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Effects of Pre- and Post-transplant Inoculation with *Glomus mosseae* on Heavy Metal (Cadmium) Absorption by Potted Tomato Plants

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Abstract: The effect of pre-and post-transplant inoculation of tomato (*Lycopersicon esculentum.* Mill.) plants with *Glomus mosseae* on the absorption of cadmium from soils amended with low and high level cadmium concentration was investigated in a pot experiment. Cadmium was supplied as cadmium sulphate to potted tomato plants previously raised in a nursery. Nursery inoculation and post transplant inoculation were combined to give four treatments i.e. pre-and post-transplant inoculated plants (A+B+); pre but not post-transplant inoculated plants (A+B-); not pre-but post-transplant inoculated plants (A-B+) and neither pre nor post-transplant inoculated plants (A-B-). The experiment was carried out at two at two levels of soil cadmium concentration. Inoculation with *G. mosseae* reduced the appearance of toxicity symptoms in tomato irrespective of soil cadmium levels. Pre-transplant inoculation alone promoted cadmium absorption as well as plant dry matter accumulation irrespective of soil cadmium contamination levels. Post-transplant inoculation alone was not as effective as pre-transplant inoculation in promoting cadmium absorption and dry matter accumulation but complimented post-transplant inoculation. Cadmium content of soils under singly inoculated and combined i.e. both pre-and post-transplant were higher than under non-inoculated. The presence of significant levels of cadmium in both soil and plant tissue was discussed.

Key words: Bio-accumulators % toxicity symptoms % *Glomus mosseae* % cadmium tolerance % pre-and post-transplant inoculation

INTRODUCTION

Mycorrhizae are symbiotic association between plants and fungi and in arbuscular mycorrhizal fungal AMF association; external hyphae extend into and colonize cortical tissues of roots during period of active plant growth [1]. This symbiosis are characterized by bidirectional movement of nutrients, where carbon flows to the fungus and inorganic nutrients move to the plant thereby providing a critical linkage between the plant root and soil. A clear symbiotic relationship has been demonstrated whereby plant roots exude photosynthates for the fungus to metabolise. In return the fungus scavenges for essential elements that are rare in particular soils [2].

In infertile soils, nutrients taken up by mycorrhizal fungi can lead to improved plant growth and reproduction. As a result, mycorrhizal plants are often more competitive and tolerate environmental stress better than non mycorrhizal plants. The most common-the Arbuscular Mycorrhizal Fungi (AMF) play potential role in finding biological solutions to soil fertility restoration and in increasing plant tolerance to heavy metal toxicity. The fact that AM fungi can stimulate metal uptakes in soils where metals are sparingly available suggest that their influence on metal uptake should be studied when the host plants are growing in soils containing potentially toxic levels of heavy metals. Such environments might include soils in the vicinity of metal mine, metalliferous bedrocks or soils downwind of metal smelters since smelters effluents often result in both acidic and metal deposition [3-5].

Some heavy metal tolerant species of arbuscular mycorrhizal fungi including Glomus mosseae had been found in soils contaminated with heavy metals including cadmium [5-8]. Likewise, some plants have been known to be tolerant to heavy metal e.g. cadmium contamination [9].

