



Decreasing Bio-toxicity of Fume Particles Produced in Welding Process by *Aloe vera* L

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ABSTRACT

As *Aloe Vera* L. can easily grow in Iran climates and due to vast probable contaminations from Iranian Natural Gas Transmission Pipelines Industry, the main purposes of this study were to decrease the risk levels for welders exposed to welding heavy metals in Iranian Natural Gas Transmission Pipelines Industry and determine the potential ability of *A. vera* for cleaning up Contaminated welding sites soil and the probable capability of this plant to phytoextract different metals (Chrome, Nickel, Copper, Cobalt, Lead and Cadmium) and also determine metal transfer factors from soil (TFS) by *A. Vera* in order to ascertain its phytoremediation potential. In winter and spring 2015, composite soil samples were collected from depth of 0-40 cm from soil samples of 5 sites from 10 sub sites Assaluyeh (Bushehr Province), Omidieh (Khuzestan Province) in Iran. The soil for the survey mixed thoroughly, then transferred to greenhouse in Tehran County. In order to assess amount of heavy metals transfer from soil to plant (shoot and root), translocation factor was determined by dividing metal concentration at shoot by its concentration at root. Metal contents were detected by Atomic Absorption Spectrophotometer by wet digestion method in Research Laboratory in Pharmaceutical Sciences Branch University. Results showed that *A. Vera* transition factors for all heavy metals in treated soil were higher than one and *A. Vera* can up-take lead and copper after 20 days ($p < 0.01$) more than other studied metals. The best result of Cadmium phytoextract was in pH of soil 6.3 by 40 days growth of plants. The maximum Cobalt, Copper and Nickel uptake rate was in pH= 6.1, 5.9 and 6.3 respectively by 40 day grown *A. Vera*.

Key words: *Aloe Vera* L., Phytoextraction, Heavy metals, welding, Clean-up soil.

INTRODUCTION

Arc welding is widespread technological process in modern manufacturing, but there are

deficiencies such as formation of aero-disperse toxic particles of the so-called welding fume. Therefore, studying of the welding fume formation is necessary to minimize the harm superimposed by the welding

process¹. Welding is the principal industrial process used for joining metals, but at the same time, it's the significant source of toxic fumes and gases emission. With the advent of new types of welding procedures and consumables, the number of welders exposed to welding fumes is growing constantly in spite of the mechanisation and automation of the process². Welding fumes contain heavy metals, such as chromium, manganese, and nickel. Worldwide industry lays down an estimated one million tons of weld metal annually. Based on an average fume production of 0.5% of weld metal, an estimated five thousand tons of fume are produced annually. With the advent of new types of welding procedures and consumables, the number of welders exposed to welding fumes is growing constantly in spite of the mechanisation and automation of the process. Presently, some 3 million persons from different professional backgrounds are directly subjected to welding fume and gas action². The materials typically found in welding fume include aluminum, beryllium, cadmium oxides, chromium, copper, fluorides, iron oxide, lead, manganese, molybdenum, nickel, vanadium, and zinc oxides which cause respiratory diseases and cancer³. Chromium is an element present in the consumables and base material of stainless steels, heat-resisting steels, some creep-resisting steels, some high nickel alloys, and armor plate. It may also be present in some consumables used for hard facing. When arc welding takes place on materials containing chromium, or by using consumables containing chromium, some of the chromium will be volatilized and escape from the protective gases in and around the arc. This metal vapor will be oxidized by the atmosphere to give particulate fume. Chromium can be present in fume in different forms: chromium in metallic form (valence state 0), trivalent form (Cr III) and hexavalent form (Cr VI). Trivalent chromium occurs widely in nature and is an essential nutrient required by the human body to promote the action of insulin in body tissues. Chromium as a pure metal has no reported human or environmental toxicity effects. Both acute and chronic toxicity of chromium are mainly caused by hexavalent chromium compounds (Cr VI). Hexavalent chromium is considered the most hazardous of all forms, and in welding fume it is a suspected human carcinogen. DNA damage in welders has been associated with hexavalent

