Impact of light on poplar rust infection

B. FABRE, F. HALKETT and P. FREY

Unity Interactions Trees-Microorganims
Lab of Ecology of forestry pathogens

Introduction

What is poplar rust ?

Poplar rusts were caused by *Melampsora* spp.

Symptoms:

- Disturbance of photosynthesis
- Loss of growth
  → Loss of productivity

- Pertubation of autumn translocation
- Sensitivity to secondary parasites
  → Mortality of young trees

Poplar planting

Poplar with rust

Species :

- *Melampsora medusae* (quarantine pest)
- *Melampsora allii-populina*
- ***Melampsora larici-populina* (*Mlp*)**

Poplar leaf with rust

Introduction

Biology of *Melampsora larici-populina*

Specificities:

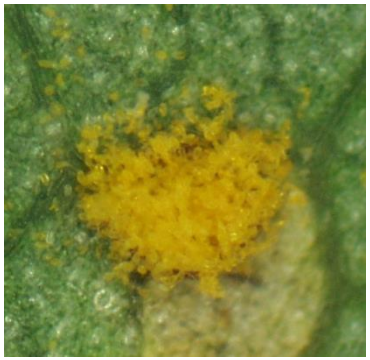
- Heteroecious and macrocyclic

  → We study mainly the asexual reproduction on poplar: the uredinia producing the urediniospores

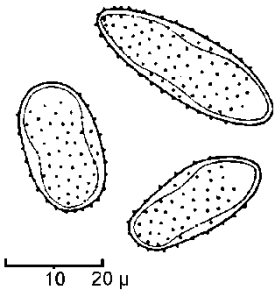
- Strict biotrophic fungus (living host is mandatory)