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Mycorrhizal fungi may protect the plant from high heavy metal concentration in soil under some circumstances. In some other observations, uptake of heavy metals by plant is increased by mycorrhizal fungi. There are some evidences that AMF facilitates cadmium uptake but this varies with soil conditions like Cadmium concentration and soil pH. Guo *et al*. [10] showed that AM inoculation increased Cadmium uptake by 37% in bean and 41and in maize. However, another study by Heggo *et al*. [11] found AM increased Cadmium uptake in soybean when soil concentration of Cadmium was low but reduced Cadmium uptake when soil Cadmium concentration was high. Weissenhorn *et al*. [4] and Joner and Leyval [12] indicated that cadmium is sequestered in the hyphae of Glomus mosseae an hence, metal transfer into the root of subterranean clover plant is inhibited. Zinc accumulation in roots of mycorrhizal plant was higher than in those of non mycorrhizal plants Banks *et al*. [13] Burke *et al*. [14] showed that the fungicide i.e. Benomyl decreased AM colonization thereby leading increased lead uptake by plants Similarly, lead has been observed to be excluded in mycorrhizal tomato plants growing in soil contaminated with spent i.e. (waste) engine sump oil [15]. This has led to the conclusion that within the limits of available information so far, the bulk of evidence tend to indicate inhibition of metal uptake by mycorrhizae [16]. It has been a habit of poor subsistence farmers who are landless to plant vegetables and annual crops like tomato, maize cassava etc on waste dump sites, highway set backs, catchments around large metal smelting plants, plastic factories and other high risk land areas with the possibility of exposure to cadmium contamination. This paper is an attempt to investigate the influence of introduced AM fungus i.e. 2 on absorption of cadmium especially with respect to time of inoculation vis-à-vis the precedence of infection compared to time of cadmium contamination and also with respect to the level of the contaminant.

MATERIALS AND METHODS

Seed collection: Certified seeds of Tomato (Ibadan Local Variety) were collected from the International Institute of Tropical Agriculture (IITA) Ibadan.

Soil preparation: Good garden soil was collected from a plot of campus location around the premises of the convocation ground of LAUTECH and was transported in steel containers to the laboratory for sterilization. Sterilization of soil was carried out using an oven; sterilization was done using metal containers after the soil was moistened to its maximum water holding capacity. Soil was placed in the oven and the temperature set at 140°C for 1 h and after wards reduced to 80°C and left for 24 h in order to ensure that all resistant nematodes, spores or their likes present in the soil were completely destroyed.

Abuscular mycorrhizae fungal inoculum AMF: This consists of soil containing spores, hyphae and root fragments i.e. *Glomus mosseae* of the trap plant maize.

The inoculum of the AM fungus *Glomus mosseae* used was prepared as described by Liasu [17].

Nursery preparation: Two planting boxes(1 m x 1 m) made of wood were constructed, each box perforated at the base to allow for easy passage of water. The boxes were surface sterilized with methylated sprit. The already sterilized soils were transferred into the boxes to be used as nursery for raising the tomato plants. The soil in the box set aside for pre-inoculation with *Glomus mosseae* was sprinkled with 50 g of the already prepared inoculum while the soil in the other box was spinkled with sterilized soil. Planting of seeds was done by broadcasting. The soil was moistened prior to seeding. The planting boxes were kept under a tree shade to reduce light and heat intensity. The growing seedlings were transplanted into planting bags after six weeks in the nursery.

Transplanting: Six-week old grown up seedlings were transplanted in the evening from the planting boxes plastic pots. (Transplanting into pots was done in the evening in order to allow the seedlings to acclimatize to their environment before sunrise there by safeguarding them from transpiration shock). Transplanting was done with care not to damage the seedlings. The soil was scooped to make holes of few centimeters on the surface of the soil. Inoculum i.e. 50 g of *G. mosseae* prepared as above was poured into the hole and seedling placed, then the roots were covered up with soil in the planting bags and labeled M+ (i.e. inoculated) while those labeled M- were left without inoculum but 50g of sterile soil was added to them (i.e uninoculated). The plants were left for one week after transplanting to enable them acclimatizes before treatment commenced.

Experimental design: The experiment was designed as a 2x2 factorial in a completely randomized block design. The first factor represented at two levels was pre-transplant inoculation with Glomus mosseae (A) i.e. pre-

inoculated (A+) and not pre-inoculated (A-). The second factor, was post-transplant mycorrhizal AMF inoculation with *G. mosseae* (B), also at two levels i.e. inoculated (B+) and uninoculated (B-).altogether giving four treatments i.e. pre-inoculated and inoculated (A+B+), pre-inoculated and uninoculated (A+B-), not pre-inoculated but inoculated (A-B+) and neither pre-inoculated nor inoculated plants (A-B-).

