

## Effects of Various Levels of Cement Dust on Seed Germination and Early Seedling Growth in 2 Cultivars of *Helianthus annuus* L.

Farnoosh Zargari and Hasan Hekmat Shoar

Department of Biology, Islamic Azad University, Marand Branch,

P.O. Box 54165-161, Marand, Iran

**Abstract:** In this study, the effects of cement dust-provided from cement factory complex of Soofian located in 30 Km north of Tabriz-Iran-on seed germination and early seedling growth including root, hypocotyl elongation and number of lateral roots in seedlings of 2 cultivars of *Helianthus annuus* L. Armavirusky and Eroflor-have been investigated. The seeds were exposed to 0, 0.42, 0.84, 1.26 and 1.68, 7 (g L<sup>-1</sup>) of cement dust solutions on based of ISTA rules for 7 days. The obtained results showed that cement dust didn't effect significantly on seed germination, root elongation and number of lateral roots in both cultivars compared to control group (p>0.01) but hypocotyl length with increasing concentration of cement dust solution showed a significantly decrease in both cultivars (p<0.01) compared control group seedlings. Hypocotyl growth was a more sensitive endpoint than root growth and number of lateral roots. There was no significant difference in seedling growth between 2 cultivars in similar treatments but it was significant in treatment of 7 g L<sup>-1</sup> of cement solution in germination percent between these cultivars (p<0.01).

**Key words:** Cement dust, heavy metals, seed germination, seedling growth, Sunflower

### INTRODUCTION

Heavy metals are important environmental pollutants that pose a major human health problem. Pollution of the biosphere with toxic metals has accelerated dramatically since the beginning of the industrial revolution (Nriogo, 1979). Some heavy metals such as Cu, Zn, Ni are essential micronutrients for plants but are toxic to organisms at high concentrations (Munzuruglu and Geckil, 2002). Plants are sometimes exposed to non-essential heavy metals, including Hg<sup>2+</sup>, Cd<sup>2+</sup> and Pb<sup>2+</sup> that are present in soil and water naturally or as contaminants from human activities such as mining and smelting of metalliferous ores, municipal wastes, fertilizers, pesticides and sewage (Kabata-Pendias and Pendias, 1989). Plant bioassays have been used to assess ecotoxicity of soil contaminated with a range of chemicals (Gorsuch *et al.*, 1991). Many studies have demonstrated that heavy metals influence the seed germination and early seedling growth of plants. Seed is a stage in the plant life cycle that is well protected against various stress. However, Soon after imbibition and subsequent Vegetative developmental processes, they become stress-sensitive in general. Therefore, seeds are thought to carefully monitor such external parameters as light, temperature and nutrient in

order to maintain the protective state until external condition become favorable for following developmental processes (Karssen, 1982; Pritchard *et al.*, 1993; Bungard *et al.*, 1997). Many studies have demonstrated that heavy metals influence seed germination and early seedling growth of plants, so a few examples are summarized: effects of Cu on *Helianthus annuus* L. (Lin *et al.*, 2003), Cu on *Brassica oleracea* (Chatterjee and Chatterfee, 2000), Cu on *Lemna trisu laca* (Prasad *et al.*, 2001), Pb on higher plants (An *et al.*, 2004), Pb on *Lemna minor* (Mohan and Hosetti, 1997), Cu and Pb on *Allium cepa* (Wierzbicka, 1999), Cu and Pb on *Avicennia marina* (Mac Farlane and Murchett, 2002), Pb, Cu on 4 crops (An, 2006), Cu, Zn, Hg and Pb on *Triticum aestivum* and *Cucumis sativus* (Munzuruglu and Geckil, 2002), Cu, Zn, Pb and Cd on *Brassica pekimensis* (Song *et al.*, 2002), Zn, Cu, Pb, Cd and Hg on *Arabidopsis thaliana* (Weiqliang *et al.*, 2005), Cd and As on *Triticum aestivum* (Liu and Zhang, 2007). It is well documented that the presence of both alkali and heavy metals in composition of cement dust-released from the cement factories-affect on growth and development of the plants by changing physicochemical properties of soil and perturbation of seed germination and metabolism in neighbouring localities plants. The porpose of this study is to assay

effect of different levels of cement dust on Seed germination and seedling growth of 2 cultivars of *Helianthus annuus*, compare these cultivars relative to this environmental stress and determine resistant cultivar.