  → All experiments are performed on excised leaves or excised foliar discs

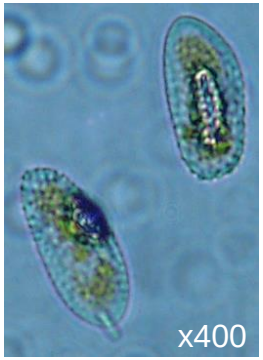
Uredinium of *Mlp*

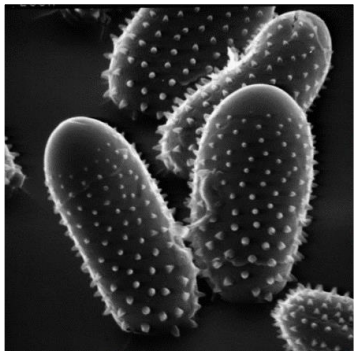


Urediniospores of *Mlp*



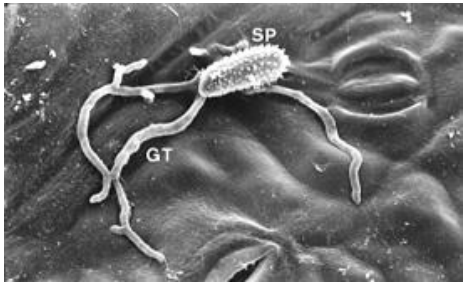
G. Dupias, 1965





Introduction

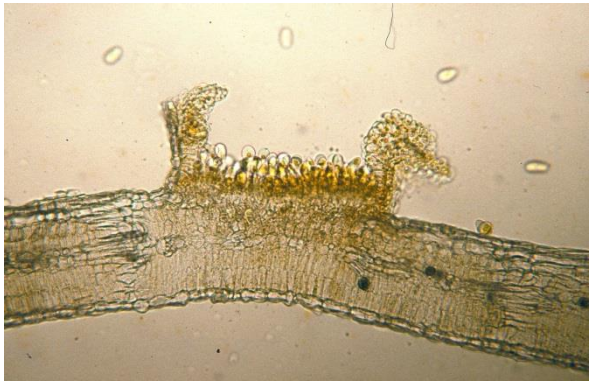
Biology of *Melampsora larici-populina*



Laurans & Pilate, 1999

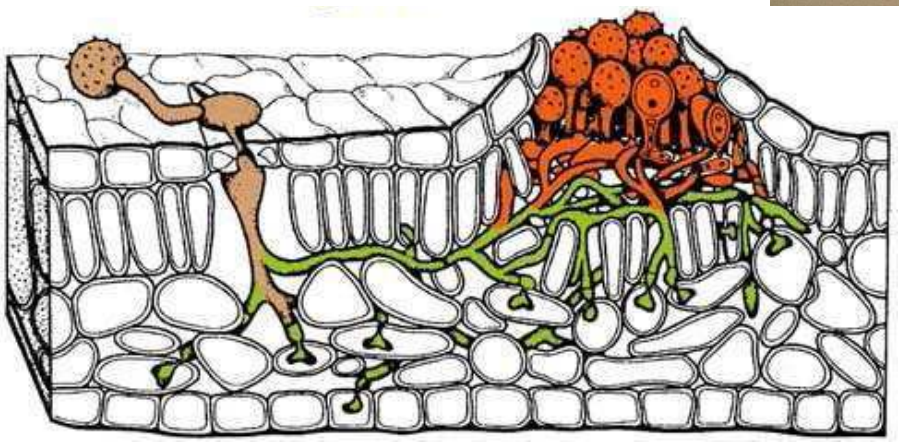
1- Penetration

2 to 6 hours



Transversal section of sporulating uredinium

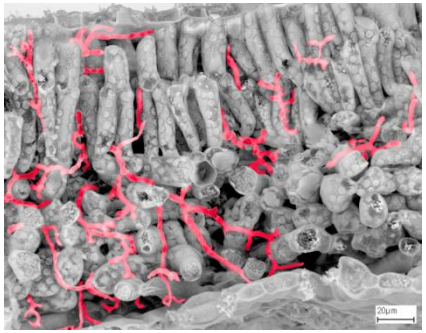
uredinium



Matthias Hahn, 2000

3- Sporulation

7th day



Rinaldi et al, 2007

2- Growth of mycelium

Introduction

Agressiveness traits

Size of the sporulating lesion

Quantity of spores produced

Latency

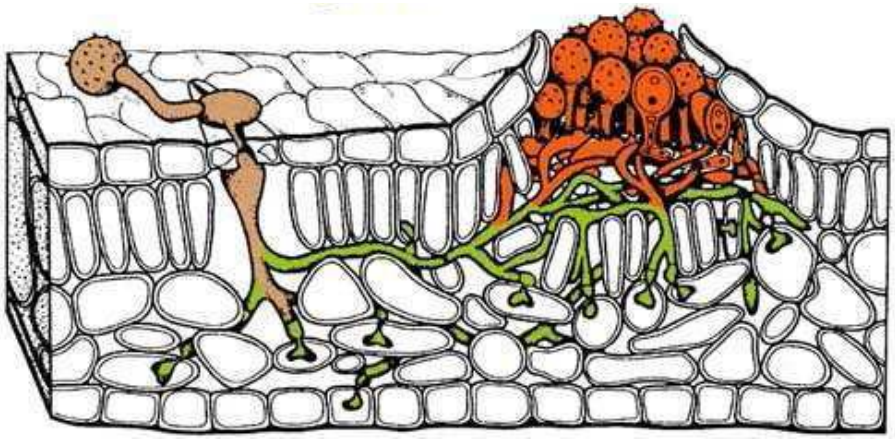
Rate of germination

Infection efficiency

1- Penetration

2 to 6 hours

uredinium



Matthias Hahn, 2000

3- Sporulation

7th day

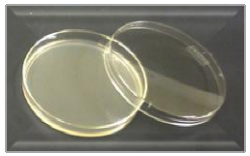
***Quantity of mycelium* in planta**

2- Growth of mycelium

Introduction

Inoculation of excised leaf discs to measure quantitative traits

18 hours

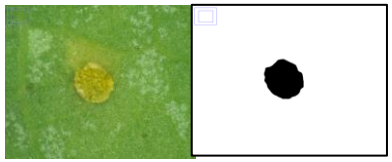


Germination rate

13 days post-inoculation

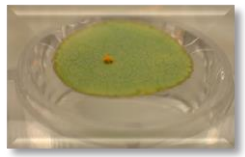
Quantity of spores produced
Size of spores

Size of the sporulating lesion



***Quantity of mycelium* in planta**

Daily observations



Infection efficiency

Latency

Introduction

Phenotyping automated system

Main purposes:

- Study many samples at the same time in order to increase statistical power
- Overcome the effects of experimenter's observations

