Screening and modes of action of antagonistic bacteria to control two fungal pathogens, Phaeomoniella chlamydospora and Neofusicoccum parvum, involved in grapevine trunk diseases
Haidar Rana, Marc Fermaud

To cite this version:
Haidar Rana, Marc Fermaud. Screening and modes of action of antagonistic bacteria to control two fungal pathogens, Phaeomoniella chlamydospora and Neofusicoccum parvum, involved in grapevine trunk diseases. 10. International workshop on Grapevine Trunk diseases, Jul 2017, Reims, France. hal-03364639

HAL Id: hal-03364639
https://hal.inrae.fr/hal-03364639
Submitted on 4 Oct 2021

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L’archive ouverte pluridisciplinaire HAL, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d’enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.
Screening and modes of action of antagonistic bacteria to control two fungal pathogens, *Phaeomoniella chlamydospora* and *Neofusicoccum parvum*, involved in grapevine trunk diseases

Haidar Rana
UMR Santé et Agroécologie du Vignoble (SAVE) (INRA / Bordeaux Sciences Agro)

10th International Workshop on Grapevine Trunk Diseases – Reims 4-7 July 2017
No efficient strategies to control GTDs

Bacteria as BCAs against GTDs

Sodium arsenite

Control with biological control agents (BCAs)

Bacteria isolated from grape berry surface

Bacteria isolated from wood tissue

46 bacterial strains

2001

Bacteria isolated from grape Berry surface

Martins et al., 2012

Bruez et al., 2015
Objectives

Screening of efficient bacteria against Pch and Np

1. Evaluation, in planta, of the antagonistic activity of 46 bacterial strains against *P. chlamydospora* and *N. parvum*

2. Evaluation of the effect of application method on biocontrol efficacy of 9 selected strains

3. Identification of the modes of action for 3 selected strains
Objectives

Screening of efficient bacteria against Pch and Np

1. Evaluation, in planta, of the antagonistic activity of 46 bacterial strains against *P. chlamydospora* and *N. parvum*

2. Evaluation of the effect of application method on biocontrol efficacy of 9 selected strains

3. Identification of the modes of action for 3 selected strains
**Experimental design:**

- **1st bioassay**
  - Screening, in planta, of 46 bacterial strains against Np and Pch.

- **Cabernet Sauvignon cuttings**
- **Bacteria/pathogen Co-inoculation**
- **Incubation in open greenhouse**
- **Measuring of necrotic lesions**
8 bacteria strains: significant reduction of necrosis length in stem cuttings between 32 and 39%
5 bacteria strains: significant reduction of necrosis length in stem cuttings between 33 and 44%
Objectives

1. Evaluation, *in planta*, of the antagonistic activity of 46 bacterial strains against *P. chlamydospora* and *N. parvum*

2. Evaluation of the effect of application method on biocontrol efficacy of 9 selected strains

3. Identification of the modes of action for 3 selected strains
2nd bioassay

1st in planta bioassay

9 strains

N. parvum

Brevibacillus reuszeri (S27)
Bacillus firmus (S41)
Pantoea agglomerans (S1, S3)

P. chlamydospora

Enterobacter sp. (S24)
Paenibacillus sp. (S18, S19)
Bacillus pumilus (S32)
Brevibacillus reuszeri (S28)

9 strains x 3 methods of bacterial application

Co-inoculation

Preventive inoculation in the hole

Preventive soil inoculation
**Résults: P. chlamydospora, N. parvum**

The effect of bacterial strain and the effect of application method was not significant.

- **bacterial efficiency was more strain dependent than inoculation method dependent**

- **bacterial efficiency dependent on the inoculation method**

- **Drenching the plant soil with the same bacterial strains was less efficient than the application in the hole**

---

**P. chlamydospora**

Haidar et al., 2016; *Microbiological Research*

---

**N. parvum**

---

**Necrosis length (mm)**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Necrosis Length (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prev-soil</td>
<td>a 80</td>
</tr>
<tr>
<td>Co-inoc</td>
<td>b 60</td>
</tr>
<tr>
<td>Prev-hole</td>
<td>c 40</td>
</tr>
</tbody>
</table>
Screening of efficient bacteria against Pch and Np

1. Evaluation, *in planta*, of the antagonistic activity of 46 bacterial strains against *P. chlamydospora* and *N. parvum*

2. Evaluation of the effect of application method on biocontrol efficacy of 9 selected strains

3. Identification of the modes of action for 3 selected strains
Bacterial strains of interest and modes of action

3 selected strains:

- *Paenibacillus sp.*
- *Bacillus pumilus*
- *Pantoea agglomerans*
- *N. parvum*
- *P. chlamydospora*

Modes of action of selected bacteria:

- Induction of grapevine defense
- Production of volatile compounds
- Production of diffusible compounds

qPCR

confrontation
Results...