### MATERIALS AND METHODS

**Plant materials and chemicals:** The seeds of 2 cultivars of *Helianthus annuus*-Armavirsky and Eroflor-were provided from agriculture organization (Tabriz-Iran). Cement dust for germination experiments was Provided from Soofian factory complex, located in 30 Km north of Tabriz-Iran. The experimental analysis of cement dust carried out by XRF analysis with using spectrophotometer Model: SRS 200 (Table 1) and Chemical analysis of Cement dust showed presence of alkali cations that are shown in Table 2. Cement dust Solutions were made in several concentrations (0.42, 0.84, 1.26, 1.68, 7) g L<sup>-1</sup> in distilled water (dH<sub>2</sub>O) and then to measure pH and EC of these solutions used a digital pH meter (Model: metrohm 827) and EC meter (Model: metrohm 712, respectively (Table 3). dH<sub>2</sub>O was used as a control treatment.

**Germination tests:** Prior to germination, dry seeds were Surface strilized with 1% Sodium hypochlorite Solution for 20 min and then rinsed with distilled water. Seed germination was tested on moist filter Paper. A piece of filter paper was placed on Petri dish and moistened with 5.0 mL of cement dust solution. Controls were maintained by moistening the filter Paper with 5.0 mL distilled water. Germination tests were carried out triplicate samples in both cultivars (each containing 25 seeds) under conditions light and dark period 12 h at 20-30°C. In the third day after starting test, the seeds were Scored as germinated when the breakage of seed coat Was visible. In 7th day some parameters of seedling growth (root, hypocotyl length and number of lateral roots were measured (Draper, 1985).

Table 1: Various of heavy metals in cement dust

<sup>1</sup> XRF <sup>2</sup> LiF 110	<sup>3</sup> XRF <sup>2</sup> LiF 100
Fe <sup>a</sup>	Fe <sup>a</sup>
Zn	Zn
Rb	Rb
Sr	Sr
Ni	Ni
Mn	Mn
Kr	Cu
Br	Kr
Cr	Br
Cd	Cr
Pb	Cd
Hg	Se
Pt	Co
Au	Mo
U <sup>b</sup>	Ag

a: High concentration,b: lack of element; 1: X-Ray Fluoresc; 2: Feloride Litium crystal with 100 (mm) plate; 3: Feloride Litium crystal with 110 (mm) plates

Table 2: Analysis of cement dust

Compounds of cement dust	Concentration (% w w <sup>-1</sup> )
Sio <sub>2</sub>	9.49
Al <sub>2</sub> O <sub>3</sub>	2.13
Fe <sub>2</sub> O <sub>3</sub>	1.88
CaO	43.72
MgO	1.33
SO <sub>3</sub>	2.14
Na <sub>2</sub> O	0.52
K <sub>2</sub> O	8.50
Cl	0.16

Table 3: EC and PH contents of cement dust solutions in various concentrations

EC (ms cm <sup>-1</sup> )	pH	Concentration of cement dust solution (g L <sup>-1</sup> )
0	7	0.00
4.81	11.09	0.42
8.56	11.23	0.84
11.92	11.39	1.26
15	11.48	1.68
17.76	11.59	7.00

**Statistical analysis:** Statistical analysis was performed by using Microsoft-Excel, spss (ver. 11). The data represent means calculated from 3 replicates. A least significant difference test (Dunken-test) was employed for comparison of the changes at p<0.01.

### RESULTS

**Determine of composition and some chemical characteristics of cement dust:** In this study, the first step to assess effect of cement dust on seed germination and early seedling growth, was, analysis of cement dust to determine its composition and some physicochemical characteristics. The result of this analysis indicated presence various heavy metals in different concentrations (Table 1) and alkali cations (Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>) are shown in Table 2. Because of these cations, cement dust had high pH and EC (Table 4).

