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pyoverdine) and enhance traits linked to chronic infection. This adaptive process responds to selective forces such as the highly oxidative environment in the CF lung. Identifying key players involved in this adaptation may aid in designing better antimicrobial treatments. In this study, we analyzed single-nucleotide polymorphisms (SNPs) in 411 human infection isolates (including 59 from CF patients) and 58 environmental habitat isolates of PA. Compared to the PAO1 reference genome, clinical and environmental isolates showed 591,087 and 467,429 SNPs, respectively, across the PA 6.4Mbp genome. Investigating SNP distribution revealed a uniform pattern of transition and transversion SNPs across environmental samples while clinical isolates displayed notably higher density of transversions specifically in the region between 2.50-2.75 Mbp. CF isolates exhibited even higher density levels (4.5-fold) than general clinical isolates in this hotspot of diversification, which includes the genes for the synthesis and uptake of pyoverdine. Further analysis focused on the identified 1378 A to C, a mutation associated with oxidative damage, within these genes. It was found that 56% of SNPs occurred at sites with adjacent 5'G and/or 3'C sequences. Notably enriched at codons encoding Glu (GAG and GAA), Asp (GAT and GAC), Ser (TCG, TCC, TCA, TCT), these mutations often resulted in the highly inactivating Ala substitution (30%). Overall, our findings reveal a mutational signature within the pyoverdine genes from clinical isolates, similar to the one our laboratory previously characterized for the low-fidelity DNA Polymerase IV, suggesting its potential role in adaptive mutation processes.

MI-15

PRELIMINARY ANALYSIS OF THE ROLE OF THE SOIL FUNGUS *Humicolopsis cephalosporioides* IN P MOVEMENT THROUGH MINERALIZATION AND SOLUBILIZATION PROCESSES.

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Humicolopsis cephalosporioides is a soil fungus associated with *Nothofagus* forests in Patagonia (South America). This fungus has been described as able to degrade several lignocellulose-related substrates, suggesting a saprotrophic role in the life cycle of *Nothofagus* ectomycorrhizae, as well as in nutrients cycling, soil development and aggregation. However, as far as we know, there is not data about its role in P movement and availability in the rhizosphere soil, nor its effect on plant growth. Therefore, we initially analyzed the capacity of three isolates of this fungus (LPSC 1155, LPSC 1157 and LPSC 1159, which were isolated from soil particles from *Nothofagus* forests with different management practices) to solubilize different inorganic P sources (including ones varying the amount of Ca atoms per molecule) under different culture conditions (agar and liquid media). We also evaluated their ability to synthesize and release organic acids, which contribute to increase P availability. We also evaluated the ability of the fungi to mineralize lecithin as a model compound containing organic P on liquid medium and to synthesize enzymes with acid and alkaline phosphatase activity. Although a differential behavior of each strain in growth and phosphate-solubilization halo was found on an agar medium containing different inorganic P forms, the ratio between the phosphate-solubilizing halo and colony diameter was mostly higher when the fungi grew on a medium containing monocalcium phosphate. However, the levels of specific organic acids liberated by the fungi, such as ascorbic, citric and malic ones were higher in a medium containing tricalcium phosphate. These results suggest that chemical nature of the P source and the main organic acids released in the environment might modulate differentially the ability of specific isolates of *Humicolopsis cephalosporioides* to mobilize this nutrient. This might be key for selecting promising isolates for increasing the P availability in rhizosphere soil, highlighting its role in P cycling.

MI-16

SURVEY AND CHARACTERIZATION OF PATHOGENIC AND MYCOTOXIGENIC MICROORGANISMS IN WALNUT (*Juglans regia* L.)

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Walnut (*Juglans regia* L.) is the most important nut crop in Argentina, with a cultivated area of about sixteen thousand hectares. The quality and safety of walnut production can be affected by phytopathogenic bacteria and/or mycotoxin-producing fungi. This study sought to explore the incidence of such microorganisms in a walnut plantation in Belén, a department in the province of Catamarca. Fallen fruit showed stilar end necrosis that extended towards the inner tissues and reached the kernel. These symptoms were consistent with those described for brown apical necrosis (BAN). Ten symptomatic fruits from each orchard in the plantation were disinfected with sodium hypochlorite (1% v/v) for 2 min and rinsed with sterile distilled water. Small pieces of affected tissue were excised and plated on potato dextrose agar (PDA) supplemented with streptomycin (200 mg/l). After 7 days, fungal species were isolated and identified on the basis of conidial morphology and through molecular techniques. The DNA of a selected strain named H1 was extracted, and internal transcribed spacer (ITS) regions were amplified and sequenced. A BLAST analysis of the isolate's nucleotide sequences indicated a 97-100% similarity with reference sequences of *Alternaria alternata* in GenBank. Its pathogenicity was studied by inoculating a 10-µl droplet of a conidial suspension (1×10^6 conidia/ml) on the stigmatic

