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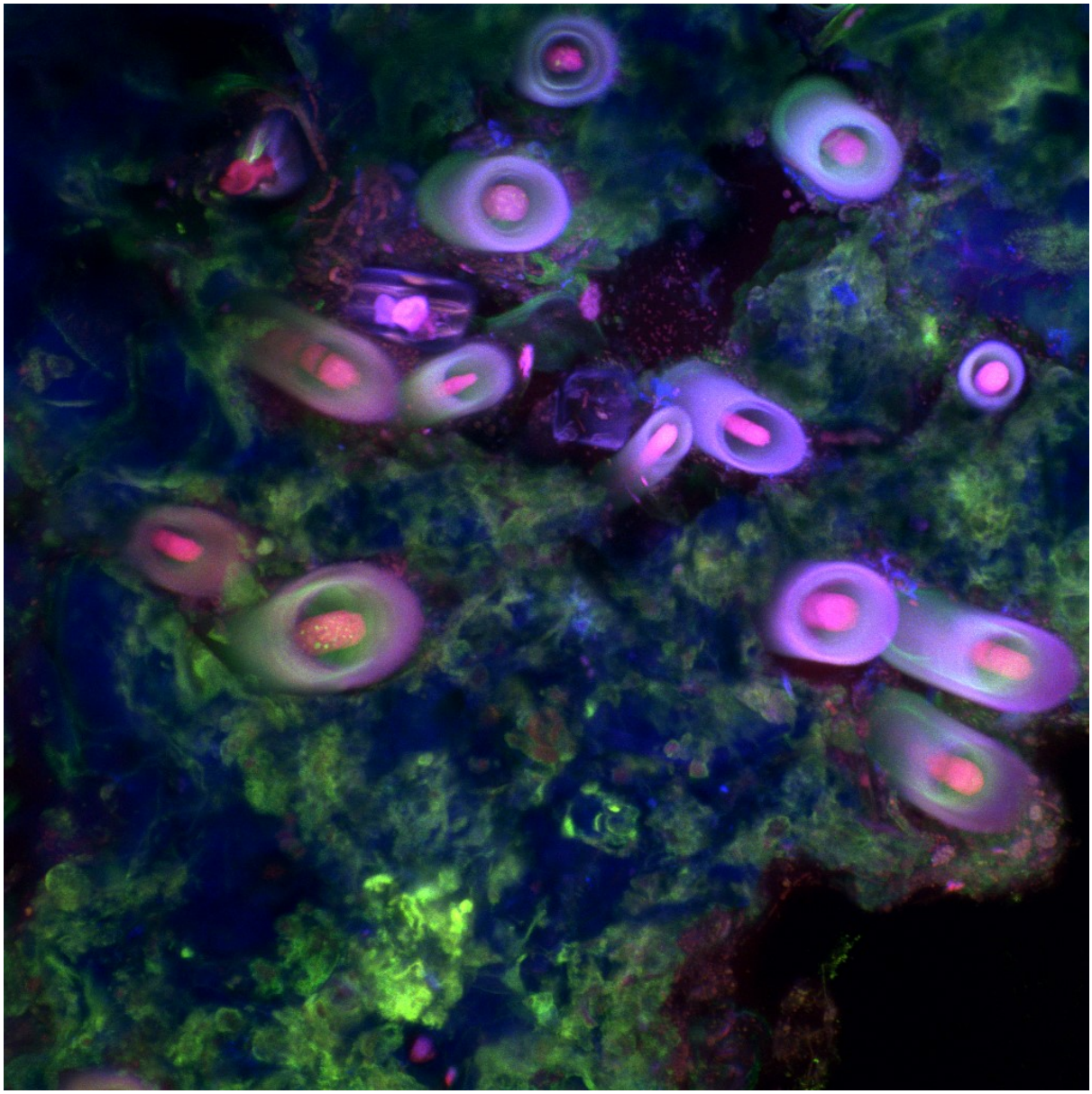

SAIB-SAMIGE Joint Meeting 2020 – *Online*

Cover image:

Mineral–microorganisms interactions

Mlewski EC[1], Gérard E[2]
[1]Centro de Investigaciones en Ciencias de la Tierra CICTERRA-CONICET-UNC. [2]Institut de Physique du Globe de Paris, IPGP.

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 Ca^{2+} ions. Diatoms and *Rivularia halophila* filaments are visible in red thanks to their photosynthetic pigments.

