Tolerance and accumulation of some heavy metals by microorganisms isolated from residues mining in Marrakech, Morocco

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Introduction

Generally, introduction of heavy metal into the environment induces to serious ecological and health problems (Cerbasi and Yetis, 2001). Therefore, there is a need to develop rapid, economical and environmentally benign technology for the removal of metals from industrial effluents. There are certain microorganisms, which can survive in high concentrations of metals and have the potential to accumulate different metals. Indeed, the contaminated sites are the sources of metal resistant microorganisms. This is achieved by the virtue of covalent interaction of metal at cell surface or within the cell by different processes. These microbes can be of immense significance in the clean up of heavy metals from the environment.

Methods

Residues mining sampling and analyses :

Mining residues samples were collected from various abandoned mining area (contaminated area) of Marrakech city in Morocco. The mining areas were: Bir nhass, Sidi Bouatman, Kettara, and Goundafa. The mining residues were analyzed for their total content of heavy metals (Pb, Cu, Zn, Cr and Cd). Conductivity and pH were also determinate for each mining residues sample.

Determination of metal accumulation by microorganisms isolates :

Microorganisms were cultured in medium agar plates without metals (Bennet for actinobacteria and Sabouraud for fungi). Microorganisms were washed once in phosphate-buffered saline (PBS) and pelleted by centrifugation. The remaining samples were resuspended in tubes containing 2 ml of metal solution (0.5 mg/ml).The samples were incubated at room temperature in a roller mixer for 3 h. The amount of residual metal present in the supernatant was measured by atomic absorption spectrophotometer. Quantities of metal removed by isolate's biomass were then calculated.

Residues mining sampling and analyses :

A disk of fungi was inoculated aseptically on Duxbury agar medium modified plates supplemented individually with different concentrations of heavy metal (Pb2+, Cu2+, Cd2+, Zn2+ and Cr6+). The effect of the heavy metal on the growth of the isolates tested was estimated by measuring the radius of the colony extension (mm) against the control (medium without metal) and the determination of the index of tolerance. The index is defined as the ratio of the extension radius of the treated colony to that of the untreated colony.

Results and discussion

Various microorganisms were isolated from mining residues in various mining area in Marrakesh city, Morocco, where the concentration of heavy metals is very high (Tab.1). 19 fungi belonged to the genera *Aspergillus*, *Penicillium* and *Cladosporium*, and 27 actinobacteria showed the high capacities to resist of heavy metal. Generally, pollution of soil and water by heavy metals may lead to a decrease in microbial diversity (Ezzouhri et al., 2009).

Mining area	Mining residues content							
	heavy metals content g/Kg							
	Pb	Cu	Zn	Cd	Cr	Fer	pH	Conductivity mS/cm
Kettara	0,028 ± 0,007	2,942 ± 0,134	0,685 ± 0,092	0 ± 0	0,017 ± 0,002	233,490 ± 34,955	2,2	10,28
Bir Nhass	1,407 ± 0,341	0,095 ± 0,024	9,989 ± 1,223	0,075 ± 0,011	0,031 ± 0,005	13,845 ± 3,142	6,52	2,21
Sidi Bouatman	10,189 ± 3,580	1,102 ± 0,029	18,870 ± 5,733	0,044 ± 0,017	0,035 ± 0,005	18,265 ± 3,794	7,42	2,08
Mine Arbar	1,619 ± 0,432	1,243 ± 0,079	14,434 ± 0,928	0,070 ± 0,007	0,006 ± 0,001	26,500 ± 2,507	7,17	2,27
Mine Tenfit	0,098 ± 0,002	0,498 ± 0,068	2,093 ± 1,020	0 ± 0	0,014 ± 0,001	10,450 ± 1,666	7,52	1,46

Table 1 : *The mining residues content.*

Quantitative accumulation results show that the actinobacteria and fungi have a high capacity to accumulate the lead and copper respectively, but this capacity depend on the isolate (fig.2). Metal accumulation or biotransformation is an alternative mechanism for metal detoxification in microorganims (Gadd, 1992).

