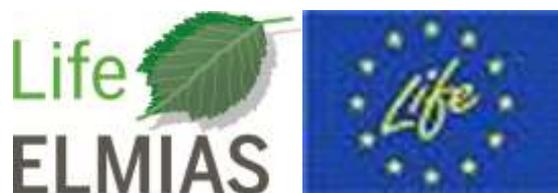


# **LIFE+ ELMIAS Ash and Elm, and IUFRO WP 7.02.01 Root and Stem Rots Conference (LIFE-IUFRO)**

26 August - 1 September 2018, Uppsala and Visby, Sweden

## **PROGRAM & BOOK OF ABSTRACTS**

*Edited by Rimvys Vasaitis*



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## 79. Control of oxidative stress as a tolerance mechanism in *Ulmus minor* against *Ophiostoma novo-ulmi*

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The high virulence of the Dutch elm disease (DED) pathogen, *Ophiostoma novo-ulmi*, has decimated European and North American elm populations during the last century. The number of native resistant elm clones available on the market is scarce, and the defense mechanisms that render a tree tolerant or resistant to the disease are not well understood. In this work we cultivated *in vitro* *Ulmus minor* plants with the aim to: i) study the pathogen dispersion rate in two clones of contrasting susceptibility level to DED, and ii) characterize plant stress responses after *O. novo-ulmi* inoculation. At 1, 3, 7, 14 and 21 days after inoculation (dai) we monitored: pathogen spread within the plant, plant oxidative stress (lipid peroxidation), content of phenolic compounds, level of chlorophylls, and plant growth. We also performed an anatomical study of the xylem to evaluate the possible role of conduit size on susceptibility. The rate of pathogen dispersal was similar in both clones, suggesting that *tolerance* rather than *resistance* mechanisms operate in the low susceptible clone. Xylem anatomy was also similar in both clones, and therefore differences in the response to the pathogen were not attributable to anatomical factors. After pathogen inoculation, the tolerant clone showed a marked increase in lipid peroxidation at 1 dai, but afterwards the levels decreased to values of control plants. The susceptible clone, on the contrary, showed enhanced values of lipid peroxidation at 1, 3, 14 and 21 dai. A peak in total phenolic compounds was detected in the tolerant clone at 3 dai, while no significant changes were observed in the susceptible clone. The susceptible but not the tolerant plantlets suffered a significant delay in apical growth and a decrease in chlorophyll content at 21 dai. The results suggest that the tolerant clone, in spite of being widely colonized by the pathogen, rapidly controlled the induced oxidative burst by producing antioxidant compounds. The susceptible clone maintained a high oxidative stress level during a long period after inoculation, which possibly led to cell damage, cessation of growth and reduced chlorophyll content. Oxidative stress homeostasis thus appears as a factor that can contribute to elm tolerance to DED. Furthermore, the *in vitro* system used in the experiment arises as plausible early-screening method of elm tolerance.

**Motivation:** The defense mechanisms that render an elm tolerant or resistant to Dutch elm disease are not well understood. We used an in vitro system to study elm stress responses after pathogen inoculation.

## Objectives :

- I. Study the pathogen dispersion rate in two clones of contrasting susceptibility level to DED
- II. Characterize plant stress responses after *O. novo-ulmi* inoculation.

## Methods

*In vitro* *Ulmus minor* plantlets were root-inoculated with *Ophiostoma novo-ulmi* spores: 20 plants of a **tolerant (T)** clone and 20 plants of a **susceptible (S)** clone. Forty additional plants were inoculated with water and served as controls.

