1

Uncovering the interactions between *Phytophthora cactorum* and the strawberry plant



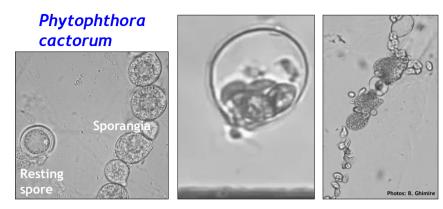
NIBIO NERVEGAN INSTITUE OF RECONSIDER RESEARCH

1



Photos: H. Eikemo and A. Stensvand

Arne Stensvand, Anupam Gogoi, Bikal Ghimire, Mandeep Poudel, Håvard Eikemo, May Bente Brurberg



Forms thick-walled resting spores (oospores) and zoospores in sporangia; zoospores move in water







Crown rot is problematic if it follows the planting material Here from imported plants with

Here from imported plants with about 30% plant and yield loss



Resistance to crown rot

Varies greatly with cultivars

+ = high resistance +++++ = high susceptibility (Eikemo et al. 2003)

Many new cultivars moderately to highly susceptible

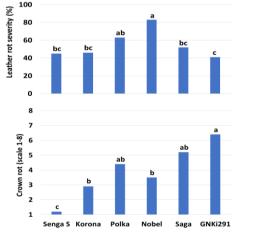
Cultivar	Susceptibility	
Senga Sengana	+	
Glima	+(+)	
Bogota	++	
Bounty	++	
Calypso	++	
Honeoye	+++	
Rita	+++	
Korona	+++	
Lambada	+++	
Zephyr	++++	
Polka	++++	
Elsanta	++++	
Evita	++++	
Tamella	+++++	

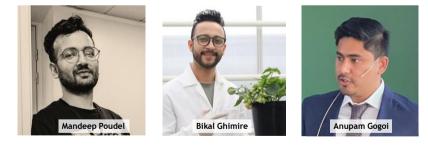
5

Crown rot vs. leather rot

Resistance to crown rot (lower graph) not correlated with resistance to leather rot (upper graph)

(Eikemo & Stensvand, 2015)





Three PhD students that have worked or are working on mechanisms of virulence ("infection strategies") of the pathogen and defence responses in the strawberry host

Infection by P. cactorum

Phytophthora species secrete effector proteins to facilitate the infection

Effector proteins enter different parts of the plant cell

Effectors interact with plant proteins to promote infection in the plant

or

Are recognised by plant resistance proteins that initiate cell death to stop the pathogen

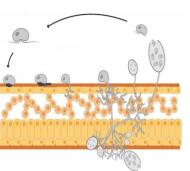


Figure: B. Ghimire

Effector candidates

RxLRs are the most studied effectors in species of *Phytophthora*

They target a wide range of cellular processes, including immune responses in plants

Specification	Numbers	
No. of transcripts with \ge 99% identity to <i>P. cactorum</i> genes	4665	
Secreted proteins	539	
Uncharacterized proteins	216	
Carbohydrate active enzymes	120	
RxLRs	40	
Proteolytic enzymes	23	
Elicitins	9	
Cysteine-rich proteins	7	
Necrosis inducing proteins	7	
Protein with kinase-like domain	5	
Transglutaminase elicitors	4	
Crinklers	3	
Others	105	

(Ghimire et al., unpublished)

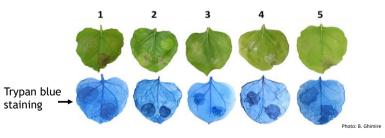
Cloning and infiltration

18 RxLRs were cloned into a plant expression vector

Agrobacterium (AGL1) containing the cloned construct infiltrated into leaves of tobacco (*Nicotiana benthamiana*) - five of the effectors induced cell death



Agroinfiltration in N. benthamiana



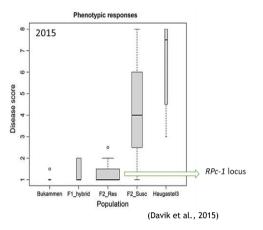
Left leaf sides: RxLR effectors (1-5) inducing cell death (= resistance reaction) Right leaf sides: Negative control (upper), positive control (lower) Positive control: A known elicitor gene for cell death of *P. infestans* Negative control: An empty vector

(Gogoi et al., unpublished)

A locus for crown rot resistance

A single major gene locus called Resistance to Phytophthora cactorum 1 (RPc-1) was identified in diploid strawberry with resistance to crown rot

It contains 69 putative disease resistance genes



Resistance in wild strawberry

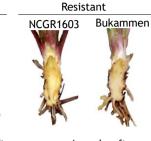
To further understand the expression level of these genes, RNA-sequencing of a susceptible and two resistant Fragaria vesca genotypes was performed after infection of P. cactorum

(Gogoi et al., 2023)

Susceptible NCGR12 NCGR1603 2 weeks after inoculation

Receptor-like proteins (RLPs)

r-like kinases (RLKs)



4 weeks after inoculation

Photos: A. Gogoi

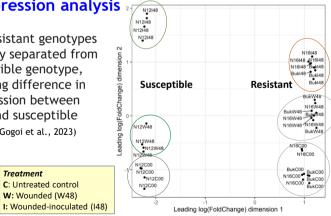
Gene expression analysis

The two resistant genotypes were clearly separated from the susceptible genotype, thus showing difference in gene expression between resistant and susceptible genotypes (Gogoi et al., 2023)

Treatment

C: Untreated control

W: Wounded (W48)



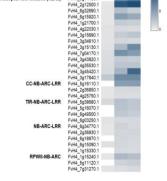
Putative disease resistance genes

Genes present in the two resistant (R) and the susceptible (S) genotypes

Darker box indicates higher expression of mRNA

Several genes with potential role in plant defence

(Gogoi et al., 2023)



FvH4 6a11080.1

FvH4 3q14180 1



Found in disease resistant genotypes of *F. vesca* Induces cell death when expressed in leaves of *N. benthamiana* (Gogoi et al. 2023)

