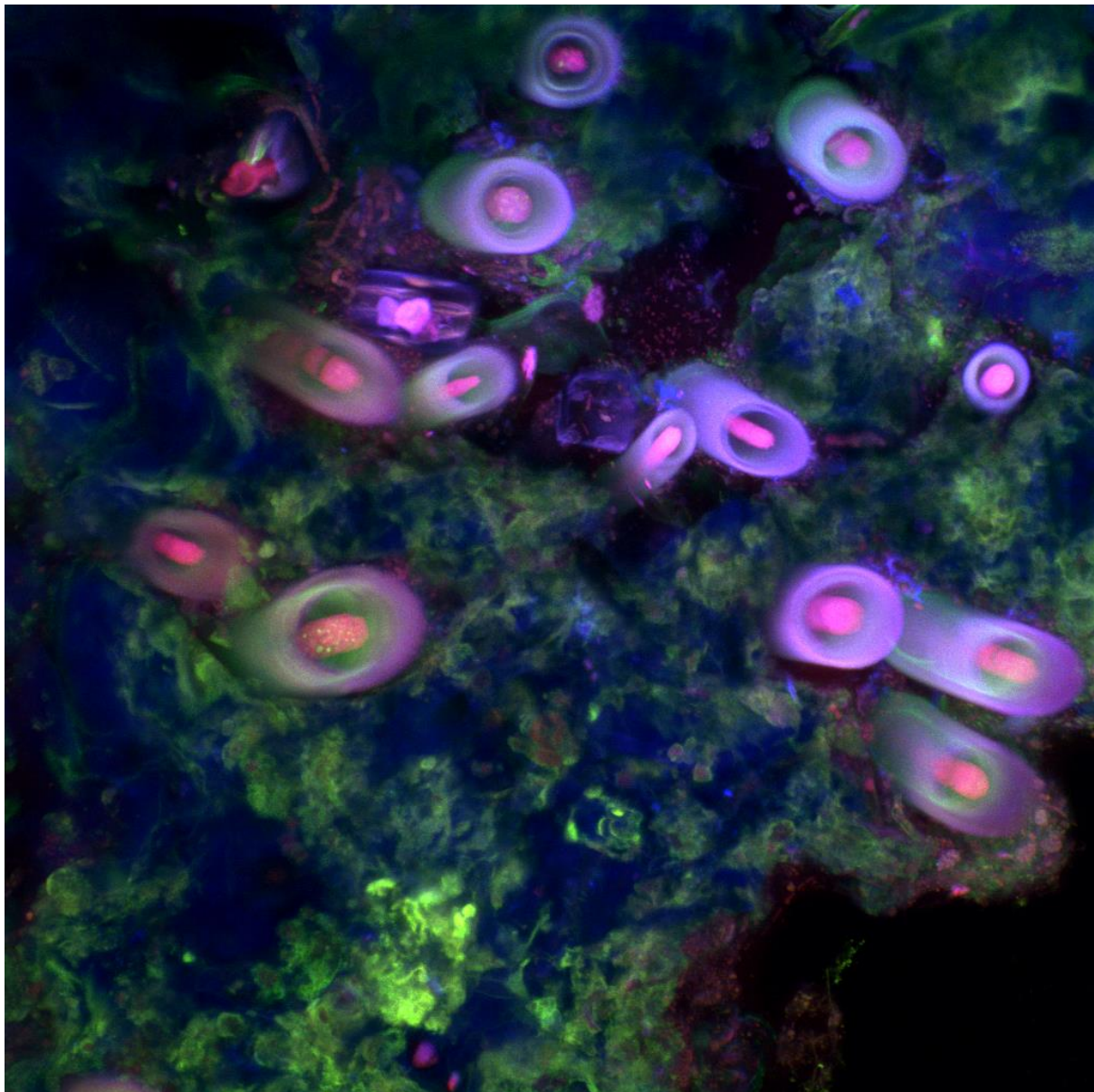




LVI SAIB Meeting – XV SAMIGE Meeting



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Cover image:

Mineral–microorganisms interactions

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A Confocal Laser Scanning Microscopy image of a resin-embedded microbialite from Laguna Negra (Puna-Catamarca), stained with calcein (a fluorescent dye that produces a stable complex in the presence of calcium and fluoresces in the green region of visible light). Mineral aggregates are observed in blue. Their surfaces are partially stained with calcein, indicate the presence of free Ca²⁺ ions. Diatoms and *Rivularia halophila* filaments are visible in red thanks to their photosynthetic pigments.

