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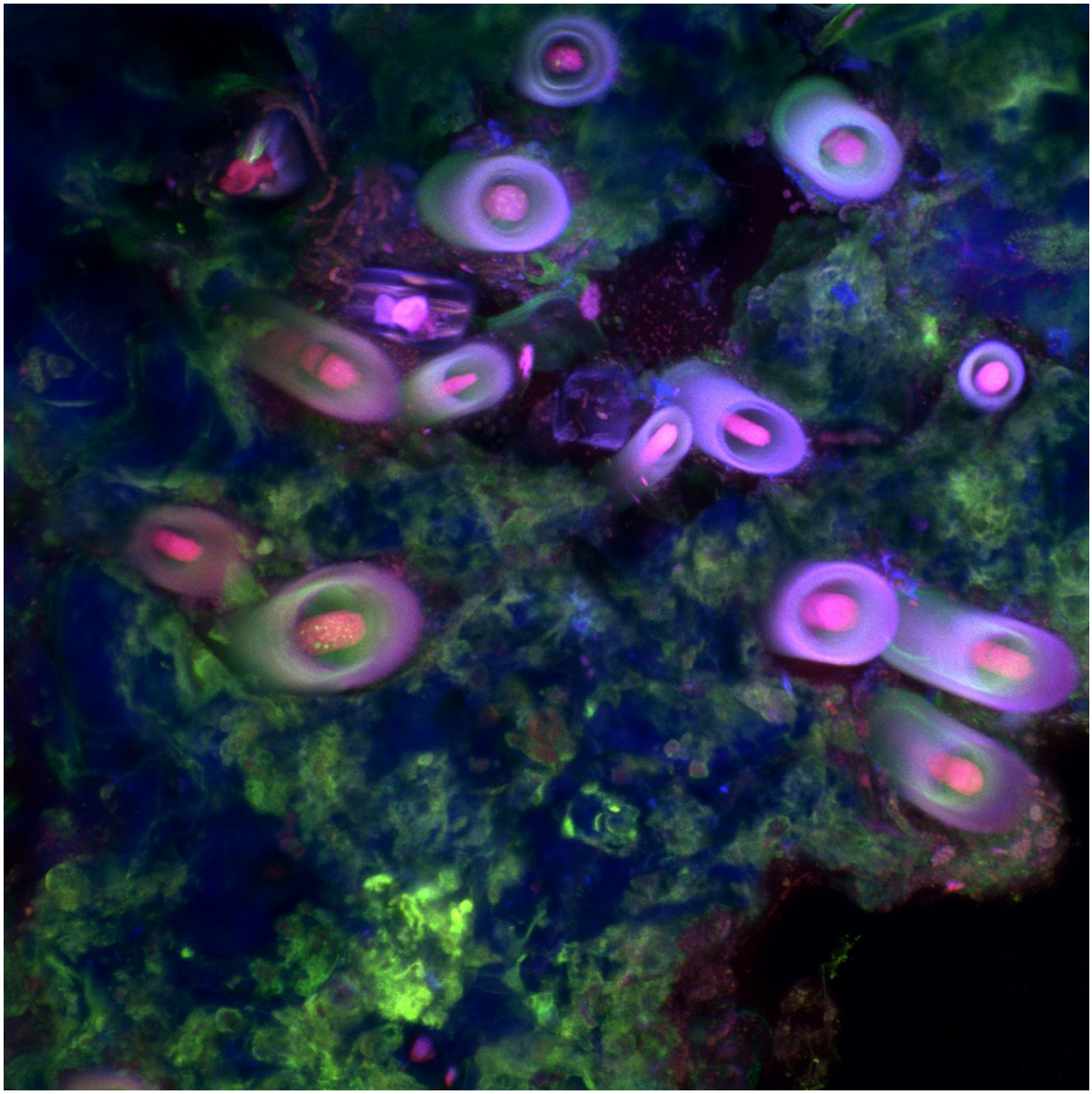
LVI SAIB Meeting – XV SAMIGE Meeting



SAIB-SAMIGE Joint Meeting 2020 – *Online*

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

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LVI Annual Meeting
Argentine Society for Biochemistry and Molecular Biology
(SAIB)

XV Annual Meeting
Argentinean Society for General Microbiology
(SAMIGE)

SAIB-SAMIGE – Online
Joint Meeting 2020

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UPRs. Finally, we are also studying the effect of Taq3-FNR and CFP-Taq3-FNR stable expression in *Arabidopsis thaliana*. Early results indicate that the phenotype of these stable lines resembles that of ClpB-overexpressing lines, which further indicate that accumulation of Taq3-FNR in the chloroplast causes an increment in the levels of chloroplastic PQC chaperones.

