IMPROVEMENT IN THE BIOENERGETICS SYSTEM OF PLANTS UNDER Hg STRESS ENVIRONMENT VIA SEAWEEDS

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Abstract

The effects of Hg and its remediation through seaweeds on seedlings were escorted in a greenhouse experiment in a randomized block design. The effects of Hg were monitored in relation with bioenergetics system of *Trigonella foenumgraecum* plant at test site scale. Plants that were exposed to Hg, showed affect in diverse ways, including affinity to suffer in morphological as well as on sugar metabolism. The stress imposed by Hg exposure also extends to chloroplast pigments that lead to the distorted photosynthetic apparatus. The outcomes of reduced contents of photosynthetic machinery related with reduced contents of glucose, sucrose, total soluble sugars and carbohydrate contents of plants. These contents plays vital rule for providing bioenergy to the plant growth regulation. It was suggested that Hg is lethal for plant bioenergetics system due to which plants fail to survive under stress. The lethal effects of Hg were tried to remediate through green seaweeds (*Codium iyengrii*). It was observed that seaweeds successfully controlled the mobility of Hg metal and improves the plant growth regulatory system at lower applied dose only. While at higher dose of Hg, seaweeds were also effective but to a certain limits. It was established that continuous addition of Hg in soil and aquatic resources execute to the plant productivity. It is demand of time to develop alternative eco-friendly remediation technologies for controlling, cleaning Hg-polluted zones.

Key words: Hg, Seaweeds, Sugar metabolism, Chloroplast pigments, Growth regulatory system.

Introduction

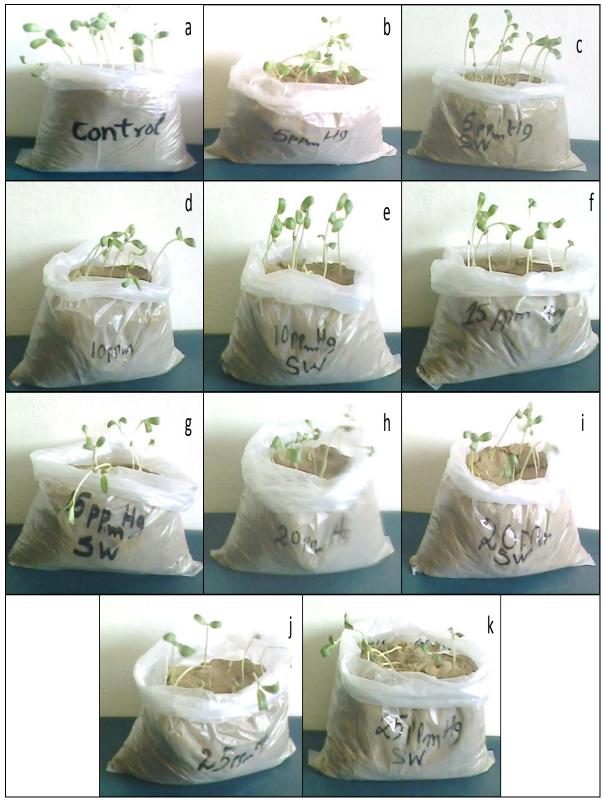
Agronomy had a major influence in hominids, being the main power behind the passage from a huntergatherer/vulture civilization to an inactive one, becoming a vital tool for social sustainability and the advancement of economics throughout the worldwide (Nagajyoti et al., 2010). However, the channels of development were accompanied by an extreme alteration of the environment and the intrinsic introduction to new risks caused directly or indirectly by those revolutions (Khalil, 1997). Among the diverse replicas accessible to search non-essential heavy metal injuries, plants reflect certain sole structures that make them stimulating issues for investigation. Initially, as main manufacturers of the food chain for humans and land animals, considering the lethal influence of heavy metal in the plant nutritional status and the risks of bio-magnifications of these toxicants for consumers is extremely significant. Furthermore, because plants misery the capability to escape from polluted areas, these organisms changed or modified the mechanisms to switch acquaintance to toxicants (Nagajyoti et al., 2010).

