Elucidation of the molecular mechanism during non-host resistance in *Arabidopsis thaliana* against *Rhizoctonia solani*

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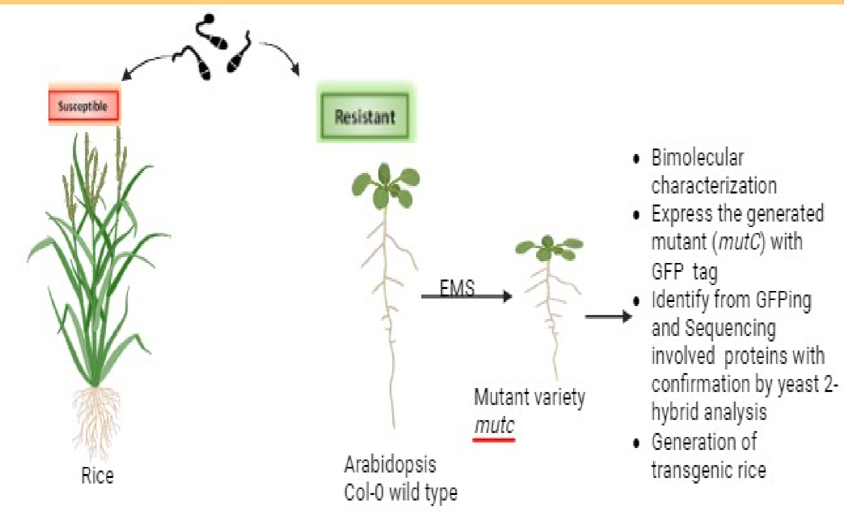
Abstract

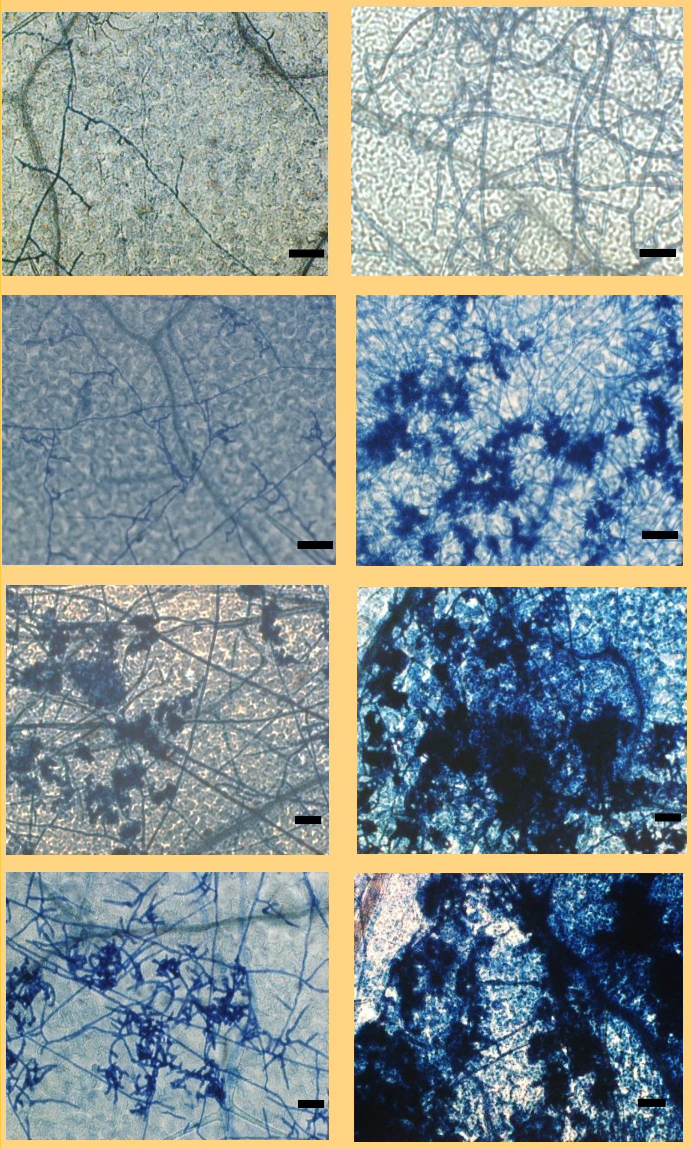
Rice (*Oryza sativa* L.) is the staple food for more than half of world's population, majorly for underdeveloped countries. Current world population is around 7.9 billion, which will be 9 billion by 2050. Feeding this huge population itself a challenge and burden on agricultural practice. Average rice crop loss is about 37.4%, rice feeds 25 people per hectare and it must be 47% per hectare by 2050. The situation is more aggravated due to shrinkage in agricultural lands and loss of crops worth of billions due to pathogens, among which fungus causes 2/3 of total damage. *Rhizoctonia solani* Kühn (teleomorph = *Thanatephorus cucumeris* Donk) is a ubiquitous soil-borne plant pathogenic fungus which causes significant yield losses in many agriculturally important crops . In nature the sheath blight is caused by *Rhizoctonia solani* and there has been no controlling measure neither any chemical pesticide that kill it. As rice is a host and Arabidopsis is a non-host to *R. solani* the causative agent of sheath blight we sought that there might be possible disease resistance mechanism that might be providing some sort of disease resistance against *R. solani*. We found that the hormone pathway mutants of *mpk4*, *pad4*, *ssi* and *jar1* behaved differentially with infection. Further, we screened the EMS generated mutant showed infection cushion at an early hour of infection depicting the fact that the gene might be very important in controlling the entry of the pathogen. Further, to understand the plausible way how it controls the disease the protein-protein interaction studies will be performed and the other molecular architecture and their regulation will be studied.