→

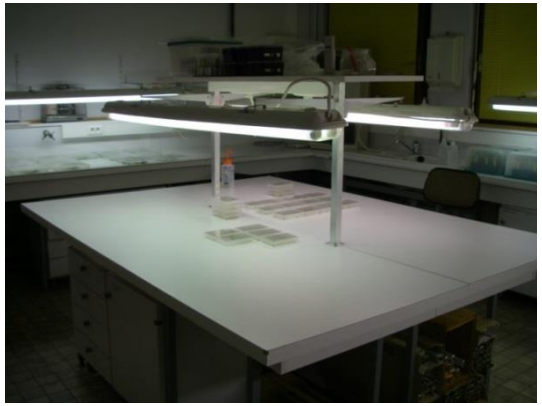
Infection efficiency

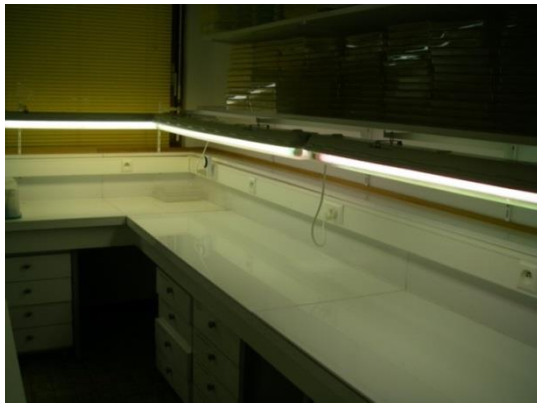
Latency

Size of the sporulating lesion

Introduction

Incubation room





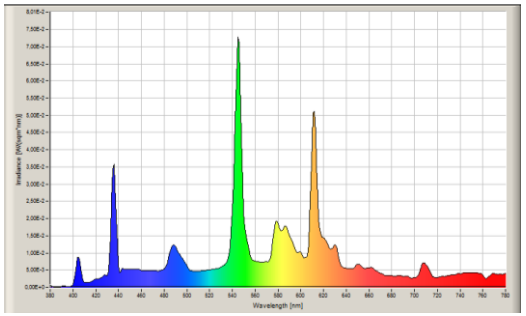
Descriptions :

- Each waterproof box contains 2 fluorescent tubes
- Mean fluence # 40 µmol/s/m^2
- Constant light regime

Main disadvantages:

- Production of heat
- Heterogeneity of the emitted light : variations in light intensity and light spectrum
- No possibility to change easily the light intensity depending on the needs

→ **Need to test other light sources in regards of quantitative traits**



Spectrum of the fluorescent tubes

1. Effects of different light spectra

Materials and methods

Tested light sources:

- Fluorescent tubes (the reference)
- 8 monochromatic LED spots:
  1. Purple (385 nm)
  2. Purple (405 nm)
  3. Blue (440 nm)
  4. Green (520 nm)
  5. Yellow (600 nm)
  6. Red (630 nm)
  7. Red (660 nm)
  8. Red + Blue (440 nm and 660 nm)

Rust strains: 3 isolates of *Mlp*

Inoculation type: Spraying of spores on agar plates

Measured quantitative trait: Germination rate

Vegeled™ lighting spots

HEPH-Condorcet, Belgium

1. Effects of different light spectra

Results of measured spore germination

Number of samples:
100 observed spores

Statistical analyses:
Germination rate → generalized linear model with binomial-raw distribution
Differences were tested with Tukey post-hoc comparaisons.

Light sources	Isolate Mlp 1	Isolat Mlp 2	Isolate Mlp 3
Fluorescent tubes	**96 %**	**84 %**	**100%**
Purple 385 nm	**16 % ***	**0 % ***	**18 % ***
Purple 405 nm	96 %	**44 % ***	**67 % ***
Blue 440 nm	99 %	98 %	100 %
Green 520 nm	99 %	100 %	100 %
Yellow 600 nm	97 %	NA	99 %
Red 630 nm	97 %	96 %	100 %
Red 660 nm	98 %	96 %	100 %
Blue 440 nm + Red 660 nm	98 %	98 %	100 %

Conclusion: Only UV radiations inhibit the spore germination.

