Metodologia (Methodology)

Phytohyperaccumulator-AMF (arbuscular mycorrhizal fungi) interaction in heavy metals detoxification of soil

Interação entre hiperacumuladores-FAM (fungos arbusculares micorrizais) na desintoxicação de solos metalíferos

AISHA UMAR

Soil fungi (micropartner) develop mycorrhizal interactions with 80% of land plant species (macropartner) (WANG & QIU, 2006). Soil composed of non-degradable heavy metals toxicity that inhibits the growth (LOMBI ET AL., 2001) and water status of plants (BARCELÓ AND POSCHENRIEDER, 1990). HMs in the soil causes vegetation free world. Phytohyperaccumulators gather more than 1% Zn or Mn, 0.1% Ni, Cu, Co or Pb and 0.01% Cd in the shoots (COBETT, 2003) than roots (VÁZQUEZ ET AL., 1994). These plants can cope with low levels of fertility and poor soil structure, which may induce mineral deficiencies (STANLEY AND ISABELLE, 2015). Mycorrhizae are the mutualistic symbiotic association (non-pathogenic) of a specific group of soil-borne fungi with the roots of higher plants (SIEVERDING, 1991). Arbuscular mycorrhizal fungi colonize the roots of approximately 65% of all known land plant food and bio energy crops.

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(Wang & Qiu, 2006) may interfere with heavy metal absorption (Liao et al., 2003). AMF is reported to enhance nutrient supply and improve water use efficiency and photosynthesis in salt marsh plant species, but the rate of AMF colonization drastically decreases at moderate salinities (Caravaca et al., 2005).

Root exudates (organic chemicals) enhance the mobility of metals and nutrients by (Pérez-Esteban et al., 2013) (i) acidification due to proton (H⁺) release or by forming organic/amino acid-metal/mineral complexes (ii) intracellular binding compounds (e.g., phytochelatins, organic acids, and amino acids) (iii) electron transfer by enzymes in the rhizosphere (e.g., redox reactions), therefore enhancing phytoremediation efficiency (Sessitsch et al., 2013). The low molecular weight organic acids (LMWOAs) e.g., oxalate, released by both mycorrhizal and non-mycorrhizal seedlings contributed to metal immobilization by stable metal complexes in soil (Johansson et al., 2008).

Establishment of Mutualistic Symbiosis b/w Phytoaccumulator-AMF

a) Phyto Chemotaxis

Root exudates are chemo attractant signals for AMF (Chaparro et al., 2013) in unique environment of rhizosphere and sources of organic and inorganic elements for fungi (Compant et al., 2010) therefore creating crosstalk in plants and fungi (Compant et al., 2011).

b) AMF Association

Zn violets enhance the rhizospheric phenomenon in polluted soil (Tonin et al., 2001). Plant-fungal association is a species specific and competitive process (Reinhold-Hurek and Hurek, 2011). Rate of colonization of roots by mycorrhizal fungi increases with increasing heavy metal content in serpentine soil (Audet and Charest, 2006). AMF needed maximum nutrients during reproductive stage so they are suitable to flourish in HMs (reproductive period (Vogel-Mikus et al., 2006) HMs from AMF (cell wall, cytoplasm, Vesicles, arbuscules, intraradical hyphae) stores in inner root parenchyma cells. Literature supported the vesicle as a storage compartment of HMs (Turnau, 1998; Weiersbye et al., 1999. AMF makes macro and micro colonies inside and outsides tissues of the plants (Ma et al., 2015; Wang et al., 2016).

Performance of Mutualistic Symbiosis

Plant–Fungal crosstalk is a significant and beneficial process to distinguish the underground communities. Metal resistant activity of AMF enhances membrane permeability and metabolic functions of phytohyperaccumulators in serpentine soil.
Nutrient-absorbing AMF facilitate the hyperaccumulators to absorb maximum minerals and nutrients from the contaminated soil as well as proficient in increasing the fertility of heavy metals contaminated soil (Navarro-Noya et al., 2012). This association prevents the plant from production of ethylene in soil stress conditions. AMF actors act as a biomodifiers and modify the original appearance of roots in prosperous style (Ahemad and Kibret, 2014). They act as biopesticides that kill phyto pathogens by releasing antifungal agents (Fig 1).

