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DATA NOTE

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

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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 organics 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

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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	*Botrytis*_data_Quality_Control	PDF (.pdf)	Zenodo (https://doi.org/10.5281/zenodo.11034395)
Data set 1	Transcriptome of *Botrytis cinerea*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 ΔpacC mutant strain were maintained on solid sporulation medium. The B05.10 and ΔpacC strains were grown at 21 °C on solid non-buffered containing Malt 2%, Glucose 2%, NH_4Cl 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 ΔpacC 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 substantively revised the work. CR, BM and NP wrote the manuscript. All authors approved the submitted version and agreed with their contributions.

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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). 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.

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