Transcriptomic Profiling Identifies a Shared Incompatibility Response in Weak and Strong Barrier Monokaryon Pairings of *Ganoderma Boninense*

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Abstract:

Basal stem rot is a disease caused by *Ganoderma boninense*, which is a white rot fungus that spreads through root contact and mycelial networks in oil palm plantations and causes disease to the oil palm. However, the barriers that determine whether two fungal genotypes fuse or reject one another remain poorly understood. Vegetative incompatibility governs this outcome, but its molecular basis in *G. boninense* is rarely described. A total of 27 RNA-seq libraries was compared from confrontations between a common monokaryon (PER2-2) and compatible (PER/19-18), weak-barrier incompatible (PER/12-11) and strong-barrier incompatible (PER/16-15) partners. The mating junction of the pairs were sampled at three time points: before interaction, during interaction and after interaction. Reads were trimmed using Trimmomatic and aligned to the *G. boninense* pseudochromosomes, GbPER71_V4 via HISAT2 with overall alignment rates of 88.0–90.8%. Differential expression was assessed with DESeq2 (padj < 0.05, $\log_2$FC > 1). A total of 1727 and 1622 genes were differentially expressed in the weak- and strong-barrier pairings, of which 920 were consistently differentially expressed for both incompatibilities. Principal component analysis (PCA) of RNA-seq data revealed that the gene expression profiles at all three pairings were different. PC1 and PC2 captured 50.7% and 40.5% of expression variance. Enrichment analysis of the 483 shared upregulated genes revealed a significant association with monooxygenase and oxidoreductase activity, heme and iron binding, proteolysis, and carbohydrate metabolism, while the 273 shared downregulated genes were associated with ATP-dependent and phosphorylation-related functions. HET-domain and laccase genes were nearly silent in the compatible pairing but strongly induced in both incompatible pairings. As HET-domain proteins

mediate non-self-recognition in fungi, these results indicate that incompatible pairings of *G. boninense* activate a shared recognition and oxidative barrier-formation programme, providing the molecular framework for vegetative incompatibility in this oil palm pathogen.

Keywords:
Somatic incompatibility, RNA-seq, white rot fungi, transcriptome.