MI-P56-270

SILVER NANOPARTICLES AS POTENTIAL ANTIBIOFILM AGENTS AGAINST *Candida albicans*, *Candida tropicalis* AND *Candida glabrata*

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Infections caused by biofilm-embedded pathogens decrease the efficacy of traditional treatments and increase antimicrobial tolerance. In addition, most of the human bacterial infections are biofilm-associated. *Candida albicans* is the yeast most frequently isolated in fungal infections, followed by *Candida glabrata* and *Candida tropicalis*. Biofilm formation is a complex process with different growth phases (early, intermediate, and maturation). Nanoparticles (NPs) are potential candidates to obtain an antifungal (ATF) activity, thus, preventing the first stages of fungal colonization and avoiding the subsequent formation of biofilms. The objective of this work was to evaluate the effect of biosynthesized silver NPs (AgNPs) against initial and mature biofilms in *Candida albicans*, *Candida tropicalis* and *Candida glabrata*. *C. albicans* SC5314, *C. tropicalis* NCPF 3111, and *C. glabrata* ATCC 2001 were studied. The minimum inhibitory concentration (MIC) and the minimum fungicidal concentration (MFC) of AgNPs were determined by the microdilution method according to Clinical & Laboratory Standards Institute M27-4th ed. The inhibitory concentration of the biofilms (%I) was evaluated over the initial stages of formation. The concentration of reduction of the biofilms (%R) was evaluated against the mature biofilms (48 h), being exposed to concentrations of AgNPs (supraMIC, MIC and subMIC). Biofilm formation was achieved through the ability of microorganisms to adhere to wells of flat-bottomed 96-well microplates and quantified by Crystal staining. The biofilms were disrupted by sonication (40 kHz, 60 s) in order to re-suspend and recover viable sessile cells and determined by plate counting (colony-forming units per mL, CFU mL⁻¹). The same MIC and MFC values were found for AgNPs in *C. albicans* 3.7 x 10⁻² pM, in *C. tropicalis* was 5.0 x 10⁻¹ pM and finally *C. glabrata* shown 1.2 x 10⁻¹ pM for AgNPs. The %I was 61 ± 8 (3.7 pM AgNPs) for *C. albicans*, 65 ± 3 (with 38.5 pM AgNPs) for *C. tropicalis*, 84 ± 4 (12.5 pM AgNPs) for *C. glabrata*. The %R was 53 ± 3 (3.7 pM AgNPs) for *C. albicans*, 84 ± 9 (38.5 pM AgNPs) for *C. tropicalis*, 69 ± 9 (77 pM AgNPs) for *C. glabrata*. Exposure to AgNPs leads to reduction in microbial biomass and surviving sessile cells, reduced by more than 10% in the three *Candida* species. This study demonstrated that AgNPs exert an antibiofilm effect against several *Candida* species, suggesting its potential application as an antibiofilm agent alone or in combination with traditional antifungal agents.

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TAXONOMIC GENE MARKER BENCHMARKING BY USING A MACHINE LEARNING APPROACH

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Bacillus cereus of Clade 2 is composed of *B. thuringiensis* and *B. cereus sensu stricto* genomospecies. The former has important agronomic applications whereas the latter is usually associated with food poisoning. Here, gene markers used for identification of these genomospecies were evaluated using "Random Forest", a machine learning technique based on decision trees. First, 2459 available genomes of *B. cereus* group were downloaded from Genbank. In order to select *B. cereus sensu stricto* and *B. thuringiensis* genomes, their average nucleotide identities (ANI) were computed against type strains *B. thuringiensis* ATCC_10792 and *B. cereus sensu stricto* ATCC 14579. 1253 out of all genomes were selected for further studies as they shared an ANI greater than the species-threshold of 96% with type strains of Clade 2. We determined as minimum quality thresholds criteria for genome exclusion based on their deviation (mean ± 2 sd) from expected genome sizes or the number of contigs (n) and N50 parameters of its assembly. Those genomes with n > 616, N50 < 28.036, size < 4.940.889 bp or size > 6.536.009 bp were classified as of low-quality and excluded. To verify their genomospecies assignments the resulting 863 sequences were further analyzed using a phylogenetic approach with 104 common ancestral genes present in all genomes under analysis including the outgroups *B. anthracis* Ames and *B. mycoides* ATCC 6462. Then, a training group with 697 strains was selected to train a forest of 10.000 trees and construct a classifier of genomospecies using the gene distances of 22 taxonomic gene markers as variables (15.334 variables). DNA gyrase subunit A (*gyrA*), pyruvate carboxylase (*pyc*), and DNA topoisomerase (*gyrB*) were found the 3 most important markers for the classifier. One-gene classifiers with a forest of 1.000 trees were constructed using the gene distance of *gyrA*, *pyc*, or *gyrB* genes (697 variables each one). Noteworthy, cross-validation analyses of these classifiers showed that the accuracy and kappa parameters were zero and one in all cases, respectively. Then, correlated variables (at 0.999) were pruned by preprocessing the data reducing the variables from 697 to 7, 17, and 8 for *gyrA*, *pyc*, and *gyrB* classifiers, respectively. Finally, the error rates were computed for the classifiers using the testing group (158 strains). No misclassifications were observed indicating that the classifiers are accurate as well as unbiased. Our pipeline could be used to select proper taxonomic markers to massively assign genomospecies identities in comparative genomic or metagenomic studies at a high-resolution level.