end of walnuts. All the inoculated fruits developed apical necrosis. To validate Koch's postulates, symptomatic tissue from the inoculated fruits was plated on PDA, and H1 was successfully reisolated. A Gram-negative bacterial strain was also isolated and named XVM1. It was characterized at the morphological and biochemical level, and identified as a *Pantoea* sp. through 16S *rDNA* gene sequencing and fatty acid profiling (FAMES). These results were corroborated by MALDI-TOF MS. As with H1, walnut fruits inoculated with this bacterial strain developed characteristic BAN symptoms. To further complement all these findings, walnut-associated mycoflora was studied in the same region. Sixty-one percent of the isolated strains were identified as potential mycotoxin producers, with a high incidence of the genera *Aspergillus*, *Alternaria*, and *Penicillium*. Although species belonging to *Aspergillus* section *Flavi* were the most frequently isolated, low levels of aflatoxins were detected (4.4 ng/g for AFB1). The presence of BAN and aflatoxins in a commercial walnut plantation in Argentina suggests a need for improved phytosanitary control programs. Nevertheless, given the limited scope of the data presented here, epidemiological studies should be conducted to better understand the impact of the disease on walnut production in Argentina.

MI-17

INSIGHTS INTO THE FUNCTIONAL PROPERTIES OF THE MAJOR COMPONENT OF MRCV VIRAL FACTORIES

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The mal de Río Cuarto virus (MRCV, *Fijivirus*, *Spinareoviridae*) causes an important maize disease in Argentina. Fijiviruses replicate and package their segmented dsRNA genomes within viral factories in a process involving RNA-RNA and RNA-protein interactions. Our previous work showed that the viral non-structural protein P9-1 is the major component of MRCV viroplasm and gives rise to dimers that multimerize into doughnut-shaped decamers and dodecamers through the protein 24 C-terminal residues (C-arm). Although both P9-1 and P9-1ΔC-arm catalyzed ATP with similar activities, an RNA-stimulated ATPase activity was only detected in the full-length protein, indicating a C-arm-mediated interaction between the ATP catalytic site and the allosteric RNA binding sites in the (do)decameric assemblies. Here, we deepened into P9-1 functional characterization. Using a classical biochemical approach, we established that P9-1 can hydrolyze GTP and ATP. In addition, we found out that both P9-1 and P9-1ΔC-arm have RNA chaperone activities since they unwind a HEX-labeled ssRNA with a known secondary structure. P9-1 RNA chaperone activity may facilitate sequence-specific, intersegment RNA-RNA interactions to ensure robust assembly of complete viral genomes. Overall, our findings suggest that P9-1 is directly involved in key viral processes, such as genome replication and RNA packaging, making it a potential target for therapeutic intervention. Future work will address the regulation of P9-1 activities.

MI-18

THE SCSABCD SYSTEM, A LINK BETWEEN COPPER AND REDOX STRESS IN THE *Salmonella* ENVELOPE

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Foodborne diseases are among the most prevalent health problems worldwide. In this regard, *Salmonella enterica* is one of the most significant pathogens of both humans and animals, that causes infections ranging from self-limited gastroenteritis to severe invasive illness in susceptible hosts. Like other bacterial pathogens, its interactions with hosts are influenced by transition metals, especially copper (Cu). Cu-sensitive *Salmonella* mutants show reduced survival in macrophages compared with the wild-type strain. Many of the Cu-distribution proteins and cuproproteins reside in the pathogen's cell envelope. The *Salmonella* *scsABCD* operon, absent in *Escherichia coli* but present in other enteropathogens, contributes to both Cu and redox stress tolerance. ScsB, ScsC and ScsD carry putative Cu-binding motifs in their periplasmic thioredoxin-like domains, while ScsA carries a peroxidase motif in its periplasmic domain. Our aim in this work is to delve deeper into the role of the Scs system in maintaining the envelope copper/redox homeostasis in *Salmonella*. Regarding the importance of periplasmic copper homeostasis for *Salmonella* virulence, we tested the intracellular survival of both WT and a *scsABCD* deleted mutant in macrophages invasion assay, both in the presence and absence of copper. We were able to determine that the removal of the whole locus *ScsABCD* is important for survival inside macrophages in the presence of 100μM CuSO₄. We assessed the intracellular copper levels of the *scsABCD* deleted mutant in *Salmonella* as well as the CpxR/A two-component system that controls *scsABCD* transcription. Our data show that, while mutation on the CpxR/A regulatory system leads to reduced intracellular Cu levels both in the presence and absence of external Cu, deletion of the *scsABCD* locus had little or no effect on intracellular Cu levels. To verify this system's expression under copper treatment, we conducted transcriptional analysis of the *scs* locus by qPCR and generated a transcriptional reporter fusing *gfp* to the *scs* promoter. Copper induction was confirmed in the WT strain, both in rich and minimal media. Preliminary results for redox stress response are also