RESEARCH ARTICLE

Changes of Photosynthetic Parameters in *Jatropha curcas* L. Leaves under Cobalt Stress

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Abstract

In the present investigation, changes of photosynthetic parameters including net photosynthetic rate (P_N); stomatal conductance (*gs*), transpiration rate (*E*), and water-use efficiency (WUE) were studied in Jatropha (*Jatropha curcas* cv. DARL-2) under different concentrations of cobalt (Co) (0, 100, 200, 400, 600, 800, and 1000 mg kg^{-1}) toxicity. The P_N, *gs*, and *E* trends were shown in increasing trends up to 200 mg kg^{-1} Co in comparison to controlled plants. However, trends of WUE and photosynthetic pigments [chlorophyll *a*, *b*, (*a*+*b*), and carotenoids] were found in decreasing order in all concentrations of cobalt stress as compared to controlled plants. The P_N value was ~1.33, and ~2.44 fold higher in 100 and 200 mg kg^{-1} but decreased ~2.61, ~6.86, and ~15.40 fold in 400, 600, and 800 mg kg^{-1} cobalt treatment, respectively. Similarly, the *gs* rate was ~1.33, and ~1.69 fold higher in 100 and 200 mg kg^{-1} but decreased ~1.89, ~3.38, and ~8.71 fold in 400, 600, and 800 mg kg^{-1} cobalt treatment. The *E* was significantly ~1.44, ~2.74, and ~1.35 fold higher in 100, 200, and 400 mg kg^{-1} decreased ~1.03 and ~1.43 fold in 600 and 800 mg kg^{-1}. The WUE was ~0.93, ~0.89, ~3.50, ~6.51, and ~10.76 fold decreased in all Co treated plants. In addition, the content of photosynthetic pigments [chlorophyll *a*, *b*, (*a*+*b*), and carotenoids] was varying in all Co treated plants. All studied plants were survived morphologically up to 800 mg kg^{-1} but seedlings were not survived due to severe cobalt toxicity stress in 1000 mg kg^{-1}. Among studied plants, Jatropha seedlings showed the best survival potential under 200 mg kg^{-1} Co stress.

Key words: Gas exchange, Net photosynthetic rate, Stomatal conductance, Transpiration rate, Water use efficiency, Photosynthetic pigments.

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Introduction

Autotrophs including photosynthetic bacteria conduct photosynthesis (Taiz and Zeiger, 2010; Pan *et al*., 2012). Combinations of different physiochemical processes manage the plant growth among which photosynthesis is an important biological interaction which play an essential role in global carbon cycle. Organism conducting photosynthesis always occupies premier trophic level in the food chain (Ashraf and Harris, 2013; Croce and Amerongen, 2014). Photosynthetic activity of plants is greatly affected by heavy metals as they affect in a multidirectional manner Krupa *et al*., 1993).

Heavy metal toxicity alters the pathway of many biological molecules by displacing the essential functional groups (Collins and Stotzky, 1989). However, lower concentration of some heavy metals like cobalt, zinc, nickel and chromium, and copper are reported essential for growth but when their concentration level increases, they adversely affect the plant growth and causes metal toxicity but toxic at higher concentration levels (Garbisu and Alkorta, 2001).

Heavy metal toxicity can also effects plant growth indirectly by altering the structure of chloroplast, decreasing the protein and lipid composition of thylakoids damaging the photosynthetic apparatus (Skórzyńska-Polit and Baszyński, 1997). These toxic metals affect two distinct parameters which are crucial for both photosynthetic activity and plant growth, *i.e*., water relations and ionic relations. During the exposure to metals the water status of leaves (water potential, transpiration rate, and relative water content) is distorted, resulting in reduction of leaf expansion (Poschenrieder and Barceló, 1999). Furthermore, disturbances in the uptake of ions and in their tissue distribution can lead to premature senescence of adult leaves, and in consequence to the reduction of the total photosynthetic area (Krupa *et al*., 1993). Additionally, changes of these parameters depend on the severity and duration of metal stress and on plant species.

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Although the effects of different heavy metals on the separate elements, such as the light or dark phase of photosynthesis, chlorophyll (Chl) content, or water relations, were intensively examined in the past, but there is still lack of complex and parallel analysis of cobalt stress on those parameters. Therefore, we simultaneously examined net photosynthetic rate, stomatal conductance, transpiration rate, and water use efficiency under different concentrations of cobalt stress on *Jatropha curcas* L. Additionally, the photosynthetic pigment synthesis as chlorophyll *a*, chlorophyll *b*, chlorophyll *a* & *b*, and carotenoids of *Jatropha curcas* L. after cobalt treatment was measured to estimate whether the negative action of heavy metals on photosynthetic apparatus was the main reason for plant growth reduction.