chromium exposure⁴. This is consistent with the classification of hexavalent chromium as a human lung carcinogen⁵. Metallic nickel and certain soluble nickel compounds as dust or fume cause sensitization dermatitis and probably produce cancer of the paranasal sinuses and the lung; nickel fume in high concentrations is a respiratory irritant⁶. Metal toxicity is influenced not only by metal concentration, but also by mineral composition and organic substances in the soil, pH, redox-potential and presence of other metals in the soil⁷. Recently appeared paper describing three basic hallmarks to distinguish hyper-accumulators from related non-hyper-accumulating species : a strongly enhanced rate of toxic metal uptake, a faster root-to-shoot translocation and greater ability to detoxify and sequester metals in the leaves⁸. An interesting breakthrough that has emerged from comparative physiological and molecular analyses of hyper-accumulators and related non-hyper-accumulators is that most key steps of hyper-accumulation rely on different regulation and expression of genes found in both kinds of plants. In particular, a determinant role in driving the uptake, translocation into the leaves, sequestration in vacuoles or cell walls of great amounts of toxic metals, is played in hyper-accumulators by constitutive overexpression of genes encoding trans-membrane transporters. Phytoremediation is the biotechnological application of plants to detoxify pollutants, and is an ideal and modern technique for environmental clean-up. Regarding the vast industrial waste materials and sewages from a lot factories and different chemical fertilizers and pesticides have caused contamination of soils in capital city Tehran⁹⁻¹³. Rapid expansion and increasing sophistication of various industries in the last century has remarkably increased the amount and complexity of toxic or hazardous substances, such are heavy metals, radionuclides, organic and inorganic wastes, pesticides, etc. which may be bio-remediated by suitable organisms (plants and microbes). This technology was termed as bioremediation or phytoremediation¹⁴. Phytoremediation is environmental-friendly, cost-effective and natural green biotechnology for the removing xenobiotic, including toxic metals, from the environment using some species of the plants. The land in Iran is very fertile, providing favorable conditions for various medicinal plants to grow. Since ancient times, local

people in every part of Iran have grown *Aloe Vera* in their home gardens, using the leaves of *A. Vera* to aid in healing wounds. *A. Vera* is cultivated in many places in Iran but not in wide range. About 90% of the plants are estimated to come from wild harvest¹⁵⁻¹⁶. *A. Vera* has been on this earth for millennia and it is a member of the Liliaceae family. The plant *Aloe vera* Linne, also known as *Aloe barbadensis* Miller, and *Aloe vulgaris* Lamarck¹⁷. According to a report from Department of the Environment Tehran, I.R. Iran in July 2005, some significant sources of Lead and cadmium are: Nickel-Cadmium Batteries, Cadmium pigments, Cadmium stabilizers, Cadmium Coating, fossil fuels, cement, phosphorous fertilizers, Lead batteries, Glasses & ceramic industries, paint manufacturers, Cadmium electronic compounds, Metal plating, Factories with the process of extraction, production and concentration of Lead ore, Industrial wastewater, solid waste and Municipal waste waters¹⁰. Nowadays, one of the most important requirements is the health aspects associated with welding and cutting and the industry is continuing its research to evaluate the effects of the welder's exposure to typical constituents of welding fumes and gases and its impact on what concerns climatic changes.

Risk assessment is the foundation for recommended occupational exposure limits designed to protect the safety and health of workers. The risk assessment process includes many phases including hazard identification, dose-response assessment, exposure assessment, and risk characterization. The risk assessment is a useful tool to improve occupational health and safety policy and decision-making process for control approaches¹⁸. Control approach is type of approach needed to achieve adequate control¹⁹. Some studies reported that Iranian Natural Gas Transmission Pipelines Industry from four cities in Iran including Assaluyeh (Bushehr Province), Omidieh (Khuzestan Province), Loshan (Gilan Province), and Borujen (Chaharmahal and Bakhtiari Province) are exposed to aero-disperse toxic particles intensively²⁰.

As *A. Vera* can easily grow in Iran climates and due to vast probable contaminations from Iranian Natural Gas Transmission Pipelines

Industry, the main purposes of this study were to decrease the risk levels for welders exposed to welding heavy metals in Iranian Natural Gas Transmission Pipelines Industry and determine the potential ability of *A. Vera* for cleaning up Contaminated welding sites soil and the probable capability of this plant to phytoextract different metals (Chrome, Nickel, Copper, Cobalt, Lead and Cadmium) and also determine metal transfer factors from soil (TFS) of *A. Vera* in order to ascertain its phytoremediation potential.