All treatments were in four replicates and they were subjected to wetting with low and high concentration of Cadmium at weekly intervals and normal wetting with water proceeded once daily until the end of the experiment.

Cadmium application: The source of artificial cadmium contamination was Cadmium Sulphate $CdSO_4$.Cadmium application commenced, six weeks after AMF inoculation to ensure proper establishment of the mycorrhizal association with the plants.

Two concentration levels of cadmium were applied to the tomato plants i.e. High concentration (above USEPA safe levels), C = 60 mg Cd SO_4 and below C = 30 mg Cd SO_4.

COLLECTION OF DATA

Growth measurement: After transplanting and introducing of inoculum to tomato seedling, the plant height was measured and number and number of leaves counted on weekly basis to determine the effect of mychorrizal inoculation on the growth of the plants.

Dry matter determination: At the termination of the experiment plants were collected and dried in the oven. Drying was done by leaving the plants in the oven at constant temperature of 70°C for two days. The dry weights of the leaves, shoot and root of the plants were determined using a weighing balance.

Procedure for drying ashing and determination of cadmium in plant: Weighed sub-sample (0.2 g) of well grinded tomato plant material from all replicates of the four treatment (Wo) were put in previously weighed crucible (W_1) and transferred into a muffle furnace set at 660°C for 3 h. After ashing, the content (i.e. crucible+ sample) was removed from the furnace and kept inside a desiccators and allowed to cool after which the whole content i.e. crucible+ash (W_2) was weighed.

Ash content was thus calculated i.e. Ash content $= \frac{W2-W1}{\text{Wt of sample}} \times 100$

The content was then digested with a ratio 1:1 of HCl and HNO_3 (i.e. 10% HCl+10% HNO_3) and 10 ml of the acid mixture was measured with the acid of measuring cylinder into the ashed sample and placed on a hot plate and allowed to boil after which it was filtered into a 50 ml volumetric flask and made up to mark with distilled water.

The solution was then read on an Atomic Absorption spectrophotometer (AAS) for the cadmium content (Cd) at 218nm wavelength. Cadmium Sulphate was used to prepare the Cadmium standard that was first read on AAS before the sample.

Calculation was done using the following mathematical formula:

Meter Reading×Slope×Dilution factor = PPMS.

$$\text{Dilution factor here} = \frac{50}{0.2} = 250$$

Cadmium content of plant was expressed as PPMS = i.e. Parts Per Million of Sample.

Determination of cadmium (Cd) in soil sample: Weighed soil sample (0.2 g) from each treatment replicate were transferred into a dry cleaned vial and 20 ml of 0.1 N HNO_3 measured into each sub sample. The mixtures were thoroughly agitated on a mechanical shaker for 45 min after which they were filtered. The filtrates were read on AAS at 218 nm wavelength. Cadmium Sulphate was used to prepare the Cd standard that was read before the samples were read.

Calculation was done using the following formula:

Meter Reading×Slope×Dilution factor

$$\text{Dilution factor here} = \frac{20}{0.20} = 100$$

Harvesting and cadmium (Cd) analysis in plant: At the end of the sixth week after transplantation, the tomato plants were harvested. The above soil portion (shoot) was cut and the root was uprooted from the soil. The shoot and root were then dried in an oven at 80°C for about 3-4 h. After drying, the shoots and roots were well grinded using a blender.

The cadmium concentration in the root and shoot was determined using dry ashing method. Well grinded

plant sample (Wo = 0.29g) weighed into a previously weighed crucible (W^1) and transferred into a muffle furnace set at 660°C for 3 h. After ashing the content (i.e. crucible+sample) was removed from the furnace and kept in a dessicator and allowed to cool after which it was weighed (W_2).

$$\text{Ash content} = \frac{\text{W2-W1}}{\text{Wt. of sample}} \times 100$$

The content was then digested with a ratio 1:1 of HCl and HNO_3. 10 ml of the acid mixture was measured with the aid of a measuring cylinder into the ashed sample and placed on a hot plate to boil. It was then filtered into 50 ml volumetric flask and made up with distilled water. The solution was then read on an Atomic Absorption spectrophotometer (AAS) for the cadmium content at nm wavelength.

