Perspective of Arbuscular Mycorrhizal Fungi Phytoremediation on Contamination and Remediation Heavy Metals Soil in Sustainable Agriculture

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Abstract: The development of modern agriculture has led to increased soils contaminated with heavy metal that is major concern global for sustainable agriculture. Arbuscular Mycorrhizal Fungi Mycorrhizae are indigenous to soil and plant rhizosphere and potential tools for sustainable agriculture. They enhance the growth of a root system and even of an entire plant and often control certain plant pathogens. It is a fascinating subject, multidisciplinary in nature and concerns scientists involved in plant health and plant protection. There have been marked advances in this field during the last few decades. Mycorrhizal fungi improve plant vigor and soil quality by using the greater surface area. They play a crucial role in plant nutrient uptake, water relations, ecosystem establishment, plant diversity and the productivity of plants. This review summarizes research carried out on role of arbuscular mycorrhizal fungi in phytoremediation of contamination and remediation heavy metals soil in sustainable agriculture. In addition, mycorrhizal fungi were found to play an important role in heavy metal detoxification and the establishment of vegetation in strongly polluted areas. The effectiveness of the bioremediation techniques depends on the appropriate selection of both the plant and the fungal partners in sustainable agriculture. Symbiotic partners selected on the basis of such research are often the best choice for future phytoremediation technologies in sustainable agriculture. Moreover, mycorrhizas of different types are also helpful in eradicating heavy metals soil toxicity monitoring.

Key words: Sustainable - Agriculture - Mycorrhizae - Phytoremediation - Metals toxicity - soil

INTRODUCTION

Issue of contamination of soil seriously threatens the environment and human health (Cao et al., 2010; Zhuang et al., 2009). In fact, Self-sustaining, low-input and energy-efficient agricultural systems in the context of sustainable agriculture have always been in the centre of attention of many farmers, researchers and policy makers worldwide (Gafsi et al., 2006). In other words, natural remediation techniques have been developed to provide more environmentally friendly and cost-effective cleanup of sites impacted by heavy metals (Adhikari et al., 2008; Khodaverdi et al., 2008). Phytoremediation is an emerging green technology, low-cost and ecologically for decontamination of soils, is defined as the process of utilizing plants to absorb, accumulate and detoxify contaminants in soil through physical, chemical and biological processes and remediate soil, sediment, surface and ground water contaminated with toxic metals and organics (Weyens et al., 2009; Xiao et al., 2008). Consequently, this technique has been shown to be effective for heavy metal contaminated soils in several laboratory and field studies (Mangkoedihardjo et al., 2008; Licht et al., 2005; Ghosh et al., 2005). Some heavy metal elements such as Cu, Fe, Mn, Ni and Zn are essential for normal growth and development of plants. These metals are required in numerous enzyme-catalyzed or redox reactions, in electron transfer and have structural function in nucleic acid metabolism (Pawlak-Sprada et al., 2011; Pawlak et al., 2009). In contrast, metals like Cd, Pb, Hg and As are not essential and may be toxic to plants at very low concentrations in soils. Heavy metals occur in terrestrial and aquatic ecosystems from both natural and anthropogenic sources and are also emitted into the atmosphere (Yang et al., 2007; Sobkowiak et al., 2004). Arbuscular mycorrhizal fungi (AMF) or endomycorrhizae,
including fungi belonging to the recently established phylum Glomeromycota, are a normal part of the root system in most natural and agroecosystems, including polluted soils (Alizadeh et al., 2013; Citterio et al., 2005). It is postulated that arbuscular mycorrhizae are the ancestral and predominant form of mycorrhizae (Frey-Klett et al., 2005). They occur in the soil rhizosphere as spores, hyphae and propagules and Arbuscular mycorrhizal fungi are considered as obligate symbiotic biotrophs, in that they cannot grow without a host plant supplying them with carbohydrates (Gaur et al., 2004). In this symbiotic association, the fungus colonizes the plant’s root hairs by entering the cortex cells and acts as an extension of the root system (Gohre et al., 2006). This type of