Phosphorous is major macronutrients unavailable for plant growth in soil (Harris and Lottermoser, 2006). The insoluble P compounds in soil can be solubilized by enzymes, organic acids and/or chelating agents excreted by both plants and fungi (Jeong et al., 2013). AMF Glomus spp. is benefited for plant growth and nutrient (N, P, and K) uptake by leguminous trees grown on Pb/Zn mine tailings. Arbuscular mycorrhizal fungi (AMF) are capable of boosting plant growth (Orlowska et al., 2013) and nutrients uptake (Guo et al., 2013), reduce metal induced toxicity (Meier et al., 2011), change metal availability through alteration of soil pH (Rajkumar et al., 2012) and affect metal translocation (Hua et al., 2009). Exudates from plant roots create intricate communication systems (nutrient and energy) with fungi of metalliferous sites called allelopathic actions, which attracts AMF. AMF induces the defense

Fig. 1. Action of AMF for phytohyperaccumulators.
mechanisms against phytopathogens directly through the solubilization of mineral nutrients (nitrogen, phosphate, potassium, iron, etc.), production of plant growth promoting substances (e.g., phytohormones—indole-3-acetic acid (IAA), gibberellic acid, abscisic acid, and secretion of specific enzymes (e.g., 1-aminocyclopropane-1-carboxylate deaminase) under stress conditions of soil (ULLAH ET AL., 2015). Insoluble specific glycoprotein glomalin produced by hyphae of AMF helps in sequestering heavy metals outside the mycelium (GONZALES-CHAVEZ ET AL., 2004). Accumulated HMs by AMF enters to plants and excreted from there by different organs.

AM fungi releases Myc factors lipochitooligosaccharide (MAILLET ET AL., 2011) that lead to root and AM symbiosis. Genes encoding proteins (e.g., metallothionein and glutathione) efficiently help in tolerance of AMF in contaminated soil. Expression of genes is done in intra and extraradical mycelium. Seven genes (SYM genes) have been identified that are required for fungal-root symbioses (Table 1).

**ROLE OF PHYTOACCUMULATORS**

Phytoaccumulators associated with AMF store and release the heavy metals in the following organs.

1: Trichome excretions

Cadmium is excreted by trichomes in a glycophyte species (tobacco) (CHOI ET AL., 2004). SARRET ET AL. (2006) reported that Zn exposure increased trichome density in young leaves but not in mature leaves of tobacco. Similarly, Cu induced the expression of genes coding for sequestering metallothionein (MT) in Arabidopsis trichomes (GUO ET AL., 2003). Glandular trichomes of the hyperaccumulating species *A. halleri* accumulate heavy metals but do not excrete them at the leaf surface.

2: Salt glands

Heavy metals especially Zn excretion takes place through glandular trichomes or salt glands (WEIS AND WEIS, 2004).

3: Leaf succulence

In some phytohyperaccumulators, leaf lusciousness help in dilution of concentrated heavy metal contents and toxic impacts in photosynthetic leaves (WANG ET AL., 2012). Absorbed salts increases leaf succulence (KATSCHING ET AL., 2013) called reservoir due to sufficient space where water contaminated with toxic ions and heavy metals stores (THOMAS AND BOHNERT, 1993).

4: Histological distribution

Heavy metal accumulated compartments exhibit less metabolic activity. Vacuoles and Cell walls can tolerate HMs concentration (CARRIER ET AL., 2003).
Table 1. Genes of the common symbiosis pathway.

