indicated in (Table 5) that highest value of vigor index in cultivar Ta-Habib after 7, 14 and 21 days has been recorded in treatment T5 (control) with 885.96, 1111.68

and 1966.68, followed by T4 (4ppm Cr^{+3} +HBA) with 772.70, 1097.55, 1346.88, whereas with same intervals highest value of vigor index in Saher has also been

recorded in treatment T5 (control) with 1173.90, 1873.04 and 2328.60, followed by T2 (2ppm Cr^{+3} +HBA) with 709.56, 1151.46 and 1700.76 respectively. While smallest value of vigor index in both wheat varieties (Ta-Habib and Saher) has been reported in treatment T3 (4ppm Cr^{+3}) after 7 days with 469.26 and 600.28, after 14 days with 807.92 and 627.60 and after 21 days with 820.80 and 1000.14, respectively, showing that Hydroxyl benzoic acid can improve vigor index in the selected varieties under heavy metal chromium treatments. Result shows in (Table 6) after 7, 14 and 21 days of treatments that highest concentration of chromium in rhizospheric soil of Ta-Habib has been recorded in the treatment T4 (4ppm Cr^{+3} +HBA) with 2.17ppm, 2.09ppm and 2.66ppm followed by T3 (4ppm Cr^{+3}) with 2.10ppm, 2.07ppm and 2.63ppm, whereas that of Saher has also found in T4 (4ppm Cr^{+3} +HBA) with chromium concentrations 2.12ppm, 2.13ppm and 2.55ppm followed by T3 (4ppm Cr^{+3}) with 2.04ppm, 2.10ppm and 2.08ppm respectively. While minimum chromium concentration in rhizospheric soil in both wheat varieties (Ta-Habib and Saher) has been founded in treatment T5 (control) after 7 days with 1.97 and 1.82 ppm after 14 days with 1.93ppm and 2.03ppm and after 21 days 2.48pp and 2.45ppm followed by T1 (2ppm Cr^{+3}) after 7 days with 2.02ppm and 1.8ppm after 14 days with 2.01ppm and 2.03ppm, after 21 days with 2.02ppm and 2.07ppm respectively. Result in (Table 6) after 7, 14 and 21 days of treatments that highest concentration of chromium in whole plants both wheat (*Triticum aestivum* L.) varieties (Ta-Habib and Saher) recorded in treatment T4 (4ppm Cr^{+3} +HBA), after 7 days with 2.60 and 2.40ppm, after 14 days with 2.67 and 2.61ppm and after 21 days with 2.71 and 2.72ppm respectively, while minimum concentrations of chromium in whole plants has been recorded in treatment T5 (control) after 7 days with 2.43 and 2.29ppm, after 14 days with 2.48 and 2.48ppm and after 21 days with 2.59 and 2.64ppm followed by T1 (2ppm Cr^{+3}) after 7 days with 2.47 and

2.29ppm, after 14 days with 2.49 and 2.50ppm and after 21 days with 2.60 and 2.65ppm respectively, showing that Hydroxyl benzoic acid can help to speedup accumulation of heavy metal chromium in plant.

Experiment held for the present study to revealed the effect of hydroxyl benzoic acid foliar spray under various concentrations of chromium (Cr^{+3}) on physiological and agronomic characteristics of *Triticum aestivum* L. (wheat) including two varieties Ta-Habib and Saher, for which three collections of agronomic & physicochemical characters with intervals of 7 days, 14 days and 21 days of chromium treatment were collected results revealed that the Hydroxyl benzoic acid has been increased the moisture content and percent field capacity while high concentrations of chromium reduced soil percent moisture content and field capacity in both wheat varieties (Ta-Habib and Sahar) significantly as shown in (Table 1). Shoot length has been reduced in the treatments which were treated with chromium in different concentrations i.e., 2ppm and 4ppm in all of the treatments, while hydroxyl benzoic acid foliar spray were found to improve the shoot characters even at high concentrations of chromium (4ppm) given in (Table 2). Similary Pati *et al.*, [24] reported that the effect of chromium stress on plants exhibited a gradual decline in the growth, total chlorophyll content and protein content along with enhanced proline content with increasing levels of chromium. Shoot weights were also illustrated in (Table 2) showed significantly reduction along the concentration gradient in cultivar Ta-Habib and Saher. Highest values of shoot weight were recorded in T5 (untreated control) followed by spray treatments T2 and T4 in comparison to other treatments such as T1 and T3 in both varieties. Saif *et al.*, [15] investigated that chromium on growth and biochemical parameters of *Triticum aestivum*, caused a significant decrease.