Materials and Methods

Plant material and Co concentrations

The *Jatropha curcas* L. plants were cultivated at open fields (Fig. 1) at Defence Institute of Bio-Energy Research, Haldwani (29.22°N and 79.52°E; 424 m asl). Seeds of Jatropha strain DARL-2 were cleaned by washing with tap water for multiple times, and soaked overnight in a solution (0.1 %, w/v) of carbendazim based broad spectrum systemic fungicide. Seeds were then allowed to dry in shed for 24 h. The dried seeds were then established on moistened filter paper placed in glass plates (Fig. 2) for possible germination at room temperature. After germination, seedlings of uniform size were selected and transplanted into pots containing autoclaved mixture of sand and soil in 1:1 ratio. Cobalt as $CoCl_2.6H_2O$ (Himedia) solution was added in the pots to obtain the cobalt concentrations of 0 (control), 100, 200, 400, 600, 800, and 1000 mg kg^{-1} soil (Fig. 3). Experiment was conducted with ten replicates of each treatment and all replication pot had three seedlings. Both control and treated pots were watered with deionized water at regular interval. All experimental pots were conducted under aseptic condition equipped with cold fluorescent light (350 µmol m^{-2} s^{-1}) with adjusted photoperiod of 12/12 h (day/night) at 500 mg l^{-1} CO_2 and 70±2% RH.

Gas-exchange measurements

All Gas exchange measurements [Photosynthetic rate (P_N), transpiration rate (E), water use efficiency (WUE) and stomatal conductance (gs)] were measured after 30 days adaptation of the plants under various cobalt concentration stress, separately, by using a portable leaf chamber analyser *(LCA-4, ADC Bio Scientific Ltd.*, Hoddesdon, UK). P_N, E, and gs were measured on uppermost, fully expanded, terminal leaflets (abaxial surface) at 11:00 h. Water-use efficiency (WUE) was calculated as ratio between photosynthetic rate (P_N) and transpiration rate (E) (A/E, mol CO_2/mol H_2O).

Estimation of total chlorophyll and carotenoids content

Chlorophyll (*a*, *b* and total *a*+*b*) and total carotenoids (Car) contents were determined from fresh leaf samples (FM). Fresh leaf discs were placed in a test tube containing 10 ml of N, N-dimethyl formamide (N-DMF) and stored for 24 h at 4°C. The absorbance of the coloured supernatant was read at 480, 647 and 666 nm in a monochromator base multimode detector (*BioTek*, *Snergy 2*, USA) with DMF as a blank. The contents of chlorophyll *a*, *b* and total chlorophyll (*a*+*b*) were calculated according to the method of Moran and Porath (1980).

Statistical analysis

All physiological measurements were carried out in triplicates by using three independent plants for each treatment. CropStat program developed at IRRI, Philippines was used for analysis of variance (ANOVA) of experiments. The treatment means were compared by least significant difference (LSD) test at a significance level of P£0.05.

Fig. 1: Open field *Jatropha curcas* L. (DARL-2) cultivation at the farm-yard of Defence Institute of Bio-Energy Research, Haldwani.

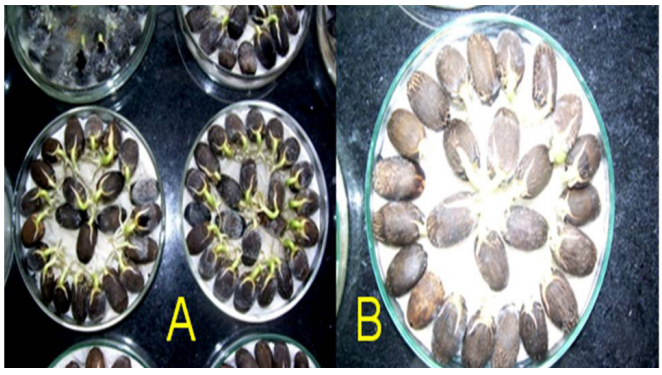

Fig. 2A-B: Germinated seeds of *Jatropha curcas* L. strain DARL-2 on moistened filter papers.

Results

As a result of increasing human activities for the fulfillment of their own necessity, toxic metal pollution has become one of the most serious environmental problems today. However, some metals like cobalt is considered a beneficial element for higher plants in spite of the absence of evidence for direct role in their metabolism (Young, 1979). But, after a permissible limit, toxic metals can act in a harmful manner by disturbing essential functional groups, displacing other metal ions, or modifying the active pathway of biological molecules (Collins and Stotzky, 1989).

Reports revealed that each plant species has its unique balanced nutrition requirements; thus, an optimum limit of nutrition or metal for one plant species is might be stressful for another one. In general, photosynthesis can active smoothly without any harm under a permissible metal limit (Ashraf and Harris, 2013).