Rationale and Objectives



Results

Infection assay with *R.solani*



Col-0

Pen2-3

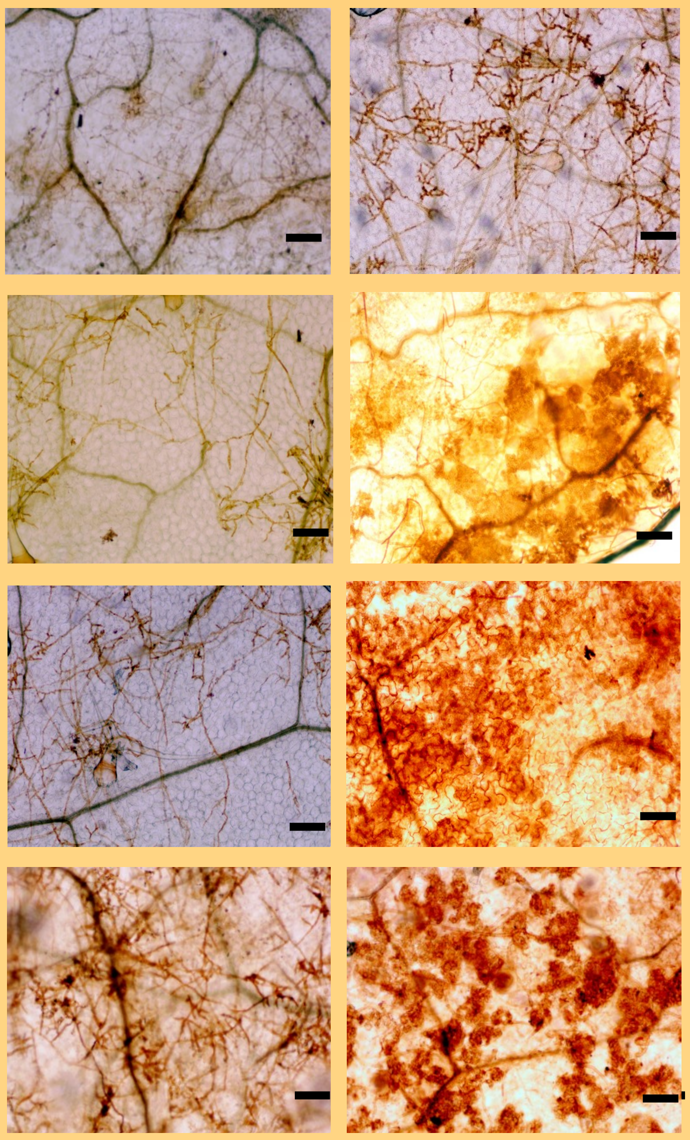
jar1

mutC

24 hpi 48 hpi

Fig. 1. Trypan blue staining and cell death analysis of *R.solani* infected leaves