<table>
<thead>
<tr>
<th>Signaling component-receptor Gene</th>
<th>Function</th>
<th>References</th>
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</thead>
<tbody>
<tr>
<td>kinase SymRK</td>
<td>Leucine-rich receptor-like kinase that plays an essential role for root</td>
<td>(GHEBER ET AL., 2008)</td>
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<td></td>
<td>endosymbiontes with Rhizobia bacteria, AM fungi and Frankia bacteria, and</td>
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<td></td>
<td>is involved in the signal transduction to the cytoplasm after the</td>
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<td></td>
<td>perception of Nod or Myc factors.</td>
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<tr>
<td>NUP95/NUP133</td>
<td>Putative components of the nuclear</td>
<td>(PARKIN 2008)</td>
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<tr>
<td></td>
<td>pore complex that are involved in the transport of macromolecules</td>
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<td></td>
<td>through the nuclear envelope.</td>
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<tr>
<td>CASTOR/POLLUX</td>
<td>Cation channels in the nuclear</td>
<td>(CHARPENTIER 2008)</td>
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<td></td>
<td>envelope that are essential for the perinuclear calcium spiking after</td>
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<td></td>
<td>the perception of Nod or Myc factors.</td>
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<tr>
<td>CCoaMK</td>
<td>Calcium and calmodulin-dependent protein kinase with three calcium</td>
<td>(KISTNER ET AL., 2005)</td>
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<td></td>
<td>binding motifs that acts as sensor of the nuclear calcium</td>
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<td></td>
<td>signatures and is involved in the phosphorylation of CYCLOPS.</td>
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<tr>
<td>CYCLOPS</td>
<td>Protein with unknown function that acts as phosphorylation target of</td>
<td>(KISTNER ET AL., 2005)</td>
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<td></td>
<td>CCoaMK downstream of the nuclear calcium spiking and is presumably the</td>
<td></td>
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<td></td>
<td>branchpoint of the common SYM pathway.</td>
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<tr>
<td>PAM1</td>
<td>intracellular hyphae in mycorrhizal roots</td>
<td>(KISTNER ET AL., 2005)</td>
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<tr>
<td>Mic1f1 and Mic2f2</td>
<td>Re-programming of root tissues during the establishment of an AM</td>
<td>(HEGGEKAMP ET AL., 2011)</td>
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<td></td>
<td>symbiosis.</td>
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<tr>
<td>(BEG 34)/(GlnZnT1)</td>
<td>Increased transcript levels of a putative Zn transporter gene and</td>
<td>(GONZALEZ GUERRERO ET AL.,</td>
</tr>
<tr>
<td></td>
<td>protecting against Zn stress.</td>
<td>2005)</td>
</tr>
<tr>
<td>GlnABC1</td>
<td>Cd and Cu detoxification in the ER(Gol) in mycorrhizes.</td>
<td>(GONZALEZ GUERRERO ET AL.,</td>
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<td></td>
<td></td>
<td>2006)</td>
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<tr>
<td>SyL67</td>
<td>Alleviating the H2O2-induced oxidative stress.</td>
<td>(URICH ET AL., 2007)</td>
</tr>
</tbody>
</table>
5: Mucilage
Polysaccharides are mixed in mucilage and found in various plant organs e.g. rhizomes, roots and seed endosperm of several plant species. Na$^+$ as a divalent cations stores in halophytes (Ghanem et al., 2010) and in heavy metal form in shoots and root tissues (Javed et al., 2013). Arsenic (Fox et al., 2012), chromium (Lakshmanraj et al., 2009) and aluminium (Miyasaka and Hawes, 2001) gather and stores in pectic polysaccharides.

MECHANISMS OF ALLEVIATION
Acidification, precipitation, chelation, complexation, and redox reactions, phytoremediation, detoxification, mobilization, immobilization, transformation, transport and distribution in hyperaccumulating plants helps to clean up metal contaminated soils through extraction (phytoextraction), stabilization (phytostabilization), and transformation (phytovolatilization) process (Lebeau et al., 2008; Glick, 2010).

PHYTOACCUMULATOR-AMF SIGNALING
Plant-released signals
Flavonoids from roots are signaling components in plant–mycorrhizal formation (Steinkellner et al., 2007). Flavonoids help in AMF spore germination, hyphal growth, differentiation, and root colonization in AMF-plant interactions (Badri et al., 2009; Mandal et al., 2010).

AMF-roots possessed intermediate levels of flavonoids pattern in early establishment, whereas, high levels of flavonoids (such as phytoalexin and medicarpin) in later stages of colonized roots (Larose et al., 2002; Badri et al., 2009). Flavonoid exerts a negative or neutral effect on different fungi due to its specificity involved in mycorrhizal symbiosis formation (Scerbino et al., 2005).

