



## SHORT COMMUNICATION

### EFFECT OF ZINC ON GROWTH AND NR ACTIVITY IN BLACK GRAM ANTAGONIZED BY MAGNESIUM AND SUCROSE

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The present investigation was designed to evaluate the effects of different doses of zinc (5.3, 6.3, 7.3, 8.3 & 9.3 mg lit<sup>-1</sup>) and combined effect of zinc with magnesium and sucrose on growth of two cultivars (PU-35 & T-9) of black gram (*Vigna mungo* L. Hepper). A concentration dependent decrease in plant height, fresh weight, chlorophyll, carbohydrate and protein content as well as NR activity was observed in both the cultivars. However, proline content increased with increase in doses of zinc. All the doses of combined treatments (zinc with magnesium and sucrose) were able to alleviate reduction caused by zinc but the alleviation was more pronounced with magnesium supplementation than sucrose.

**Key words:** Antagonism, magnesium, sucrose, zinc.

Zinc is a micronutrient and is a component of several enzymes of carbohydrate and nitrogen metabolism, thus involved in various physiological processes of plants (Marschner 1995). The higher concentration of zinc inhibits plant growth and development (Sutter *et al.* 2002, Manivasagaperumal & Vijayaregen 2005, Sinhal 2005, Sinhal *et al.* 2006). Zinc in combination with magnesium and sucrose increases the NR activity. (Verrappa *et al.* 1999, Singh *et al.* 2005). In the city waste water of Bareilly, toxic amount of zinc was detected (Sinhal 2005). The present investigation was therefore, undertaken to study the individual and combined effect of zinc with magnesium and sucrose on growth, NR activity, protein content and proline content in black gram.

The analysis of city waste water of Bareilly city through atomic absorption spectrophotometer shows the presence of Zn<sup>2+</sup> in toxic amount (7.3 mg lit<sup>-1</sup>) (Sinhal 2005). Therefore two lower and two higher doses than that of existing concentration of zinc present in city waste water were taken.

The seeds of two cultivars of *Vigna mungo* (L.) Hepper, PU- 35 & T-9 were presoaked in distilled water and were sown in three separate plots. In first, the irrigation of different doses of zinc (5.3, 6.3, 7.3, 8.3, & 9.3 mg lit<sup>-1</sup>) and in second and third plot, the combined treatment of different doses of zinc with magnesium and sucrose in ratio of 1:1, (5.3, 6.3, 7.3, 8.3 & 9.3 mg lit<sup>-1</sup> of each) was given at the interval of 15 days respectively. The effects of zinc alone and in combination with magnesium and sucrose was studied on plant height, fresh weight, chlorophyll, carbohydrate content, NR activity, protein and proline content of 45 days old plants. The chlorophyll content was determined following the method described by Arnon (1949), carbohydrate content by Morris (1948), protein content by Lowry *et al.* (1951), NR activity by Srivastava (1974) and proline content by Bates *et al.* (1973).

A concentration dependent decrease was noticed in plant height and fresh weight in both the cultivars of *Vigna mungo* (Table 1). The maximum reduction in

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**Table 1.** Effect of zinc and its combined treatment with magnesium and sucrose on growth parameters of black gram