The cadmium comment in the soil was determined by weighing 0.2 g soil sample into a dry clean vial and 20 ml of 0.1N HNO_3 was measured into it. It was then shaken on a mechanical shaker for 45 min and then filtered and the filterate was read on an Atomic Absorption Spectrophotometer.

Statistical analysis: The data were subjected to analysis of variance (ANOVA) using single classification analysis to determine whether variation in mean value among the tested treatments were significant or not. Means of treatment means were separated by Duncan's Multiple Range Test (DMRT) at $p>0.05$

RESULTS AND DISCUSSION

Pre-transplant inoculation of tomato seedlings i.e. nursery inoculation with Glomus mosseae promoted growth vigour of the emerging young seedlings (Table 1). The shoot height, no of leaves, stem diameter and number of nodes of inoculated tomato seedlings were much higher than those of uninoculated plants. AM fungi hve been established as facilitators of improved plant growth particularly in infertile soils [2]. Equally, pre-inoculated seedlings were not as adversely affected by the toxic effects of cadmium contamination of soils at both low and high cadmium concentration. Hence pre-inoculation more than post transplant inoculation of tomato plants appear to mitigate the harmful effect of cadmium contamination as presented by the reduced level of symptom manifestation in pre-transplant inoculated tomato plants when compared with post-transplant inoculated ones (Table 2). The severity of toxicity symptoms in tomatoes planted in soil with high cadmium concentration was higher than that of those planted in low cadmium concentration when considered against plants within corresponding pre-inoculation and post-inoculation treatment. Mature fruits were observed only in pre-and post-transplant inoculated tomatoes growing in soils with high level of cadmium concentration while fruiting though initiated was not completed in pre-inoculated but not post transplant inoculated tomato plants. However, in soils with low levels of cadmium concentration, mature fruits were observed in all pre-inoculated plants irrespective of post-transplant inoculation. Neither post-

Table 1: Effect of mycorrhizal inoculation with *Glomus mosseae* on the growth of tomato plants at the point of transplanting

Treatment mycorrhizal inoculation	Growth level just before transplant			
	Shoot height (cm)	No of leaves	Plant diameter (cm)	No of nodes
M^+ (Inoculated)	35.0a	8.0a	1.0a	20.0a
M- (Uninoculated)	14.5b	6.0b	0.9b	12.0b

Mean separation within columns by Duncan's Multiple Range Test, ($p>0.05$)

Table 2: Toxicity symptom rating (based on the physical appearance of Tomato plants growing in soils supplemented with high and low Cadmium concentration

Treatment	60 mg kgG^1 of $CdSo_4$	30 mg kgG^1 $CdSo_4$
A^+ B^+	+	+
A^+ B^-	++	+
A-B^+	+++	++
A-B^-	++++	+++
Symptom	Classification	Key appearance (Morphpology)
Key to symptom rating		
++++	Lethal toxicity	Withering of leaves resulting in the death of plants.
+++	Severe toxicity	Plants were about drying off while fruiting was aborted.
++	Partial toxicity	Leaves of plants were observed but fruiting was still in progress.
+	No toxicity	Plants were growing normally and fruiting was in process.