**Effects of cement dust on germination:** To assess effects of cement dust on germination, we made cement dust solution in several concentrations (0-7 g L<sup>-1</sup>) and determined their pH and EC. With increasing of solutions, the pH and EC increased (Table 3). The seeds of both cultivars were treated with these different concentration solutions. Seed germination percent and average of germination rate were measured at the end of third day of starting test. The obtained results of analysis variencie didn't show significant changes of germination in treated seeds compared to control group seeds in any concentration of solution in both cultivars (Fig. 1a and b) but there was significant difference in treatment of 7 g L<sup>-1</sup> of cement dust solution in germination percent between these cultivars (p<0.01).

Table 4: Some characteristics of cement dust

pH	EC(ms cm <sup>-1</sup> )	Density	Cement dust 1 (g)	
			Insoluble substances	Soluble substances
11.59	17.76	0.64	0.86	0.14

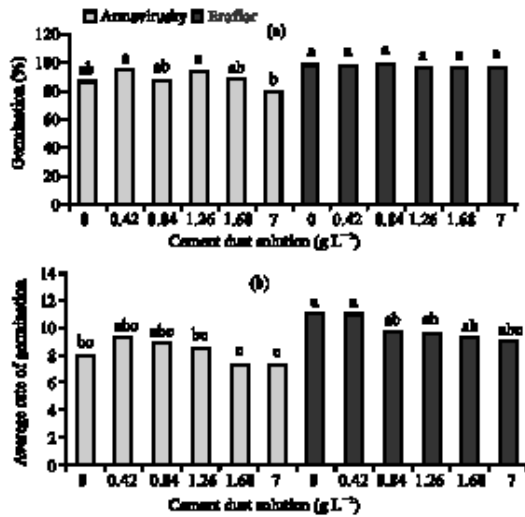


Fig. 1: Seed germination (a) and average of rate germination (b) of 2 cultivars of *Helianthus annuus*

These results similar to results of previous works by (Gyana *et al.*, 2000; An, 2006; Lyle, 2003; Weiqiang *et al.*, 2005; Song *et al.*, 2002).

**Effects of cement dust on early seedling growth:** Root and hypocotyl elongation of seedlings in 7th day of treatment were measured (Fig. 2). The results indicated that with increasing concentration of cement dust solutions, root and hypocotyl length decreased in both cultivars compared to control group seedlings. This reduction in hypocotyl length was significant ( $p < 0.01$ ) but non-significant in root length (Fig. 3a and b). The previous works by (Song *et al.*, 2002; Lyle, 2003; Liu and Zhang, 2007; Muzuroglu and Geckil, 2002; An, 2006), showed similar results with a difference in more sensitivity of hypocotyl relative to root. In this study, the number of lateral roots in control and treated seedlings also were measured. In all of treatments a non-significant reduction in number of lateral roots compared to control seeds was observed. An increase in number of lateral roots in treatments of 1.26, 1.68, 7 g L<sup>-1</sup> of cement dust solutions relative to treatments of 0.42 and 0.84 g L<sup>-1</sup> of solutions was observed in Armavirsky and this increase in Eroflor was observed in treatments of 1.68 and 7 g L<sup>-1</sup> relative to lower concentration treatments (Fig. 3c).

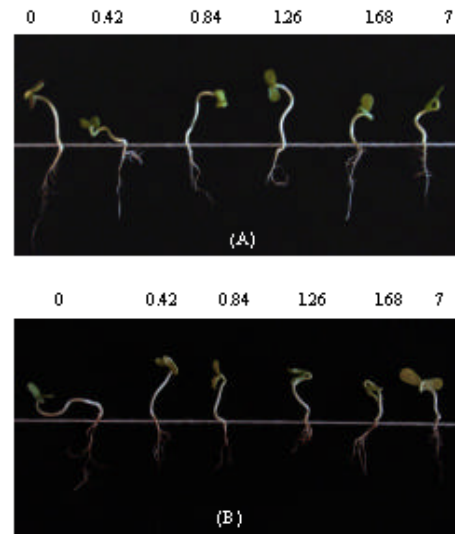


Fig. 2: Treated seedlings with different concentrations of cement dust solution compared to control seeds in 7th day of starting germination test (A): Armavirsky (B): Eroflor