Detailed literature search showed that the mechanistic change due to toxicity of heavy metals is originated through a complex pattern of interactions between cellular macromolecules and the metal ions. The ingress of the metal in the plant cell through roots or areal parts can activate numerous metabolic and signal transduction pathways and genetic processes to neutralize the source of toxicity (Patra & Sharma, 2000). Metal toxicity adversely effects on carbohydrate metabolism. Many researchers reports that the decrease in total sugar content of metal stressed leaves probably corresponded with the photosynthetic inhibition or stimulation of respiration rate. Higher starch accumulation in damaged leaves of *Tilia argentea* and *Quercus cerris* may result both in the higher resistance of their photosynthetic apparatus and low starch export from the mesophyll. The negative effect of heavy metals on carbon metabolism is a result of their possible interaction with the reactive centers of ribulosebisphosphate carboxylase (John *et al.*, 2008; Volesky & Holen, 1995; Kupper *et al.*, 1996). The aims and objectives of the present paper were to remediate the metal toxicity through credible of seaweeds on edible plants. The most important and socioeconomically crop were selected for investigation to check the effects of Hg on their bioenergetics system and improvement of injured C metabolism of plants by nutrients of seaweeds. This paper also discussed the help of seaweeds in improving the bioenergetics system of plant under Hg stress.

Material and Method

The fresh green seaweeds were collected from Bullaygi coastal area of Karachi early in the morning from free floating tides in Feb. 2010. The collected seaweeds were washed thoroughly using sea water and transported to the Laboratory. The seaweeds now washed here using Tap water for 1h to avoid any contamination during collection and transportation. They were identified by usual method as Codium iyengrii (Askari et al., 2007). They were spread for drying purpose on mat at room temperature. Then they were grinded after complete dryness and used as it is without further treatment. Pants of Trigonella foenumgraecum were grown in complete randomized design in six separate 1kg bags with three replicate containing Hg and seaweeds and presented in Fig. 1. The plants were harvested after a 30 D and subjects to biochemical analysis.

Estimation of chlorophyll contents: 98% Acetone were prepared for analysis and extract were prepared by methods recording optical density of pigments using UV-Visible spectrophotometer (Azmat *et al.*, 2006).



Germination and visual symptoms of *Trigonellafoenum-graecum* under Hg and seaweeds treatments

Fig. 1a; Control plant exhibiting normal growth. 1b; Weak and slightly bent plants. 1c; Erect plants with improved rate of germination. 1d; Weak and bent plants. 1e; Upright healthy plants. 1f; Thin stems with low rate of germination. 1g; A slight enhancement in vigor and germination. 1h; Very poor germination and growth. 1i; Erect plants with improved germination and growth. 1j and k; At high mercury concentration the plants were not survived even in the presence of seaweeds.

Free carbohydrates: were determined by method described by Yemm & Willis (1954). Optical density was recorded at 620 nm against a reagent blank. Standard curve of glucose was prepared to estimate free carbohydrate. Working standard of 100 μ gm/ml solution was further diluted from 10-100 μ gm/ml. The contents were determined using formula:

Sample OD x CF x TDF / Mol. Wt = μ mol/gm fresh weight.

where, Sample OD= sample's optical density. CF= curve factor (OD x Concentrations of glucose). TDF= total dilution factor (Sum of all dilutions, when DF = Total solvent / sample or extract)

Glucose contents were estimated by the method of Riazi *et al.* (1985). Absorbance was recorded at 630nm. Standard curve was obtained by making different dilutions of pure glucose with distil water from 100μ g/ml stock solution. Glucose range was $10-100\mu$ g/ml.

Sucrose contents were determined by the method described by Riazi *et al.*, 1985. Absorbance was recorded at 620nm. Standard curve was obtained by making different dilutions of pure sucrose with distil water from 100μ g/ml stock solution. Sucrose range was $10-100 \mu$ g/ml.

Total soluble sugars were estimated by the method of Riazi *et al.*, 1985. Absorbance was recorded at 625nm. Standard curve was made by different dilutions of pure glucose with distil water from 100μ g/ml stock solution. Glucose range was $10-100\mu$ g/ml.

Statistical analysis: Statistical analysis was based on twoway ANOVA. The effects of Hg and its treatment through seaweeds were considered statistically significant when p<0.05. Data are presented as mean \pm standard errors (n = 3).