A+ B+ = Pre-inoculated; inoculated, A+B- = Pre-inoculated; not-inoculated, A-B+ = Not pre-inoculated; inoculated, A-B- = Not pre-inoculated; not inoculated

Table 3: Influence of mycorrhizal inoculation on the shoot height and stem diameter of tomato plants exposed to different cadmium level at the end of the experiment

Cadmium concentration	Mycorrhizal inoculation	Plant shoot height	Plant diameter
C_1 (60 mg kg^{-1} of $CdSO_4$)	A^+B^+	26.00a	3.60a
	A^+B^-	26.00a	3.10ab
	A^-B^+	21.00b	2.80b
	A^-B^-	12.00c	1.60c
C_2 (30 mg kg^{-1} of $CdSO_4$)	A^+B^+	38.00a	3.50a
	A^+B^-	37.00a	3.00b
	A^-B^+	30.00b	2.60c
	A^-B^-	25.00c	2.10d

Mean separation within columns by Duncans's Multiple Range Test. (p>0.05), A+B+ = Pre-inoculated; inoculated, A+B- = Pre-inoculated; not inoculated, A-B+ = Not pre-inoculated; inoculated, A-B- = Not pre-inoculated; not inoculated

Table 4: Cadmium content of plant tissue and planting soil exposed to mycorrhizal inoculation, varying cadmium levels

Cadmium concentration	Treatment	Cadmium concentration in plant tissue (mg kg^{-1})	Cadmium concentration in planting soil (mg kg^{-1})
C_1 (60 mg kg^{-1} of $CdSO_4$)	A^+B^+	0.180a	1.210a
	A^+B^-	0.170a	1.200a
	A^-B^+	0.110b	1.100b
	A^-B^-	0.090c	1.000c
C_2 (30 mg kg^{-1} of $CdSO_4$)	A^+B^+	0.062a	0.170a
	A^+B^-	0.055b	0.150b
	A^-B^+	0.039c	0.100c
	A^-B^-	0.020d	0.090c

Mean separation within columns by Duncan's Multiple Range Test, (p>0.05), A+B+ = Pre-inoculated; inoculated, A+B- = Pre-inoculated; not-inoculated, A-B+ = Not pre-inoculated; inoculated, A-B- = Not pre-inoculated; not inoculated

transplant inoculation (only) nor non inoculation of soils supported formation of mature fruits. However fruit initiation was observed in post-transplant inoculated (only) tomatoes growing in soils with low level of cadmium concentration.

Tomato plant height in soils with both high and low cadmium concentration was generally promoted by pre-inoculation with a complimentary effect by post inoculation even though there was no significant difference between the height of plants in A+B+ and A+B- (Table 3). The complimentary interaction between pre-inoculation and post inoculation can be deduced from the fact that though plant height of post-inoculated (only) tomato plants is significantly lower than all pre-inoculated, it is higher than that of uninoculaed plants. Plant diameter also followed a similar trend except that the diameter of A+B+ was significantly higher than A+B-.

Cadmium content of both soil and tomato plants were higher in pre-inoculated than in non pre-inoculated plants with no significant differences between the levels in A+B+ and A+B- in tomatoes growing in soils with high levels of cadmium concentration (Table 4). The trend is similar for those growing in soils with low levels of cadmium concentration except that the cadmium content of A+B+ tomatoes were lower than A+ B-tomatoes.

Dry matter accumulation in tomato plants was promoted by pre-inoculation with *Glomus mosseae* while the promotive effect was complimented by post-transplant inoculation as total dry weights as well as the weight of the various plant components i.e. of pre and post-transplant inoculated tomato were significantly higher than those of pre-transplant inoculated (only) plants. The conditional behaviour of inoculated tomato plants with respect to cadmium intake as shown in this work appears to conform with the opinion of some workers [10, 11] while the two faced behaviour could be due to the fact that while promoting cadmium absorption by tomato plants a significant proportion of the metal may have may get sequestered within extraradical fungal hyphae in the rhizospheric soil matrix. With those in the extraradical hyphal mass been analysed along with the rhizosphere soil samples, an increase in detectable cadmium content of the soil is imminent as there is bound to be continuity between the extra-radical and the intra-radical portions of the fungal mycelium which may create further difficulties in interpretation. Separation of the fungal mass from rhizosphere soil may offer a solution to this problem [12]. Similar observation had been reported by Weissenhorn and Leyval [4], Weissenhorn *et al*. [6] and Joner and Leyval [18]. The