Therefore, effect of different concentrations of cobalt stress was performed with leaves of Jatropha growing at 0 (control),

Fig. 3: Showing effect of different concentration [0 (control, 100, 200, 400, 600, 800, and 1000 mg kg^{-1} soil] of cobalt stress on *Jatropha curcas* L. strain DARL-2.

100, 200, 400, 600, 800, and 1000 mg kg^{-1} soil of cobalt. In the case of net photosynthetic rate (P_N), stomatal conductance (*gs*), and transpiration rate (*E*), trends were shown in an upgrade manner up to 200 mg kg^{-1} soil cobalt treatment in comparison to controlled plants (Fig. 4A-C). However, trends of water use efficiency (WUE) and photosynthetic pigments as Chl a, Chl b, and carotenoids were found in decreasing order in all concentrations of cobalt stress as compared to controlled plants (Table 1, Fig. 4D).

In our case, all studied plants were survived morphologically up to 800 mg kg^{-1} soil of cobalt, but when the cobalt concentration was increased up to 1000 mg kg^{-1} soil of cobalt, seedling were not survived due to severe cobalt toxicity stress (Fig. 2). Among studied plants, cobalt treatment with 200 mg kg^{-1} soil showed the best survival potential (Fig. 2).

Effect of cobalt stress on gas-exchange measurements

Under Co stress, P_N increased significantly up to 200 mg kg^{-1} soil in comparisons to controlled plants (Fig. 4A). It was suddenly decreased from 400 to 800 mg kg^{-1} in comparison to controlled plants (Fig. 4A). Among all studied cobalt treatments, 200 mg kg^{-1} showed maximum P_N of 10.21 µmol (CO_2) $m^{-2}s^{-1}$ followed by 100, 0, 400, 600, and 800 mg kg^{-1} with P_N value of 5.55, 4.19, 1.60, 0.60, and 0.27 µmol (CO_2) $m^{-2}s^{-1}$, respectively (Fig. 4A). The P_N value was ~1.33, and ~2.44 fold higher in 100 and 200 mg kg^{-1} cobalt treatment, respectively whereas, it was decreased ~2.61, ~6.86, and ~15.40 fold in 400, 600, and 800 mg kg^{-1} cobalt treatment, respectively, as compared to controlled plants (Fig. 4A). It was maximum in 200 mg kg^{-1} and minimum in 800 mg kg^{-1} of cobalt treatment with P_N value of 10.21 and 0.27 µmol (CO_2) $m^{-2}s^{-1}$, respectively, as compared to other studied plants (Fig. 4A).

The *gs* was also strongly increased up to 200 mg kg^{-1} soil in comparison to controlled plants (Fig. 4B). It was again decreased drastically from 400 mg kg^{-1} to 800 mg kg^{-1} in comparison to controlled plants (Fig. 4B). Among all studied cobalt treatments, the plants which treated with 200 mg kg^{-1} showed maximum stomatal conductance rate of 0.114 mol (H_2O) $m^{-2}s^{-1}$ followed by 100, 0, 400, 600, and 800 mg kg^{-1} with stomatal conductance value of 0.09, 0.07, 0.04, 0.02 and 0.27 mol (H_2O) $m^{-2}s^{-1}$, respectively (Fig. 4B). The *gs* rate was ~1.33, and ~1.69 fold higher in 100 and 200 mg kg^{-1} cobalt treatment, respectively, whereas, it was decreased ~1.89, ~3.38, and ~8.71 fold in 400, 600, and 800 mg kg^{-1} cobalt treatment as compared to controlled plants.

E for control plants was about 0.26 mmol (H_2O) $m^{-2}s^{-1}$, whereas, it was ~0.38, ~0.71, ~0.35, ~0.25, 0.18 mmol (H_2O) $m^{-2}s^{-1}$ in the cobalt treatment of 100, 200, 400, 600, and 800 mg kg^{-1} soil, respectively (Fig. 4C). *E* value was maximum in 200 mg kg^{-1} and minimum in 800 mg kg^{-1} cobalt treatment. It was significantly ~1.44, ~2.74, and ~1.35 fold higher in 100, 200, and 400 mg kg^{-1} cobalt treatment but after 400 mg kg^{-1}, *E* value was ~1.03 and ~1.43 fold decreased in 600 and 800 mg kg^{-1} treatment as compared to controlled plants (Fig. 4C).