Concentration of treatment (mg lit <sup>-1</sup> )	Plant height (cm)	Weight plant <sup>-1</sup> (g)	Chlorophyll content (mg g <sup>-1</sup> fw)	Carbohydrate content (mg g <sup>-1</sup> fw)
<b>Variety PU-35</b>				
Control	20.35±0.35	19.85±1.22	1.39±0.45	75.25±1.85
Zn <sup>2+</sup> 5.3	18.25±0.46	18.13±0.48	1.25±0.92	68.13±2.69
Zn <sup>2+</sup> 6.3	16.10±0.55	16.65±0.48	1.11±1.25	60.19±1.12
Zn <sup>2+</sup> 7.3	14.45±0.97	14.14±1.25	1.01±2.45	54.49±1.25
Zn <sup>2+</sup> 8.3	12.13±1.25	11.23±2.13	0.85*±1.10	46.61*±2.13
Zn <sup>2+</sup> 9.3	09.37±2.15	9.17±0.52	0.61±1.32	37.29*±1.38
Zn <sup>2+</sup> 5.3 + Mg <sup>2+</sup> 5.3	25.19±1.35	26.66±0.86	2.19±08.95	95.52*±1.92
Zn <sup>2+</sup> 6.3 + Mg <sup>2+</sup> 6.3	24.65±3.17	25.16±0.96	2.01±0.85	91.61*±1.33
Zn <sup>2+</sup> 7.3 + Mg <sup>2+</sup> 7.3	20.19±1.15	23.16±1.29	2.01±0.71	84.62*±3.01
Zn <sup>2+</sup> 8.3 + Mg <sup>2+</sup> 8.3	18.65±1.39	20.11±1.11	1.55±0.8	78.92±3.18
Zn <sup>2+</sup> 9.3 + Mg <sup>2+</sup> 9.3	17.80±3.29	18.10±3.13	1.19±1.13	73.19±1.26
Zn <sup>2+</sup> 5.3 + sucrose 5.3	24.13±3.13	25.15±1.25	2.11±0.82	93.13±2.13
Zn <sup>2+</sup> 6.3 + sucrose 6.3	23.65±2.12	24.24±3.16	1.93±1.85	88.19±4.13
Zn <sup>2+</sup> 7.3 + sucrose 7.3	19.85±3.16	22.13±0.97	1.81±0.81	82.12±2.67
Zn <sup>2+</sup> 8.3 + sucrose 8.3	17.97±0.98	19.54±2.15	1.41±1.89	75.62±1.28
Zn <sup>2+</sup> 9.3 + sucrose 9.3	17.10±0.69	17.13*±3.01	1.02±1.33	71.72±3.93
C.D. at 5%	2.306	1.434	.0234	4.564
C.D. at 1%	3.193	1.984	.0322	6.320
<b>Variety T-9</b>				
Control	20.12±0.87	19.05±1.45	1.33±0.69	73.32±2.85
Zn <sup>2+</sup> 5.3	18.05±1.18	17.85±1.43	1.12±1.92	66.29±1.10
Zn <sup>2+</sup> 6.3	15.96±2.05	16.16±2.41	1.02±1.08	58.21±2.18
Zn <sup>2+</sup> 7.3	13.85±0.93	13.81±1.62	0.88±3.41	52.24±3.12
Zn <sup>2+</sup> 8.3	11.17±3.13	10.19±1.89	0.59±0.93	42.16±3.01
Zn <sup>2+</sup> 9.3	08.15±1.98	08.11±2.11	0.38±1.03	32.19±2.82
Zn <sup>2+</sup> 5.3 + Mg <sup>2+</sup> 5.3	25.03±2.19	25.69±2.49	2.01±0.93	94.14±3.92
Zn <sup>2+</sup> 6.3 + Mg <sup>2+</sup> 6.3	23.95±1.99	24.23±1.96	1.91±1.05	89.29±1.88
Zn <sup>2+</sup> 7.3 + Mg <sup>2+</sup> 7.3	18.98±0.98	22.12±1.17	1.62±0.99	82.29±4.03
Zn <sup>2+</sup> 8.3 + Mg <sup>2+</sup> 8.3	17.67±3.38	19.91±0.89	1.41±1.81	76.13±3.40
Zn <sup>2+</sup> 9.3 + Mg <sup>2+</sup> 9.3	16.99±3.45	17.25±2.63	1.09±1.37	71.47±2.19
Zn <sup>2+</sup> 5.3 + sucrose 5.3	24.10±0.94	24.34±1.33	2.03±0.97	91.98±2.01
Zn <sup>2+</sup> 6.3 + sucrose 6.3	23.05±1.88	23.13±1.93	1.81±1.15	86.18±1.95
Zn <sup>2+</sup> 7.3 + sucrose 7.3	18.91±3.45	21.18±0.97	1.51±1.28	79.26±2.97
Zn <sup>2+</sup> 8.3 + sucrose 8.3	17.17±1.32	17.10±1.15	1.19±1.33	72.36±1.08
Zn <sup>2+</sup> 9.3 + sucrose 9.3	16.65±0.97	17.11±3.21	1.13±0.83	70.28±1.66
C.D. at 5%	2.479	2.507	.0626	6.015
C.D. at 1%	3.432	3.470	.0867	8.329