Table 5: Dry matter accumulation of inoculated and pre-inoculated plants exposed to different treatments at the end of the experiment

Treatment	Inoculation treatment	Total dry weight (g)	Dry weight of leaves (g)	Dry weight of shoot (g)	Dry weight of root (g)	Dry weight of fruit (g)	Shoot/Root ratio
C_1 (60 mg kgG^1 of $CdSO_4$)	A^+B^+	1.90a	0.51a	0.96a	0.21a	-	3.8:1a
	A^+B^-	1.10b	0.17b	0.81b	0.20a	-	3.1:1b
	$A-B^+$	1.06bc	0.12b	0.76b	0.18a	-	2.3:1a
	$A-B^-$	1.01c	0.01c	0.11c	0.06b	-	1.6:1b
C_2 (30 mg kgG^1 of $CdSO_4$)	A^+B^+	1.60a	0.61a	0.84a	0.73a	0.40a	1.15:1a
	A^+B^-	1.01b	0.51b	0.60b	0.44b	0.30b	0.73:1b
	$A-B^+$	1.00b	0.33c	0.21c	0.37c	-	0.57:1c
	$A-B^-$	1.00c	0.10d	0.08d	0.11d	-	0.73:1b

Means within the same inoculation treatment at each cadmium concentration level not followed by same letters are significantly different according to Duncan's Multiple Range Test. ($p>0.05$), A+B+ = Pre-inoculated; inoculated, A+B- = Pre-inoculated; not-inoculated, A-B+ = Not pre-inoculated; inoculated, A-B- = Not pre-inoculated; not inoculated

mycorrhiza induced dry matter accumulation by tomato in spite of cadmium accumulation and the absence of significant demonstration of toxicity symptoms when compared with non mycorrhizal tomato plants growing in soils with same level of cadmium contamination suggests that AM inoculation can assist the plant in adjusting to the toxic effects of cadmium accumulation in the plants system. This is without bias to the reported resistance of most bio-accumulator plants to heavy metal toxicity [16]. The internal detoxification mechanisms i.e. cellular sequestration and extrusion [19] could have been modified by mycorrhizal symbiosis as AM infection had been known to alter host physiology and anatomy including membrane permeability [20]. The low level of detectable cadmium from the rhizosphere soils of non mycorrhizal tomato could be due partly to leaching, or conversion into unavailable forms. It is however not clear whether tomato in the non symbiotic state is an excluder of cadmium since plant cadmium level is lower in the non mycorrhizal than in the mycorrhizal tomato plants. However, we found out that tomato with the aid of mycorrhizal inoculation excluded lead when grown in soils ammened with spent motor engine oil but bio-accumulated the metal in the absence of the symbiotic partner (Liasu *et al*. in press).

CONCLUSIONS

Pre-transplant inoculation more than post-transplant inoculation of tomato potted plants with *Glomus mosseae* promoted cadmium absorption from soils amended with both high and low concentration of cadmium. Post-transplant inoculation complimented pre-transplant inoculation in promoting growth, dry matter accumulation and in reducing cadmium toxicity symptom manifestations when compared to the non mycorrhzal plants. The exact mechanism of cadmium absorption by tomato(both mycorrhizal and non mycorrhizal) is not yet fully understood. Further experiment needs to be carried out in order to understand the process.

