Introduction

Colombian cacao has a growing demand due to its appreciated flavor and aroma; this crop is affected by pests and diseases that mitigate its production. Different species of the oomycete Phytophthora produced the Black pod disease worldwide. Phytophthora palmivora Butler is present in all cacao-producer countries and causes losses of 20 to 30% of the production and 10% of the death of cacao trees (Bailey y Meinhardt, 2016).

In Colombia, pod production and the generation of seedlings in nurseries are severely affected by this disease. This study’s purpose was to understand better the defense mechanisms related to the plant response during the pathogen infection. We used a transcriptomic approach to obtain information about the genes involved in the resistance to diseases. Dual RNA-sequencing permitted the simultaneous capture of pathogen-specific transcripts and the plant defense-expressed genes during infection, providing a complete view of the interaction. Our goal was to identify the pathogen and plant genes expressed during the early stages of black pod infection (0h, 24h, 48h, and 96h), comparing the expression patterns in two cacao genotypes with contrasting responses.

Methodology

Three Biological replicates

<table>
<thead>
<tr>
<th>Time</th>
<th>Control (Sandra)</th>
<th>1x10⁷ UFC/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>0h</td>
<td>180</td>
<td>90</td>
</tr>
<tr>
<td>24h</td>
<td>180</td>
<td>90</td>
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<tr>
<td>48h</td>
<td>180</td>
<td>90</td>
</tr>
<tr>
<td>96h</td>
<td>180</td>
<td>90</td>
</tr>
</tbody>
</table>

24 RNAseq libraries sequenced on Hiseq X system (150 PE, Illumina) in three independent lanes by biological replicate

P. palmivora TOCHA 325

A

CCN-51 and SCA-6

1x10⁷ UFC/ml

P. palmivora TOCHA 325

B

DNA extraction

Library Nextera flex

Hiseq X 150 PE (Illumina)

P. palmivora TOCHA 325

C

Figure 1: Methodology for conducting dual transcriptomics in P. palmivora T. cacao. A. Experimental design for the RNAseq assay. B. Workflow for obtaining P. palmivora genome C. Bioinformatic workflow to conduct P. palmivora assembly and dual RNAseq assembly.

Conclusions

We present preliminary results in which some candidate defense response genes were identified. A new genome for P. palmivora was assembled and annotated. The next step will be to use this genome to identify pathogen effectors and continue to explore the transcriptomic data to identify the associations between the plant defense response and the pathogen effectors. These results are an essential tool to identify genes responsible for the resistance and generate improved varieties with resistance to black pod disease to reduce losses due to diseases caused by Phytophthora sp.

References


Acknowledgements

This study was funded by the Ministerio de Agricultura de Colombia. We thank Leonnora Rodríguez, Martha Carrera, Mauricio Soto, Esperanza Torres and Bayardo Parra for their guidance and help in the experimental work.