2. Effects of different light sources

Materials and methods

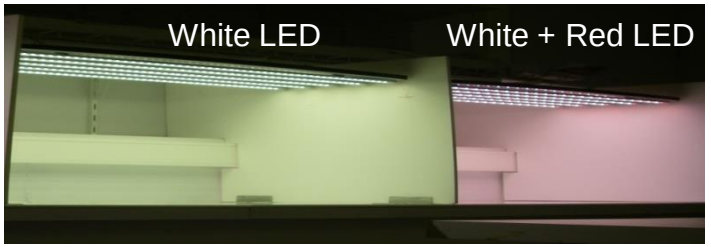
Tested light sources:

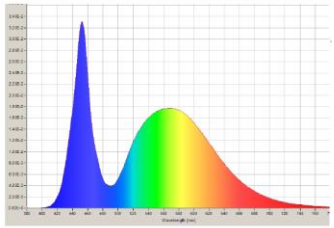
- Fluorescent tubes (the reference)
- White LED profiles
- White and Red LED profiles

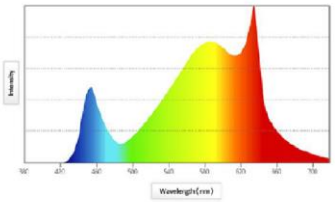
Mean fluence # 40 µmol/s/m^2

A light map was performed for each light sources and before each test.

Vegeled™ lighting profiles







Rust strain: 1 isolate of *Mlp*

Inoculation type: monospore inoculations on excised foliar discs

Measured quantitative traits:

- Germination rate
- Infection efficiency
- Latency, defined as the time elapsed between the inoculation day and the date of lesion appearance

2. Effects of different light sources

Results

Number of samples: 96 inoculated droplets per light modality

Statistical analyses:

- Germination rate and infection efficiency → generalized linear model with binomial-raw distribution
- Latency → generalized linear model with gamma-raw distribution

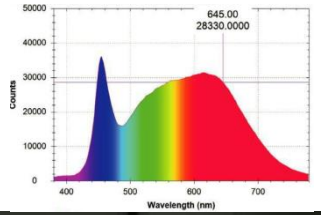
Differents were tested with Tukey post-hoc comparaisons.

Light sources	Mean Fluence	Rate of spore germination	Infection efficiency	Latency
Fluorescent tubes	**40 µmol/s/m2**	**100%**	**0.21**	**8.0 days**
White LED profiles	40 µmol/s/m2	100%	0.29	**7.6 days ***
White + Red LED profiles	45 µmol/s/m2	100%	0.24	**7.8 days ***

Conclusion: The mesured quantitative traits were similar to the reference.

3. Effect of different fluences

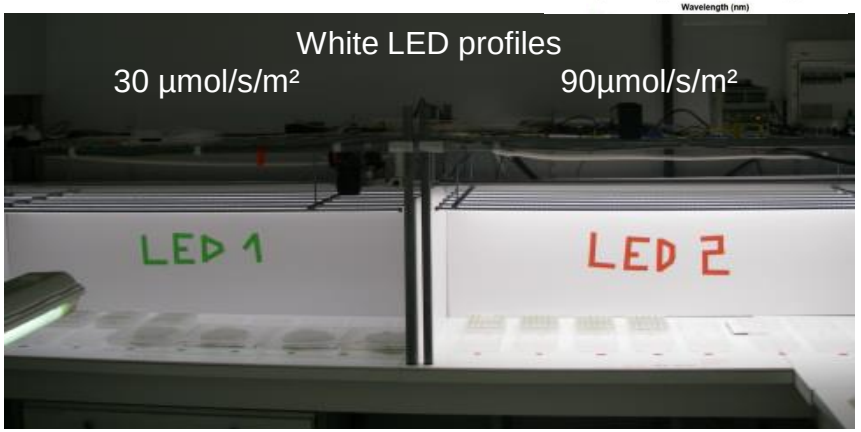
Materials and methods



Tested fluences:

- 30 µmol/s/m²
- 90 µmol/s/m²

A light map was performed for each fluence.