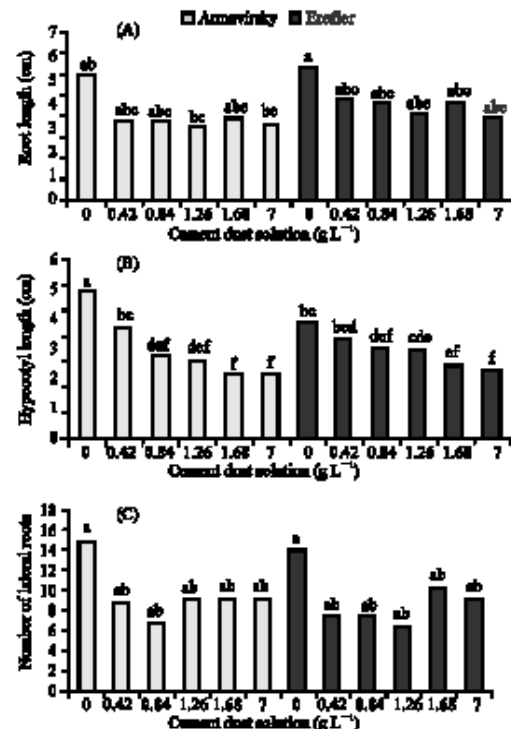


Fig. 3: Root length (A), hypocotyl length (B) and number of lateral roots (C) of 2 cultivars of *Helianthus annuus* (Armavirsky and Eroflor) in the presence of different concentrations of cement dust solutions in the 7th day of germination test

## DISCUSSION

Heavy metals and alkali cations in cement dust can influence the plants of vicinity areas cement companies. This effect can be occurred in different stages of growth and development of life cycle in plants.

Our results about germination support the idea that tissues covering the embryo play a role in selective penetration of different heavy metals into seeds (Weiqiang *et al.*, 2005). This was first suggested by the fact that seeds still germinated in the presence of high concentrations ( $7 \text{ g L}^{-1}$ ) of cement dust solution but the subsequent seedling growth was severely inhibited at higher concentration of cement dust solution.

With the increases in the concentration of solutions, hypocotyl length reduced more significantly than the other endpoints and the reduction followed the order: hypocotyl length > root length, number of lateral roots > average rate of germination > germination percent.

It has been thought that heavy metals are taken up by the cell through metal transporters. Several such transporter proteins have recently been identified (Clemens, 2001; Kjar *et al.*, 1998). It is at moment not clear which transporters contribute to the uptake of metals such as  $\text{Pb}^{2+}$  and  $\text{Zn}^{2+}$  in seeds and cations such as  $\text{Ca}^{2+}$  how affected their toxicities.

## CONCLUSION

Our results indicated that seed germination in a heavy metal environment was not a reliable indicator for metal tolerance of the plants. Regarding to results of this study, we might be get a conclusion that Eroflor in compared to Armavirusky is tolerant and could be suitable for cultivation in vicinity areas of cement company.

## ACKNOWLEDGEMENT

This study is the part of thesis of the first author. We thank management Tabriz University and Soofian cement company for their assistance with laboratory experiments. (Amavirusky and Eroflor) in the peresence different concentrations cement dust solutions in third day of germination test.

## REFERENCES

An, J.Y., Y.M. Kim, T.I. Won and S.W. Jeong, 2004. ombined effect of copper, cadmium and lead upon *Cucumis sativus* growth and bioaccumulation. *Sci. Total Environ.*, 29; 326 (1-3): 85-93.