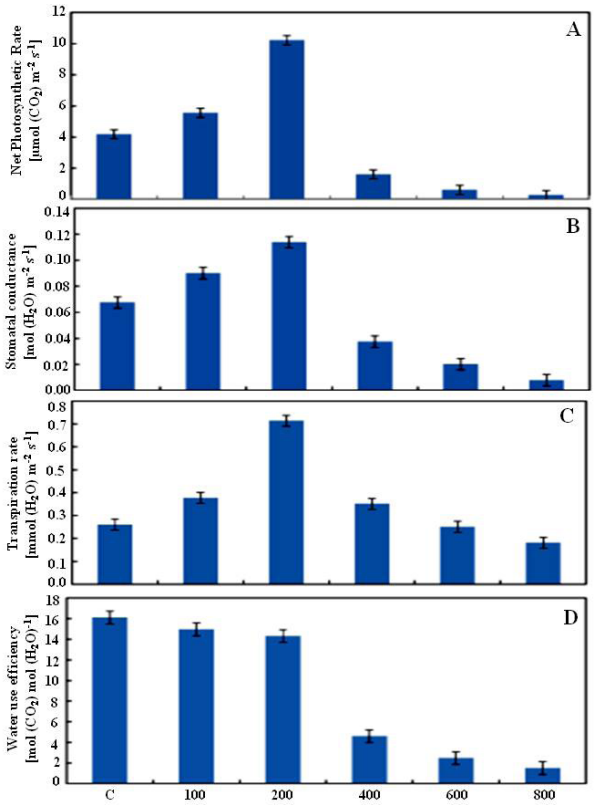

Fig. 4A-D: Measurement of gas exchange parameters [A. Photosynthetic rate (P_N), B. Stomatal conductance (*gs*), C. Transpiration rate (*E*), and D. Water use efficiency (WUE) under different cobalt concentrations in *Jatropha curcas* L. strain DARL-2

WUE was continuously decreased in all plants of cobalt treatment due to higher *E* as compared to controlled plants (Fig. 4D). The value of WUE was ~16.10, ~14.96, ~14.31, ~4.60, ~2.47 and 1.50 mol(CO_2)mol(H_2O)$^{-1}$ in 0, 100, 200, 400, 600, and 800 mg kg^{-1} treated plants, respectively. It was ~0.93, ~0.89, ~3.50, ~6.51, and ~10.76 fold decreased as compared to controlled plants, respectively (Fig. 4D).

Effect of cobalt stress on photosynthetic pigments

The bio-synthesis of photosynthetic pigments [Chl *a*, *b*, and total Chl (*a*+*b*)] and carotenoids content in all plants was affected by different concentrations of cobalt stress (Table 1). The Chl a value was found

Table 1: Effect of different concentration of Cobalt stress (100, 200, 400, 600, and 800 mg kg^{-1} soil) on alteration of photosynthetic pigments in *Jatropha curcas* L. Different letters in each column indicate significant differences at P ≤ 0.05, as determined using least significant difference (LSD) test. Standard errors of the two treatment means for each parameter are given in the last row.

Cobalt mg kg^{-1} Soil	*Chl a (mg g^{-1} fw)*	*Chl b (mg g^{-1} fw)*	*Total chlorophyll (a+b) (mg g^{-1} fw)*	*Carotenoids (mg g^{-1} fw)*
Control	36.148	37.512	73.658	0.299
100	32.108	28.514	60.621	0.234
200	29.538	32.820	62.357	0.212
400	27.671	35.024	62.694	0.155
600	24.288	26.225	50.542	0.155
800	18.366	13.153	31.518	0.048
SE (M)	1.251	2.826	2.4270	0.039
LSD	3.773	8.519	7.3160	0.119

~36.15, ~32.11, ~29.54, ~27.67, ~24.29, and ~18.37 mg g^{-1} FM in 0, 100, 200, 400, 600, and 800 mg kg^{-1} cobalt treatment, respectively (Table 1). It was reduced ~1.13, ~1.22, ~1.30, ~1.49, and ~1.97 fold in 100, 200, 400, 600, and 800 mg kg^{-1} treated plants, respectively, as compared to controlled plants (Table 1). Also, Chl b value was found in decreasing pattern. It was observed ~37.51, ~28.51, ~32.82, ~35.02, and ~26.22 mg g^{-1} FM in 0, 100, 200, 400, 600, and 800 mg kg^{-1} cobalt treatment, respectively (Table 1). The Chl b accumulation was found ~1.31, ~1.14, ~1.07, ~1.43, and ~2.85 fold decrease in 100, 200, 400, 600, and 800 mg kg^{-1} cobalt treated plants, respectively, as compared to controlled plants (Table 1). Similarly, total Chl (a & b) content was also found in decreasing trend (Table 1). It was ~73.66, ~60.62, 62.35, 62.69, 50.54, and 31.52 mg g^{-1} FM in 0, 100, 200, 400, 600, and 800 mg kg^{-1} cobalt treatment, respectively (Table 1). It was decreased ~1.21, ~1.18, ~1.17, ~1.45, and ~2.34 fold in 100, 200, 400, 600, and 800 mg kg^{-1} treated plants, respectively, as compared to controlled plants (Table 1).