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**Table 2.** Effect of zinc and its combined effect with magnesium and sucrose on NR activity, protein content and proline content in black gram

Concentration of treatment (mg lit <sup>-1</sup> )	NR activity (μmol NO <sub>2</sub> <sup>-</sup> hr <sup>-1</sup> g <sup>-1</sup> fw)	Protein content (mg g <sup>-1</sup> fw)	Proline content (μmol g <sup>-1</sup> fw)
<b>Variety PU-35</b>			
Control	18.29±1.85	51.96±1.55	0.010±0.83
Zn <sup>2+</sup> 5.3	16.85±1.62	47.81±1.32	0.068±0.93
Zn <sup>2+</sup> 6.3	15.19±2.63	40.21±2.66	0.075±0.83
Zn <sup>2+</sup> 7.3	13.18±3.13	34.19±1.88	0.081±0.99
Zn <sup>2+</sup> 8.3	10.12±1.24	30.69±1.92	0.091±0.81
Zn <sup>2+</sup> 9.3	08.46±1.65	24.62±2.77	0.098±0.49
Zn <sup>2+</sup> 5.3 + Mg <sup>2+</sup> 5.3	26.69±1.82	69.85±1.82	0.010±0.91
Zn <sup>2+</sup> 6.3 + Mg <sup>2+</sup> 6.3	25.49±1.48	67.81±1.49	0.015±0.79
Zn <sup>2+</sup> 7.3 + Mg <sup>2+</sup> 7.3	23.75±1.80	60.12±2.05	0.028±0.96
Zn <sup>2+</sup> 8.3 + Mg <sup>2+</sup> 8.3	20.16±2.29	53.16±1.92	0.032±0.65
Zn <sup>2+</sup> 9.3 + Mg <sup>2+</sup> 9.3	17.98±2.11	49.98±1.32	0.039±0.88
Zn <sup>2+</sup> 5.3 + sucrose 5.3	25.56±1.13	68.10±1.66	0.011±1.02
Zn <sup>2+</sup> 6.3 + sucrose 6.3	24.77±2.98	66.39±1.88	0.017±0.89
Zn <sup>2+</sup> 7.3 + sucrose 7.3	22.02±1.84	57.45±2.25	0.031±0.78
Zn <sup>2+</sup> 8.3 + sucrose 8.3	19.14±1.44	51.69±1.81	0.035±0.88
Zn <sup>2+</sup> 9.3 + sucrose 9.3	16.87±1.00	49.43±3.01	0.040±1.16
C.D. at 5%	1.468	3.871	.000720
C.D. at 1%	2.032	5.361	.000990
<b>Variety T-9</b>			
Control	18.03±0.93	50.85±1.69	0.011±0.69
Zn <sup>2+</sup> 5.3	16.19±1.06	46.43±1.63	0.078±0.92
Zn <sup>2+</sup> 6.3	15.01±2.12	39.26±2.15	0.081±0.81
Zn <sup>2+</sup> 7.3	12.40±1.70	32.16±2.69	0.090±0.99
Zn <sup>2+</sup> 8.3	09.21±3.19	28.83±1.65	0.097±1.19
Zn <sup>2+</sup> 9.3	07.72±1.25	23.29±2.09	0.109±0.88
Zn <sup>2+</sup> 5.3 + Mg <sup>2+</sup> 5.3	25.26±1.32	68.19±3.03	0.012±0.96
Zn <sup>2+</sup> 6.3 + Mg <sup>2+</sup> 6.3	24.81±1.43	66.61±1.92	0.017±0.87
Zn <sup>2+</sup> 7.3 + Mg <sup>2+</sup> 7.3	22.71±2.80	58.63±2.49	0.029±0.84
Zn <sup>2+</sup> 8.3 + Mg <sup>2+</sup> 8.3	19.81±1.81	50.53±1.03	0.033±0.87
Zn <sup>2+</sup> 9.3 + Mg <sup>2+</sup> 9.3	17.62±2.02	48.45±3.10	0.040±1.19
Zn <sup>2+</sup> 5.3 + sucrose 5.3	24.60±1.20	67.42±1.86	0.013±0.86
Zn <sup>2+</sup> 6.3 + sucrose 6.3	23.69±2.18	65.81±1.91	0.019±0.74
Zn <sup>2+</sup> 7.3 + sucrose 7.3	21.01±1.66	55.44±2.78	0.033±0.79
Zn <sup>2+</sup> 8.3 + sucrose 8.3	18.81±0.92	50.05±1.88	0.037±1.25
Zn <sup>2+</sup> 9.3 + sucrose 9.3	16.88±1.39	48.18±1.19	0.042±1.19
C.D. at 5 %	1.410	4.178	.000375
C.D. at 1 %	1.953	6.192	.000517