MATERIAL AND METHODS

Collection of sample

Mature healthy and fresh leaves of 150 *A. Vera* samples were collected from in Agricultural Center Library in Iran. Leaves were taken and washed with fresh water.

Sample Preparation

The leaves were cut in to small pieces and their thick epidermis was removed. The solid gel of the leaf was homogenized and dry ashing method was adopted by placing the properly dried and ground plant sample in to the vitresile crucible overnight in an electric muffle furnace maintaining the temperature between 410-440 °C. This ash sample destroys all the organic material from the sample. The ash was removed from crucible and allowed to dry in desiccator. The yield of ash was approximately 5 g/ 100 g.

Study Area

In winter and spring 2015, composite soil samples were collected from depth of 0-40 cm from soil samples of 5 cities from 10 sub cities Assaluyeh (Bushehr Province), Omidieh (Khuzestan Province) in Iran. The soil for the survey mixed thoroughly, then transferred to greenhouse in Tehran County. This research was conducted of industrial pollution from Iranian Natural Gas Transmission Pipelines Industry in Khuzestan and Bushehr provinces due to the soil contamination from welding processes which they probably contaminated by very high concentrations of Cadmium, Nickel, Cobalt, lead and chrome in these areas (20). According to the heavy metal contents in the soil of this area, a shallow composite soil samples (depth of 0-40 cm) were collected of

heavy metal contaminated soil of polluted area and this soil sample put in a greenhouse in the county of Tehran and plants grown and not grown in it by different pH's after every 10 days in 60 day studying.

Analysis of Samples

A. Vera (one month old plants) were grown in a local nursery until transplant into the research study. For metal analysis 2g of shoots along with leaves and roots of 50 numbers of plants sample were taken separately in every ten day in this study during 60 days. 8ml of concentrated nitric acid (65%) and 1 ml of perchloric acid (70%) was added and 1 ml hydrogen peroxide^{12,14}. Application of concentrated HNO₃ along with thirty percent hydrogen peroxide H₂O₂ (Merck) for mineralization of samples to the complete digestion of samples (12-14, 21) following Environmental Protection Agency (EPA) Method 3052 was done.

All glassware and plastic containers used were washed with liquid soap, rinsed with water, soaked in 10% volume/volume nitric acid at least overnight, and rinsed abundantly in deionized water and dried in such a manner to ensure that any contamination does not occur. Five-point calibration curves (five standards and one blank) were constructed for each analyte. The calibration curve correlation coefficient was examined to ensure an $r^2 \geq 0.998$ before the start of the sample analysis. The digested samples were diluted with 10% HNO₃ and brought up to 50 mL and analyzed by a graphite furnace atomic absorption spectrophotometry, (GFAAS). The measurements were performed using a Perkin Elmer Pin A Aclé 900 T atomic absorption (AA) spectrophotometer and using at least five standard solutions for each metal. All recoveries of the metals studied were greater than 95%. Digestion followed by the measurement of total concentrations of Cd using Atomic Absorption Spectrophotometer using an air-acetylene flame for heavy metals: Pb, Cd, Co, Cu, Cr, Ni and Cu. All necessary precautions were taken to avoid any possible contamination of the sample as per the AOAC guidelines Physical and chemical properties and concentrations of heavy metals (Chrome, lead, Cadmium, Copper, Cobalt and Nickel) in soils before and after planting *A. Vera* and also after the growth period (in every 10 days) measured²¹⁻²⁶. In order to assess amount of heavy metals transfer from soil to plant (shoot

and root), translocation factor was determined by dividing metal concentration at shoot by its concentration at root²⁸. Different parts of plant samples (roots and leaves) were separated and washed and digested by wet method according the standard protocol for measuring Cadmium and Lead. The uptake rate is given by the following equation (12, 14,21).

$$U = (TSCF) (T) (C) \quad (1)$$

Where

U = uptake rate of contaminant, mg/day

TSCF = transpiration stream concentration factor, dimensionless

T = transpiration rate of vegetation, L/day

C = aqueous phase concentration in soil water or groundwater, mg/L.

