



HAL
open science

Transcriptomic changes in the PacC transcription factor deletion mutant of the plant pathogenic fungus *Botrytis cinerea* under acidic and neutral conditions

Christine Rascle, Bastien Malbert, Isabelle R Gonçalves, Mathias Choquer,
Christophe Bruel, Nathalie Poussereau

► To cite this version:

Christine Rascle, Bastien Malbert, Isabelle R Gonçalves, Mathias Choquer, Christophe Bruel, et al.. Transcriptomic changes in the PacC transcription factor deletion mutant of the plant pathogenic fungus *Botrytis cinerea* under acidic and neutral conditions. *BMC Genomic Data*, 2024, 25 (1), pp.87. 10.1186/s12863-024-01257-3 . hal-04814248

HAL Id: hal-04814248

<https://hal.science/hal-04814248v1>

Submitted on 11 Dec 2024

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.

DATA NOTE

Open Access



Transcriptomic changes in the PacC transcription factor deletion mutant of the plant pathogenic fungus *Botrytis cinerea* under acidic and neutral conditions

Christine Rasclé¹, Bastien Malbert², Isabelle Goncalves¹, Mathias Choquer¹, Christophe Bruel¹ and Nathalie Poussereau^{1*}

Abstract

Objectives *Botrytis cinerea*, the causal agent of gray mold, is a necrotrophic fungus that can infect a wide variety of plant species and plant tissues. During infection, this pathogen modulates the pH of its environment by secreting organic acids or ammonia. Deletion of the gene encoding the pH-responsive transcription factor PacC revealed the importance of this regulator in different steps of the infection process and particularly in the secretion of organic acids, reactive oxygen species and plant cell wall degrading enzymes. This study aimed to identify the genes controlled by this fungus-specific transcription factor when the fungus is placed under acidic or neutral conditions.

Data description *Botrytis cinerea* B05.10 and the knock-out *BcpacC* mutant strains were grown on solid non-buffered medium for 3 days on the surface of cellophane membranes before transfer for 4 h onto the surface of liquid medium buffered at pH 5.0 or 7.0 followed by mycelium collection. After RNA sequencing, differentially expressed genes according to strains or pH conditions were listed. These data will be useful in understanding the adaptation process of *B. cinerea* during plant infection.

Keywords *Botrytis cinerea*, pH regulation, PacC transcription factor, Necrotrophic fungus

Objectives

Botrytis cinerea, the causal agent of gray mold, is a necrotrophic fungus capable of colonizing a wide variety of plant species and plant tissues with acidic or neutral tissues [1]. During infection, this pathogen modulates the pH of its environment by secreting organic acids or

ammonia [2]. This ability to change and adapt to different pHs suggests an efficient regulatory mechanism. The transcription factor PacC is considered as a critical component in the regulation of pH-responsive genes in fungi [3]. PacC activates genes expressed under alkaline conditions and represses genes expressed under acidic conditions. Consequently, *B. cinerea* modulates the expression of pacC during its infectious process. The investigation of a course of sunflower infection revealed a low expression of pacC during the first days of interaction when the pH decreases until acidic pH and an increase of expression when the pH reaches a neutral value. Deletion of the gene encoding PacC in *B. cinerea* revealed the importance of

*Correspondence:

Nathalie Poussereau
nathalie.poussereau@univ-lyon1.fr

¹Universite Claude Bernard Lyon 1, INSA Lyon, Bayer SAS, MAP, UMR5240, Villeurbanne 69100, France

²BayerCropScience Division Centre de Recherche de la Dargoire, Lyon 69009, France



© The Author(s) 2024. **Open Access** This article is licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License, which permits any non-commercial use, sharing, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if you modified the licensed material. You do not have permission under this licence to share adapted material derived from this article or parts of it. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by-nc-nd/4.0/>.