Microbial Signals:
Myc factor of AMF (Maillet et al., 2011) modulates root system architecture (e.g., lateral root branching, formation of new organs), facilitating symbiotic infections (Maillet et al., 2011), alter the chemical composition of root exudates, plant physiology and plant defense via releasing of various signaling molecules, such as volatile organic compounds (VOCs) are enabling host plants to colonize nutrient (e.g., sulfur and iron) in poor soils (Bailly and Weisskopf, 2012).

METAL DETOXIFICATION
AMF are capable of metal translocation and distribution in inner root parenchyma cells (Kaldorf et al., 1999). The mechanisms involved (1) cell surface biosorption/precipitation of metals; (2) active efflux pumping of metals out of the cell via transporter system; (3) sequestration of
Roots release exudates called LMWOAs (succinic acid, citric, oxalic, malic) (MEIER ET AL., 2012) that encourage AMF growth. Chelators in rhizosphere dissolves sparingly soluble mineral, nutrients (Zn, Fe, P), and detoxify metals and metalloids (Pb, As, Cd, Cu) (LI ET AL., 2013) and facilitate apoplast/symplastic movement of metal ions (MAGDZIAK ET AL., 2011).

a) Bioaccumulation

Intracellular accumulation of HMs called Bioaccumulation, a complex process. Bioaccumulation process concentrates the toxic metals in non-living (biosorption) (MA ET AL., 2013) or living (bioaccumulation) cells (RAJKUMAR ET AL., 2012).

Physical and chemical Bonding of metals with waste materials of fungi called passive biosorption. It is not dependent on metabolism of fungi. But the metabolism-dependent active bioaccumulation involved carrier mediated ion pumps, complex permeation and endocytosis (CHOJNACKA, 2010).

S-layer proteins entrap metal ions either in bio or non bio cells called biosorption (GADD, 2004), while facilitator or carrier proteins are involved in bioaccumulation of P and S essential nutrients (SUAREZ AND REYES, 2002; ELANGOVAN ET AL., 2005).

b) Bioleaching

Penicillium, Aspergillus, Trichoderma and Fusarium are (MULLIGAN AND GALVEZ-CLOUTIER, 2003) uses their metabolic machineries in alleviating metal phytotoxicity bydissolution, complexation, adsorption and Redox reactions (PATHAK ET AL., 2009). This reaction helps in bioleaching of heavy metals (Cd, Zn, Cu, Cr, Fe, Pb) from sediments, soils and sludge.
Metal Mobilization

Strong binding of metals to soil particles accounts for the insolubilization of metals in soil and contributes to their unavailability for plant uptake phytoextraction (Ma et al., 2013). Metal mobilizing fungi modify the rhizosphere and mobility in soil through biogeochemical cycling processes of heavy metals by acidification, protonation and chelation (Rajumar et al., 2012; Sessitsch et al., 2013).

c) Biogeochemical cycling of heavy metals

The influence of fungal activity on metal mobilization/immobilization, translocation and transformation improve metal bioremediation processes (Sessitsch et al., 2013; Ahemad and Kibret, 2014). Acidification, chelation and protonation lead to mobilization of metals, whereas precipitation, alkalinization, and complexation cause metal immobilization whereas chemical transformation can cause metal mobilization or immobilization of heavy metals.

d) Acidification

Mobility of metals decreases with increasing soil pH (Richards et al., 2000) has influenced on the activities of both plants and fungi. The hydrogen ions excreted by plant roots can displace heavy metal cations adsorbed on soil particles, leading to acidification of the rhizosphere. Root exudates can lower the pH of rhizosphere (Sheoran et al., 2011), which enhancing soil metal mobility and plant metal bioavailability in soil solution (Alford et al., 2010).

e) Chelation

Natural organic chelators are known as metal-binding compounds, organic acid anions, siderophores, biosurfactants, and metallophores (Sessitsch et al., 2013) are released from both plants and fungi which scavenge metal ions (Gadd, 2004) and reactive oxygen species (Leitenmaier and Küpper, 2013).

