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***Bacillus subtilis* PTA-271 mediated-ISR in *Arabidopsis* requires a primed SA-dependent camalexin response upon pathogen challenge**

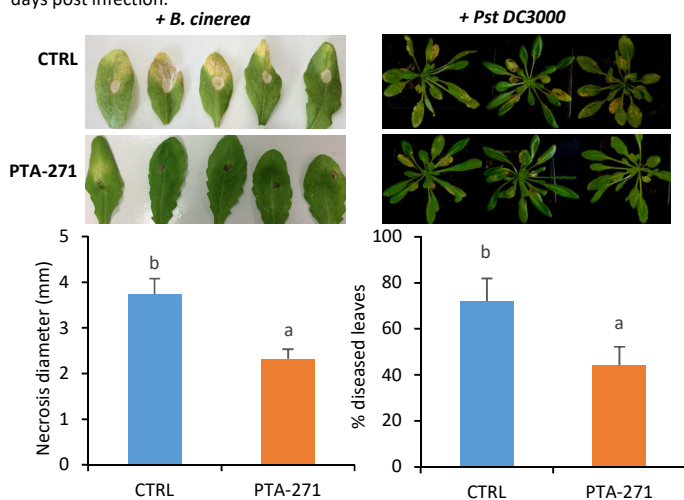
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Bacillus subtilis PTA-271 has been shown to induce ISR against the necrotroph *Botrytis cinerea* (Verhagen et al., 2011) and the hemibiotroph *Neofusicoccum parvum* (Trotel-Aziz et al., 2019) in grapevine. However, the mechanisms by which the bacterium activates the host's immune response underlying ISR remain to be elucidated. In this study, we examined whether the extent of priming state could contribute to the PTA-271-induced ISR in *Arabidopsis*, by focusing on the camalexin response and *CYP71A12* gene expression after *B. cinerea* and *Pst DC3000* infection. Our data provide evidence that PTA-271 triggers ISR against both pathogens through priming camalexin synthesis in SA- and NPR1-dependent manner. This response was further compromised in *pad3* mutant after pathogen infection, highlighting that camalexin plays a prominent role in *B. subtilis*-ISR against both necrotroph and hemibiotroph.

RESULTS

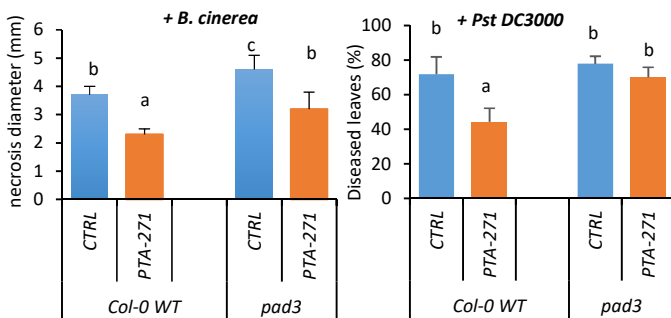
PTA-271 triggers ISR against *B. cinerea* and *Pst DC3000* in *Arabidopsis*

Roots of wild-type plants Col-0 were inoculated with PTA-271 for 2 weeks, then leaves were infected with *B. cinerea* and *Pst DC3000*. ISR was determined at 4 days post infection.



➤ PTA-271 significantly reduces the symptoms and disease incidence caused by *B. cinerea* and *Pst DC3000*.

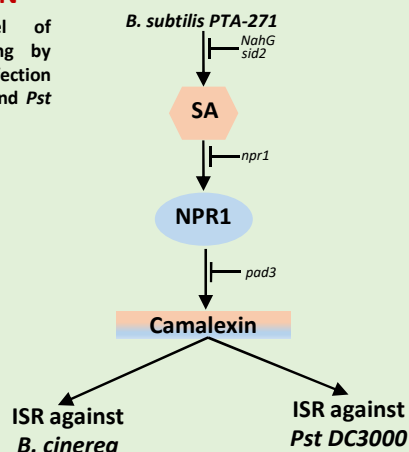
Loss of camalexin synthesis in *pad3* mutant compromises PTA-271-ISR against both *B. cinerea* and *Pst DC3000*



