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For quality of life

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Analysis of Tomato spotted wilt virus effector-triggered immunity

Dryas de Ronde

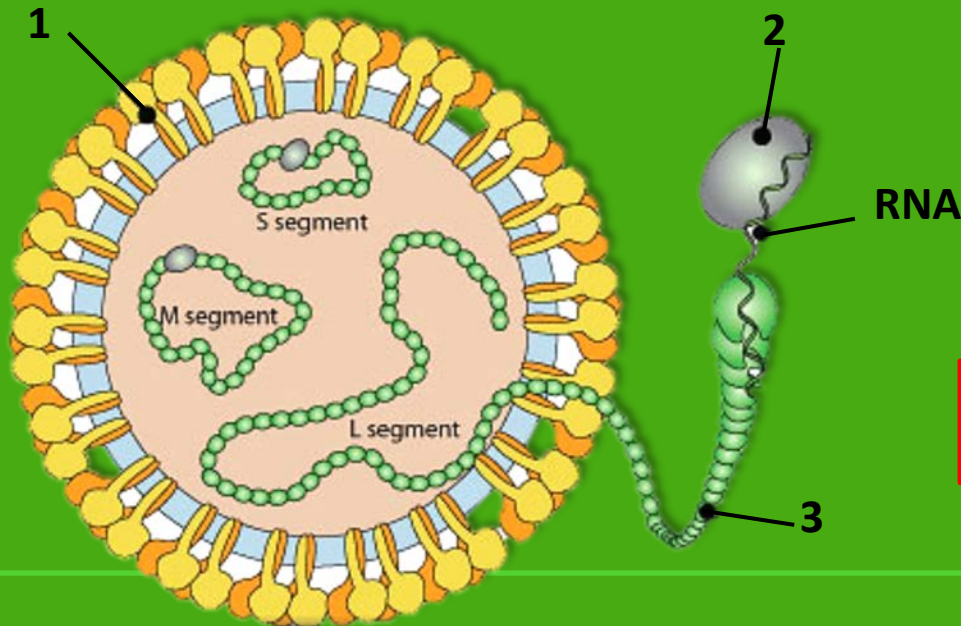
2013

Analysis of *Tomato spotted wilt virus* effector-triggered immunity

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Tomato spotted wilt virus (TSWV)

- Tomatenbronsvlekkenvirus (NL)
- *Bunyaviridae* family
 - Animal infecting viruses: e.g. *Smallpox virus*, *Rift Valley fever virus*
 - Tospovirus genus
- Host range > 1000 species of plants
 - Pepper, tomato, tobacco, potato, lettuce, etc.



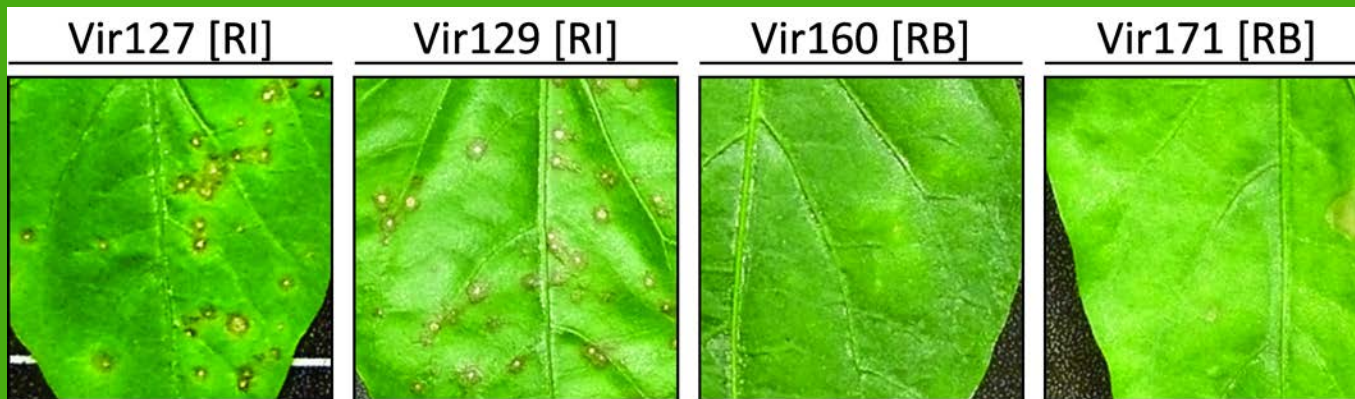
Resistance against TSWV

- Single dominant Resistance (*R*)-gene
 - Tomato: *Sw5b*
 - Pepper: *Tsw*
 - Specific against TSWV only
 - Triggers a typical Hypersensitive response (HR)