Experimental work revealed that the root length after 7 days, 14 days and 21 days was adversely affected by chromium in treatments T1 (2ppm Cr⁺³) and T3 (4ppm Cr⁺³) compared to spray treatments i.e., T2 (2ppm Cr⁺³+HBA) and T4 (4ppm Cr⁺³+HBA) while maximum root length in both varieties were recorded in T5 (untreated control) and minimum root length were reported in T3 (4ppm Cr⁺³) as described in (Table 3). Similar results have been reported previously Jamal *et al.* [25] and Gang *et al.* [28] where high Cr⁺³ concentrations significantly reduced root growth in crop plants. Root weight has been increased in control treatments, for example T5 (untreated Control) has maximum root weight (Table 3). Root weight get decline

along the concentration gradient of heavy metal chromium, in both varieties. The best treatments for both varieties were treatment T5 (untreated control) at each intervals. Highest root weight was recorded in cultivar Ta-Habib compared to cultivar Saher it means that cultivar Saher was more affected compared to cultivar Ta-Habib. Shanker *et al.*, [29] suggested the reason of the high accumulation in roots of the plants could be because Cr⁺³ is immobilized in the vacuoles of the root cells, thus rendering it less toxic, which may be a natural toxicity response of the plant. Das *et al.*, [22] reported that Cr⁺³ leads to decrease of root growth and biomass, chlorosis, photosynthetic impairment and finally plant death.

Table 6. Analysis of chromium in rhizospheric soil and in wheat plants under various concentrations of heavy metal chromium

Treatments	Rhizospheric Soil						Whole Plant					
	7 days		14 days		21 days		7 days		14 days		21 days	
	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2	V1	V2
T1	2.02±0.06	1.89±0.04	2.01±0.04	2.03±0.04	2.02±0.02	2.07±0.06	2.47±0.04	2.29±0.05	2.49±0.08	2.50±0.09	2.60±0.06	2.65±0.01
T2	2.05±0.0	1.98±0.07	2.02±0.09	2.04±0.09	2.65±0.02	2.08±0.10	2.47±0.05	2.30±0.0	2.58±0.11	2.53±0.09	2.70±0.07	2.70±0.03
T3	2.10±0.0	2.04±0.07	2.07±0.07	2.10±0.06	2.63±0.11	2.54±0.08	2.52±0.10	2.33±0.042	2.58±0.03	2.54±0.08	2.63±0.10	2.71±0.02
T4	2.17±0.137	2.12±0.12	2.09±0.10	2.13±0.09	2.66±0.04	2.55±0.04	2.60±0.05	2.40±0.09	2.67±0.08	2.61±0.03	2.71±0.03	2.72±0.07
T5	1.97±0.0	1.82±0.07	1.93±0.08	2.03±0.06	2.48±0.01	2.45±0.05	2.43±0.11	2.29±0.01	2.48±0.01	2.48±0.07	2.59±0.02	2.64±0.05

V1= (Ta-Habib), V2= (Saher), T1= (2ppm Cr⁺³), T2= (2ppm Cr⁺³+HBA), T3= (4ppm Cr⁺³), T4= (4ppm Cr⁺³+HBA), T5= (Control)

The present study revealed that the leaf length has been decreased with chromium even at 2ppm concentration in all of the three treatments in both varieties (Ta-Habib and Saher) (Table 4). Results showed that

leaf weight also has been reduced with chromium, relatively the leaf weight has been lowered with chromium even at low concentration indicated that significantly chromium decreased the weight of leaf

(Table 4). The best treatments for both varieties were T5 (untreated control). In comparison to cultivar Saher highest values of leaf length and weight were recorded in cultivar Ta-Habib. However with increase in Cr^{+3} concentrations gradual decline were recorded in all growth parameters (Table 4). Mady, [18] indicated that, an applied treatment of foliar spray significantly increased all growth parameters as number of branches and leaves per plant, leaf area per plant and leaves dry weight. Result showed that number of leaves and leaf width decreases with increase in chromium concentration while hydroxyl benzoic foliar spray showing positive role in this category (Table 5). Result of vigor index were given in (Table 5) showing that highest vigor index were recorded in treatment T5 (untreated control) while reduce vigor index were noted in treatment T3 (4ppm Cr^{+3}) compared to other treatments, whereas hydroxyl benzoic spray have role to increase seedling vigor index in both varieties of *Triticum aestivum* L. (wheat) including Ta-Habib and Saher (Table 5). Saddiqe *et al.* [15] reported that seedling vigor index showing a significant decrease in seedling vigor due to the presence of metal. Result regarding to chromium concentrations in rhizospheric soil and whole plant of wheat (*Triticum aestivum* L.) varieties (Ta-Habib and Saher) that Cr^{+3} concentrations in wheat plant and rhizospheric soil significantly increased along the concentration gradient, highest chromium concentration was found in treatment T4 followed by T3 and T2 in both varieties of *Triticum aestivum* L. (wheat) while minimum Cr^{+3} concentrations in rhizospheric soil and whole plant were founded in T5 (untreated control) after 7 days, after 14 days and after 21 days (Table 6). Similarly, Rafique *et al.* [10] suggested that heavy metals accumulate in ecological food chain through uptake at primary producer (plants) level and then through consumption at consumer levels.