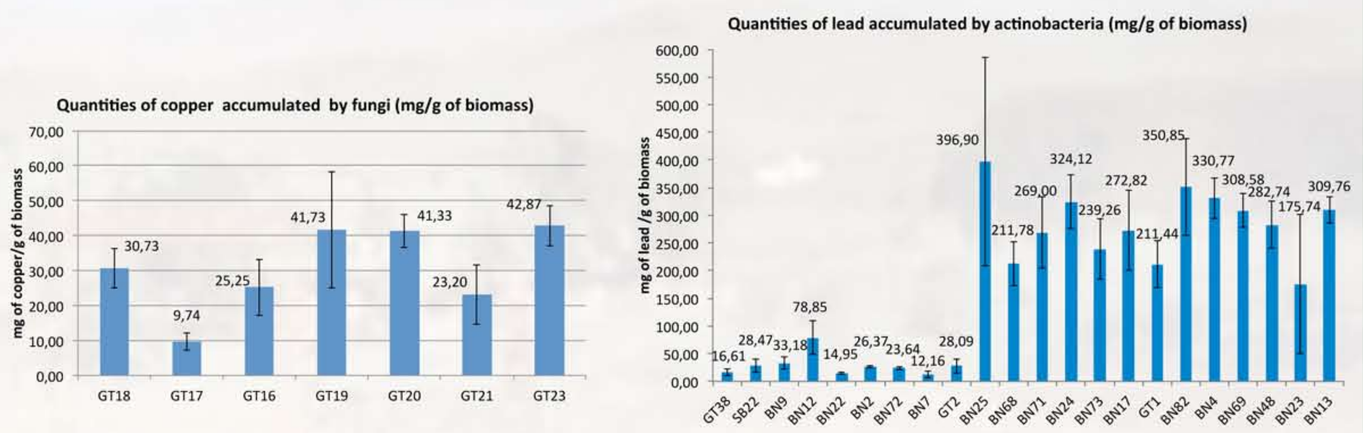
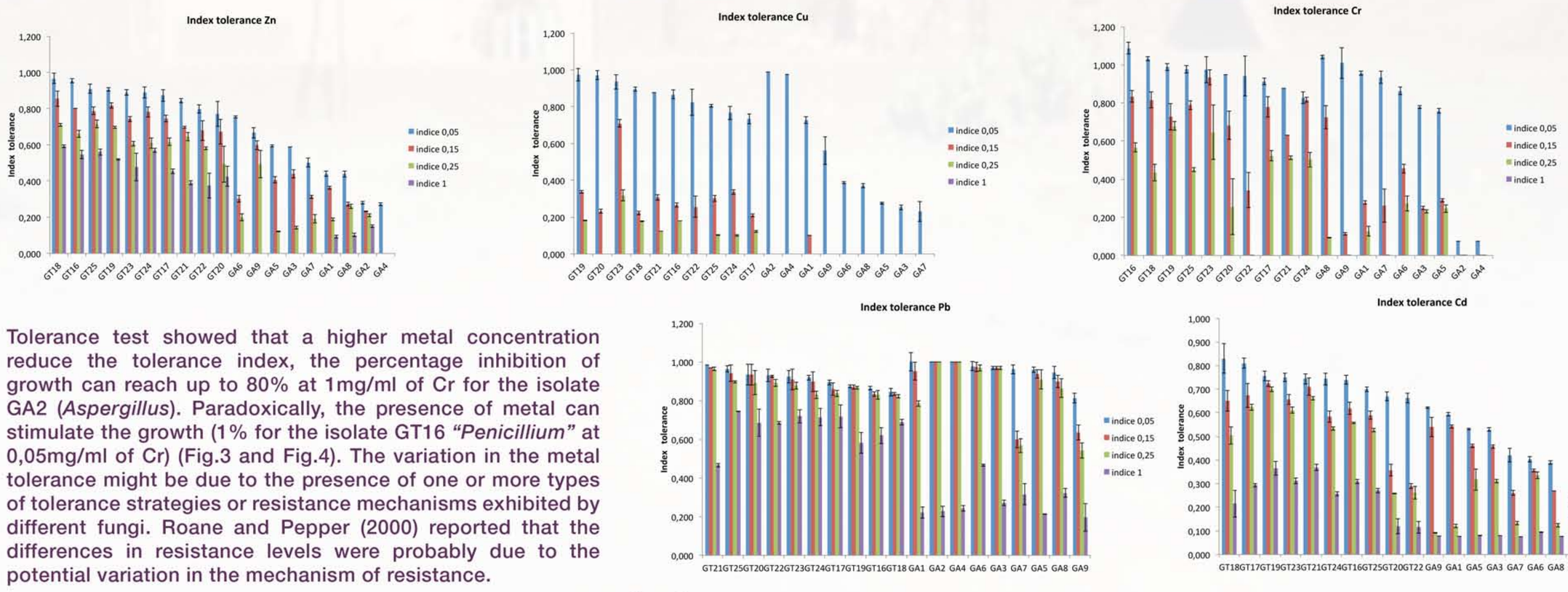


Figure 2 : *Quantities of heavy metal removed.*



Tolerance test showed that a higher metal concentration reduce the tolerance index, the percentage inhibition of growth can reach up to 80% at 1mg/ml of Cr for the isolate GA2 (*Aspergillus*). Paradoxically, the presence of metal can stimulate the growth (1% for the isolate GT16 *"Penicillium"* at 0,05mg/ml of Cr) (Fig.3 and Fig.4). The variation in the metal tolerance might be due to the presence of one or more types of tolerance strategies or resistance mechanisms exhibited by different fungi. Roane and Pepper (2000) reported that the differences in resistance levels were probably due to the potential variation in the mechanism of resistance.

Figure 3 : *Index tolerance of each metal for different isolates tested.*

Figure 4 : *Picture showing difference in the growth after exposure to different concentrations of heavy metals for 15 days.*

Conclusion

The removal of toxic components from industrial effluents is of great importance, not only because of the decontamination effect but also because this removal protects the environment. The microorganisms isolates described here may be of use for removing heavy metals from contaminated effluents.

References

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