Reactive oxygen species (ROS- O$_2^-$, H$_2$O$_2$, and –OH) are generated under stress conditions and bear strong oxidizing activities that can attack all types of biomolecules and represent intermediates emerging during the successive reduction of O$_2$ to H$_2$O$_2$ (Mittler 2002; Panda 2008). Plants experience certain heavy metal ions during the conversion plant accumulation of H$_2$O$_2$ into highly reactive –OH molecule in a metal-catalyzed reaction via the Fenton reaction. The oxidized metal ions undergo are-reduction in subsequent reaction with superoxide radicals (O$_2^-$).

Fenton reaction

\[
\text{H}_2\text{O}_2 + \text{Fe}^{2+} + \text{Cu}^+ \Rightarrow \cdot\text{OH} + \text{OH}^- + \text{Fe}^{3+} + \text{Cu}^{2+} + \text{O}_2^- + \text{Fe}_3^{2+} + \text{Cu}^{2+} \Rightarrow \text{Fe}_2^{2+} + \text{Cu}^{2+} + \text{O}_2
\]
Haber Weiss reaction

\[ \text{H}_2\text{O}_2 + \text{O}_2 \rightarrow \cdot \text{OH} + \text{OH}^- + \text{O}_2 \]

The ·OH molecule is one of the most reactive species known to initiate radical chain reactions leading to irreversible chemical modifications of various cellular components. Protonated form of ·O2· in lipid peroxidation Heavy metals (HM) like Cu, Zn, Cd, Cr, Pb, Hg, As, Fe etc under toxic concentration inactivates enzymic antioxidant defense system in plants resulting into increased ROS signaling generally leading to death of a plant including membrane permeability, chlorosis, growth retardation, browning of roots, effects on both photosystems, cell cycle arrest and others can be observed (Mittler, 2002; Tiwari et al., 2008). Plants and AM fungi have evolved several mechanisms to maintain ion homeostasis under elevated HM concentrations (Clemens 2001; Hall 2002). The basic principles of detoxification mechanisms include the extracellular HM chelation by root exudates and/or binding of HM to the rhizodermal cell walls uptake of HM avoiding. Active plant efflux systems control cytosolic concentrations of HMs.

Intracellularly the plant cell produces chelating agents such as phytochelatins and metallothioneins, which have high-affinity HM binding properties. The resulting complex can finally be exported from the cytoplasm across the tonoplast and become sequestered inside the vacuole (Hall, 2002). Proteins are involved in linkages with metals, thereby forming complex biochemical compounds called metal-proteins, metallothionein and peptides like phytochelatins. Besides these, organic acids and amino acids take active part in detoxification of heavy metals in plants (Pal and Rai, 2009).

Metal-binding peptides (MTs-cysteine-rich) can eradicate the phytoxic effect of free metal ions and allow metal uptake, sequestration, compartmentation, xylem loading, and transport within the plants (Cai and Ma, 2002). Phytochelatins (PCs) are synthesized from the tripeptide glutathione under PCs synthase (Solanki and Dhankhar, 2011). These PCs is immediately induced by heavy metal exposure in plant tissues (Pal and Rai, 2010).

MTs also occur in AMF and those genes encoding several enzymes for PCs synthesis in mycorrhizal roots exposed to metal stress and enhancing photosynthetic activity (Rivera-Becerril et al., 2005).

Siderophore is producing in fungi due to Fe acquisition by different plant species (Ganorkar and Bhosle, 2013). Chelation through binding toxic metals to siderophores triggers the enhancement of plant Fe uptake capacity and the decrease of free metal ion concentration (Dimkpa et al., 2008). Fe is a micronutrient and its low concentration in soil is
necessary to support robust plant and fungal life due to its low solubility under metal stress. Plants acquire sufficient Fe through three mechanisms, 1: (Fe solubilization by all dicots and monocots via rhizosphere acidification) 2: Secretion of phytosiderophores (PSs) and uptake Fe$^{3+}$-PS, 3: uptake Fe$^{3+}$-microbial siderophores by plants. Many studies have shown that PSs are able to solubilize and transport metals by chelation, and thus being secreted into the rhizosphere through a potassium-mugenic acid symporter (SAKAGUCHI ET AL., 1999).