Chapter 2

- Problem: resistance breaking isolates

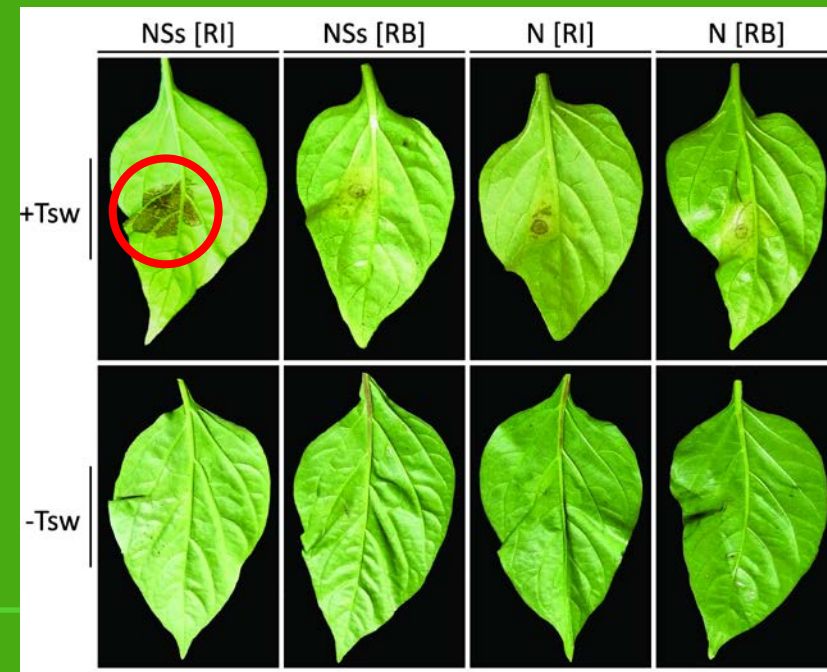
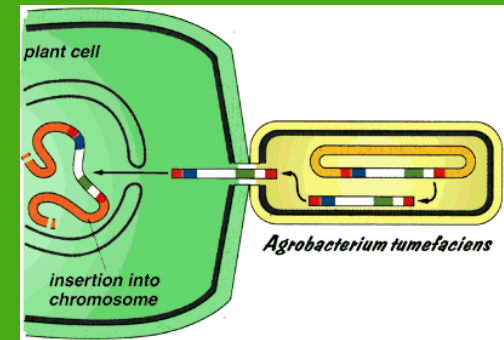


- Avirulence protein remained unknown



Identification avirulence protein

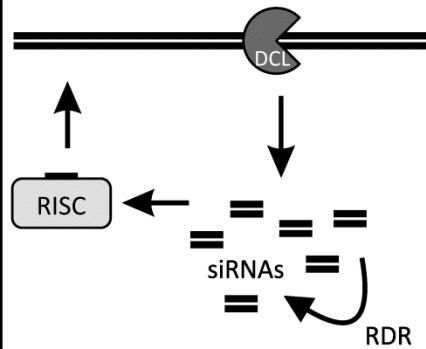
- Candidate proteins:
 - N and NSs
 - Expression in pepper
 - *Agrobacterium tumefaciens*
 - HR response only with NSs protein from RI- isolate on *+Tsw Capsicum* plants