association is characterized by the formation of arbuscules (finely branched hyphal structures) in the region of the root cortex that may function as nutrient organs (or nutrient exchange sites between the symbionts) and also for fungal multiplication (Govindarajulu et al., 2005). The AMF genera Gigaspora and Scutellospora produce only arbuscules and extensive intraradical and extraradical hyphal networks, whereas Glomus, Entrophospora, Acaulospora and Sclerocystis also produce vesicles (formerly known as vesicular-arbuscular mycorrhizal (VAM) fungi (Hibbett et al., 2000). The genes involved in arbuscular mycorrhizae and rhizobial symbioses are common in both infection processes. The formation of mycorrhizae induces great changes in the physiology of the roots, in the internal morphology of the plant and in the mycorrhizosphere, i.e., the soil surrounding the roots (Hossain et al., 2007). The symbiotic association of AMF and plant roots has been considered to be the oldest symbiosis of plants and is suspected to ecologically be the most important symbiotic relationship between microorganisms and higher plants (Jeffries et al., 2000). Arbuscular mycorrhizal associations are reported to occur in about 80% of terrestrial plants including trees, shrubs, forbs and grasses and many plants are able to establish symbiotic relationships with AMF. The plants are called mycorrhizal crops. However, crop plants from Brassicaceae, Chenopodiaceae and Polygonaceae do not form mycorrhizal associations (Javot et al., 2007a). The present review was mainly aimed to investigate the potential use of Arbuscular mycorrhizae fungi phytorextraction of heavy metals from soil polluted in sustainable agriculture. The roots of terrestrial plants are in immediate contact with soil metal ions. Essential heavy metals are transferred into the root by specific uptake systems, but at high concentrations they also enter the cell via nonspecific transporters. At high concentrations heavy metals interfere with essential enzymatic activities by modifying protein structure or by replacing an essential element, resulting in deficiency symptoms. As a consequence toxicity symptoms such as chlorosis, growth retardation, browning of roots, effects on both photosystems, cell cycle arrest and others are observed (Alizadeh, 2010; Turnau et al., 2002). Anthropogenic soil contamination resulting from mining activities, industrial processes, agriculture and military activities has resulted in high localized concentrations of heavy metals. Conventional soil remediation practices in most countries rely primarily on the excavation of the contaminated soil. However, physical displacement, transport and storage, or alternatively soil washing are expensive procedures which leave a site devoid of soil microflora. AM fungi are significant in the remediation of contaminated soil as accumulation. The external mycelium of AM fungi allows for wider exploration of soil volumes by spreading beyond the root exploration zone (Alizadeh et al., 2007), thus providing access to greater quantities of heavy metals present in the rhizosphere. Higher concentrations of metals are also stored in mycorrhizal structures in the root and in fungal spores. AM fungi can also increase plant establishment and growth despite high levels of soil heavy metals due to improved nutrition (Alizadeh et al., 2007), water availability (Alizadeh et al., 2013) and soil aggregation properties (Alizadeh, 2010) associated with this symbiosis. AM fungi occur in the soil of most ecosystems, including polluted soils. By acquiring phosphate, micronutrients and water and delivering a proportion to their hosts they enhance the host nutritional status. Similarly, heavy metals are taken up via the fungal hyphae and can be transported to the plant. Thus, in some cases mycorrhizal plants experience enhanced heavy metal uptake and root-to-shoot transport while in other cases AM fungi contribute to heavy metal immobilization within the soil. The result of mycorrhizal colonization on remediation of contaminated soils depends on the plant- fungus-heavy metal combination and is influenced by soil chemical and physical conditions. The significance of AM fungi in soil remediation has been recognized (Alizadeh et al., 2013). A vast amount of literature is available on the effects of mycorrhizal colonization on plants under heavy metal stress but contradictory observations and wide variations in results are reported (Alizadeh & Nadian, 2010). Enhanced understanding of heavy metal tolerance of plants and AM fungi has defined valuable parameters for improving phytoremediation, i.e., the engineered use of