Infection assay with *R.solani*



Col-0

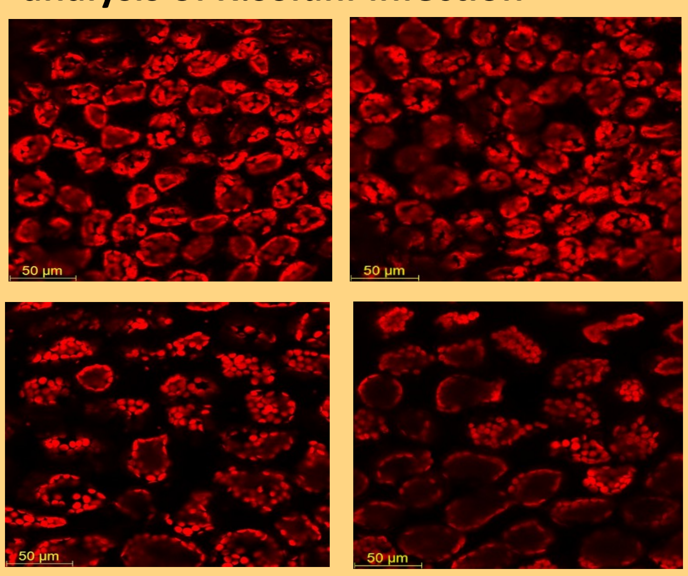
Pen2-3

jar1

mutC

24 hpi 48 hpi

Fig. 2. DAB staining and ROS accumulation analysis of *R.solani* infection



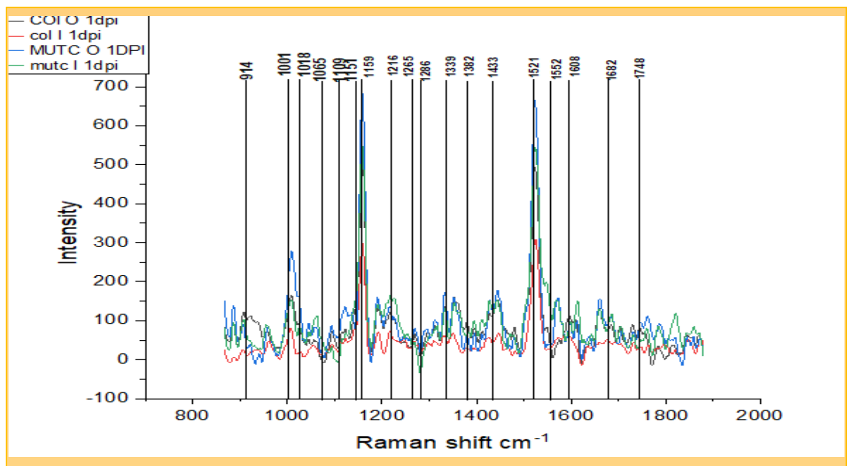
Col-0

mutC

Water control 2Dpi

Infected 2 Dpi

Fig.3. Evaluation of chlorophyll autoflurorescence by R.solani infection in wildtype Col-0 and mutant *mutc*



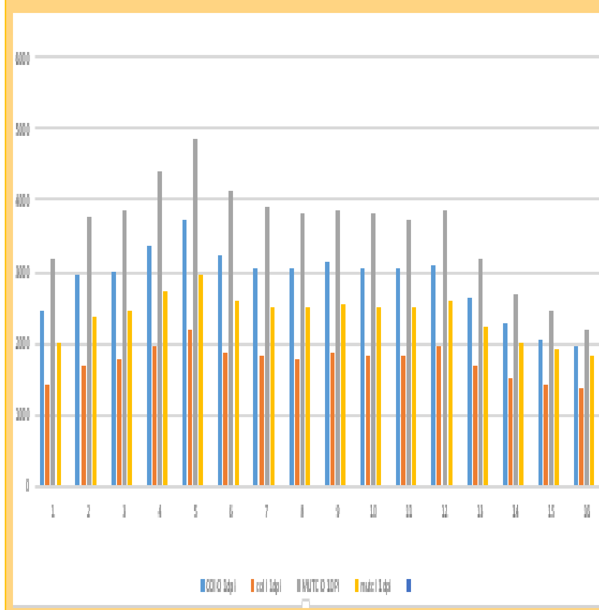


FIG .4: Raman spectra of the infected leaf samples with *R.solani* (*infected leaf sample subjected to raman measurement under conditions* 800-1800 cm^{-1} of spectral range,10s of acquisition time,20mW laser power and 20 X magnification objective)

Conclusion

- EMS mutant *mutc* shows formation of infection cushion at early hours of infection
- Chlorophyll degradation in *mutc* at 48 hpi
- Change in raman spectra upon infection of *mutc* shows changes in compounds compared to wild type

Future Aspects

- Cytological studies for hypersensitive response
- Express the generated mutant (*mutC*) with GFP tag
- Identify from GFPing and Sequencing involved proteins with confirmation by yeast 2-hybrid analysis
- Study spatio-temporal expression of the NHR gene wit relevance to immunity
- Generate double and triple mutants and study patho physiology
- Generate rice transgenic using *mutC* from *A. thaliana* and test it pathophysiology in field condition

References

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Publications

- Pati, D., Kesh, R., Mohanta, V., Pudake, R. N., Sevanthi, A. M., & Sahu, B. B. (2022). Genome-Editing Approaches for Abiotic Stress Tolerance in Small Millets. In *Omics of Climate Resilient Small Millets* (pp. 259-273). Singapore: Springer Nature Singapore