Results

Photo-biological reactions in photosynthetic pigments: The effect of Hg on chloroplast pigments of Trigonella foenumgraecum were reported in the Table 1. Results provide the experimental evidences of Hg toxicity on photosynthetic apparatus of Trigonella foenumgraecum. The distorted photobiology of plant was reflected as an increase in concentration of Chlorophyll 'a' at all experimental levels of Hg viz., 0.106 \pm 0.00011, 0.111 \pm 0.00008, 0.152 \pm 0.00014, 0.163 \pm 0.000057, 0.112 \pm 0.00008 respectively where as it was 0.068 \pm 0.0023 in control while the contents of Chlorophyll 'b' showed an augmentation up to 0.0605 ± 0.0001 , 0.0786 ± 0.0001 , $0.0945 \pm 0.0002, \, 0.1075 \pm 0.0002$ and 0.0924 ± 0.0001 respectively over control which was 0.0552 ± 0.0001 . These results were similar to that of earlier work of Barman & Bera (2002) who also reported an increase in chlorophyll content of leaves under Hg stress. The lethal effects of Hg on Chlorophyll 'a' were remediated through addition of seaweeds in metal contaminated soil. It was found that working capability of photosynthetic apparatus increased significantly and appeared as 0.091 \pm 0.00011, 0.098 \pm 0.00014, 0.101 \pm 0.00014, 0.111 \pm 0.00015, 0.078 \pm 0.00011 respectively (Table 1). Results reported in Table 1

showed a significant increment in total chlorophyll content of Trigonellafoenum-graecum up to 0.0945 ± 0.00014 , $0.1134 \pm 0.00012, 0.1141 \pm 0.000088, 0.1814 \pm 0.000088$ and 0.1167 ± 0.000088 respectively at all experimental levels of applied mercury over control which was $0.0637 \pm$ 0.00034. Application of green seaweed to mercury polluted soil exhibited a significant improvement in total chlorophyll content as 0.0774 ± 0.00017 , $0.0872 \pm$ $0.00012, 0.1008 \pm 0.00012, 0.1152 \pm 0.000088$. It was interesting to note that the carotenoid contents of Trigonella foenumgraecum increased at all elevated concentration of Hg (Table 1) as 0.0139 ± 0.0001 , $0.0804 \pm$ 0.0001, 0.1710 \pm 0.001, 0.1189 \pm 0.0001 and 0.0876 \pm 0.0001 respectively, where control was 0.0048 ± 0.0001 mg/gmf.wt. Plants manufacture carotenes from fats and other organic metabolic building blocks. This condition was improved by the application of green seaweeds as $0.0075 \pm 0.0001, 0.0086 \pm 0.00001, 0.0761 \pm 0.0001,$ 0.0573 ± 0.00001 and 0.0846 ± 0.0021 respectively. The seaweeds act as ligands with mercury and let environment free from metal (Volesky & Holan, 1995; Prasad & Prasad 1987b; Azmat et al., 2006; Askari et al., 2007; Igwe et al., 2008; Kumar et al., 2009). This increasingly pattern was brought down significantly by using green seaweeds up to $0.0657 \pm 0.002, 0.0742 \pm 0.0002, 0.0743 \pm 0.0002, 0.0743$ \pm 0.0017 and 0.0672 \pm 0.0001 respectively at all applied concentrations of mercury. It was found that working capability of photosynthetic apparatus increased significantly as 0.091 ± 0.00011 , 0.098 ± 0.00014 , $0.101 \pm$ $0.00014, 0.111 \pm 0.00015, 0.078 \pm 0.00011$ respectively (Table 1). Chlorophyll content in treated plants of *Trigonella foenumgraecum* at 25ppm Hg (0.078 ± 0.00011) were very close to the control value (0.068 ± 0.0023) which indicate the binding of Hg into the seaweeds.

