ACCLIMATIZATION OF ASSOCIATION BETWEEN *Festuca arundinacea* AND *Lewia* sp. (endophyte) ENHANCES PHYTOREMEDIATION OF PYRENE CONTAMINATED SOIL

Caliope Mendarte Alquisira, Araceli Tomasini Campocosio, David Manuel Díaz Pontones[1], Mariano Gutiérrez Rojas.
Departamento de Biotecnología; [1]Departamento de Ciencias de la Salud. Universidad Autónoma Metropolitana-Iztapalapa. San Rafael Atlixco 186, Col. Vicentina, Iztapalapa 09340, D.F.
utm_2811@hotmail.com

***Key words**: Festuca arundinacea, Lewia sp., endophyte, acclimatization, phytoremediation.*

Introduction. In Mexico, oil pollution affects around 26,000 km^2 of soils. Phytoremediation is an environmentally friendly technology that allows recovers sites like these, through the degradation and/or stabilization of pollutants (1). This process can be improved by combining plants with entophytic fungi that increase the tolerance against contaminants (1). These associations may be more efficient through the acclimatization to the polluted environment to be used in.

The aim of this work was to evaluate the effect of the *in vitro* acclimatization between *Festuca arundinacea* and its fungal endophyte *Lewia* sp. to a hydrocarbon mixture (HM) on pyrene (PYR) removal in an artificially contaminated soil.

Methods. We used 15-days old plants of *F. arundinacea* obtained from surface-sterilized seeds. *Lewia* sp. was isolated from seeds of the same plant (2). The association was induced in lab-tubes with Murashige & Skoog (MS) medium with 10 g/L sucrose and pH 5.8. Each tube was inoculated with 20 mg (dry basis) of fungal biomass. After 20 days of contact, the association was corroborated by multiphoton confocal microscopy. The acclimatization assays were performed in tubes with MS medium and different initial concentrations of a HM (0, 500, 1000 and 1500 mg/L), which was prepared with hexadecane, phenanthrene and pyrene (2:1:1 p/p). All tubes were incubated at 25 °C with 16 h photoperiods for 0, 15, 30 and 45 days of acclimatization. The association was transplanted to pots containing the polluted soil during 60 days more. At the end of this period, the residual PYR was quantified by CG-FID. Each treatment was evaluated with 3 replicates.

Results. The endophyte association between *F. arundinacea* and *Lewia* sp. with the above described methodology was demonstrated to be induced (Fig. 1).

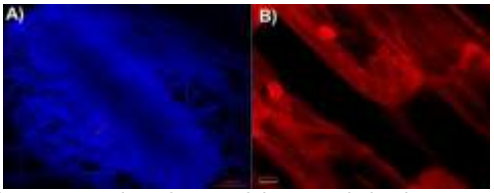

Figure 1. Section of roots of the association between *F. arundinacea* and *Lewia* sp. A) Chitin and cellulose. B) Hyphae compartmentalized within cells of the plants.

The initial concentration of the HM during the acclimatization had no effect on the PYR removal. In contrast, when the acclimatization increased up to 45 days, the association was able to remove 97.5% of PYR (Fig. 2). Our results can be attributed to the fact that increased acclimatization days may induce the production of enzymes and compounds involved in the degradation and detoxification of hydrocarbons, such as peroxidases and glutathione, among others (3).

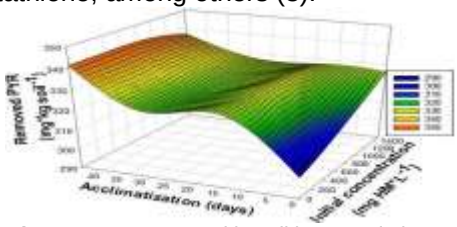

Figure 2. PYR removal in soil by association *F. arundinacea-Lewia* sp. with respect to acclimatization days and initial HM concentration.

Conclusions. Increasing the acclimatization time in the association of *F. arundinacea-Lewia* sp. enhanced the removal of PYR in contaminated soil up to 97.5% extent.

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