Total carotenoids content was not much disturbed by cobalt stress. It was found ~0.30, ~0.23, 0.21, ~0.15, ~0.16, and 0.05 mg g^{-1} FM in 0, 100, 200, 400, 600, and 800 mg kg^{-1} cobalt treatment, respectively (Table 1). Total carotenoids content was not significantly decreased in 100 and 200 mg kg^{-1} treated plants as compared to controlled plants. It was decreased ~1.28 and ~1.41 fold in 100 and 200 mg kg^{-1} treated plants as compared to controlled plants but carotenoids content was significantly decreased ~1.92, ~1.93, and ~6.23 fold in 400, 600, and 800 mg kg^{-1} treated plants as compared to controlled plants (Table 1).

Discussion

Heavy metal affects plants in two ways. Firstly, it alters reaction rates and influences the kinetic properties of enzymes leading to changes in plant metabolism. Secondly, excessive heavy metals lead to oxidative stress (Almodovar *et al*., 2014). It is known that excessive heavy metal exposure may increase the generation of reactive oxygen species (ROS) in plants, and oxidative stresses arise if the balance between ROS generation and removal is broken. The toxicity of heavy metals may arises a result of the generation of ROS that may cause wide ranging damage to proteins, nucleic acids, and lipids, eventually leading to cell death (Mittler, 2002). The ability of *Jatropha curcas* L. to cope with heavy metal stress depends on oxidative stress defence mechanisms (Gao *et al*., 2008, 2009).

It has been established that Co like other pollutant elements are relatively toxic to plants when given in supranormal doses (Chatterjee and Chatterjee, 2000; Pandey and Sharma, 2002; Gopal *et al*., 2003). According to Vrtoch *et al.* (2007), after 50 µM Co treatment, tobacco showed interveinal chlorosis on young leaves and significant suppression of growth. Similar toxic effect of cobalt on tomato plants was described by Gopal *et al.* (2003). Chlorosis is considered as indirect effect caused by alterationsin the concentrations of essential mineral nutrients, a decrease in net photosynthesis as a consequence of stomatal closure, reduced intercellular spaces and by alterations within chloroplasts (Vazquez *et al*., 1987; Chatterjee and Chatterjee, 2000). Also, cobalt and other heavy metals affect photosynthesis and the activities of related enzymes (Austerfeld, 1979; Blaylock *et al*., 1986).

However, present study showed that the value of all gas exchange measurement parameters as P_N, *gs,* and *E* was observed in increasing pattern in Jatropha plants treated with 100 and 200 mg kg^{-1}. Although, cobalt has been categorized as a beneficial element for plants (Marschner, 2003; Pilon-Smits *et al*., 2009). Due to the lack of the evidence for bioactive forms of cobalt, it is not easy to understand the mechanism for the beneficial effects of cobalt on plants. Presumably cobalt exerts indirect effects on plant metabolism at low concentrations. Evidence indicates that these effects may result from the role of cobalt in cross-linked interactions with other elements. The beneficial effects of cobalt on plants are also attributed to its inhibition of the production of ethylene production, which regulates such processes as germination, growth, ripening, senescence, and stress resistance (Yu and Yang, 1979).

Similar beneficial effects of cobalt to other crops include senescence retardation in lettuce (Tosh *et al*., 1979), germination improvement in sunflower (Singh and Rao, 1993), tomato and cucumber (Helmy *et al*., 1994), prolongation of the vase life of cut flowers, maiden hair ferns (Fujino and Reid, 1983), marigold (Chandra *et al*., 1981) and roses (Venkotarayappa *et al*., 1980), ozone and drought tolerance (Young, 1979; Wenzel *et al*., 1995), and wilt disease control in guava (Dwivedi, 1991) were also reported.

On molecular level, it was found that Co in supranormal concentrations caused inhibition of catalase activity, decrease of chlorophyll content connected with chlorosis (Chatterjee and Chatterjee, 2000; Pandey and Sharma, 2002). Toxicity of cobalt at C0 50 µM resulted in both decrease of tobacco transpiration rates and cobalt uptake.

In present study, after a particular cobalt dose, cobalt treatment showed toxic effects on Jatropha plants. All the selected parameters of net gas exchange, Chl and carotenoids content under higher dose of cobalt was in decreasing trend. Similar trends were reported with plants treated with Cd^{2+} were able to photosynthesize although not to the extent that the control plants did, but those plants treated with high Cd^{2+} concentrations were unable to photosynthesize showing the lowest P_N values of all treatments.