Carbohydrates component as a Photo biological products: It was observed that carbohydrate contents in shoots and roots of Trigonella foenumgraecum declined significantly at all experimental levels up to 0.36 ± 0.01 , $0.39 \pm 0.01, 0.39 \pm 0.02, 0.45 \pm 0.01, 0.49 \pm 0.01,$ respectively over control which was 0.686 ± 0.003 (Table 2) and in roots were 0.273 ± 0.03 , 0.336 ± 0.02 , $0.350 \pm$ 0.01, 0.393 ± 0.02 and 0.436 ± 0.01 over control which was 7.38 ± 0.04 . There was a significant increase in glucose content as 3.386 ± 0.0011 , 3.389 ± 0.0011 , 3.647 ± 0.0011 , 4.149 ± 0.0011 and 4.159 ± 0.0011 respectively at all concentrations of applied mercury, as compare to control which was 3.217 ± 0.0011 . Results reported in the Table 2 showed that non-reducing sugar, sucrose increases at all experimental levels of mercury in Trigonella foenumgraecum up to 50.61 ± 0.0493 , 49.70 ± 0.0088 , $40.75 \pm 0.0264, \ 32.5 \pm 0.0208$ and 28.77 ± 0.0176 respectively when control was 14.41 ± 0.0145 . Results of total soluble sugars (Table 1) obtained from Trigonella foenumgraecum exhibited some interesting results as 8.28 ± 0.0288 , 8.22 ± 0.0132 , 7.37 ± 0.049 , 6.69 ± 0.0461 and 6.23 ± 0.0173 over control which was 7.38 ± 0.04 . At 5ppm and 10ppm an increase in total soluble sugars was recorded, at 15ppm it was almost same as control but it decreased at 20 and 25ppm of Hg. It means at 5, 10 and 15ppm plant tried to enhance the tolerance capacity under mercury stress (Datta et al., 2011). Whereas at 20ppm and 25ppm of Hg plant could not cope with the toxic effects of

metal and showed a significant decrease in total soluble sugar contents. Biosorption of mercury through seaweeds showed a significant improvement in TSS contents of shoot as 8.28 \pm 0.0288, 8.22 \pm 0.0132, 7.37 \pm 0.049, 6.69 \pm 0.0461, 6.23 \pm 0.0173 at all experimental levels of mercury over control which was $0.686 \pm 0.003 \mu mol/gm$ F.wt. Carbohydrate contents of root were significantly enhanced as 8.21 \pm 0.0033, 8.1 \pm 0.0288, 7.35 \pm 0.023, 7.61 \pm 0.0152, 6.74 \pm 0.08 at all experimental levels of mercury over control which was $7.38 \pm 0.04 \ \mu mol/gm$ F.wt. The glucose was significantly improved by seaweeds application as 3.066 \pm 0.0011, 3.076 \pm 0.0011, 3.220 \pm $0.0011, 3.550 \pm 0.0011, 3.970 \pm 0.0011$ when control was $3.217 \pm 0.0011 \,\mu$ mol/gm F.wt. Substantial improvement in sucrose content of Trigonella foenum-graecum growing under seaweed amended soil was found to be as 16.82 \pm $0.0288, 15.02 \pm 0.012, 14.25 \pm 0.0233, 12.9 \pm 0.0233,$ 11.98 ± 0.0145 when control was 14.41 ± 0.0145 µmol/gm F.wt. Whereas total soluble sugars of Trigonella foenumgraecum was 8.21 ± 0.0033 , 8.1 ± 0.0288 , 7.35 ± 0.023 , 7.61 \pm 0.0152, and 6.74 \pm 0.08 μ mol/gm F.wt. respectively (Davis et al., 2003).