**Metal Immobilization**

Some AM fungi also reduce plant metal uptake or translocation to aerial plant parts by decreasing metal bioavailability in soil via precipitation, alkanilization, and complexation processes.

a) Alkanilization

Some AMF exhibit the ability to absorb metals through substratum alkanilization activity by release of OH$, therefore affecting the metal stability in soil (Büdel ET AL., 2004) and a reduction in metal phytoavailability in the rhizosphere by secreting glomalin (GIASSON ET AL., 2008). AMF can act as metal sinks to reduce the mobile and available metal cations in soil suitable environment for plants growth in metal contaminated soils (Göhre and PASZKOWSKI, 2006) (Fig 3).

b) Complexation

The excretion of EPSs by plant associated (HOU ET AL., 2013) metal biosorption onto to EPS include metal ion exchange, complexation with negatively charged functional groups, adsorption and precipitation (ZHANG ET AL., 2006). AMF can reduce metal mobility in soil by excreting glomalin (insoluble metal-sorbing glycoprotein) that reduces metal mobility or sequesters metals and metal biostabilization in soil (VODNIK ET AL., 2008) (Fig 3).

Glomalin-related soil proteins (GRSP) are sequestering Pb (0.21–1.78%) and Cd (0.38–0.98%) in an in situ field experiment consisted humic and fulvic acids in soil (WU ET AL., 2014).

c) Klebsiella planticola precipitated cadmium (SHARMA ET AL., 2000).

**Metal Transport and Distribution**

Different metals are differently mobile rate within a plant, e.g., Cd and Zn are more mobile than Cu and Pb. The metal chelation with ligands (e.g., organic acids, amino acids, and thiols) facilitates the metal movement from roots to shoots (ZACCHINI ET AL., 2009). Due to the high cation exchange capability of the xylem cell, the metal movement is severely retarded when the metals are not chelated by ligands (SHENG ET AL., 2008)
The future research should be focused on: (1) the mechanism of plant-microbe-metal interactions under stressful environmental conditions; (2) the effectiveness of co-inoculation of PGPB and AMF response to multiple biotic and/or abiotic stress; (3) the identification of functional genes of beneficial microbes for growth enhancement and metal metabolism; (4) the optimization of techniques for application in large scale polluted fields; and (5) the exploration of commercial production of bioinoculants for use in metal decontamination.

Fig. 3. Mechanism of heavy metals detoxification.
Fig. 4. Pathway for movement of heavy metals from soil to phytohyperaccumulators.
Table 2. Examples of AMF in HM's detoxification (literature citation).

