SAMIGE / Sociedad Argentina de Microbiología General

# VII CONGRESO ARGENTINO DE MICROBIOLOGÍA GENERAL "SAMIGE DEL BICENTENARIO"

"Dedicado a la presentación de trabajos de investigación básica sobre microorganismos (bacterias, arqueas, hongos y levaduras)"

SAMIGE

Sociedad Argentina de Microbiología General

**18 al 20 de Mayo, Centro Cultural "Ing. Eugenio F. Virla"**
**Universidad Nacional de Tucumán,**
**San Miguel de Tucumán, Tucumán, Argentina**
**2011**

## Bioremediation and Biocontrol

### BB P01. Cr(VI) bioremediation by Streptomyces sp. MC1: Effect on Zea mays

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In natural water and subsurface soils, chromium (Cr) occurs in two major oxidation states: III and VI. Cr(VI) is the major chromium species used in industry and is the common pollutant in soil and waste water, while Cr(III) is a relatively insoluble and non-toxic. Cr(VI) produce toxicity acute and chronic, neurotoxicity, dermatotoxicity, genotoxicity, carcinogenicity and immunotoxicity. Cr(VI) compounds are 1,000 fold more toxics than Cr(III) compounds. Biological transformation of Cr(VI) to Cr(III) by enzymatic reduction is a means of decontamination. This biological reduction may provide a less costly and environmentally friendly approach to remediation. On the other hand, vegetables can be used as contamination level markers because they can accumulate these compounds.

A Cr(VI) resistant actinobacteria strain, Streptomyces sp. MC1, previously isolated, in our lab, from sugar cane, has the ability of reducing Cr(VI) in sterile soil samples. The aim of this work was to evaluate the ability of this strain to bioremediate non sterile soil samples contaminated with Cr(VI), using Zea mays as bioindicator. Streptomyces sp. MC1 was grown in Tryptic Soy Broth (g $L^{-1}$: Tryptone 15, Soy Peptone 3, NaCl 5, $K_2HPO_4$ 2.5 and glucose 2.5) during 3 days at 30 °C. Glass pots were filled with 200 g of soil and kept at 20% humidity with distilled water. $K_2Cr_2O_7$ solution was added at final Cr(VI) concentration of 200 mg $kg^{-1}$ of soil. Later these soil samples were inoculated with pre-cultured Streptomyces sp. Mc1.

Zea mays seeds were sterilized and sown on plates with agar. Maize young plants were potted at the same time of Streptomyces inoculation (t0), 14 days (t14) and 28 days (t28) after the inoculation, and cultivated during 14 days. Chromium bioavailability was measured by atomic absorption spectrophotometry (AAS) after centrifugation 1 g of soil at 5540 g. Maize biomass was estimated as dry weight. Chromium accumulate by plants was measured by AAS after plant treatment with concentrated $H_2SO_4$. Streptomyces sp. MC1 was able to reduce Cr(VI) bioavailability up to 73% after 42 days. Similarly, Zea mays reduced up to 70% of chromium bioavailability, in absence of Streptomyces sp. Mc1.

On the other hand, Zea mays biomass decreased up to 88% with Cr(VI) and without Streptomyces sp. MC1. However, Zea mays biomass decreased only 32% in presence of Cr(VI) and Streptomyces sp. MC1, when the sow was made after 28 days of inoculation, and Cr(VI) bioavailability was reduced up to 97%.

This is the first report where Streptomyces sp. MC1 and Zea mays show a synergic effect that could be useful to bioremediate Cr in soil samples.

### BB P02. Biosurfactant production by autochthonous bacteria from Reconquista River basin: screening and extraction.

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Biosurfactans are natural surface-active compounds with a hydrophilic head and a hydrophobic tail. They are involved in the emulsification of non-aqueous phase liquids, allowing the degradation of several organic compounds. Moreover, biosurfactans can act as metal ligands decreasing their bioavailability in the environment. Biosurfactants can be produced by a large group of microorganisms and are mostly glycolipids, phospholipids, polysaccharide-lipid complexes, lipoproteins-lipopeptides, hydroxylated and cross-linked fatty acids. The aim of our work is to make a screening for surfactant production of the isolates: Pseudomonas veronii 2E, Delftia acidovorans AR, Ralstonia taiwanensis M2, Klebsiella ornithinolytica 1P and Klebsiella oxytoca P2 to study complexing capacity with Cu(II), Cd(II) and Zn(II) for their application in future metal bioremediation techniques. Surfactant production was evaluated by three methods: a) Blue Agar (BA) plates (g/L: $(NH_4)_2HPO_4$ 1.5, $KH_2PO_4$ 4, yeast extract 0.4, CTAB 0.2, glucose 20, methylene blue 0.015, $MgSO_4.7H_2O$ 1.97, agar 3) were inoculated and surfactant production was evidenced by the development of a dark blue halo after 24-48 hs at 32°C; b) Blood Agar (BLA) plates were inoculated to test for haemolytic activity, which is related to biosurfactant production and c) Surface Tension (ST) of bacterial cultures´ supernatants was measured using a stalagmometer. In addition, haemolytic activity was tested in cultures´ supernatants in BLA plates. Surfactant production was examined in the presence of two different carbon sources in a minimal broth: sunflower (SF) oil (5%) and glucose (20 g/L). Our results indicated that SF oil was a better carbon source for surfactant production stimulation. Supernatants of 2E and M2 cultures presented a decrease in ST values (68.03 and 60.59 dyn/cm respectively), compared to medium (71.73 dyn/cm) or water (72.75 dyn/cm) and haemolytic activity was registered when SF oil was the carbon source. Persistent emulsification of SF oil in aqueous phases was observed in M2 cultures. Although P2 presented low ST value (68.61 dyn/cm) when glucose was the carbon source (78.55 dyn/cm in control medium), no haemolytic activity was observed in its culture supernatant. However, culture supernatants obtained with SF oil developed haemolysis in BLA. Neither AR nor 1P cultures decreased ST comparing to control. In accordance, no haemolytic activity was detected in culture supernatants of AR, eventhough haemolytic activity was observed by growing in BLA plates. These evidences indicate that 2E and M2 are biosurfactant producers, and extraction techniques from cultures had to be developed for a proper chemical identification of the compounds involved. These molecules with emulsificating properties are potential ligands for further metal complexing capacity experiments.