- An, Y.J., 2006. Assessment of comparative toxicities of lead and copper using plant assay. *Chemosphere*, 62: 1359-1365.
- Bungard, R.A. *et al.*, 1997. Effects of chilling, light and nitrogen containing compounds on germinations, rate of germination and inhibition of *Clematis vitalba* L. *Ann. Bot.*, 79: 643-645.
- Chatterjee, J. and C. Chaterfee, 2000. Phytotoxicity of cobalt, chromium and copper in cauliflower. *Environ. Pollut.*, 109: 69-74.
- Clemens, S., 2001. Molecular mechanisms of plant metal tolerance and homeostasis. *Planta.*, 212: 475-786.
- Draper, S.R., 1985. *Inter. Rules Seed Testing*. *Seed Sci. Technol.*, 13 (2): 322-323.
- Gyana, R.R., S. Sanghamitra and D. Premananda, 2000. Effects of chromium and nickel on germination and growth in tolerant and non-tolerant populations of *Echinochloa colona* (L.). *Link. Chemosphere*, 40: 855-859.
- Gorsuch, J.W. *et al.*, 1991. *Plants for Toxicity Assessment*. 2, ASTM STP 1115. ASTM, Philadelphia.
- Kabata-Pendias, A. and H. Pendias, 1989. *Trace elements in the soils and plants*. Florida: CRC Press.
- Karssen, C.M., 1982. Seasonal Patterns of Dormancy in Weed Seeds. In: Khan, A.A. (Ed.). *The physiology and Biochemistry of seed Development, Dormancy and germination*. Elsevier Biomedical Press, Amesterdam. The Netherlands, pp: 243-270.
- Kjar, C., M.B. Pedersen and N. Elmegaard, 1998. Effect of soil copper on black bindweed (*Fallopia convolvulus*) in the laboratory and in the field. *Arch. Environ. Cont. Toxi.*, 35: 14-19.
- Lin, J., W. Jiang and D. Liu, 2003. Accumulation of copper by roots, hypocotyls, cotyledons and leaves of sunflower (*Helianthus annuus* L.). *Bioresour. Technol.*, 86: 151-15.
- Liu, X.L. and S.Z. Zhang, 2007. Intraspecific differences in effects of co-contamination of cadmium and metal uptake by Wheat. *Environ. Sci.*, 19: 1221-1227.
- Lyle, E.C., 2003. Seed germination of Anise, Caraway and Fennel in heavy metal contaminated solutions. *J. Herbs Spices Med. Plants*, 10 (3): 83-93.
- MacFarlane, G.R.M. and M.D., Urchet, 2002. Toxicity, growth the grey mangrove *Avicennia marina* (Forsk). *Vierh. Marine Environ. Res.*, 54: 65-84.
- Mohan, B.S. and B.B. Hosetti, 1997. Potential phytotoxicity of lead and cadmium to *Lemna minor* grown in sewage stabilization ponds. *Environ. Pollut.*, 98: 233-238.
- Munzuroglu, O. and H. Geckil, 2002. Effects of metals on seed germination, root elongation and coleoptile and hypocotyls growth in *Triticum aestivum* and *Cucumis sativus*. *Arch. Environ. Contam. Toxi.*, 43 (2): 203-213.

- Nirogo, G.O., 1979. Global inventory of natural and anthropogenic emissions of trace metals to the atmosphere. *Nature*, 279: 409-411.
- Pitchard, H.W. *et al.*, 1993. Influence of temperature on seed germination and the nutritional requirements for embryo growth in *Arum maculatum* L. *New Phytol.*, 123: 801-809.
- Prasad, M.N.V. *et al.*, 2001. Physiological responses of *Lemna trisulaca* L. (duckweed) to cadmium and copper bioaccumulation. *Plant Sci.*, 161: 881-889.
- Song, Y., H. Xu, L. Ren, P. Gong and Q. Zhou, 2002. Eco-toxicological effects of heavy metals on the inhibition of seed germination and root elongation of Chinese cabbages in soils. *Huan Jing Ke Xue*, 30, 23 (1): 103-107.
- Weiqiang, L., M. Khan, S.H. Yamaguchi and Y. Kamiya, 2005. Effects of heavy metals on seed germination and early seedling growth of *Arabidopsis thaliana*. *Plant Growth Regulation*, 46 (1): 45-50 (6).
- Wierzbicka, M., 1999. Comparison of lead tolerance in *Allium cepa* with other plant species. *Eviron. Pollut.*, 104: 41-52.