The ratios were higher than one it was considered as suitability of plant at that condition for use in phytoremediation. The enrichment factor (EF) has been calculated to derive the degree of soil contamination and heavy metal accumulation in soil and in plants growing on contaminated site with respect to soil and plants growing on uncontaminated soil^{12,14,27}. Metal contents were detected by Atomic Absorption Spectrophotometer by wet digestion method in Research Laboratory in Pharmaceutical Sciences Branch University.

Statistical analyses

SPSS (V. 18) software was used for all statistical analyses. The level of significance was taken as $p < 0.05$.

RESULTS

Chemical extraction of the soil profile before and after planting *A. Vera* samples during 60 days is shown in the figure 1 in pH= 5.6-7.1, as plant availability of certain heavy metals depends on soil properties such as soil and contains exchange capacity and on the distribution of metals among several soil fractions. All the soil data are expressed on a dry basis.

Results showed that *A. Vera* transition factors for all heavy metals in treated soil were higher than one and *A. Vera* can up-take lead,

Table 1: Cadmium content (mg/kg DW) remaining in contaminated soil A. Vera cultivated

Time	Cd content(mg/kg WD) in soil	Cd content (mg/kg DW) in Aloe Vera Leaves	Cd content (mg/kg DW) in Aloe Vera Root
0	8.001	0.0287	0.2218
10	6.8871	1.0022	2.4444
20	5.1032	3.0176	3.0562
40	3.2211	3.8876	3.6667
60	2.8642	3.897	3.7891

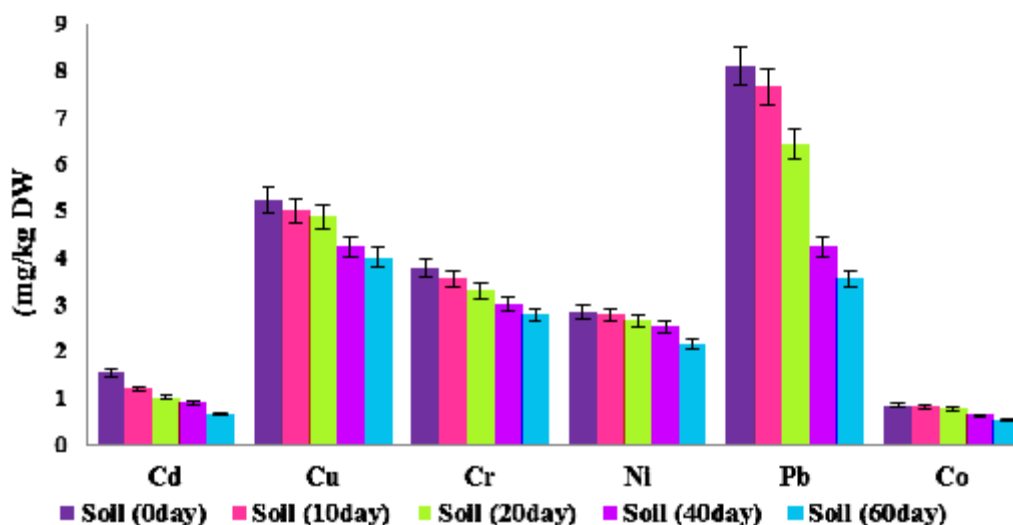


Fig. 1: Heavy metal contents (mg/kg DW) in soil treated by *A. Vera* L. during 60 days

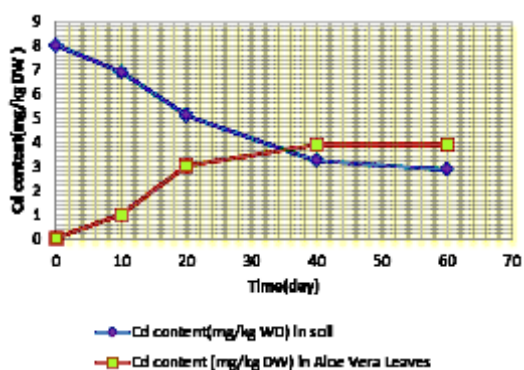


Fig. 2: Cadmium content (mg/kg DW) remaining in contaminated soil *A. Vera* treated