Table 1 Overview of data files/data sets

Label	Name of data file/data set	File types (file extension)	Data repository and identifier (DOI or accession number)
Data file 1	Data processing file	Bash script (.sh)	Zenodo (https://doi.org/10.5281/zenodo.11033333)
Data file 2	<i>Botrytis_data_Quality_Control</i>	PDF (.pdf)	Zenodo (https://doi.org/10.5281/zenodo.11034395)
Data set 1	Transcriptome of <i>Botrytis cinerea</i> WT and BcpacC deletion mutant at pH 5 and 7	FASTQ files (fastq.gz)	NCBI GEO (https://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE265877)
Data set 2	Lists of differentially expressed genes for the different conditions	Text files (.txt)	Zenodo (https://doi.org/10.5281/zenodo.11034476)

this regulator in different steps of the infection process of the pathogen particularly in the secretion of oxalic acid, reactive oxygen species, plant cell wall degrading enzymes [4] and regulation of secondary metabolism [5]. The virulence defect of the mutant was observed in plant hosts characterized by neutral tissues (cucumber cotyledons, zucchini fruits, romaine lettuce) and not in acidic plant tissues (tomatoes, grape berries) [4, 5]. To identify the genes controlled by this transcription factor, the *pacC* mutant and the parental strain were cultured under acidic or neutral conditions to investigate gene expression.

Data description

Fungal strains and growth conditions

Botrytis cinerea strain B05.10 (WT) and the deletion Δ *pacC* mutant strain were maintained on solid sporulation medium. The B05.10 and Δ *pacC* strains were grown at 21 °C on solid non-buffered containing Malt 2%, Glucose 2%, NH₄Cl 100mM and Agar 1.5%, for 3 days on the surface of cellophane membranes inoculated with 4-mm mycelial plugs. The membranes were then transferred for 4 h onto the surface of 2 ml of liquid Gamborg medium buffered at pH 5.0 or 7.0. The mycelium was then collected from the membranes, frozen in liquid nitrogen and lyophilized. For each strain (WT and mutant) and each pH condition (5.0 and 7.0), 15 plates were used for RNA extraction. Three independent experiments were assessed. A total of 12 RNA samples were sequenced.

RNA extraction and sequencing

Total RNA was extracted using the RNeasy midi kit (Qiagen). RNA was then treated by DNase (Ambion) and RNA quality was verified using the Bioanalyzer RNA 6000 Nano kit (Agilent).

1.5 µg of each sample was sent to Fasteris SA (Switzerland) for library construction and sequencing. Libraries were prepared using the Illumina TruSeq Stranded mRNA Library Prep kit and were sequenced for 1×125 bp single-end reads on an Illumina HiSeq 2500. Reads were trimmed using Trimmomatic (v0.33) [6] and then aligned on *Botrytis cinerea* B05.10 reference genome (ASM14353v4 from NCBI) using STAR (v2.6) [7]. FeatureCounts (Subread v2.0.2) [8] was then used with the -t

exon parameter to count alignments overlapping exons and the -g gene_id to summarize the counts for each gene [9]. The differential gene expression was performed using DiCoExpress [10, 11]. P-values were adjusted using the Benjamini-Hochberg method. Genes were considered differentially expressed when having an adjusted p-value strictly inferior to 0.05. Data file 1 (Table 1) presents all scripts used to process the data. Data file 2 (Table 1) contains the analysis of RNA-Seq quality on raw and normalized counts. Data set 1 (Table 1) provides the raw data of gene expression levels of *Botrytis cinerea* WT at pH 5.0 and at pH 7.0 and BcpacC deletion mutant at pH 5.0 and pH 7.0. The lists of the differentially expressed genes according to strains or pH conditions are recorded in Data set 2 (Table 1). Briefly, for WT strain, 1196 genes were differentially expressed (689 genes were up-regulated at pH 5.0 while 507 genes were up-regulated at pH 7.0). Regarding Δ *pacC* strain, 910 genes were differentially expressed (488 genes up-regulated at pH 5.0 and 422 at pH 7.0). At pH 7.0 (optimal expression of BcpacC) 980 genes were down-regulated in *pacC* deleted strain compared with WT strain, whereas 934 were up-regulated.