<table>
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<tr>
<th>Host Plant</th>
<th>Colonizing AMF</th>
<th>Soil Condition</th>
<th>Mechanism</th>
<th>Experiment</th>
<th>Reference</th>
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<tr>
<td>Sunflower</td>
<td>Rhizophagus irregularis</td>
<td>Saline contaminated</td>
<td>Phytoextraction of Cd</td>
<td>Greenhouse</td>
<td>Oga et al., 2013</td>
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<td></td>
<td></td>
<td>soil with three different Cd concentrations</td>
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<tr>
<td>Sunflower</td>
<td>Funneliformis mossae</td>
<td>HM Contaminated Soil</td>
<td>Phytochelatization of Cd and Zn</td>
<td>In Field</td>
<td>Oga et al., 2013</td>
</tr>
<tr>
<td>Litchi chinensis</td>
<td>Glomus intraradices and Gigaspora margarita</td>
<td>HM Contaminated Soil</td>
<td>Increased endogenous phytohormones level (IAA and isopentyl adenine), Phyt Growth</td>
<td>In Field</td>
<td>Yac et al., 2006</td>
</tr>
<tr>
<td>Aspergillus and Rhizopus</td>
<td>Glomus versiformis melanosporium</td>
<td>HM Contaminated Soil</td>
<td>Bisorption of Cr and Cd</td>
<td>In Field</td>
<td>Grazzini et al., 2007</td>
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<tr>
<td>Yeast</td>
<td>Glomus intraradices melanosporum</td>
<td>HM Contaminated Soil</td>
<td>Polyphenol MTS (TmPMT) and PC synthase (TmPCS) enhanced tolerance to essential (Cu and Zn) and non-essential (Cd, As, and Hg), biophilic metal ions</td>
<td>In Field</td>
<td>Grazzini et al., 2011</td>
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<td>S. affinis,</td>
<td>Glomus caldonicum and G. mossae</td>
<td>Cd-contaminated acidic soil</td>
<td>Decreased soil DTPTA-extractable Cd by 21–30% via alkalinization process</td>
<td>In Field</td>
<td>Gyi et al., 2013</td>
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<td>Salix viminalis</td>
<td>Neoblima cristuliforme</td>
<td>Cd 55 and 62%, respectively and Zn</td>
<td>In Field</td>
<td>Gyi et al., 2009</td>
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<td>S. caprea</td>
<td>Brgia Carophora</td>
<td>Cd and Zn</td>
<td>In Field</td>
<td>Grazzini et al., 2011</td>
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<td>Sorghum bicolor</td>
<td>Glomus intraradices and G. sparsorum (AMP)</td>
<td>Cd- and Zn-contaminated soil</td>
<td>Phytophosphatization reduced Cu and Zn under elevated Cu-Zn conditions, increased Cu and Zn uptake and translocation under normal Cu-Zn concentrations</td>
<td>In Field</td>
<td>Tag et al., 2009</td>
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<td>Thlaspi sp.,</td>
<td>Calaminula tricoloris</td>
<td>Cd, Cu or Zn-contaminated soil</td>
<td>In Field</td>
<td>Tomson et al., 2007</td>
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<td>M. truncatula</td>
<td>P. syringae</td>
<td>Citrine handling and glutathione S-transferease alleviation of damage caused by reactive oxygen species</td>
<td>In Field</td>
<td>Tag et al., 2002</td>
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<td>Tomato</td>
<td>Glomus intraradices</td>
<td>&quot;Breisingberg&quot; seed-Zn</td>
<td>In Field</td>
<td>Tomson et al., 2002</td>
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<td>Thlaspi protein</td>
<td>Highly Cd, Zn and Pb-contaminated</td>
<td>Elevated nutrient demands on heavy metal uptake and tolerance</td>
<td>Greenhouse experiment</td>
<td>Kottkeba et al., 2006</td>
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<td>Thlaspi polyphax</td>
<td>Glomus intraradices</td>
<td>Zinc contamination</td>
<td>In Field</td>
<td>Whitfield et al., 2004</td>
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<tr>
<td>Thlaspi polyphax</td>
<td>(AMF fungi)</td>
<td>Zinc contamination</td>
<td>In Field</td>
<td>Whitfield et al., 2004</td>
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Table 2 (cont.)
Plants and fungi coexist in serpentine soil (enrich with heavy metals) and their cohesive interactions play a vital role in metalliferous soil. Metallophytes colonized by AMF helps to alleviate the toxicity of metalliferous environments. In this work phytoremediation-AFM interaction heavy metals (HMs) stress called biology-based technology is examined. Phytohyperaccumulator includes phytovolatilization, phytostabilization, phytoextraction, chelate-enhancement strategy. AMF enhance the efficiency of hyperaccumulator by different mechanism. Acidification, precipitation, chelation, complexation, redox reactions, phytoremediation, detoxification, mobilization, immobilization, transformation, transport and distribution in hyperaccumulating plants helps to clean up metal contaminated soil through extraction (phytoextraction), stabilization (phytostabilization), and transformation (phytovolatilization) processes.