Discussion

Seaweeds are very fascinating plants and abundantly available in beaches of the coastal areas all around the world. Seaweeds having a suitable content of nitrogen and potassium are traded as soil additives. They function as both fertilizer and soil conditioner. They are much better than from any traditional animal composts and the typical N: P: K ratios in chemical manures. They contain the enormous amounts of insoluble carbohydrates which act as a soil conditioners (improve aeration and soil structure, specifically in clay soils) and have good moisture retention properties or increase the water holding capacity of soil (Askari et al., 2007). Their effectiveness as fertilizers is also sometimes attributed to the trace elements they contain, but the actual contribution they make is very small compared to normal plant requirements. The application of seaweeds as a soil conditioner for controlling losses of soil due to the Hg, results in a healthy growth of the plant in contaminated environment (Zerouala et al., 2003; Meena et al., 2004; Same et al., 2002). It was proposed that alginate; a carbohydrate composed of long chains plays a crucial rule for controlling the mobility of Hg metal into the plant. It was visually observed that bags containing seaweeds when watered become swelled up. That may form a strong gel around the root exudates which hold water in it and provide water and nutrients under stress as reported by Same et al. (2002). The improvements in bioenergetics system have included higher seed germination, higher yields (Schiewer & Wong., 2000) increased uptake of soil nutrients, increased resistance to Hg toxicity, enhanced photosynthetic rate and more resistance to frost (Ahalya et al., 2003). It was found that the plant under Hg stress showed growing capability when seaweeds were used as a medical treatment for plant safety. The plants photobiological reactions were improved that result in a correction of carbon metabolism of plant. It was established that Hg was accumulated or biosorbed through enormous amounts of insoluble carbohydrates or OH group of carbohydrate which bind the metal on its

surface and check the mobility of metal through roots of plant to areal parts of plant.

Results suggested that Hg stimulates chlorophyll content at early stages of Trigonella foenumgraecum similar to that of wheat growth as observed by Liu (2010). The increase in concentration of pigments may be attributed to toxic mercury complex with pigment proteins which is nonfunctional chlorophyll due to which chlorophyllus pigments were also considered as stress indicator in various plant species (De-Filippis et al., 1981; Assche & Clijsters, 1990; Ciobanu & Babeanu, 2006). The nonfunctional chlorophyll or Hg containing porphyrin ring rather than Mg, failed to perform photosynthesis and results in failure of photobiological reactions. It was suggested that nonfunctional chlorophyll fails to perform function of absorption of solar energy radiation into leaves. The solar energy absorbed always used for photolysis of water. But distorted surface structure of leaves due to Hg may be responsible in decline of photosynthesis as reported earlier by Askari & Azmat (2013). This results in failure of photolysis of water. Several researchers reports that plant under Hg stress produce excess of chlorophyll up to certain period of time but due to shortage of water and other proteinecious enzymes it might not perform photosynthesis properly and plants fails to survive in stress (Datta et al., 2012). Current experiments suggest that Trigonella foenumgraecumis a tolerant plant and produce large amount of stress proteins under mercury stress. But these stress compounds are meant for survival and not for the production of biosynthetic compounds. Increase in total chlorophyll may be due to stress of heavy metal mercury faced by plants (Norama et al., 1972; Barman & Bera, 2002a; Liu et al., 2010). The increase in carotenoids may attribute to the mercury stress for absorption of solar radiation. Carotenoids occurring naturally in the chloroplasts and chromoplasts are typically deliberated to execute two main functions in photosynthesis. They work for as an auxiliary light reaping pigments, extending the range of wavelengths over which light can drive photosynthesis, and they act to protect the chlorophyllous pigments from the harmful photo destructive reaction which occurs in the presence of oxygen (Tantrey & Agnihotri, 2010). The elevated concentration of carotenoids under Hg stress showed protective role of this important pigment (Table 1). High contents of carotenoids in the plant species showed their organization for prevention of both portions of the photosynthetic unit (the light harvesting antenna and the reaction center). They absorb blue light for use in photosynthesis, and protect chlorophyll from photodamage. In the present investigation an increase in concentration of carotenoids may be attributed to absorb extra energy which was not utilized by chlorophyll and help in continuation of photosynthesis. The accessory pigment role is significant in this investigation which is a singlet-singlet energy transfer from the carotenoid to the bacteriochlorophyll, while the protective role is a triplettriplet energy transfer from the bacteriochlorophyll to the carotenoid. It was reported that the inhibition of photosynthesis in Hg-stressed cumber leaves is more likely a consequence of an altered source-sink relationship due to distorted C metabolism, rather than due to toxic effects of Hg on the photosynthetic apparatus.