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MI-P23-139
FROM ALFALFA TO SOYBEAN: IDENTIFYING BENEFICIAL MUTATIONS TO EDIT MULTIPLE BACTERIAL LOCI BY CRISPR-CAS9 SYSTEM

Frare R[1,2], Galindo Sotomonte L[1,2], Pascuan C[1,2], Liebrenz K[1,2], Soto G[1,2], Ayub N[1,2]
[1]IABIMO-CONICET, [2]IGEAF-INTA, Buenos Aires, Argentina. E-mail: romina.frare@gmail.com

Alfalfa is the main forage worldwide due to its high biomass production, excellent nutritional qualities, and adaptation to a wide range of environments. In addition to its indirect impact on human nutrition, alfalfa is also the most important legume crop in cultivated areas worldwide after soybean (around 30 and 120 million ha, respectively). The evolutionary and economic success of the legumes species is principally due to their symbiotic association with nitrogen-fixing bacteria (i.e., rhizobia). We recently have shown that commercial alfalfa inoculants (strains B399 and B401), which are closely related to the model alfalfa symbiont *Sinorhizobium meliloti* 1021 (99.99% of nucleotide identity at genomic level), have lost an extensive number of genes during rhizobial domestication, and that this complex evolution process determines the effectiveness of legume–rhizobia symbiotic interaction under field conditions (*J Mol Evol* 2017 PMID: 28828631; *J Biotechnol* 2017 PMID: 29050878, *Microbiol Ecol* 2018 PMID: 29330647; *Microbiol Ecol* 2020 PMID: 31828388). To understand this process deeply and to extend the benefits of this knowledge to other public legume breeding programs (mainly, the massive production of knockout soybean rhizobia via CRISPR-Cas9 genome-editing system), we performed bioinformatics analyses for the identification and functional classification of missing genes (both pseudogenes and deleted genes) and studied their occurrence in other important rhizobia including soybean inoculants.

MI-P24-140
STRUCTURAL AND FUNCTIONAL ANALYSES OF Pr-INDUCED VARIANTS OF *Xcc*BphP BACTERIOPHYTOCHROME FROM *Xanthomonas campestris*

Conforte VP[1], Otero LH[2,3], Toum L[1], Sirigu S[4], Antelo G[2], Rinaldi J[2], Klinke S[2,3], Chavas L[4], Vojnov AA[1], Goldbaum FA[2], Malamud F[5], Bonomi HR[2]
[1]ICT MILSTEIN-FPC-CONICET. [2]FIL, IIBBA-CONICET. [3]PLABEM. [4]Synchrotron SOLEIL, France. [5]Dto. de Cs. Básicas, UNLU-CONICET.

Photoreceptors are able to detect light and transduce that signal generating a cellular response. Among them are the red/far-red light sensing bacteriophytochromes (BphP). These bilin-binding proteins have the ability to photoswitch between two states, a red-absorbing (Pr) and a far-red-absorbing (Pfr), by the isomerization of the bilin chromophore and generating structural changes that result in the transduction of the light signal into biochemical signaling. The genome of *Xanthomonas campestris* pv. *campestris* (*Xcc*), the causative agent of black rot in crucifers, codes for a functional BphP (XccBphP) which was extensively described in previous studies. It has been defined as a negative regulator of several light-mediated mechanisms involved in its virulence. Here, we deepened the analysis of the XccBphP structure and function. In this work, we designed and constructed three different variants with single amino acid changes that affect XccBphP photocycle favoring its Pr state: L193Q, L193N and D199A. We purified the recombinants full-length mutants, crystalized them and solved their crystal structures, showing a Pr conformation almost identical to the wild-type previously obtained. We also examined their UV-Vis absorption spectroscopic properties, proving that Pr is their preferred state. After establishing the effect of each mutation on the structure of XccBphP, we tested the effects of altering the *Xcc*BphP photocycle using exopolysaccharide (EPS) production and stomatal aperture assays as indicators of its bacterial signaling pathway. Null mutant complementation assays showed that L193Q or L193N Pr-stabilized versions decreased bacterial EPS production in darkness or under far-red light and increased it under red light in an amplified manner compared to the complementation with the wild-type version. Furthermore, strains expressing Pr-favored *Xcc*BphP versions could not promote stomatal reopening at *Xcc* null mutant levels when tested in *Arabidopsis* epidermis. Taken together, our results highlight the relevance of the *Xcc*BphP Pr-Pfr balance in physiological processes in *Xcc*.

MI-P25-141
IN SILICO PREDICTION AND *IN VITRO* DETECTION OF PROPHAGES AND CRISPR-CAS IN BACTERIAL GENOMES OF ANTARCTIC ENVIRONMENTS

Napolitano N[1,4,5], Pelliza LA[2], Aran M[2], Quiroga C[3], Mac Cormack WP[4,5], López JL[1]
[1]IBAVIM, UBA. [2]IIBBA-CONICET. [3]IMPaM-CONICET-UBA. [4]NANOBIOTEC, CONICET- UBA. [5]IAA.
E-mail: nicolasnapolitano1@gmail.com

Bacteriophages play a key role in microbial evolution, biogeochemical cycles, and human diseases. Phages are genetically diverse, and the particular architecture of their genomes is the result of constant genetic exchange with other phages and the genome of their hosts, which highlights their complex evolutionary history and generates a wide spectrum of genomic diversity. On the other hand, bacteria encode in their genome defense mechanisms to avoid infection. Our hypothesis is that the Antarctic environmental bacteria, scarcely explored in their content of prophages and defense systems, could show a new genetic spectrum of viruses and defenses. Therefore, the aim of this work is to predict *in silico*, induce *in vitro* with Mitomycin C and characterize the presence of prophages and CRISPR-Cas systems in Antarctic bacteria. For this purpose, three isolates whose complete genome was obtained in the laboratory were explored *in silico* in search of prophages (ACLAME Prophinder