green plants to remediate an affected site. The utility of AM fungi in soil remediation is also important for sustainable agriculture. Application of these fungi is generally useful to overcome heavy metal problems and to alleviate soil stress and ultimately increases agricultural production (Turnau et al., 2003). In many cases AM fungi serve as a filtration barrier against transfer Siddiqui and Pichtel of heavy metal ions from roots to shoots. The protection and enhanced capability of mineral uptake result in greater biomass production, a prerequisite for successful remediation. AM isolates existing naturally in heavy metal-polluted soils are more metal-tolerant than isolates from non-polluted soils and are reported to efficiently colonize plant roots in heavy metal-stressed environments (Turnau et al., 2005; Van der Heijden et al., 1998). Thus, it is important to screen indigenous and heavy metal-tolerant isolates in order to guarantee the effectiveness of AM symbiosis in restoration of contaminated soils. The potential of phytoremediation of contaminated soil can be enhanced by inoculating metal hyperaccumulating plants with mycorrhizal fungi at the contaminated site. However, there is a need to optimize the conditions to grow AM fungi in large quantities and to characterize and screen a large number of AM fungal species for tolerance to metals (Sobkowiak et al., 2003; Turnau et al., 2001; Weissenhorn et al., 1996). In nature, some plants hyperaccumulate heavy metals. Heavy metal complexes in hyperaccumulators plants are mainly associated with carboxylic acids like citric, malic and malonic acids. These organic acids are implicated in the storage of heavy metals in leaf vacuoles. Amino acids like cysteine, histidine glutamic acids and glycine also form heavy metal complexes in hyperaccumulators (Yu et al., 2010). These complexes are more stable than those with carboxylic acids. They are mostly involved in heavy metal transport through xylem. Moreover, hyper-accumulator plants can increase availability of metals like Fe and also Zn, Cu and Mn by releasing chelating phytosiderophores. Hyperaccumulation mechanisms may then be related to rhizosphere processes such as to the release of chelating agents (phytosiderophores and organic acids) and/or to differences in the number or affinity of metal root transporters (Xu et al., 2008). Although hyperaccumulator plants are widely used in phytoextraction, they are generally of low biomass, inconvenient for phytoremediation. However, arbuscular mycorrhizae fungi (AMF), especially Glomus intraradices, colonized Festuca and Agropyron species have shown higher heavy metal (Zn, Cd, As and Se) content than non-colonized controls (Trotta et al., 2006). As for hyperaccumulators, fungi can synthesize cysteine-rich metal binding proteins called metallothioneins. AMF might therefore be directly implicated in heavy metal hyperaccumulation in plants (Tonin et al., 2001). Phytoremediation has already proven its potential in numerous applications around the world (Turnau et al., 2006). There are several processes associated with phytoremediation of heavy metal polluted soils. Phytostabilization is the reduction of the mobility, bioavailability and/or toxicity of the pollutant in the rhizosphere, while the process of phytoaccumulation is the sequestration, by plant roots, of the contaminants, typically heavy metals and then translocation to their aerial parts. The most common heavy metals found in polluted soils are Pb, As, Cr, Cd, Ni and Zn. In phytoremediation, the contaminant mass is not destroyed but ends up in the plant shoots and leaves, which can then be harvested and disposed of safely. The relatively low potential cost of phytoremediation allows for the decontamination of many sites that cannot be treated with currently available methods. In addition, it has aesthetic advantages and long term applicability: it preserves the topsoil and reduces the amount of hazardous materials generated during cleanup (Vázquez et al., 2008). However, research in this field must be pursued to enhance biomass and heavy metals accumulation in plants. In this way, mycorrhizal fungi may be very helpful (Vivas et al., 2005). Arbuscular mycorrhizae have often been reported to sequester and to accumulate metals in their biomass as well as in the roots of host plants (Vogel-Miku et al., 2009). It is reported that intracellular and extraradical mycelium of AM and ectomycorrhizal (ECM) fungi would have