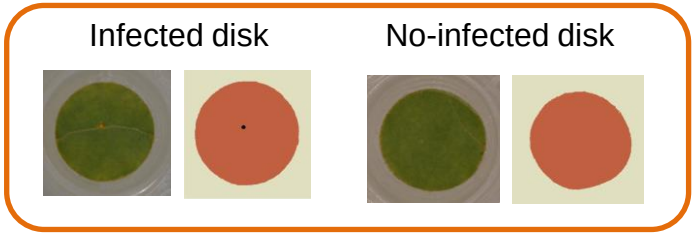


Rust strain: 2 isolates of *Mlp*

Inoculation type: Monospore inoculation on excised foliar discs

Measured quantitative traits:

- Germination rate
- Infection efficiency
- Latency, defined as the time elapsed between the inoculation day and the date of the lesion detection by the automaton



3. Effect of different fluences

Results

Number of samples: 1 536 inoculated droplets per light modality and isolate

Statistical analyses:

- Germination rate and infection efficiency → generalized linear model with binomial-raw distribution
- Latency → generalized linear model with gamma-raw distribution

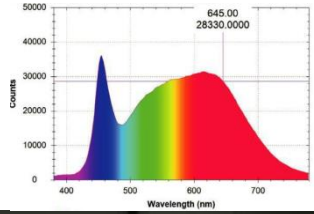
Differents were tested with Tukey post-hoc comparaisons.

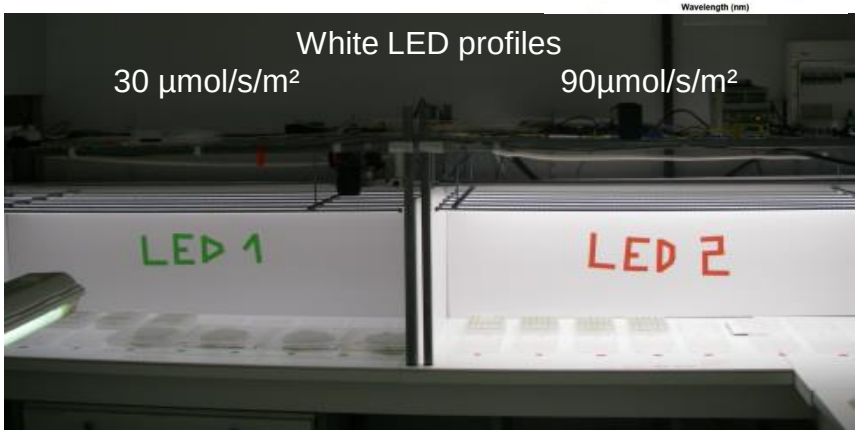
Strains	Light sources	Mean Fluence	Rate of spore germination	Infection efficiency	Latency
Isolate Mlp 1	**White LED profiles**	**30 µmol/s/m²**	**100%**	**0.46**	**8.8 days**
Isolate Mlp 1	White LED profiles	90 µmol/s/m²	98%	0.41 *	8.7 days
Isolate Mlp 2	**White LED profiles**	**30 µmol/s/m²**	**97%**	**0.44**	**8.5 days**
Isolate Mlp 2	White LED profiles	90 µmol/s/m²	91%	0.35 *	8.4 days

Conclusion : As the fluence increases, the infection efficience decreases.

3. Quid of poplar reponses ?

Materials and methods





Tested fluences:

- 30 µmol/s/m2
- 90 µmol/s/m2

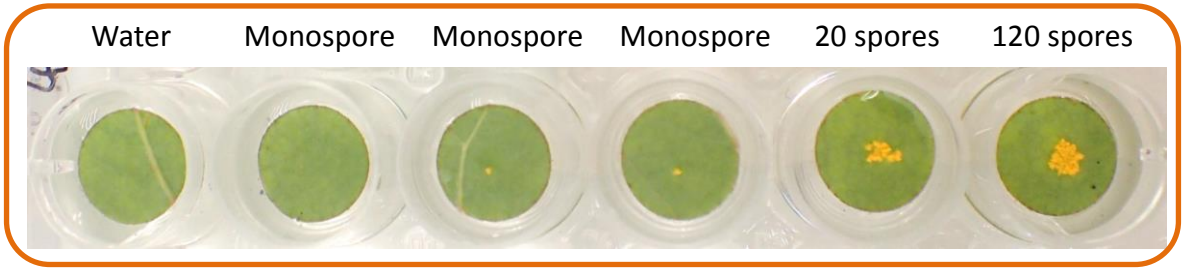
A light map was performed for each fluence.

