"INFLUENCE OF *Bacillus subtilis* USED AS BIOLOGICAL CONTROL AGENT,IN THE BACTERIAL DIVERSITY IN AGRICULTURAL SOIL WITH PRESENCE OF HEAD SMUT OF CORN".

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Introduction. The biological control is the use of organisms to reduce the population density of another organisms, weeds and diseases, this practice has been increasing in agriculture to be friendly with environment[(1)]. The bacteria commonly used in the management of plant diseases caused by fungi is *Bacillus subtilis*[(2)], this microorganism has a positive effect on the control of the fungus *Sporisorium reilianum*, agent causal of head smut of corn, which is a systemic infection in where the symptoms become obvious at flowering time[(3,4)]. It has been reported that rhizospheric microbial communities have a positive effect on the crops, since they can promote plant growth and protect against attack by phytopathogenic fungi[(2)].

In this regard, studies of microbial diversity helps know and culture conditions the health of an ecosystem[(5)], the objective in this project is to analyze the effect of the use of *B. subtilis* as biological control agent of head smut, in the diversity of bacterial communities in agricultural soils using molecular techniques.

Methods. Experimental model was established in Mixquiahuala, Hgo., on demonstration plots with two tillage systems (traditional and conservation). Tests were performed as follows; seeds of commercial hybrid maize susceptible to the disease (AS1601) were inoculated with *B. subtilis* ($1X10^8$ cells/mL) using vacuum. Seeds were planted in plots of 1.20 X 5 m with four grooves in which was added two seeds each 17.5 cm until a total of 80. Were subsequently covered with 1g prepared teliospores in soil (30.5 g of teliospores / kg soil). The control treatment was performed without the addition of bacteria. After 15 days we applied a reinforcement and two months later was thinned to leave in each groove a total of 30 plants. Soil rhizospheric samples were taken every 15 days from treated and untreated plants with biological control agent, which were used for metagenomic DNA extraction. The DNA samples are being used for amplification of the 16S rDNA, to measure bacterial diversity by DGGE.

Results. The experimental model was established in corn crops in where were taken a total of 160 samples rhizospheric soil, 58 correspond to the traditional tillage plots and 102 correspond to the conservation tillage plots. The purification method was standardized for DNA extracted from soil samples, which allowed the amplification of 16S rDNA gene and using as control the DNA extracted from *Escherichia coli* (Fig 1). Optimal conditions have been established for carrying out the DGGE electrophoresis of fragments genes 16S rDNA amplified by PCR.

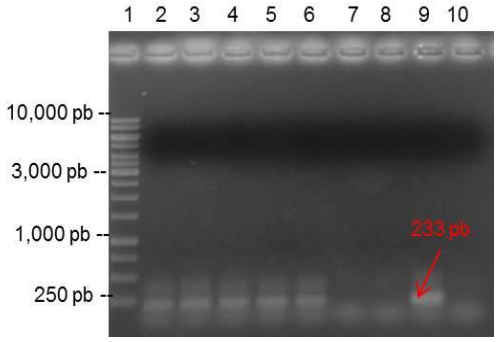

Fig.1 PCR amplification of DNA extracted from agricultural soils (lane 2-8), positive-control (lane 9), negative-control (lane10).

Conclusions. Methodological development made possible to obtain DNA from soil with the necessary quality for amplification of genes of interest, and the analysis by DGGE.

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