Conclusion

The study was aimed to evaluate the effect of heavy metal (Cr^{+3}) on wheat (*Triticum*

aestivum L.) in response to Hydroxyl benzoic acid (HBA) foliar spray. On the basis of agronomic and physicochemical characteristics it has been concluded that high concentration of chromium adversely affected on both the selected wheat (*Triticum aestivum* L.) varieties (Ta-Habib and Saher). It has also been concluded that exogenous application of Hydroxyl benzoic acid can reduce the toxic effect of chromium, indicating that Hydroxyl benzoic acid can acts as bio-regulator against the phototoxic effect of chromium. From agronomic and physicochemical characteristics it was known that cultivar Ta-Habib up to some extent show tolerances against toxic effect of chromium compared to Saher. In the present study it is shows that 2ppm Cr^{+3} concentration has negative effects on growth parameters while 4ppm Cr^{+3} concentration is more toxic and adversely affects the agronomic and physicochemical characteristics of the selected varieties of wheat (*Triticum aestivum* L.). Results of the experiments explicitly showed that chromium is potentially toxic in different concentrations at different phases of plant growth and development; moreover, the toxicity of chromium in plants depends on concentration and varies in different species.

Authors' contributions

Conceived and designed the experiments: S Ullah, Performed the experiments: M Khan & M Idrees, Analyzed the data: M Khan & S Ali, Contributed materials/ analysis/ tools: S Ali & MN Khan, Wrote the paper: M Khan & MN Khan, Proof reading: M Adnan & FA Ozdemir.

References

1. Pandey SK & Pandey SK (2008). Germination and seed- ling growth of field pea *Pisum sativum* Malviya matar-15 (HUDP-15) and Pusa prabhat (DDR-23) under varying level of copper and chromium. *Ame J Sci* 4(3): 28-40.
2. Paksoy M & Acar B (2009). Effect of organic fertilizers on yield components

- of some tomato cultivars. *Asian J Chem* 21(8): 6041- 6047.
3. Nazir R, Khan M, Masab M, Rehman HU, Rauf NU, Shahab S, Ameer N, Sajed M, Ullah M, Rafeeq M & Shaheen Z (2015). Accumulation of heavy metals (Ni, Cu, Cd, Cr, Pb, Zn, Fe) in the soil, water and plants and analysis of physicochemical parameters of soil and water. *J Pharm Sci & Res* 7(3): 89-97.
4. Dan T, Johnson BHD, Conard B, Stiebe & Veska E (2008). Toxicity thresholds for oat (*Avena sativa* L.) grown in niimpacted agricultural soils near Port Colborne, Ontario, Canada. *Can J Soil Sci* 88: 389-398.
5. Fernanda A, Quinteros AHB, Poliandri LI, Machiavelli JP, Cabilla, Wei BH, Xi ZB, Zhao Y, Wang S, Liu E & Jiang Y (2007). Effect of inoculating microbes in municipal solid waste composting on characteristics of humic acid. *Chemosphere* 68: 368-374.
6. Akinci IE & Akinci S (2010). Effect of chromium toxicity on germination and early seedling growth in melon (*Cucumis melo* L.). *Afr J Biotech* 9: 4589-4594.
7. Scoccianti V, Crinelli R, Tirillini B, Mancinelli V & Speranza A (2006). Uptake and toxicity of Cr (Cr³⁺) in celery seedlings. *Chemosphere* 64: 1695-1703.
8. Saif MS, Haq MU & Memon KS (2005). Heavy metals contamination through industrial effluent to irrigation water and soil in Korangi area of Karachi (Pakistan). *Int J Agric Biol* 7(4): 646-648.
9. Latif MI, Lone MI & Khan KS (2008). Heavy metals contamination of different water sources, soils and vegetables in Rawalpindi area. *Soil & Environ* 27(1): 29-35.
10. Rafique U, Ashraf A, Khan AK, Nasreen S, Rashid R & Mahmood Q (2010). Toxic chromium from tanneries pollutes water resources and soils of Pakistan. *J Chem Soc Pak* 32: 644-649.
11. Hussain SI, Ghafoor A, Ahmad S, Murtaza G & Sabir M (2006). Irrigation of crops with raw sewage: hazard and assessment in effluent, soil and vegetables. *Pak J Agric Sci* 43(3-4): 97-101.
12. Khan MJ, Bhatti AU, Hussain S & Ullah W (2007). Heavy metal contamination of soil and vegetables with industrial effluents from sugar mills and tanneries. *Soil Environ* 26(2): 139-145.
13. Ali S, Farooq MA, Yasmeen T, Hussain S, Arif MS, Abbas F, Bharwana SA, & Zhang G (2013). The influence of silicon on barley growth photosynthesis and ultra-structure under chromium stress. *Ecotoxic. Environ Safety J Homep* 89: 66-72.
14. Khan MY, Asghar HN, Jamshaid MU, Akhtar MJ & Zahir AZ (2013). Effect of microbial inoculation on wheat growth and phytostabilization of chromium contaminated soil. *Pak J Bot* 45(S1): 27-34.
15. Saddiqe Z, Farooq A, Khan F, Khalid H & Javeria S (2015). Effect of chromium (VI) on physical growth and biochemical parameters of wheat (*Triticum aestivum* L.) seedlings. *Biologia* 61(2): 219-226.
16. Hayyat MU, Mahmood R, Akram S, Haider Z, Rizwan ST, Siddiq Z & Nawaz R (2015). Use of alleviators to reduce toxicity of chromium in germination of common agricultural crops. *J of Environ & Ecol Res* 3(2): 44-49.
17. Hamid R, Parray JA, Kamili AN & Zafar MU (2012). Chromium stress in *Brassica juncea* L. cv. 'Pusa Jai Kissan' under hydroponic culture. *Afr J of Biotech* 11 (90): 15658-15663.
18. Mady MA (2009). Effect of foliar application with salicylic acid and vitamin e on growth and productivity of tomato (*Lycopersicon esculentum* Mill.) plant. *J Agric Sci Mansoura Uni* 34(6): 6735-6746.