Higher dose of cobalt as from 400 to 800 mg kg^{-1} treatment also reduce the gas exchange parameters. Similar hazardous effects of increasing cobalt level in the reductions of net photosynthesis was also reported by Gad *et al.* (2011). Another toxic metal like cadmium (Cd^{2+}) also reduce the stomatal opening (reductions in stomatal conductance), thereby reducing mesophyll CO_2 availability. Alternatively, Cd^{2+} could have induced a reduction in the synthesis or activity of Calvin cycle enzymes reducing the demand for CO_2 and, resulting in reductions in stomatal conductance (Krupa *et al*., 1993; Dong *et al*., 2005). Similar results were found by Chaffei *et al.* (2004) where *Lycopersicum esculentum* plants, exposed for one week to Cd^{2+}, showed a decrease in the rate of net photosynthesis due to stomata closure, Rubisco inactivation and chlorophyll degradation (Mendoza *et al*., 2007).

Photosynthesis is one of the important physiological processes most susceptible to heavy metal toxicity. Metals toxicity affects it at multiple levels such as pigment biosynthesis/degradation, stomatal functioning, enzyme inhibition, alteration in chloroplast membranes structure/function and photosystems (Mysliva-Kurdziel *et al*., 2004). Heavy metal stress causes the activation of chlorophyll degrading enzyme, chlorophyllase (Abdul-Basset *et al*., 1995) and in particular Co inhibits those involved in chlorophyll biosynthesis, such as 5- aminolevulenic acid (ALA) synthase, ALA dehydratase, prophobilinogenase and unporphyrinogen III decarboxylase (Shalygo *et al*., 1999). Therefore, in present report, the concentration of chlorophyll exhibited a decrease in response to Co stress (Table 1).

The Co also caused the decrease in stomatal conductance and consequently the intercellular CO_2 concentration and carbonic anhydrase (CA) activity (Ali *et al*., 2010). The Carbonic anhydrase (CA) catalyzes the interconversion of CO_2 and HCO_3^-, and its activity is to a large extent regulated by CO_2, besides zinc and light (Tiwari *et al*., 2005). Thus, the decreased CO_2 concentration was accompanied

with a concomitant decline in CA activity. The decreased chlorophyll concentration, stomatal conductance, intercellular CO_2 concentration and CA activity together with decreased Hill reaction activity (Chatterjee *et al.*, 2006) ultimately hampered the net photosynthetic rate (Fig. 4A) which is also confirmed by positive correlation between CA activity and net photosynthetic rate as well as between chlorophyll and net photosynthetic rate.

The decrease in chlorophyll content and catalase activity might be due to impaired incorporation of iron for synthesis of chlorophyll and protoporphyrin in excess cobalt conditions similar to some earlier reports (Agarwala *et al*., 1977; Miller *et al*., 1982). Interference of heavy metals including cobalt with iron in plant metabolism is known to induce disturbances creating physiological iron deficiency and decrease in chlorophyll synthesis (Samarkoon and Rauser, 1979).

In conclusion, the results presented here suggest that Co at lower concentration (>200 mg kg^{-1}) favors the gas exchange parameters and photosynthetic pigmentation in Jatropha. The higher concentration (<200 mg kg^{-1}) generated oxidative stress, which was noticed on the basis of decreasing trends in all studied photosynthetic parameters. All studied plants of *Jatropha* were survived morphologically up to 800 mg kg^{-1} but seedlings were not survived under 1000 mg kg^{-1} due to severe cobalt toxicity stress. Among studied plants, Jatropha seedlings showed the best photosynthetic potential under 200 mg kg^{-1} Co stress.

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References

Abdul-Basset, R., Iss, A.A. and Adam, M.S. 1995. Chlorophyllase activity: Effects of heavy metals and calcium. *Photosynthetica* **31**: 421-425.

Agarwala, S.C., Bisht, S.S. and Sharma, C.P. 1977. Relative effectiveness of certain heavy metals in producing toxicity and symptoms of iron deficiency in barley. *Canadian Journal of Botany* **55**: 1299-1307.

Ali, B., Hayatb, S., Hayatb, Q. and Ahmad, A. 2010. Cobalt stress affects nitrogen metabolism, photosynthesis and antioxidant system in chickpea (*Cicer arietinum* L.). *Journal of Plant Interactions* **3**: 223-231.

Almodovar, C.G., Gimeno, V., Nieves, M., Lopez, L.D., Simon, I. and Sanchez, F.G. 2014. Improving the tolerance of *Jatropha curcas* L. plants to abiotic stresses. *CAB Reviews Perspectives in Agriculture Veterinary Science Nutrition and Natural Resources* **9**: 01-10.

Ashraf, M. and Harris, P.J.C. 2013. Photosynthesis under stressful environments: An overview. *Photosynthetica* **51**(2): 163-190.