	Chlorol	Chlorophyll 'a'	Chlorol	Chlorophyll 'b'	Total Ch	Total Chlorophyll	Caro	Carotenoids
Hg [ppm]	Experimentals	Treated	Experimentals	Treated	Experimentals	Treated	Experimentals	Treated
0	0.068 ± 0.0023	0.068 ± 0.0023	0.0552 ± 0.0001	0.0552 ± 0.0001	0.0637 ± 0.00034	0.0637 ± 0.00034	0.0048 ± 0.0001	0.0048 ± 0.0001
2	$0.106^{*}\pm0.00011$	$0.091^{**} \pm 0.00011$	$0.0605^{*}\pm0.0001$	0.0657 ± 0.002	$0.0945^{*} \pm 0.00014$	$0.0774^{**}\pm 0.00017$	$0.0139^{\pm0.0001}$	$0.0075^{*\pm0.0001}$
10	$0.111^{*}\pm0.00008$	$0.098^{**} \pm 0.00014$	$0.0786^{*}\pm0.0001$	$0.0742^{**} \pm 0.0002$	$0.1134^{*} \pm 0.00012$	$0.0872^{**} \pm 0.00012$	$0.0804^{*}\pm0.0001$	$0.0086^{**\pm0.00001}$
15	$0.152^{*}\pm0.00014$	$0.101^{**}\pm 0.00014 0.0945^*\pm 0.0002$	$0.0945^*\pm 0.0002$	$0.0743^{**}\pm0.0002$	$0.1141^{*}\pm0.000088$	$0.1008^{**}\pm 0.00012$	$0.1710^{*\pm0.001}$	$0.0761^{**\pm0.0001}$
20	$0.163^{*}\pm0.000057$	$0.163^{*} \pm 0.000057$ $0.111^{**} \pm 0.00015$	$0.1075^{*} \pm 0.0002$	$0.0743^{**} \pm 0.0017$	$0.1814^{*}\pm0.000088$	$0.1152^{**} \pm 0.000088$	$0.1189^{*\pm0.0001}$	$0.0573^{**\pm0.00001}$
25	$0.112^{*}\pm0.00008$	$0.078^{**}\pm0.00011$	$0.078^{**}\pm 0.00011 0.0924^{*}\pm 0.0001$	$0.0672^{**}\pm0.0001$	$0.1167^{*}\pm0.000088$	$0.0984^{**}\pm 0.00015$	$0.0876^{*}\pm0.0001$	$0.0846^{*\pm0.0021}$

Table 1. Effect of Hg and seaweed on Photosynthetic pigments of Trigonellafoenum-graecum (mg / gmf.wt).

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Hg [ppm] 0 0.686 \pm 0.01 0 0.686 \pm 0.01	tal plant Root 0.626 ±	Carbohydrates Carbohydrates Treated plants s Treated plants s Shoots R	plants						
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		Shoots		LOIAL SOLUDIC SUGALS	ule sugars	NIIC	20101056	5	GIRCOSE
$\begin{array}{ccc} 0 & 0.686 \pm 0.0 \\ \\ 5 & 0.36^{*} \pm 0.0 \end{array}$		7 30 L 0 0	Roots	Experimental plants	Treated plants	Experimental plants	Treated plants	Experimental plants	Treated plants
5 0.36* ± 0.0		+0.0 ± 00.1	7.38 ± 0.04	7.38 ± 0.04	7.38 ± 0.04	14.41 ± 0.0145	14.41 ± 0.0145	3.217 ± 0.0011	3.217 ± 0.0011
	$01 0.273^* \pm 0.03$	$8.28^{*} \pm 0.0288$	8.21 ± 0.0033	$8.28^{*} \pm 0.0288$	8.21 ± 0.0033	$50.61^{*} \pm 0.0493$	$16.82^{**}\pm0.0288$	$3.386^* \pm 0.0011$	$3.066^{**} \pm 0.0011$
$10 0.39^* \pm 0.0$	$0.39^{*}\pm0.01 0.336^{*}\pm0.02$	$8.22^{*} \pm 0.0132$	$8.1^{**} \pm 0.0288$	$8.22^{*} \pm 0.0132$	$8.1^{**}\pm 0.0288$	$8.1^{**}\pm 0.0288 49.70^{*}\pm 0.0088$	$15.02^{**} \pm 0.012$	$3.389^{*}\pm0.0011$	$3.076^{**}\pm 0.0011$
$15 0.39^* \pm 0.0$	$0.39^{*}\pm0.02 0.350^{*}\pm0.01$	7.37 ± 0.049	7.35 ± 0.023	7.37 ± 0.049	$\textbf{7.35} \pm 0.023$	$40.75^* \pm 0.0264$	$40.75^{*}\pm0.0264 14.25^{**}\pm0.0233$	$3.647^* \pm 0.0011$	$3.220^{**}\pm0.0011$
20 $0.45^* \pm 0.01$	$0.393^{*}\pm0.02$	$6.69^{*} \pm 0.0461$	$7.61^{**} \pm 0.0152$	$6.69^{*} \pm 0.0461$	$7.61^{**}\pm 0.0152$	$32.5^{*}\pm0.0208$	$12.9^{**}\pm 0.0233$	$4.149^{*}\pm 0.0011$	$3.550^{**}\pm0.0011$
25 0.49* ± 0.0	$0.49^{*}\pm 0.01 0.436^{*}\pm 0.01 6.23^{*}\pm 0.0173$	$6.23^{*} \pm 0.0173$	$6.74^{**} \pm 0.08$	$6.23^{*} \pm 0.0173$	$6.74^{**} \pm 0.08$	$28.77^* \pm 0.0176$	$28.77^{*}\pm0.0176 11.98^{**}\pm0.0145$	$4.159^{*}\pm0.0011$	$3.970^{**} \pm 0.0011$