Limitations

In this study, our data result from two genotypes and two pH conditions (pH 5.0 and pH 7.0). Previous studies revealed that *pacC* expression level increases from pH 3.0 to pH 7.0. Additional RNA-seq analysis could be performed at pH 3–4 to strengthen the data obtained under acidic conditions.

Abbreviations

WT wild type

Author contributions

NP conceived and supervised the research project. CR designed the experiments and obtained the data. BM and IG processed the data. MC, CB and CR substantially revised the work. CR, BM and NP wrote the manuscript. All authors approved the submitted version and agreed with their contributions.

Funding

This work was supported by the French National Research Center (CNRS).

Data availability

The data described in this Data note can be freely and openly accessed on the NCBI (GEO) under (<https://www.ncbi.nlm.nih.gov/geo/query/acc>).

[cgi?acc=GSE265877](https://doi.org/10.5281/zenodo.11034395)). Quality assessment is available through Zenodo (<https://doi.org/10.5281/zenodo.11034395>). Data processing script can be accessed through Zenodo (<https://doi.org/10.5281/zenodo.11033333>). The lists of Differentially expressed genes can be obtained from Zenodo (<https://doi.org/10.5281/zenodo.11034476>).

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

Received: 22 May 2024 / Accepted: 23 July 2024

Published online: 09 October 2024

References

1. Manteau S, Aboua S, Lambert B, Legendre L. Differential regulation by ambient pH of putative virulence factor secretion by the phytopathogenic fungus *Botrytis cinerea*. *FEMS Microbiol Ecol*. 2003;43:359–66.
2. Billon-Grand G, Rasclé C, Droux M, Rollins JA, Poussereau N. pH modulation differs during sunflower cotyledon colonization by the two closely related necrotrophic fungi *Botrytis cinerea* and *Sclerotinia sclerotiorum*. *Mol Plant Pathol*. 2012;13(6):568–78.
3. Peñalva MA, Tilburn J, Bignell EM, Arst HN Jr. Ambient pH gene regulation in fungi: making connections. *Trends Microbiol*. 2008;16(6):291–300.
4. Rasclé C, Dieryckx C, Dupuy JW, Muszkieta L, Souibgui E, Droux M, et al. The pH regulator PacC: a host-dependent virulence factor in *Botrytis cinerea*. *Environ Microbiol Rep*. 2018;10(5):555–68.
5. Zhuo R, Li G, Peng H, Zong Y, Wang X, Lu S, Chen Y, Zhang Z, Tian S, Li B. Extensive regulation of pH-responsive transcription factor PacC on secondary metabolism contributes to development and virulence of *Botrytis cinerea*. *Postharvest Biology and Technology*. 2023; 197:112219.
6. Bolger AM, Lohse M, Usadel B. Trimmomatic: a flexible trimmer for Illumina sequence data. *Bioinformatics*. 2014;30(15):2114–20.
7. Dobin A, Davis CA, Schlesinger F, Drenkow J, Zaleski C, Jha S, et al. STAR: ultrafast universal RNA-seq aligner. *Bioinformatics*. 2013;29(1):15–21.
8. Liao Y, Smyth GK, Shi W. FeatureCounts: an efficient general purpose program for assigning sequence reads to genomic features. *Bioinformatics*. 2014;30(7):923–30.
9. Malbert B, Rigaiil G, Brunaud V, Lurin C, Delannoy E, 2018. https://doi.org/10.1007/978-1-4939-8654-5_19.
10. Lambert I, Paysant-Le Roux C, Colella S, Martin-Magniette ML. DiCoExpress: a tool to process multifactorial RNAseq experiments from quality controls to co-expression analysis through differential analysis based on contrasts inside GLM models. *Plant Methods*. 2020;16(1):68.
11. Baudry K, Paysant-Le Roux C, Colella S, Castandet B, Martin ML. Analyzing multifactorial RNA-Seq experiments with DicoExpress. *J Vis Exp*. 2022;185:e62566.

Publisher's Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.