REFERENCES

1. Stahl, E., 1990. Der Sinn der Mykorrhizenbildung Jahrb. Wiss. Bot., 34: 540-668.
2. Bethlenfalvay, G.J. and R.G. Lindermann, 1992. Mycorrhizae and Crop Productivity. In: Berthlenfalvay, G.J. and R.G. Linderman (Eds.). Mycorrhizae in Sustainable Agriculture. Am. Soc. Agron., ASA Special Publication, 54: 1-27.
3. Killham, A. and M.K. Firestone, 1983. Vesicular arbuscular mycorrhizae mediation of grass response to acidic and heavy metal deposition. Plant and Soil, 72: 39-48.
4. Weissenhorn, I. and C. Leyval, 1995. Root colonization of maize by a Cd-sensitive and a Cd-tolerant *Glomus mossae* and cadmium uptake in sand culture. Plant Soil, 175: 223-238.
5. Weisenhorn, I., C. Leyval and J. Berthelin, 1993. Cadmium tolerant *Arbusuclar mycorrhizal* (AM) fungi from heavy metal polluted soil. Plant and Soil, 157: 247-256.
6. Weissenhorn, I., C. Leyval, G. Belgy and J. Berthelin, 1995a. Arbuscular mycorrhizal contribution to heavy metal uptake by maize (Zea mays) in pots with contaminated soil. Mycorrhiza, 5: 245-251.
7. Weissenhorn, I., C. Leyval and J. Berthelin, 1995. Bioavailability of arbuscular mycorrhizal in a soil polluted by atmospheric deposition from a smelter. Biol. Fert. Soils, 19: 22-28.

8. Gildon, A. and P.B. Tinker, 1983. Interactions of vesicular-arbuscular mycorrhizal infection and heavy metals in plants. 1. The effects of heavy metals on the development of vesicular-arbuscular mycorrhizas. New Phytol., 95: 247-261.
9. Ebbs, S.D., M.M. Lasat, D.J. Brady, J. Cornish, R. Gordon and L.V. Koehian, 1997. Heavy metals in the environment: Phyto-extraction of Cadmium and zinc from a contaminated soil. J. Environ. Qual., 26: 1424-1430.
10. Guo, Y., E. George and H. Marshner, 1996. Contribution of an arbuscular mycorrhizal fungus to the uptake of cadmium and nickel in bean and maize plants. Plant and Soil, 184: 195-205.
11. Heggo, A., J.S. Angle and R.L. Chaney, 1990. Effects of vesicular arbuscular mycorrhizal fungi on heavy metal uptake by soybeans. Soil Biol. Biochem., 22: 865-869.
12. Joner, E.J. and C. Leyval, 1997. Uptake of 109Cd by roots and hyphae of a *Glomus mosseae*/*Tripholium subterraneum* mycorrhiza from soil amended with high and low concentration of cadmium. New Phytol., 135: 353-360.
13. Banks, M.K., A.P. Schwab, G.R. Fleming and B.A. Hetrick, 1994. Effects of plants and soil microfauna on leaching of zinc from mine tailings. Chemosphere, 29: 1691-1699.
14. Burke, S.C., J.S. Angle, R.L. Chaney and S.D. Cunningham, 2000. Arbuscular mycorrhizae effects on heavy metal uptake by corn. Intl. J. Phytol., 2: 23-29.
15. Liasu, M.O., A.F. Ogundola and M.O. Atayese, 2006. Influence of mycorrhizal inoculation on growth and development of potted tomato in a soil contaminated with spent oil. Science Focus, In Press.
16. Lasat, M.M., 2002. Phytoextraction of Toxic Metals: A Review of Biological Mechanisms. J. Environ. Qual., 31: 109-120.
17. Liasu, M.O., 2001. The Effects of Mycorrhizal Inoculation on Productivity of Cassava an Hedgerow Trees in Continuing Alley Cropping Systems in Ajibode Village. Nigeria. Ph.D Dissertation. University of Ibadan, Ibadan, pp: 104.
18. Joner, E.J. and C. Leyval, 2001. Time course of heavy metal uptake in maize and clover as affected by root density and different mycorrhizal inoculation regime. Biol. Fert. Soils, 33: 351-357.
19. Baker, J.M., 1981. Accumulators and excluder strategies in the response of plants to heavy metals. J. Plant Nutr., 3: 643-654.
20. Paulitz, T.C. and R.G. Linderman, 1989. Interactions between fluorescent pseudomonands and VA mycorrhizal fungi. New Phytol., 113: 37-45.