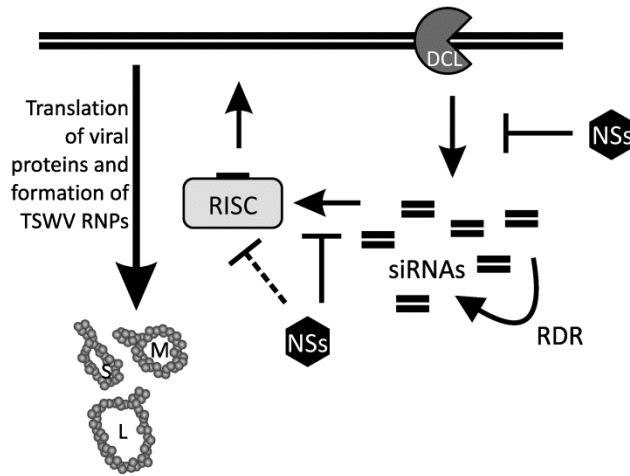
Chapter 3

- Endogenous function of NSs:

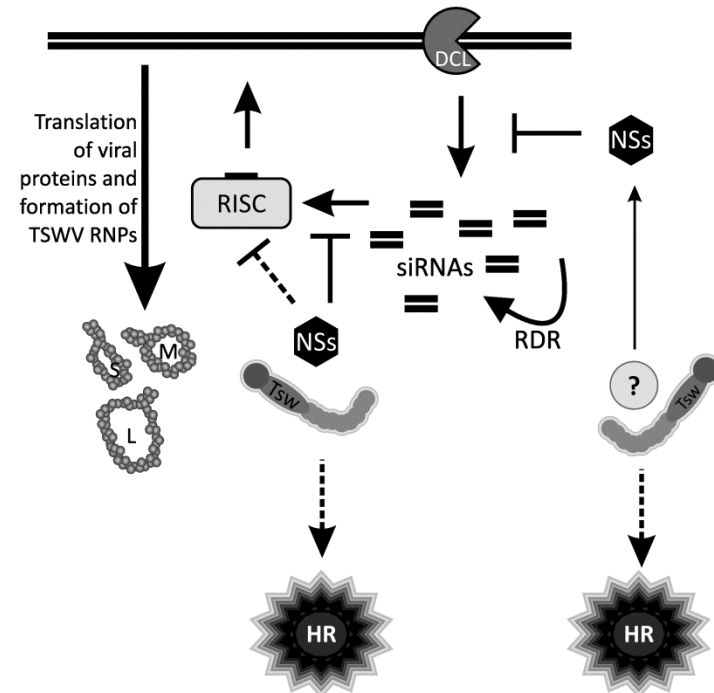
Basal defence



Silencing suppression



Resistance



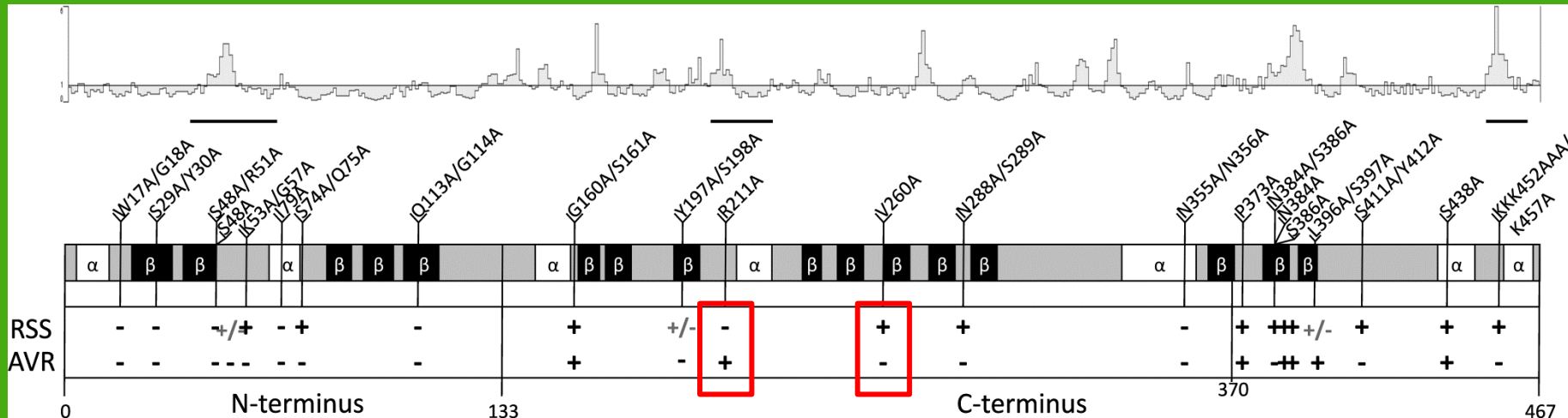
dsRNA produced triggers the RNA interference pathway, leading to breakdown of the viral RNA

