



CHLOROPHYLL CONTENT OF GRAM (*CICER ARIETINUM L.*) UNDER CADMIUM AND MERCURY TREATMENTS

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ABSTRACT

The influence of Cd and Hg on Chlorophyll 'a', Chlorophyll 'b', total chlorophyll of *Cicer arietinum L.* plants was evaluated grown with or without nitrogen. Cadmium and mercury treatments at 10 and 25 $\mu\text{mol/L}$ affected chlorophyll of gram as compared to control. However, the addition of nitrogen (4 mM/L) somehow minimized the effect of heavy metals. Cadmium and mercury at 10 $\mu\text{mol/L}$ produced significant effect on chlorophylls. While higher concentrations (25 $\mu\text{mol/L}$) significantly reduced chlorophyll content of plants. Nitrogen increased the chlorophyll content of metal treated plants.

Key words: *Cicer arietinum*, Cadmium mercury, Chlorophyll.

INTRODUCTION

Heavy metal contamination affects the biosphere in many places worldwide¹. Metal concentration in soil ranges from less than 1 mg/Kg to high as 100,000 mg/Kg, whether due to geological origin of the soil or as a result of human activity². Excess concentrations of some heavy metals in soils such as Cd (II), Cr (VI), Cu (II), Ni (II) and Zn (II) have caused disruption of natural aquatic and terrestrial ecosystems³.

Use of industrial effluent and sewage sludge on agricultural land has become a common practice in India as a result of which these toxic metals can be transferred and concentrated into plant tissues from the soil. These metals have damaging effects on plants themselves and become a health hazard to man and animals. Above certain concentrations and over narrow range, the heavy metals turn in to toxins⁴. Some heavy metals at low doses are essential micronutrients for plants, but in higher doses they may cause metabolic disorders and growth inhibition for most of the plant species⁵.

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Photosynthesis is highly sensitive to Cd imposition, Chlorophyll being one of the targets as well as enzymes involved in CO₂ fixation^{7,8}.

EXPERIMENTAL

Materials and methods

The seeds of *Cicer arietinum* var. Rachana were obtained from National Seed Corporation, Varanasi (U.P.). The seeds were surface sterilized with dilute solution of sodium hypochloride to prevent any fungal contamination. The sand was thoroughly washed with water and then treated with 2% sodium hypo chloride and dried. The seeds were sown in earthen pots containing equal quantities (2 Kg) of washed and acid treated loamy sandy soil. The soil was treated with long Ashton nutrient solution⁹. Metal treatments of Cd and Hg were prepared using cadmium nitrate and mercuric chloride with concentrations of 10 and 25 µmol/L. Nitrogen was prepared using ammonium nitrate at 5 mM/L concentration. Cd and Hg (control, 10 and 25 µM) were provided along with nutrient solution twice a week. Nitrogen was also provided along with nutrient solution. Three identical sets were maintained during the whole experiment and experiments were conducted in green house to provide controlled conditions.

The samples were taken from three week old seedlings for biochemical analysis. The chlorophyll was estimated by Arnon's method¹⁰ technique using Beckman Du-2 spectrophotometer.

RESULTS AND DISCUSSION

Effect of heavy metals on chlorophyll

Data presented in Table 1 clearly show the effect of Cd and Hg (10 and 25 µM/L) and their interaction with nitrogen (5 mM/L) on chlorophyll content of gram plants. Chlorophyll 'b' was affected more than Chlorophyll 'a'.

Chlorophyll a was reduced up to 1.993 and 1.972 mg/g Fw Chlorophyll 'a' respectively, at 1 µM Cd and Hg treatments in comparison to control (2.190).

Chlorophyll b was reduced up to 1.451 and 1.436 mg/g Fw Chlorophyll b as compared to control (1.614) under Cd and Hg treatments at 1 µM concentrations.

Table 1: Effect of cadmium (Cd) and mercury (Hg) on chlorophyll 'a' content in g (*Cicer arietinum* L.) grown with or without nitrogen (N)

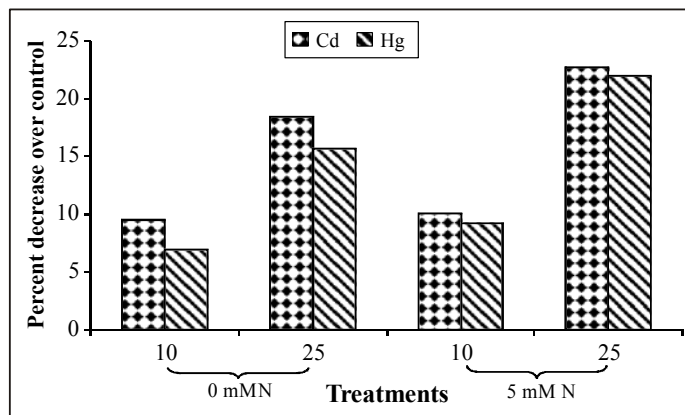
Metals	Concentration (μM)	Chlorophyll 'a'			
		0 mM N	% Age decrease	5 mM N	% Age decrease
	Control	2.240	0	2.456	0
Cd	10	2.027	9.51	2.286	6.92
	25	1.827	18.44	2.072	15.64
Hg	10	2.014	10.09	2.229	9.24
	25	1.730	22.73	1.916	21.99

Table 2: Effect of cadmium (Cd) and mercury (Hg) on chlorophyll 'b' content in g (*Cicer arietinum* L.) grown with or without nitrogen (N)