Austerfeld, F.A. 1979. The effect of nickel, cobalt and chromium on net photosynthesis of the primary and secondary leaves of *Phaseolus vulgaris*. *Photosynthetica* **13**: 434-438.

Blaylock, A.D., Davis, T.D., Jolley, V.D. and Walser, R.H. 1986. Influence of cobalt and iron on photosynthesis, chlorophyll, and nutrient content in regreening chlorotic tomatoes and soybeans. *Journal of Plant Nutrition* **9**: 823-838.

Chaffei, C., Pageau, K., Suzuki, A., Gouia, H., Ghorbel, M.H. and Masclaux-Daubresse, C. 2004. Cadmium toxicity induced changes in nitrogen management in *Lycopersicum esculentum* leading to a metabolic safeguard through an amino acid storage strategy. *Plant and Cell Physiology* **45**: 1681-1693.

Chandra, G., Reddy, K.S. and Mohan-Ram, H.Y. 1981. Extension of vase-life of cut marigold (*Tagets patula*) Chrysanthemum flowers by use of cobalt chloride. *Indian Journal of Experimental Biology* **19**: 150-154.

Chatterjee, C., Gopal, R. and Dube, B.K. 2006. Physiological and biochemical responses of French bean to excess cobalt. *Journal of Plant Nutrition* **29**: 127-136.

Chatterjee, J. and Chatterjee, C. 2000. Phytotoxicity of cobalt, chromium and copper in cauliflower. *Environmental Pollution* **109**: 69-74.

Collins, Y. E., and Stotzky, G. 1989. Factors affecting the toxicity of heavy metals to microbes. In: Beveridge, T.J. and Doyle, R.J. (Eds.), Melal Ions and Bacteria, Wiley, New York, pp. 3 I-90.

Croce, R. and Amerongen, H.V. 2014. Natural strategies for photosynthetic light harvesting. *Nature Chemical Biology* **10**: 492-501.

Dong, J., Wu, F.B. and Zhang, G.P. 2005. Effect of cadmium on growth and photosynthesis of tomato seedlings. *Journal of Zhejiang University Science B* **6**: 974-980.

Dwivedi, S.K. 1991. Effect of some heavy metals on growth of *Fusarium oxysporium* fsp-psiddii causing guava wilt disease. *International Journal of Tropical Diseases* **9**: 127-130.

Fujino, D.W. and Reid, M.S. 1983. Factors affecting the vase life of fronds of maidenhair fern (*Adiantum raddianum*). *Scientia Hortculturae* **21**: 181-188.

Gad, N., Abd El-Moez, M.R. and Kandil, H. 2011. Barley response to salt stress at varied levels of cobalt I: Growth and mineral composition. *Journal of Applied Sciences Research* **7**(11): 454-1459.

Gao, S., Li, Q., Ou-Yang, C., Chen, L., Wang, S.H. and Chen, F. 2009. Lead toxicity induced antioxidant enzyme and phenylalanine ammonia-lyase activities in *J. curcas* radicles. *Fresenius Environmental Bulletin* **5**: 811-815.

Gao, S., Yan, R., Cao, M., Yang, W., Wang, S. and Chen, F. 2008. Effects of copper on growth antioxidant enzymes and phenylalanine ammonia-lyase activities in *J. curcas* seedlings. *Plant, Soil and Environment* **54**: 117-122.

Garbisu, C. and Alkorta, I. 2001. Phytoextraction: A cost effective plant-based technology for the removal of metals from the environment. *Bioresource Technology* **77**: 229-236.

Gopal, R., Duke, B.K., Sinha, P. and Chatterjee, C. 2003. Cobalt toxicity effects on growth and metabolism of tomato. *Communications in Soil Science and Plant Analysis* **34**: 619-628.

Helmy, Y.H., El-Abd, S.O. and Singer, S.M. 1994. Seed germination of tomato and cucumber in salinized condition and prevention of its effect. *Egyptian Journal of Horticulture* **21**: 121-131.

Krupa, Z., Öquist, G. and Huner, P.A. 1993. The effects of cadmium on photosynthesis of *Phaseolus vulgaris* chlorophyll a fluorescence analysis. *Physiol Plantarum* **88**: 626-630.

Marschner, H. 2003. Mineral nutrition of higher plant. 2nd ed. Amsterdam. *Academic Press*, pp. 201-228, 405-435.

Mendoza, D.G., Gil, F.E., Jorge, M., Santamaria, J.M. and Perez, O.Z. 2007. Multiple effects of cadmium on the photosynthetic apparatus of *Avicennia germinans* L. as probed by OJIP Chlorophyll Fluorescence Measurements. *Zeitschrift für Naturforschung* **62c**: 265-272.