TSWV NSs (RSS) binds long dsRNA and siRNAs, thereby inhibiting the RNA interference mechanism and induce a successful infection

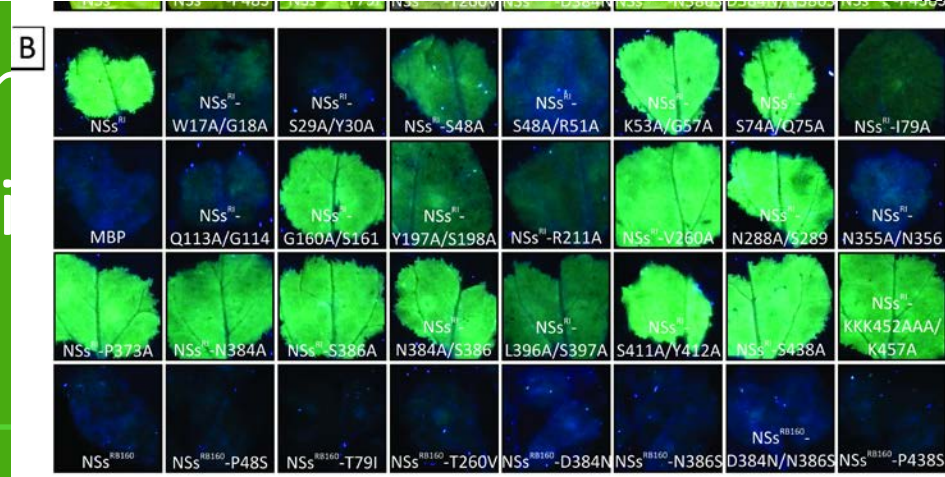
The Tsw-gene product recognises (direct or indirect) the AVR-protein (NSs), triggering a downstream Hypersensitive response (HR)

Functional link RSS and AVR

- Mutant-screen of NSs



- Uncoupling of function
- Importance of N-termini



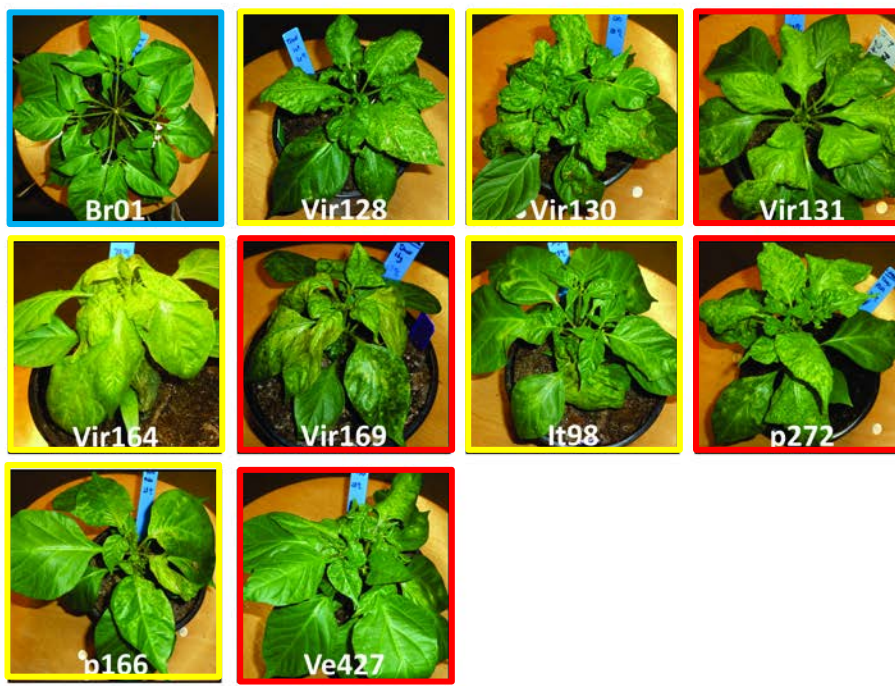
A

22 °C



B

28 °C



Positive: $T_{sw} \geq 32 \text{ } ^\circ\text{C}$
 Susceptible to TSWV: $\geq 28 \text{ } ^\circ\text{C}$