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The lethal effects of Hg on photosynthetic microorganism were remediated through addition of seaweeds in Hg contaminated soil. Seaweeds prevent movement of mercury to the plant as they contain polysaccharides and other organic substances which provide a suitable surface to Hg to form a complex (Azmat *et al.*, 2006; El-Sheekh *et al.*, 2000).

Carbohydrates contents are main energy provider to plant growth regulatory system and involve in metabolism to produce energy and other compounds. Our findings expressed a little increase in carbohydrate content along with increased concentrations of mercury as because Trigonella foenumgraecum is a proteinecious and resistant plant but CHO contents remained less at all concentrations of applied mercury as compared to control (Table 2). Reduced carbohydrate contents influence the overall growth of plants, this may attribute to reduced relative water content, number of open stomata as reported earlier by Azmat & Askari (2013) and chlorophyll contents (Table 1)under Hg stress, which ultimately reduced the photosynthetic rates as reported earlier (Jianjun et al., 2008; Xin et al., 2006; John et al., 2008; Yemm & Willis, 1954). It may be attributed that increase in concentration of carbohydrate at all applied doses may be for in improving bioenergetic system of plant under stress.

Decreased CHO content is an indicator of disturbed carbon metabolism in mercury treated plants. This may attributed to an oxidative stress in plants as reported earlier (Ciobanu & Babeanu, 2006), expressed by enzymatic and non-enzymatic antioxidant activity. That is why plant produced excess chlorophyll, carbohydrates and stress proteins to lessen mercury toxicity (Table 1). This increase in CHO contents at elevated concentration of Hg may be due to higher energy demand which may meet with the expenses of some other important contents like that of protein. It is an established fact that Hg may inhibit the water translocation from the shoot or exchange of gases from the surface of leaf. This reduction in carbohydrate may be related with an increase concentration of glucose, which is a simple and easily soluble sugar, raw material of photosynthesis and end product of cellular respiration. It was severely affected by Hg, both in shoot and root of Trigonella foenumgraecum plants. The higher concentration of glucose (Table 2) in leaves content showed that it is not utilizing for providing energy or in polymerization into sucrose or carbohydrates. It may featured to the inactivation of proteins containing -SH group associated with glucose uptake (Ghosh et al., 2007; Berman & Bera, 2002a). This increase in glucose contents molecules may be related to protective role or high carotenoid concentration to carry on the process of photosynthesis under stress or it may be the alternative pathway that results in an increase in glucose contents. Result of seaweed treated plants indicated an increase in root carbohydrate of Trigonella foenumgraecum.