PL-P26-288
SERRALYSIN-LIKE PROTEIN OF *Candidatus liberibacter asiaticus* AFECTS EXTRACELULAR MATRIX IN HETEROLOGOUS SYSTEMS AND VIRULENCE IN HOST PLANTS

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Huanglongbing (HLB) is the major threat for *Citrus* species and it is caused by intracellular alfa-proteobacteria belong to the genus *Candidatus* Liberibacter (*Ca*L), being '*Ca*. L. asiaticus' (*Las*) the prevalent specie. This bacterium is psyllid-transmitted and resides within the citrus phloem. Infected trees symptoms -blotchy and mottled leaves, small fruits, as well as shorter lifespan and premature fruit drop- have been associated with an increase of callose deposition and starch accumulation that constricts symplastic transport producing a source-sink imbalance. The inability to culture *Ca*L has hampered progress in studying the pathogen and its virulence mechanisms, our limited understanding relies on transcriptome and proteomic data and on the expression of putative virulence proteins in surrogate models. Those studies highlight the idea that *Las* actively manipulate plant defense response. Extracellular proteases which belong to the Serralysin metalloprotease family are wide known virulence factors exported through the secretion system type I (SST1). A putative serralysin gene was identified in *Ca*L genomes and its expression levels was increased when *Las* changed it environment from the psyllid to the citrus leaf, suggesting a function in the infection process. Here, we study the function of the Las-serralysin (hereafter Las_1345) as virulence factor. First, we decipher whether Las_1345 could be secreted through bacteria that express SST1-secreted proteases such as *Serratia marcescens* (*Smc*) and *Xanthomonas citri and X. campestris* (*Xac* and *Xcc,* respectively). Las_1345 only was detected in the pellet fraction of every surrogate bacteria, nevertheless, we measured intracellular protease activity. Las_1345 expression increased the proteolityc activity of *Xcc* by more than 50% and partially restored (5–10%) the protease activity of protease deficient *Smc* strain (*prtA*). Las_1345 purified from *E. coli* also showed low proteolytic activity. Then, we analyzed whether Las_1345 can exert its activity on plant proteins. Las_1345-GFP was localized at the cell membrane in *Nicotiana benthamiana* and tissue integrity was not affected. Moreover, protease activity measured in tissue cells expressing Las_1345 arose similar levels as in control tissue, suggesting that Las_1345 may not act as protease in plant tissue. Interesting, Las_1345 modify biofilm structure in surrogate bacteria, which was associated with less virulence in citrus plants. We propose that Las_1345 is a virulence factor contributing with Las colonization in the citrus phloem.

SIGNAL TRANSDUCTION

ST-P01-23
α,25(OH)$_2$D$_3$ PROMOTES OXIDATIVE STRESS IN ENDOTHELIAL CELLS TRANSFORMED BY vGPCR

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The infectious cause of Kaposi's sarcoma (KS) neoplasm is KS-associated Herpesvirus (KSHV or human herpesvirus 8). Furthermore, virally G Protein-coupled Receptor (vGPCR) is one of the molecules from the lytic phase able to induce KS-associated cellular modifications through paracrine oncogenesis. We have previously demonstrated that 1α,25(OH)$_2$D$_3$ exerts antiproliferative effects on endothelial cells that stably express vGPCR by inhibiting NF-κB pathway and promoting apoptosis and autophagy. Oxidative stress is frequent in many types of cancer where reactive oxygen species (ROS) can act as a promoting or suppressing agent. In this work, our goal was to study the involvement of ROS as part of the antineoplastic mechanisms triggered by 1α,25(OH)$_2$D$_3$ in vGPCR cells. By a spectrofluorimetric method using the H2-DCF-DA probe, ROS levels were detected higher than control conditions after 1α,25(OH)$_2$D$_3$ (10 nM, 24 or 48 h) treatment. When VDR expression was knocked down by shRNA against VDR (vGPCR-shVDR cell line), ROS increase was found to be VDR dependent (48 h). Our previous reports indicated that vGPCR cells proliferation decreases at 80% after 1α,25(OH)$_2$D$_3$ treatment, triggering cell cycle arrest and apoptosis by a mechanism dependent on the caspase-3 cleavage. In this case, Western blot studies showed an increase expression of pro-apoptotic proteins like BIM and caspase-3 cleavage by 1α,25(OH)$_2$D$_3$ (10 nM, 48 h) and no reversal effect by N-Acetyl-cysteine (1 mM) antioxidant was observed. Altogether, these preliminary results suggest that ROS levels promotion by 1α,25(OH)$_2$D$_3$ through VDR, triggers apoptosis-related mechanisms on vGPCR cells.