19. Yastreb TO, Kolupaev YY & Vayner AO (2012). Induction of heat resistance in wheat coleoptiles by 4-hydroxybenzoic acid: connection with the generation of reactive oxygen species. *J Stress Physio & Bioche* 8(3): 72-81.
20. Amin H, Arain BA, Amin F & Surhio MA (2013). Phytotoxicity of chromium on germination, growth and biochemical attributes of *Hibiscus esculentus* L. *Ameri J P Sci* 4: 431-2439.
21. Oliveira H (2012). Chromium as an environmental pollutant: insights on induced plant toxicity. *J Bot* 10: 1155-11560.
22. Das BC, Panda P, Sahoo PK, Jena S & Padhi P (2014) Effect of chromium (VI) on wheat seedlings and the role of chelating agents. *Curr Sci* 106(10): 1387-1393.
23. Allen SE, Grimshaw HM, Parkinson JA & Quarmby C (1974). Chemical analysis of ecological materials. Blackwell Scientific Publications.
24. Pati S, Ghadei A, Arzoo A, Nayak SK, Mohapatra A & Satapathy KB (2014). Physiological responses induced by chromium toxicity to *Cucumis sativus* L. and *Macrotyloma uniflorum* Lam. *IOSR- J of Environ Sci Toxic & Food Tech* 8(12): 58-63.
25. Jamal SN, Iqbal MZ & Athar M (2006). Phytotoxic effect of aluminum and chromium on the germination and early growth of wheat (*Triticum aestivum*) varieties Anmol and Kiran *Int J Env Sci Tech* 3: 411-416.
26. Dey SK, Jena PP & Kundu S (2009). Antioxidative efficiency of *Triticum aestivum* L. exposed to chromium stress. *J Environ Biol* 30: 539-544.
27. Datta JK, Bandhyopadhyay A, Banerjee A & Mondal NK (2011). Phytotoxic effect of chromium on the germination, seedling growth of some wheat (*Triticum aestivum* L.) cultivars under laboratory condition. *J Agric Tech* 7: 395-402.
28. Gang A, Vyas A & Vyas H (2013). Toxic effect of heavy metals on germination and seedling growth of wheat. *J Environ & Dev* 8: 206-213.
29. Shanker AK, Djanaguiraman M, Sudhagar R, Chandrashekar CN & Pathmanabhan G (2004). Differential anti-oxidative response of ascorbate glutathione pathway enzymes and metabolites to chromium speciation stress in green gram (*Vigna radiata* (L) R Wilczek, cv CO4) roots. *Plant Sci* 166:1035-1043.