Temperature shift assays

TSWV-RI @ 32 °C -> 22 °C

A



12 dpi, day 0
temp-shift



13 dpi, day 1
temp-shift



14 dpi, day 2
temp-shift



15 dpi, day 3
temp-shift



17 dpi, day 5
temp-shift

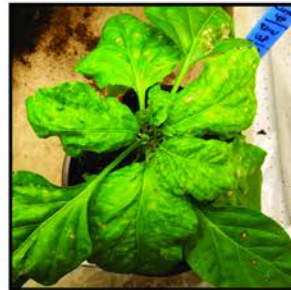
B



12 dpi, day 0
temp-shift



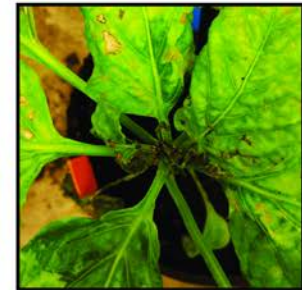
13 dpi, day 1
temp-shift



17 dpi, day 5
temp-shift



19 dpi, day 7
temp-shift



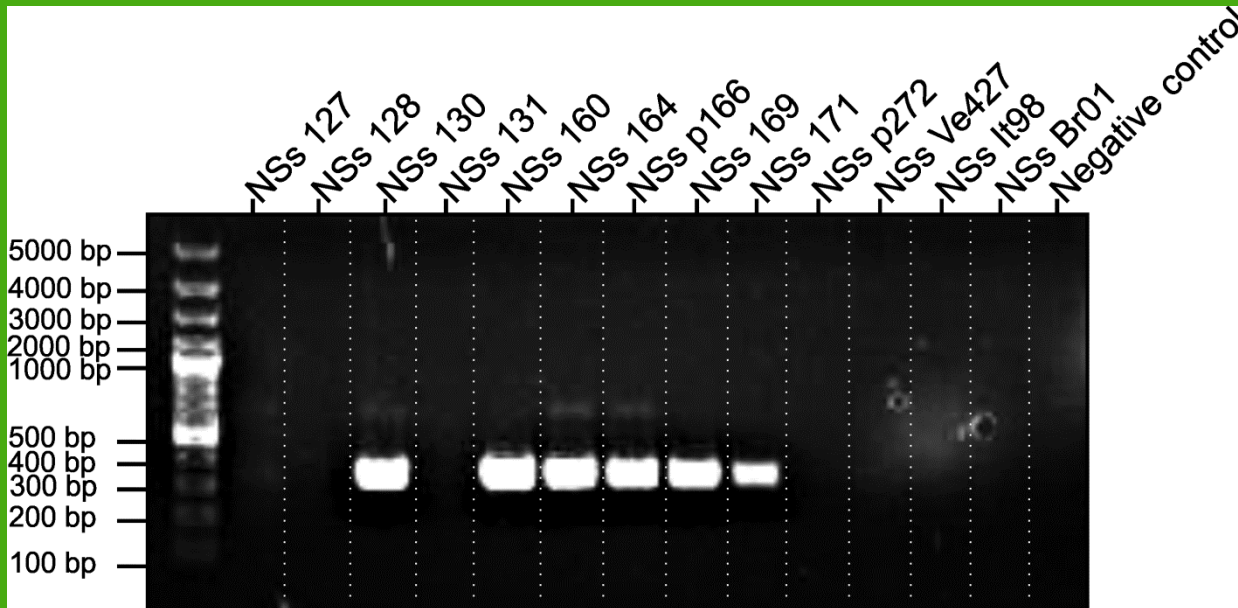
22 dpi, day 10
temp-shift

TSWV-TempRB @ 28 °C -> 22 °C



Diagnostic tool

- Detect resistance breaking isolates



Questions??



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