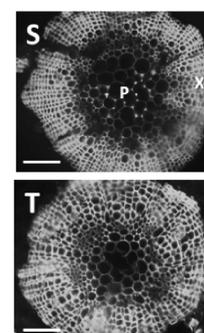
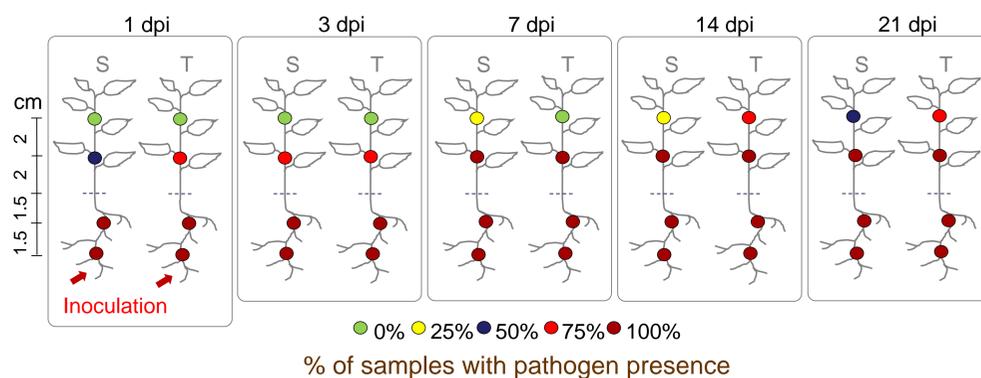


At 1, 3, 7, 14 and 21 days after inoculation (dai) we monitored:

- Pathogen spread within the plant (PCR)
- Plant oxidative stress (lipid peroxidation)
- Content of phenolic compounds
- Level of chlorophylls
- Plant growth
- Xylem anatomy

## Results

No significant differences in plant colonization rate between the T and S clone were detected

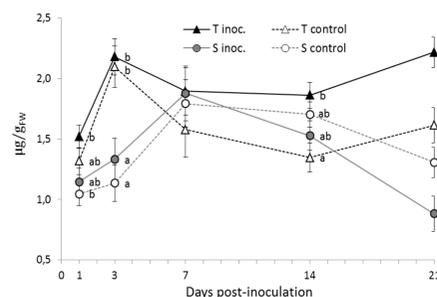
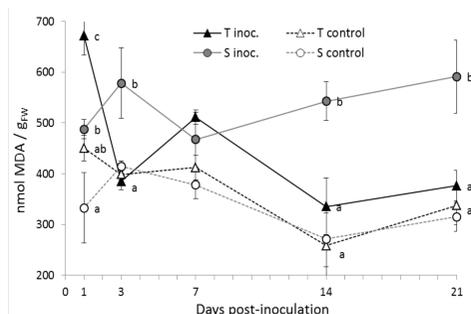


Xylem anatomy (vessel diameter, theoretical hydraulic conductivity) was similar in both clones

P: pith X: xylem  
Bar = 40 μm

### Lipid peroxidation:

The T clone showed a marked increase at 1 dai. The S clone, showed enhanced values at 1, 3, 14 and 21 dai.

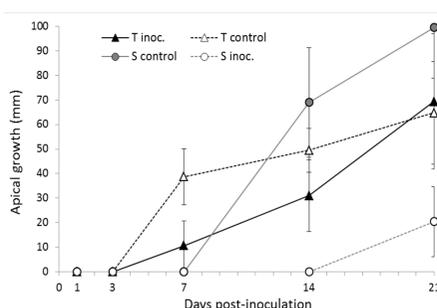
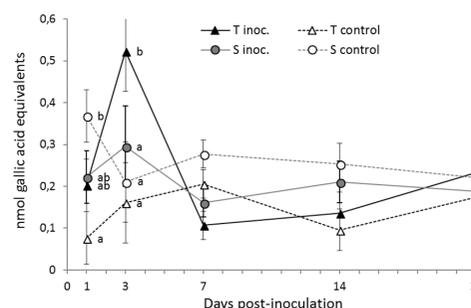


### Chlorophyll content:

The S but not the T plantlets, suffered a significant decrease at 21 dai.

### Total phenolics:

A peak was detected in the T clone at 3 dai, while no significant changes were observed in S clone.



### Apical growth:

The S but not the T plantlets, suffered a significant delay in growth at 21 dai.

**Conclusions** The results suggest that the tolerant clone, in spite of being widely colonized by the pathogen, rapidly controlled the induced oxidative burst by producing antioxidant compounds. The susceptible clone maintained a high oxidative stress level during a long period after inoculation, which possibly led to cell damage, cessation of growth and reduced chlorophyll content. Oxidative stress homeostasis thus appears as a factor that can contribute to elm tolerance.