potential for metal sorption (Or1owska, et al., 2005). Most of the metals were demonstrated to be bound to the cell wall components like chitin, cellulose, cellulose derivatives and melanins of ecto-and endomycorrhizal fungi (Or1owska, et al., 2008). Research revealed that AM fungi promote increased yield in crops due to increased nutrient uptake especially in marginal soils (Alizadeh et al., 2013; Antosiewicz et al., 2008; Al Agely et al., 2005). This root fungus facilitates resistance to soil borne pathogens and also promotes resistance to soil pollutants including heavy metals and hydrocarbons in some cases (Bai et al., 2008; Chaney et al., 1978). Researchers also reported that AM fungi promotes the uptake of metal ions or decreased uptake and or having no effects on metal uptake (Or1owska et al., 2008;
While conflicting reports have been given on the effects of AM fungi on phytoextraction of metal including heavy metals from polluted soil (Orłowska et al., 2011; Chojnacka et al., 2005). Da-Silva et al., (2006) also illustrated that AM fungi increased the capacity of plants to extract contaminant from soil. Similarly, Shevyakova et al., (2011) exhibited that exogenous application of putrescine increases Ni\(^{2+}\) accumulation in rape shoots, improving potential for phytoremediation of contaminated soil. Mathur et al., (2007) also postulated that mycorrhiza enhances the uptake of Cu\(^{2+}\). On the contrary, Galli et al., (1995) reported that no differences in Cu\(^{2+}\) uptake were detected between mycorrhizal and non-mycorrhizal plants. However, Cd\(^{2+}\) exclusion particularly in the uncontaminated control soil is probably mediated by AM inoculation through the agency of fungal hyphae which adsorb the metal and keep it sequestered on active sites of the hyphal wall (Lasat, 2002). Although the mycorrhizal mechanisms for enhancing uptake are not entirely known, some of them could be the following: Transfer of metals to the hyphae by cation exchange and chelation (non-metabolic binding of metals to cell walls). Interacting with hyphal synthetized products or metabolites that act as biosorption agents such as chitin and glomalin, an insoluble glycoprotein. The thin hyaline layer of the spore wall of Glomus geosporum AMF is composed mainly of chitin. Chelation of metals inside the fungus. Intracellular precipitation with phosphate (PO4) (Jeffries et al., 2000). Uptake of metals is controlled by or depends on different factors including the following: 1) AM species, 2) Metabolite composition, 3) Fungal biomass CEC, 4) Edaphic and environmental conditions, 5) Metal pools, 6) Metal electrochemical properties, 7) Competition between metals for mycorrhizal surface adsorption sites, 8) Nature of the host plant, 9) Root exudation patterns. (Harrier et al., 2004) observed that the effect of AMF associations on metal root uptake appears to be metal and plant specific. Greater root length densities and presumably more hyphae enable plants to explore a larger soil volume thus increasing access to cations (metals) not available to nonmycorrhizal plants (Johansson et al., 2004). As related by other studies (Xu et al., 2008) AMF alters the pattern of Zn translocation from root to shoot in Festuca arrundinaceae. Zinc hyphal uptake and translocation are known to be similar to P transport. In their in vitro experiment, (Vogel-Miku et al., 2009) observed that zinc adsorption at spore propagules was weak - approximately 9.6 µg of Zn per gram of spore in the 500 µg/g Zn treatment because mycorrhizal hyphae vacuoles and arbuscules contain phosphorus in the form of polyphosphate. Additionally, Zn is transferred to the plant host though AMF hyphae and arbuscules. Arbuscules are involved in this transfer by providing a considerable increase in fungus and plant contact surface area (Liasu et al., 2006). Frequent degeneration of fungal arbuscules in the root thus allows Zn content to be transferred directly into the host cell (Mathur et al., 2007) reducing Zn concentrations in fungi. (Ogbo et al., 2010) found that well-developed mycorrhization, containing arbuscule formations, increased the metal content in plant shoots. Zn can then be accumulated in leaves as a citrate complex in the vacuole (Liao et al., 2003). Phosphate is central to mycorrhizal symbiosis. In P deficient soils, plant roots exude chemical signals to attract AMF. In such environments, AMF have developed an active phosphate transporter (Garg et al., 2010). Arsenate (As(V)) is chemically similar to phosphate and can enter cells via arsenite (As(III)) translocating