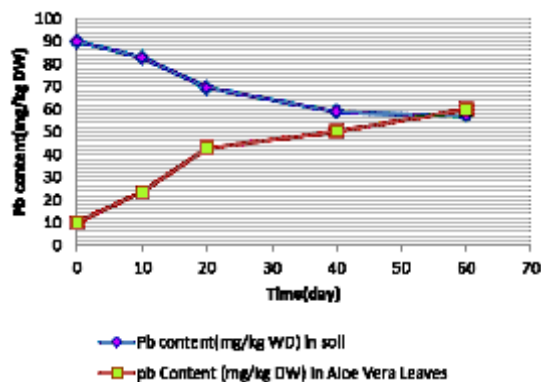


Fig. 3: Lead content (mg/kg DW) remaining in contaminated soil *A. Vera* treated

Cadmium and copper after 20 days ($p < 0.01$) more than other studied metals. In figures 2 and 3, the phytoremediation of lead and Cadmium trend by this plant indicates that *A. vera* cultivated in the soil can be considered as a suitable hyper-accumulator by its relatively large ratio of biomass concentration of the contaminant to soil concentration.

As expected the additional number of plants cultivated in sample soils increased the portion of Pb, Cd, Zn, and Cr. The heavy metals uptake rate by this plant is significantly affected by number of plants cultivated as for lead uptake ($p < 0.005$) while for Cd and chrome the p -value was less than 0.01. The percentage of cadmium uptake by leaves and roots of this plant has been demonstrated in table 1, due to number of harvestings in soil pH = 6.3 which shows that in Figure 2 it has been indicated that younger plants had more potential to uptake Nickel and Copper and concentrate them than older plants as the translocation factor in third harvest and after 50 days conditions were decreased which indicates that metal concentrations in shoots were higher than roots and the plant is suitable for phytoremediation. A highly significant, although low, positive correlation ($r = 0.52$, $p = 0.01$, $n = 200$) was found between Lead and Cadmium and chrome of the greenhouse-grown *A. Vera*, compared to a non-significant and much lower correlation between the three other heavy metals: Nickel, Cobalt and Copper.

CONCLUSION

Plants respond to stress induced by cadmium using some defense reactions such as immobilization, exclusion, accumulation of cadmium ions in vacuoles, synthesis of phytochelatins and stress proteins as well as production of ethylene²⁸. Immobilization in both, cell wall and extra cellular polysaccharides (mucilage, callose) is the first barrier against Cd^{2+} transport into the root²⁹.

Shanker *et al.* in 2005 published review article devoted to the chromium toxicity in plants. It was stressed that toxicity of Cr to plants depends on its valence state: Cr(VI) is highly toxic and mobile whereas Cr(III) is less toxic. Since plants lack a specific transport system for this metal, it is taken up by varied transporters of essential ions such as sulphate or iron. Toxic effects of Cr on plant growth and development include alterations in the germination process as well as the growth of roots, stems and leaves, which may affect total dry mass production and yield. Cr also causes deleterious effects on plant physiological processes such as photosynthesis, water relations and mineral nutrition. Metabolic alterations due to Cr exposure have also been described in plants by a direct effect on enzymes or other metabolites or by its ability to generate reactive oxygen species which may cause oxidative stress. These authors noticed that Cr, in contrast to other toxic metals like Cd, Pb, Cu or Al, has received little attention from plant scientists. Its complex electronic chemistry has been a major hurdle in unraveling its toxicity mechanism in plants. Our results for chrome translocation indicate that *A. Vera* has high potential for taking up this toxic heavy metal and Translocation factor in all conditions were higher than one which proves that metal concentrations in shoots were higher than roots and the plant is suitable for phyto-remediation and the heavy metals uptake rate by this plant is significantly affected by time of growing plant as for lead uptake ($p < 0.01$) while for Cd and Nickel the p -value was less than 0.03.

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Conflicts of Interest

None of the authors have any conflicts of interest associated with this study.

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