Sucrose molecule is a disaccharide, composed of glucose and fructose. The augmentation in sucrose content of plant related to the stress by mercury toxicity. This increase may also be the alternative activated protective system of carotenoids which developed for the absorption of solar radiation due to the nonfunctional chlorophyll. As carbohydrate is a polymer composed of numerous monomers, increase in sucrose contents may be due to reduction in polymerization of carbohydrate. It is evident that sucrose and starch are the end products of photosynthesis and mostly sucrose is formed in the leaf cytosol, and transported through the phloem to the sink tissues where it is metabolized or stored in the vacuole (Yong, 2012). Increase in sucrose concentration indicated that Hg fails the source and sink system of the plant and fail long distance signaling system that balances metabolism between different organs. The experimental evidences are quite strong. The sucrose concentration in any plant organ is an integral of the recent patterns of supply and consumption. Failure of translocation of sucrose is due to the Hg accumulation is responsible root disorientation or defoliation leads to a progressive decline in metabolism activity in roots (Patra & Sharma., 2000). Changes in sucrose content in a plant species may correlate with these developmental and metabolism changes. As well as specific alterations in gene expression, altered sucrose contents have been implicated in changes in both the rate of cell division and in the patterns of morphogenesis (Fig. 1). During the night, sucrose is formed from starch, produced and stored in the chloroplasts during the day. The high contents of sucrose in chloroplast may involve in the absorption of energy through OH ions but this may be responsible for the increase in bio-energetic, correspond to decline in growth and survival. The investigation on plant proved that sucrose metabolism was the key factor which was disturb under Hg toxicity and elevation in contents also prove that the sucrose was not used properly and ultimately plants fail to survive (Jean-Philippe et al., 2012).

A vital role of total soluble sugars (TSS) (reducing and non-reducing) including sucrose, glucose, and fructose, is evident in plant structure (Fig. 1) and metabolism and act as molecule signals which regulate genes involved in photosynthesis, sucrose metabolism and osmolyte synthesis etc (Rosa et al., 2009; Couée, 2006; Azad & Kafilzadeh, 2012; Couee et al., 2006). In stress situations where soluble sugars are involved, such as metal toxicity chilling, herbicide injury, or pathogen attack, are related in the formation of reactive oxygen species. These converging or antagonistic relationships between soluble sugars, and reactive oxygen species (ROS) production, are related to the control of oxidative stress (Ahmed, 2003; Chandra & Garg, 1992; De et al., 1985). All these links place soluble carbohydrates in a pivotal role in the prooxidant and antioxidant balance, and must have constrained the selection of adaptive mechanisms involving soluble sugars and preventing de-regulation of reactive oxygen species production. The results of present investigation showed that initially there was an increase in the total soluble sugar which may be attributed with the adaption of plant metabolism in stress condition or scavenging the ROS species produced in stress condition but at higher condition, decrease may be linked with failure of scavenging of ROS species due to which plant could not stay alive.

Mechanism of plant adaptation in stress conditions: Above investigation suggests that the mechanism of toxicity of Hg in plant may have several ways including i) having ability to form complex with water, ii) accumulation in prophyrine ring, iii) accumulation in cell wall, iv) inhibit water movement in the plant, v) inhibition in processes of photosynthesis, vi) production of ROS and vii) distort the biochemical pathway. It was established that Hg may forms complex with water within the plant due to which water molecule will not be available for photolysis. Consequently synthesis of glucose during processes of photosynthesis will adversely effect. Reactive oxygen species is also another aspect of Hg toxicity which cannot be rule out in plant growth. All plants/organisms need energy for their existence that involved with reproduction, growth, or other activities. In plant, photosynthetic apparatus use light energy to produce carbohydrate (glucose) which usually used later on to supply the energy for cell function. Photosynthesis is therefore a process in which the energy in sunlight is stored in the bonds of glucose for later use. The altered concentration of various biochemical contents showed adaptation for survival but lethal effects of Hg were dominants which approximately remediated via adsorption on seaweeds surface.

Conclusion

It was concluded from current investigation that the plants prepared their food in presence of sunlight, stored in the bonds of glucose for later use. But under stress this system fails which was successful repaired at low dose of Hg through the nutrients of seaweeds. Therefore it is strongly recommended that polluted water must be treated before using for irrigation.

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