ATP’ase (Ogbo et al., 2010). The presence of AMF can therefore enhance both phosphate and arsenate uptake in such conditions (Mathur et al., 2007). Also, at high levels of P, mycorrhizal colonization may be reduced with consequent reductions in uptake and cause deficiencies of essential metals like Cu and Zn. Interactions such as these may be involved in the apparent alleviation of Zn toxicity in polluted sites (Khan, 2005). If the sites are P deficient, then mycorrhizal P uptake can result in increased growth and dilution of Zn in the tissues (Garg et al., 2010). In an in vitro study using transformed carrot roots (Daucus carota L.) growing in a phytogel (M media), (Khan, 2006) found that even without pressure, AMF hyphae passed from the proximal to the distal side of the Petri dish into the M media containing low and high concentrations of Zn and Cd. The hyphal network was well developed and sporulation was high in the low heavy metal level side (100 µg/g Zn and 5 µg/g Cd). More than 16,000 spores per half Petri plates were counted for the low Cd and Zn treatments. Metalloids and some metals (e.g., As, Se, Hg, Sn, Pb) can be transformed by fungi into their methylmetal form which causes their volatilization in soil gasses and eventually in the atmosphere. In a greenhouse study, (Lasat, 2002) suggested that phytoaccumulation of As and Se can slightly diminish because of phytovolatilization. As showed by (Janos, 2007) and (Jeffries et al., 2003), Se may be lost in part by phytovolatilization in the dimethyl diselenide (CH\_3SeSeCH\_3) form. Dimethyl arsenic (AsO(CH\_3)\_2(OH)), methyl mercury (CH\_3Hg\_2) and tetramethyl lead (Pb(CH\_3)\_4) are the most common methylated forms of As, Hg and Pb that can also be phytovolatilized.
CONCLUSION

Although usually considered important primarily for P uptake, AMF can improve assimilation of other non-metallic nutrients such as N, K, S, B as well as of metallic nutrients (Zn, Cu, Mn and others), particularly in unpolluted soils of low nutrient status. It has been suggested that mycorrhizae may benefit plant growth by increasing the availability of P from non-labile sources. The response to AMF colonization may vary among the different plant species. However, it should be considered to introduce mycorrhizae inoculums tolerant to metallic nutrients (e.g., Zn, Cu, Mn or others) into low-input agricultural soils in order to facilitate the recycling of organic, industrial and urban wastes on agricultural fields that would otherwise be extremely dangerous to agricultural ecosystems. For environmental considerations, mycorrhizal associations should be managed to attenuate the possibility of contaminating the soil and surface water. In order to exploit microbes as biofertilizers, bio-stimulants and bioprotectants against pathogens and heavy metals, ecological complexity of microbes in the mycorrhizosphere needs to be taken into consideration and optimization of rhizosphere/mycorrhizosphere systems need to be tailored. There is interspecific variation between AMF regarding translocation of metals to plants. Effect of AMF associations on metal root uptake appears to be metal and plant specific. Greater root length densities and presumably more hyphae, enable plants to explore a larger soil volume thus increasing access to cations (metals) not available to non-mycorrhizal plants. Arbuscular mycorrhizal fungi have great potential in the remediation of disturbed land and low fertility soil but the use of these mycorrhizae and other beneficial microbial communities, by farmers in their fields is still lacking. Further experiments are needed to assess the ability of AMF to continue growing in the presence of multiple toxic metal or metalloid cations, either alone or in combination. The understanding of interactions occurring between AMF and its biotic and abiotic environment is still in its infancy. The characterization of the composition of AMF exudates and the effects of these compounds on soil microbial community, plant nutrition, metal accumulation in plant shoots and shoot biomass production have implications for sustainable soil management and land rehabilitation. The role of AMF in As uptake by plants strongly depends on fungal isolates and therefore suitable AMF strains must be carefully considered when AMF are employed in agriculture or in remediation techniques. All tested isolates could be used to stabilize highly As polluted soils, however, the best adapted are those originating from polluted areas.

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