**KEYWORDS**: Arbuscular; mycorrhizal fungi; glomalin; detoxification

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### SUMMARY

<table>
<thead>
<tr>
<th>Deschampsia flexuosa (Cu grass)</th>
<th>AMF soil biota</th>
<th>Zn contamination</th>
<th>Reduced Zn concentrations in shoots of D. flexuosa</th>
<th>In Field</th>
<th>(SYDNEY and BRINDA, 2013)</th>
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<tr>
<td>Sorghastrum nutans(Cu grass)</td>
<td>AMF + Soil microbes</td>
<td>low-contaminated (LC) soil</td>
<td>Increased the efficacy of AMF from LC soils but decreased the efficacy of AMF from HC soils in promoting plant growth.</td>
<td>In Field</td>
<td>(SYDNEY and BRINDA, 2012)</td>
</tr>
<tr>
<td>Phytolacca americana,</td>
<td>AMF communities</td>
<td>Heavily polluted microphoric soils</td>
<td>Glomus show all effect on D. ambrosioides, Kulikspora and Ambispora show D. ambrosioides and the rhizosphere of P. americana.</td>
<td>In Field</td>
<td>(LIANG et al., 2010)</td>
</tr>
<tr>
<td>Retusa glutinosae</td>
<td></td>
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<tr>
<td>Potentilla frutescens</td>
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<tr>
<td>Liriodendron tulipifera</td>
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<tr>
<td>Dryopteris aemusoides</td>
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<td></td>
<td>Glomus etulatum and G. confluens infraradicis</td>
<td>Cu, Pb, and Zn</td>
<td>D. ambrosioides more sensitive than G. confluens infraradicis</td>
<td>In vitro</td>
<td>(TEVESIA et al., 2004)</td>
</tr>
<tr>
<td>Aster tripolium</td>
<td>Glomus geosporum</td>
<td>Cu and Cu concentrations</td>
<td>AMF enhance metal accumulation in the root system of A. tripolium</td>
<td>In Lab</td>
<td>(LIYS et al., 2008)</td>
</tr>
<tr>
<td>Aster tripolium</td>
<td>Contaminated Soil</td>
<td>AMF</td>
<td>AMF clearly increased Cu accumulation</td>
<td>In Field</td>
<td>(CONTINUIDI et al., 2009)</td>
</tr>
</tbody>
</table>

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**Table 2 (conclusion)**
Plantas e fungos coexistem em solos serpentinos (ricos em metais pesados) e suas interações têm um papel fundamental nesses solos metalíferos. Metalófitos colonizados por fungos arbusculares micorrizais auxiliam a aliviar a toxicidade dos ambientes metalíferos. Neste trabalho, as interações de hiperacumuladores, FAM (fungos arbusculares micorrizais), que se desenrolam em condições de estresse por metais pesados, e que constituem uma parte da biologia tecnológica, são discutidas. Essas interações incluem estratégias de: fitovolatilização, fito-estabilização, fito-extração, aprimoramento quelático. FAM confere eficiência aos hiperacumuladores por meio de diferentes mecanismos. E.g., acidificação, precipitação, quelação, complexificação, reações-redox, fitoremediação, detoxicação, mobilização, imobilização, transformação, transporte, distribuição, i.e., processos que em plantas acumulantes ajudam a limpar a contaminação metálica mediante mecanismos de extração e estabilização.

PALAVRAS-CHAVE: Arbuscular; fungos micorrizais; glomalina; detoxificação

BIBLIOGRAPHY


CHAPARRO, J. M.; D. V. BADRI; M. G. BAKKER; A. SUGIYAMA; D. K. MANTER; J. M. VIVANCO. 2013. Root exudation of phytochemicals in *Arabidopsis* follows specific patterns that are developmentally programmed and correlate with soil microbial functions. *PLoS ONE* 8:e55731 10.1371/journal.pone.0055731.


FOX, D. I; T. PICHLER; D. H. YEH; N. A. ALCANTAR. 2012. Removing heavy metal in water: the interactions of cactus mucilage and arsenate (As (V)). *Environmental Science and Technology* 46: 4553–4559.


during senescence and in response to copper. *New Phytologist* **159**: 369–381.


S. SAkAGUCHI, T.; N. K. NISHIZAWA; H. NAKANISHI; E. YOSHIMURA; S. MORI. 1999. The role of potassium in the secretion of mugineic acids family


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VAZQUEZ, M. D.; C. POSCHENRIEDER; J. BARCELO; A. J. M. BAKER; P. HATTON; G. H. COPE. 1994. Compartmentation of zinc in roots and


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