3 selected strains:

- Pantoea agglomerans
- Paenibacillus sp.
- Bacillus pumilus
- N. parvum
- P. chlamydospora

Modes of action of selected bacteria

Induction of grapevine defense

Production of volatile compounds

Production of diffusible compounds

Days post inoculation:
- 0
- 15
- 90

Bact/Path Prev hole
Bact
Bact/Path Co-inoc
Path

confrontation
Results

Production of volatile compounds

<table>
<thead>
<tr>
<th>Bacterial strain</th>
<th>Volatile compound</th>
<th>Retention time (minute)</th>
<th>Molecular weight (g/mol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>S19 (Paenibacillus sp.)</td>
<td>Compound of pyrazine type</td>
<td>12.8</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td>2,6-Bis (2-methylpropyl) pyrazine</td>
<td>12.4</td>
<td>192.3</td>
</tr>
<tr>
<td></td>
<td><strong>1-Octen-3-ol</strong></td>
<td>6.9</td>
<td>128.22</td>
</tr>
<tr>
<td>S32 (Bacillus megaterium)</td>
<td><strong>2,5-dimethyl Pyrazine</strong></td>
<td>5.4</td>
<td>108.14</td>
</tr>
<tr>
<td></td>
<td>3-octanone</td>
<td>6.6</td>
<td>123.21</td>
</tr>
<tr>
<td></td>
<td>trimethyl-pyrazine</td>
<td>6.8</td>
<td>122.17</td>
</tr>
<tr>
<td></td>
<td>2-ethyl-3,5-dimethyl pyrazine</td>
<td>8.1</td>
<td>136.19</td>
</tr>
<tr>
<td>S1 (P. agglomerans)</td>
<td>Phényl éthyl alcohol</td>
<td>8.6</td>
<td>22.16</td>
</tr>
</tbody>
</table>

1-Octen-3-ol: Inhibition of Pch >96%

2,5-dimethyl pyrazine
3 selected strains:

- **Pantoea agglomerans**
- **Paenibacillus sp.**
- **Bacillus pumilus**

**Modes of action depend on pathogen**

- **Induction of grapevine defense**
- **Production of volatile compounds**
- **Production of diffusible compounds**

**Results**

Bacterial strains of interest and modes of action
Conclusions

1. The most efficient strains: Enterobacteriales
   - Some bacterial strains increase *N. parvum* necrosis

2. Bacterial efficiency dependent on the inoculation method

3. No
   - *Paenibacillus* sp. inhibits *Pch* by production of volatile compounds

- The most efficient strains: Bacillales

- No

- *Paenibacillus* sp. inhibits *Np* by the induction of grapevine defense

- No
Marc Fermaud, Alain Deschamps
(Supervisors)
Patrice Rey
Jean Roudet
Emilie Bruez
Jessica Vallence
Thanks for your attention
<table>
<thead>
<tr>
<th>PR proteins</th>
<th>Gene Symbol</th>
<th>Function</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PR proteins</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VvPR1</td>
<td>PR protein class1</td>
<td></td>
</tr>
<tr>
<td>VvPR10</td>
<td>PR protein class10</td>
<td></td>
</tr>
<tr>
<td>VvCHIT3</td>
<td>Chitinase class III</td>
<td></td>
</tr>
<tr>
<td>VvGLU</td>
<td>β-1,3glucanase</td>
<td></td>
</tr>
<tr>
<td><strong>cell wall reinforcement</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VvCALS</td>
<td>Calloosesynthase</td>
<td></td>
</tr>
<tr>
<td><strong>Redox status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VvGST</td>
<td>Glutathione S-transferase</td>
<td></td>
</tr>
<tr>
<td><strong>Indole and phenylpropanoid pathways</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>VvANTS</td>
<td>Antranilatesynthase</td>
<td></td>
</tr>
<tr>
<td>VvSTS</td>
<td>Stilbenesynthase</td>
<td></td>
</tr>
<tr>
<td>VvCHS</td>
<td>Chalconesynthase</td>
<td></td>
</tr>
<tr>
<td>VvPAL</td>
<td>Phenylalanineammonialyase</td>
<td></td>
</tr>
</tbody>
</table>