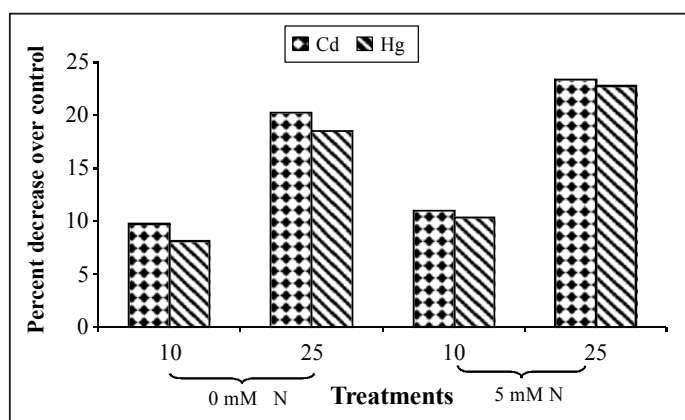
Metals	Concentration (μM)	Chlorophyll 'b'			
		0 mM N	% Age decrease	5 mM N	% Age decrease
	Control	1.664	0	1.893	0
Cd	10	1.501	9.80	1.740	8.08
	25	1.298	20.19	1.544	18.44
Hg	10	1.482	10.94	1.696	10.41
	25	1.275	23.38	1.462	22.77

Table 3: Effect of cadmium (Cd) and mercury (Hg) on Total chlorophyll content in g (*Cicer arietinum* L.) grown with or without nitrogen (N)

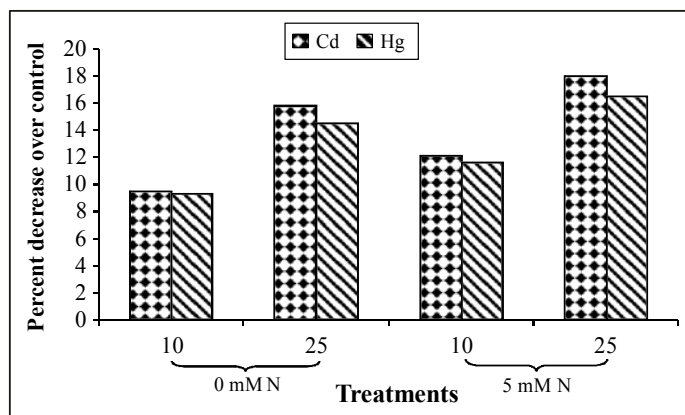
Metals	Concentration (μM)	Total Chlorophyll			
		0 mM N	% Age decrease	5 mM N	% Age decrease
	Control	1.540	0	1.67	0
Cd	10	1.393	9.55	1.514	9.34
	25	1.296	15.84	1.428	14.49
Hg	10	1.353	12.14	1.477	11.56
	25	1.263	17.99	1.394	16.53



Chlorophyll 'a'



Chlorophyll 'b'



Total Chlorophyll

Fig. 1: Effect of heavy metals and nitrogen interaction on chlorophyll content in g plants

At higher concentrations (25 $\mu\text{M/L}$) Chlorophyll a was reduced up to 18% and 23% as compared to control under Cd and Hg treatments. Chlorophyll b was reduced up to 20% and 24% as compare to control under Cd and Hg at 25 $\mu\text{M/L}$ concentration.

Total chlorophyll was reduced up to 10% and 13% under Cd and Hg treatments at 10 $\mu\text{mol/L}$ concentrations. At 25 $\mu\text{mol/L}$ concentrations of Cd and Hg total chlorophyll was reduced up to 16% and 19% as compare to control.

On the contrary, additional supply of nitrogen in nutrient medium some low minimized the effect of heavy metals and proved beneficial for all pigments studied. Increase in Chlorophyll 'b' was more as compared to Chlorophyll 'a'.

Similar results have been obtained by several workers on various crops. Oncel et al.¹¹ found that Cd reduces Chlorophyll 'a' and chlorophyll 'b' in wheat. Heavy metal Cd severely inhibits plant growth and even causes plant death by disturbing the uptake of nutrients and inhibiting photosynthesis via degradation of chlorophyll. Zhang-Pei, et al.¹² reported the reduction in Chlorophyll 'a', Chlorophyll 'b' and total chlorophyll in groundnut by industrial effluents containing heavy metals.

The influence of Cd and Hg on proline content of Gram and their interaction with nitrogen. As compared with control (0.372 mg/g proline), plants showed an increase up to 0.438 and 0.446 mg/g proline under Cd and Hg treatments at 10 $\mu\text{mol/L}$ concentrations. At higher concentrations (25 $\mu\text{mol/L}$) proline content was further enhanced up to 36% and 38% under Cd and Hg treatments.

In this case nitrogen application reduced proline content in plants. At higher concentrations (25 $\mu\text{mol/L}$) proline was reduced upto 15% and 14% over control under Cd and Hg treatment. Our results are in agreement with several workers who obtained similar results on several crops. Accumulation of proline may be due to increased synthesis from glutamate, lower rate of protein oxidation and slowed incorporation of proline in to protein.

CONCLUSION

Based on the results, we conclude that chlorophylls and proline is seriously affected by heavy metals. Chlorophyll 'b' was reduced more as compared to Chlorophyll 'a'. The amount of proline increased in plants under stress caused by metals. Mercury proved more

toxic than Cd at both concentrations. The deleterious effects of heavy metals may be alleviated in plants if provided with appropriate concentration and form of nitrogen in nutrient medium. Further research is needed in order to evaluate the effect of different heavy metals on various crops.

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