Priming of camalexin accumulation contributes to PTA-271-ISR against *B. cinerea* and *Pst DC3000*

CONCLUSION

Proposed model of camalexin priming by PTA-271 upon infection with *B. cinerea* and *Pst DC3000*

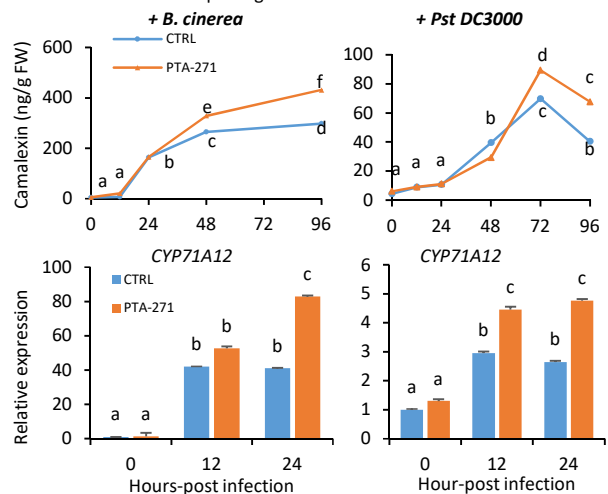


➤ PTA-271 induces ISR against necrotrophic and hemi-biotrophic pathogens by priming camalexin synthesis through *CYP71A12* expression

➤ PTA-271-primed camalexin is dependent on SA and NPR1 pathway

PTA-271 primes camalexin synthesis upon pathogen challenge

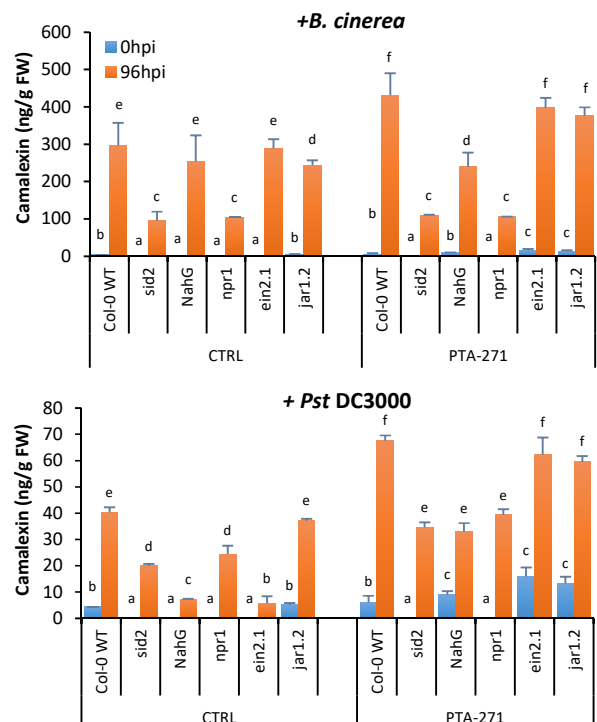
Priming of camalexin production and *CYP71A12* gene expressions were analysed by UPLC and RT-qPCR, respectively. The samples were selected in different time of the pathogens infection.



➤ PTA-271 primes camalexin accumulation and *CYP71A12* gene expression in response to the pathogens

➤ Stronger level of camalexin and *CYP71A12* expression are primed in *B. cinerea*-infected leaves

SA and NPR1 are required for priming camalexin after pathogen infection



➤ Camalexin accumulation is dependent on SA and NPR1, but not on JA and ET upon *B. cinerea* and *Pst DC3000* challenge.

References

- Trotel-Aziz, P., Abou-Mansour, E., Courteaux, B., Rabenoelina, F., Cl ment, C., Fontaine, F., et al. (2019). Front. Plant Sci. 10
- Verhagen BWM, Trotel-Aziz P, Jeandet P, Baillieul F, Aziz A (2011). Phytopathology 101:768-777