Rust strains: 1 isolate of *Mlp*

Inoculation types:

On excised foliar discs, 3 inoculum densities are performed:

- only one spore
- 20 spores
- 120 spores



Measurements:

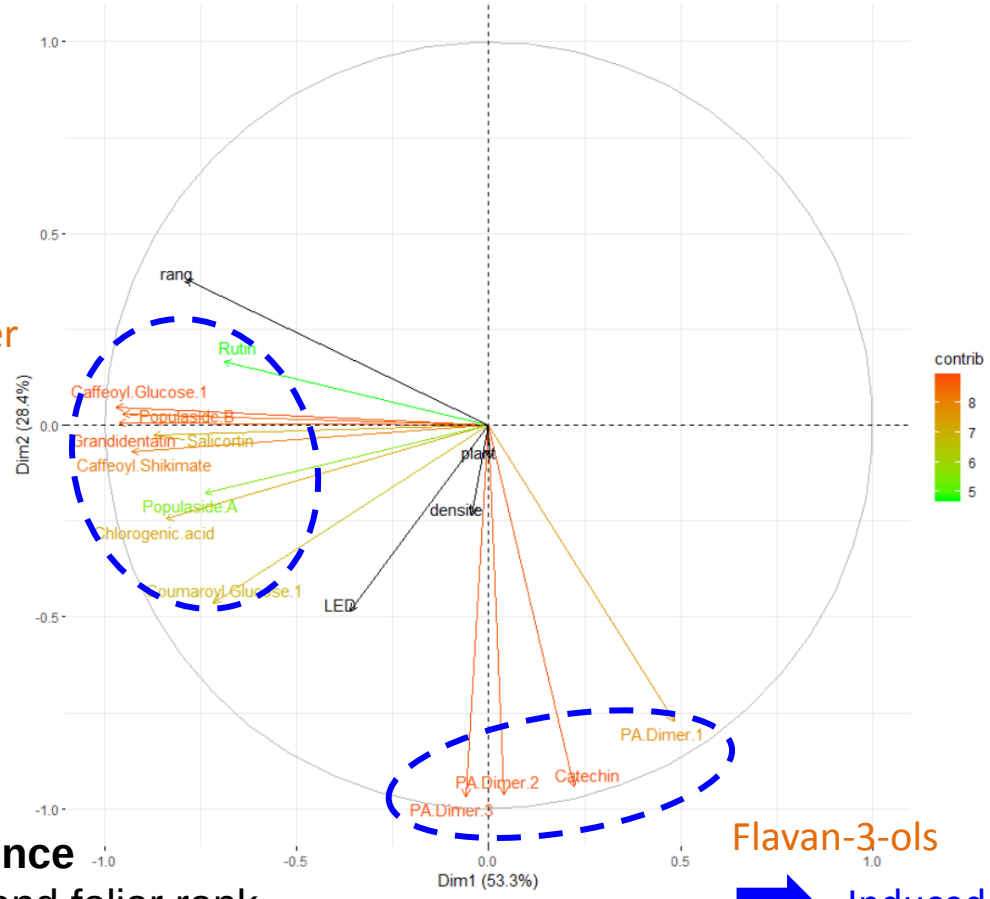
- Sampling at 7 days post-inoculation
- 12 infected foliar discs per modality
- Extraction and quantification of phenolic compounds (Ultra-High Performance Liquid Chromatography)

3. Quid of poplar reponses ?

Correlations circle of the phenolic compounds

Flavanols
Salicinoids
Hydroxycinnamic acid ester

Constitutive
phenolic compounds



Flavan-3-ols

→ Induced
phenolic compounds

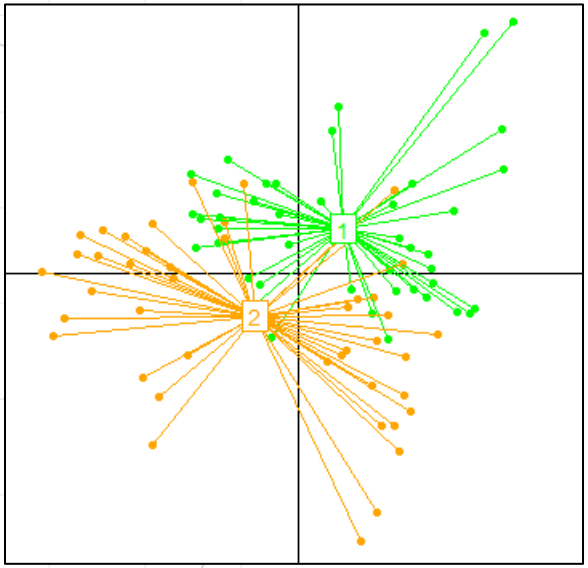
Axis 1: 53,3 % : constitutive defence
→ effect of light fluence and foliar rank

