

Temporal Gene Expression Analysis Reveals a Synergistic
Effect of Combined Drought and Heat Stress in Grapevine

(Vitis vinifera L.)

By

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Declaration

I declare that this thesis is a record of original work and contains no material which has been accepted for the award of any other degree or diploma in any university. To the best of my knowledge and belief, this thesis contains no material previously published or written by another person, except where due reference is made in the text.

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Date

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Preface

This research was performed over 10 months as part of a Master of Biotechnology (Plant Biotechnology). In accordance with the requirements of the program, the research is presented in the format of a manuscript for submission to a peer-reviewed scientific journal. I have chosen to follow the format of *Journal of Experimental Botany*. My co-authors for the manuscript are Carlos Rodriguez Lopez, Kiflu Gebremicael Tesfamicael, Penny Tricker, Ute Baumann and Everard Edwards. Carlos Rodriguez Lopez developed the novel experimental design that I used in this research and supervised me in the other aspects of the data analysis. Kiflu Gebremicael Tesfamicael helped to carry out the glasshouse experiment and assisted me with lab experiment and data collection. Na Sai, Konstantinos Bogias and Jimmy Breen helped me with bioinformatics application. Carlos Rodriguez Lopez suggested the project, supervised my research and reviewed drafts of the manuscript. The manuscript in this thesis is intended as the first draft of a manuscript for future publication. The final word count for the manuscript (excluding references and supplementary materials) is 7804.

The appendix contains the supplementary materials. I have followed the Author Guide except that I used double-spaced line and lack of line numbering for which my manuscript differs from the journal's instructions to satisfy the thesis guidelines for the Master of Biotechnology (Plant Biotechnology) program.

Research Manuscript

Title: Temporal Gene Expression Analysis Reveals a Synergistic Effect of Combined Drought and Heat Stress in Grapevine (*Vitis vinifera* L.)

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Abbreviations

Abscisic Acid (ABA)

Counts Per Million (CPM)

Diethylpyrocarbonate (DEPC)

Differentially Expressed Genes (DEGs)

False Discovery Rate (FDR)

Gene Ontology (GO)

Hierarchical Indexing for Spliced Alignment of Transcripts (HISAT)

Log Ration Fold Change (LogFC)

Nitric Oxide (NO)

Nitrate Reductase (NR)

Reactive Oxygen Species (ROS)

Ribosomal RNA (rRNA)

RNA Quality Indicator (RQI)

RNA Sequencing (RNA-Seq)

Vapour Pressure Deficit (VPD)

Abstract

Grapevine (*Vitis vinifera* L. cv. Cabernet Sauvignon) is widely used for winemaking all over the world. Drought and heat stresses are two of the major abiotic stresses reducing grape quality and yield. However, drought and heat tolerance are still poorly characterized in perennial crops such as grapevine. During this study, stomatal conductance, stem water potential and leaf temperature were measured to determine plant physiological status. RNA-seq technology was used for the analysis of differentially expressed genes (DEGs) of leaf samples between the control and three treatments, which were drought, heat and a combined treatment. Gene expression profiles were grouped by treatments and timepoints. The great majority of unique DEGs were found to be induced by the combined drought and heat treatment. 169 up-regulated genes were induced by drought, 85 by heat and 1218 by the combined treatment; 78 down-regulated genes were induced by drought, 72 by heat and 1427 by the combined treatment. Three potential and significant regulation pathways of stress response were identified based on Gene Ontology (GO) analysis i.e. cytokinin-activated signalling pathway, ion transport pathway and Nitric Oxide-mediated pathway. This study provides preliminary insights into the transcriptomic response to drought and heat stress in grapevine.

Key words: Grapevine, RNA-seq, Drought, Heat, Differentially Expressed Genes, Gene Ontology