ISSS-ISTA Webinar on Seed pathology - the good and the bad microbes

Louna Colaert - Sentenac and Ruud Barnhoorn have answered all the questions that were sent in during the session in detail below.

You showed us that the microbiota diversity in seedlings varied from the one in the initial inoculum and seeds. The experiment was done using non-sterilised soil for sowing the seeds; did you have a look at the soil community before sowing/after sowing at the time you investigated the seedling-associated communities? What about the possible impact of the soil on the changes in the seed-to-seedling evolution of the microbiota?

LCS: *Thanks! Indeed, seedling microbiota is assembled by the plant from the seed microbiota and the environment, including soil microbiota. We characterized the soil community both before and after sowing. Our inoculated strains are not found back in the soil, but the inoculation seems to impact the rhizosphere community during the first developmental stages of the plant (Arnault et al. 2024 FEMS). We are currently working on the conditions impacting the robustness of Synthetic communities' inoculation effect on plant, and soil microbiota is definitively one factor we'll consider.*

About the Seed Health Image Database platform, you'll be launching. How will ISTA include everyone involved in the seed pathology and verify if pictures correlate with the identified names? And seeing that seeds are used mostly by farmers, how is this going to be shared with them?

RB: *The seed health picture database (DB) is a DB which has the objective to function as 1 an education DB for scientists but also as a reference DB for seed scientists. Monitoring of good pictures is done with the help of experts and reviewers who help whenever a submission is entered. Please enlarge picture on the right for more information.*

Do you do pathogenic tests with Pantoea agglomerans isolate detected?

RB: *We do not test for Pantoea agglomerans ourselves. There is interest from the beet seed producing companies but this most likely will be for P. a. pv. betae. Maybe this article can help you. https://www.researchgate.net/publication/358649664_Validation_of_species-specific_PCR_assays_for_the_detection_of_Pantoea_ananatis_P_agglomerans_P_allii_and_P_stewartii*

Do you do a pathogenic test with Pseudomonas isolates?

LCS: *No, we didn't test the pathogenic effect of the isolated Pseudomonas. We were interested in the strains able to improve our selected traits, but we'll keep the idea!*

I would like to know, were any of the isolates pathogenic? And lastly, were there any tests done on the isolates you used to confirm them as beneficial microorganisms?

LCS: *Some isolates we selected had a detrimental effect on seedling emergence when inoculated in single-strains inoculation, but we didn't conduct pathogenicity tests. The selected strains were used to design Synthetic Communities that we further tested in inoculations: we assess their effect on seed germination, seedling emergence and phenotype, and are studying their effect on the plant metabolism to understand their action. We will continue to assess their beneficial impact in various conditions.*

Is seed soaking better overnight or is just 2-3h with agitation enough?

RB: *This depends on the type of seed. Open field crops seed (i.e. carrot, cabbage) contain a lot of saprophytes on them and then 2-3 soaking is best. For in fruit seeds (i.e. tomato, bean) overnight soaking is better for the pathogen is most likely be located inside the seed.*

Can you explain the association of seed microbiota with seed borne diseases?

RB: *The question is not completely clear to me. But in short it might be the case that the presence of certain organisms in the microbiota of the seed can help certain pest to either benefit from or have a negative effect in pest appearance. Demonstrating this is very hard and not so much studied as I know.*

I would like to know which type of quick tests are being used to detect pathogens in seeds and nurseries?

RB: *Quick tests I assume are tests that deliver you results within a day. Immuno strips, Lamp or RPA assays can be used but they should be developed for the pest of interest and lack possibility to discriminate between viable and non-viable pests. Direct screening of seed by molecular testing is very sensitive and quick 2days but again no discrimination between viable and non-viable is possible. This also accounts for ELISA testing which is although less sensitive again. For bacteria dilution plating and pathogenicity tests are the best option but take longer than a week and semi-selective media are needed.*

By what criteria were eight soybean varieties selected for the experiment? How did they take into account the fact that varieties were initially resistant or susceptible to certain pathogens?

LCS: *The eight common bean varieties were selected to represent the diversity of cultivated bean in France. They also display various resistance/susceptibility to pathogens* Globisporangium ultimum *(previously* Pythium ultimum*), responsible for damping off.*

How did Louna characterised seed quality? She talked about emergence percentage rates, low and high emergence percentage. I need more understanding about this.

LCS: *I use this definition: seed quality is the ability of the seed to germinate and emerge quickly and uniformly once sown. In this study, we used germination rate, emergence final percentage and normal to abnormal seedling ratio as proxies of the seed quality. For the varieties of common bean we study, about half of the varieties display final emergence percentage above 50% and half below 50%. We then use those groups to study microorganisms associated with good quality.*

According to you how to differentiate good microbe from bad microbe?

LCS: *Depending on the context (environment, conditions, microbiota composition and diversity,), microorganisms can display various roles in their interaction with the plant.*

Inoculation tests in various conditions would allow to identify microorganisms that have robust « good » effect.

Ruud talked about various characteristics along with the sensitivity talking about the concentration of pests. Can you explain how to determine this concentration and the threshold to know that this kind of pathogen become a threat to eradicate or to control?

RB: To answer your question I like to refer you to the link on the ISTA website with additional info on method validation. *Link: https://www.seedtest.org/api/rm/XS74H86BYJ87N33/validation-of-seed-health-methods-and-organisation.pdf*