Axis 2: 28,4 % : induced defence
→ effect of light fluence and inoculum density

3. Quid of poplar reponses ?

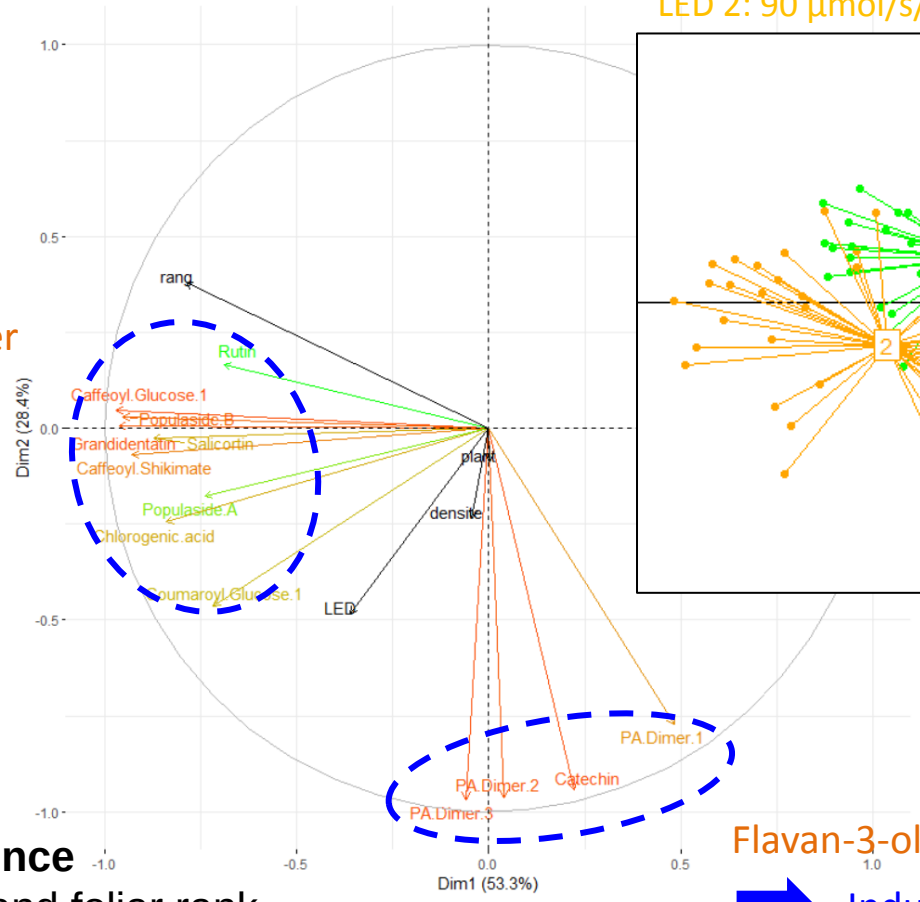
Correlations circle of the phenolic compunds

LED 1: 30 µmol/s/m²
LED 2: 90 µmol/s/m²



Flavanols
Salicinoids
Hydroxycinnamic acid ester

Constitutive
phenolic compounds



Flavan-3-ols

→ Induced
phenolic compounds

Axe 1 : 53,3 % : constitutive defence
→ effect of light fluence and foliar rank

Axe 2 : 28,4 % : induced defence
→ effect of light fluence and inoculum density

Thanks

UMR 1136 – Interactions Arbres-Microorganismes

Fabien Halkett
Pascal Frey
Jérémy Pétrowski
Christophe Bailly
Delphine Vrydagh (2013-2014)
Pierre Santerre Filleux d'Arrentières (2018)
Manon Paineau (2018)

UR 1138 – Biogéochimie des Ecosystèmes Forestiers

Jérôme Demaison

UMR 1121 – Laboratoire Agronomie et Environnement

Romain Larbat

Samuel Colasse
Professor at HEPH-Condorcet

Manuel Colasse