plant height and fresh weight was at high dose of zinc (9.3 mg lit<sup>-1</sup>) in both the cultivars. In the combined treatment of zinc with magnesium and sucrose, all the doses (5.3, 6.3, 7.3, 8.3, & 9.3 mg lit<sup>-1</sup>) showed promotary effect on plant height and fresh weight as compared to the zinc treatments (Table 1). The higher concentrations of heavy metals have been reported to retard cell division, elongation, differentiation and affect plant growth and development (Kastori *et al.* 1998, Tomar *et al.* 2000). A similar trend of reduction was also noticed in case of chlorophyll and carbohydrate contents. The reduction in chlorophyll and carbohydrate contents by Zn have been noticed by other workers (Tomar *et al.* 2000, Bakiarogloce & Karatagli 2000). Prasad and Prasad (1987) concluded that there are two enzymes, i.e.  $\delta$ -Aminolevulinic acid (ALA) dehydratase and Protochlorophyllide reductatase which are involved in chlorophyll biosynthesis in higher plants are inhibited by heavy metals because these metals binds with functional sulphhydryl (-SH) group of the enzymes. Pedler *et al.* (2004) found that the combined treatment of zinc with magnesium showed promotary effect in wheat and radish.

The NR activity and protein content were also decreased with the increase of concentration of zinc. The alleviation of zinc toxicity was more pronounced in combined treatment of zinc with magnesium as compared to the zinc with sucrose in both the cultivars (Table-2). Inhibition of NR activity under the influences of heavy metals may be multifacial, eg. due to reduced supply of NADPH, disorganization of chloroplast, less NO<sub>3</sub><sup>-</sup> supply to the site of synthesis caused by water stress and direct effect of heavy metals on protein synthesis because they have a strong affinity for functional sulphhydryl (-SH) group of the enzyme (Singh *et al.* 1998).

Puranik and Srivastava (1983) and Vyas and Puranik (1993) observed that sucrose enhances the stability of NR and mobilize endogenous nitrate pool for enzyme activity. Sugars may provide carbon skeleton for amino acid biosynthesis, hence they should increase the activities of the enzymes involved in primary amination. Bose and Mishra (1992) concluded that application of magnesium to soil increases the nitrogen status of plants and ultimately the protein content.

The zinc concentration dependent increase was noticed in proline content. The higher proline content was observed with the highest dose of zinc (9.3 mg lit<sup>-1</sup>). In the combined treatments (zinc with magnesium & sucrose) the proline content was decreased with increase of magnesium and sucrose (Table 2). The accumulation of proline content in heavy metal stressed plants is associated with reduced damage to membranes and proteins (Alia and Mohanty 1997 & Verma 1999).

Thus the present investigation showed that zinc exert toxic effect on all the parameters studied in *Vigna mungo* (L). The concentration dependent increase in proline content is the indication of increase of metal toxicity. Thus the accumulation of proline in plants under the influence of different doses of zinc may be due to defense mechanism but at higher concentration it is not able to alleviate zinc toxicity. In combined treatments (zinc with magnesium & sucrose), the proline content was decreased as compared to zinc treatments. It is an indication of alleviation of zinc toxicity through magnesium and sucrose. The decrease in proline content was more in combined treatment of zinc with magnesium as compared to zinc with sucrose. It shows that magnesium is able to alleviate zinc toxicity more than sucrose.

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