Miller, G.W., Denney, A., Pushnik, J. and Ming-Ho, Y. 1982. The formation of delta-aminolevulinate a precursor of chlorophyll in barley and the role of iron. *Journal of Plant Nutrition* 5: 289-300.

Mittler, R. 2002. Oxidative stress, antioxidants and stress tolerance. *Trends in Plant Science* **7**: 405-410.

Moran, R. and Porath, D. 1980. Chlorophyll determination in intact tissues using *N, N* Dimethyl formamide. *Plant Physiology* **65**: 478-479.

Mysliva-Kurdziel, B., Prasad, M.N.V. and Strzalka, K. 2004. Photosynthesis in heavy metal stressed plants. In: Prasad, M.N.V. (Ed.) Heavy metal stress in plants: From biomolecules to ecosystems. Berlin: Springer-Verlag, pp. 146-181.

Pan, J., Lin, S. and Woodbury, N.W. 2012. Bacteriochlorophyll excitedstate quenching pathways in bacterial reaction centers with the primary donor oxidized. *The Journal of Physical Chemistry B* **116**: 2014-2022.

Pandey, N. and Sharma, C.P. 2002. Effect of heavy metals Co_2^+, Ni_2^+ and Cd_2^+ on growth and metabolism of cabbage. *Plant Science* **163**: 753-758.

Pilon-Smits, E.A.H., Quinn, C.F., Tapken, W., Malagoli, W. and Schiavon, M. 2009. Physiological functions of beneficial elements. *Current Opinion in Plant Biology* **12**: 267-274.

Poschenrieder, C.H. and Barceló, J. 1999. Water relations in heavy metal stressed plants. In: Heavy metal stress in plants. Springer, Berlin, Heidelberg, pp. 207-229.

Samarkoon, A.B. and Rauser, W.E. 1979. Carbohydrate level and photoassimilate export from leaves of *Phaseolus vulgaris* exposed to excess cobalt, nickel and zinc. *Plant Physiology* **63**: 1165-1169.

Shalygo, N.V., Kolesnikova, N.V., Voronetskaya, V.V. and Averina, N.G. 1999. Effect of Mn^{2+}, Fe^{2+}, Co^{2+} and Ni^{2+} on chlorophyll accumulation and early stages of chlorophyll formation in greening barley seedlings. *Russian Journal of Plant Physiology* **46**: 496-501.

Singh, B.G. and Rao, G.R. 1993. Effect of chemical soaking of sunflower (*Helianthes annuus*) seed on vigour inder. *Indian Journal of Agricultural Sciences* **63**: 232-233.

Skórzyńska-Polit, E. and Baszyński, T. 1997. Differences in sensitivity of the photosynthetic apparatus in Cd-stressed runner bean plants in relation to their age. *Plant Science* **128**: 11-21.

Taiz, L. and Zeiger, E. 2010. Plant Physiology. 5th Edition, Sinauer Associates Inc., Sunderland, pp. 782.

Tiwari, A., Kumar, P., Singh, S. and Ansari, S.A. 2005. Carbonic anhydrase in relation to higher plants. *Photosynthetica* **43**: 1-9.

Tosh, S., Choudhuri, M.A. and Chatterjee, S.K. 1979. Retardation of lettue (*Lactuca sativa* L.) leaf senescence by cobalt ions. *Indian Journal of Experimental Biology* **17**: 1134-1136.

Vazquez, M.D., Poschenrieder, C. and Barcelo, J. 1987. Chromium (VI) induced structural and ultrastructural changes in bush bean plants (*Phaseolus vulgaris* L.). *Annals of Botany* **59**: 427-438.

Venkotarayappa, T., Tsujita, M.J. and Murr, D.P. 1980. Influence of cobaltous ion on the post-harvest behavior of rose (*Rose hyorida* cultivar Samantha). *Journal of the American Society for Horticultural Science* **105**: 148-151.

Vrtoch, L., Pipíška, M., Horník, M. and Augustín, J. 2007. Bioaccumulation of $^{60}Co^{2+}$ ions in Tobacco plants. *Nova Biotechnologica et Chimica* **VII-I**: 41-49.

Wenzel, A.A., Schlautmann, H., Jones, C.A., Kupper, K. and Mehlhom, H. 1995. Aminoethoxyvinylglycine, cobalt and ascorbic acid all reduce ozone toxicity in mung beans by inhibition of ethylene biosynthesis. *Physiologia Plantaum* **93**: 286-290.

Young, R.S. 1979. Cobalt in biology and biochemistry. Academic Press, London, pp. 177.

Yu, Y.B. and Yang, S.F. 1979. Auxin induced ethylene production and its inhibition by aminorthoxyglycine and cobalt ion. *Plant Physiology* **64**: 1074-1077.