BIODIVERSITY AND FUNCTIONAL ACTIVITIES OF MICROORGANISMS FROM MANGROVE OF SÃO PAULO STATE

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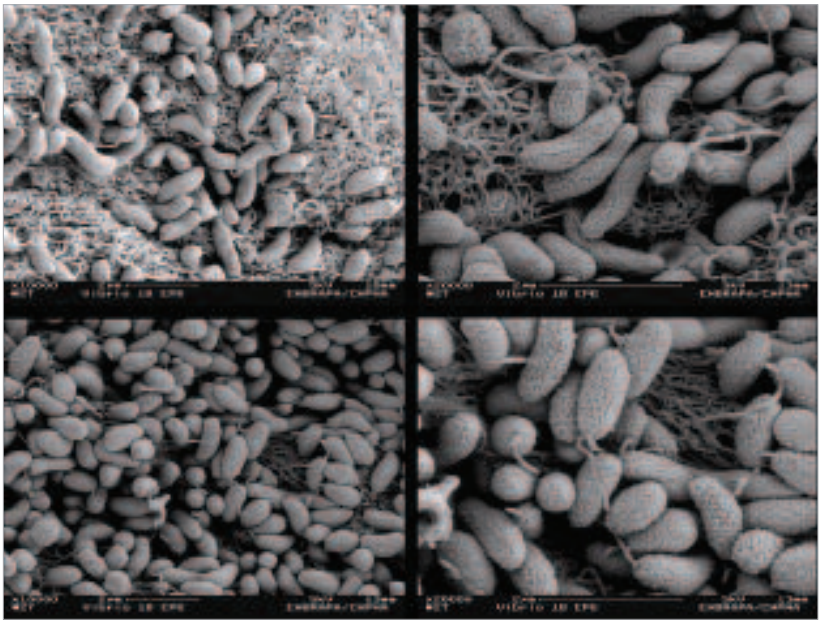
Mangrove communities are recognized as highly productive ecosystems that provide large quantities of organic matter to adjacent coastal water in the form of detritus. This ecosystem is rich in organic matter, in general are nutrient-deficient, especially of nitrogen and phosphorus. Microbial activity is responsible for major nutrients transformations within a mangrove ecosystem. So it is open up new areas of biotechnological explorations, which drive the necessity to isolate and culture these organisms. The biochemical versatility and diversity of rare microorganisms represent an enormous variety of genes that are still unknown. The objectives of this project were: to study the biological diversity of microorganisms from sediments, rhizosphere and endophytes, especially, cyanobacterias, bacteria, fungi, actinobacterias and archeobacteria and to study the molecular diversity present within a species and genus. In this study, so far, abort 2,100 bacteria, 56 actinobacteria, 850 fungi and 27 cyanobacteria were isolated from sediments, philosphere and rhizosphere of red, white and brown mangroves.53 endophytic bacteria, isolated from Rhizophora mangle, were able to fix atmospheric nitrogen. Most of these diazotrophytic bacteria were identified as *Bacillus spp.*, *Pseudomonas spp.* and *Vibrio sp*. Some of the cyanobacteria strains isolated produced secondary metabolites against Micrococcus luteus, and some of them, identified as Phormidium, Synechococcus and Leptolyngbya synthesized polyketides. Evidence for the *in vitro* antibiosis of 86 fungi was demonstrated. Organic extracts from five strains were inhibitory to the human fungal pathogens, *Aspergillus fumigatus* and *Candida albicans*. Two fungal strains completely inhibited the growth of *Trypanossoma cruzi*. Bioassay studies showed that compounds produced by two *Streptomyces* strains, isolated from rhizosphere of red mangrove, had antifungal activity against *Pythium*, an important plant pathogen in hydroponic system. In greenhouse conditions, these strains reduced the disease in cucumber. These actinobacteria may be useful for biocontrol and other applications.



Endophytic strains of Vibrio sp. *isolated from red mangrove*

SUMMARY OF RESULTS TO DATE AND PERSPECTIVES

In this research, it was found a great diversity of fungi and bacteria from mangrove sediments. So far, 2.100 bacteria, 850 fungi, 56 actinobacteria and 27 cyanobacteria were isolated. At the same time, the project investigates the non-cultured diversity of bacterial communities. ARDRA (Amplified Ribosomal DNA Restriction Analysis) revealed the presence of 10 ribotypes, further identified in the following genera: *Vibrio, Listonella, Aeromonas, Microbacterium, Dermabacter, Brevibacterium, Paenibacillus, Staphylococcus, Kurthia, Bacillus, Nesteronkonia, Kytococcus, Kocuria* and *Rothia*. Additionally, DGGE technique applied to access non-culture bacteria determined that Alphaproteobacteria responded to seasonal variation, while Betaproteobacteria communities were different in the summer and winter.

The biodiversity of cellulolytic bacteria was also studied. Mangrove leaves and wood are mainly of lignocellulose components that are degradable by microorganisms, and bacteria are responsible for most of the carbon flux in tropical mangrove sediments. Cellulolytic activity was found in 31 rhizobacteria that produced high amount of endoglucanase (1,4-β-D-glucan). The confirmation of endoglucanase activities was proved by amplification of the *EglA* gene. The production of cellulase was restricted to the genera *Bacillus* and *Paenibacillus*. In high concentration of salt the strains produced significant amount of biofilms suggesting the function of this polymer as mechanisms of tolerance to saline habitats.

In this study, 53 endophytic bacteria strains, most of them belonging to the genera *Bacillus* and *Pseudomonas* and *Vibrio* were isolated from *Rhizophora mangle*. All strains, isolated in selective nitrogen free broth medium, showed the potential in fixing N_2. This is the first report of endophytic strains of *Vibrio*. These results indicate that endophytic bacteria from mangrove may provide an advantage as nitrogen – fixer agents.

Bacterial strains, isolated from contaminated mangrove of Bertioga, São Paulo, produced biosurfactants that reduced the surface tension. One *Pseudomonas stutzeri* strain which produced water/oil emulsifications was able to grow in the presence of petroleum and its derivates.

Bioprospection studies are in course involving microorganisms from mangrove. Some fungal strains produced antifungal antibiotics against *Trichophyton rubrum, Candida albicans, Aspergillus fumigates* and *Criptococcus neoformans*. Extracts of two fungi also were effective against *Trypanossoma cruzi*. Some cyanobacteria, identified as *Phormidium*, *Synechococcus* and *Leptolynybya*, amplified the PKS gene (polykeitide synthase). These antibiotics were active against *Micrococcus luteus*.

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