Analysis of the Stress-inducible Promoter of *TdDHN8/WCOR410* from Wheat Using Transient Expression Assays

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[1].

Signature of Student

Student Name

Date

[1] If a manuscript should be published, this declaration page will also need to include the percentage of contribution by the student and be signed by all others in addition.

Preface

The master research project went through eight months from February 5th to September 27th, and has successfully been completed as we planned at the beginning. The research was mainly focusing on the analysis of a new wheat promoter, pTdDHN8/WCOR410, which was originally presumed as a drought-inducible promoter. As a backup research plan, we also tested nine lines of the T1 transgenic barley plants transformed with the pRab17-*GUS* fusion under 250mM salt stress.

We spent nearly five months to optimize the experimental conditions of transient expression assay using cell suspension cultures that are rarely used for the analysis of inducible promoter in plants. The effort included: 1) which plant tissue was optimum to characterize the activity and inducibility of the *TdDHN8/WCOR410* promoter; 2) how to minimize the factors that affected the transformation efficiency in cell suspension cultures via particle bombardment; 3) how to precisely induce the osmotic stress in the growth medium. Finally, we efficiently optimized the experiment conditions, paving the way to further dissect the *TdDHN8/WCOR410* promoter activity using transient expression assay in cell suspension cultures. In term of the backup research, we got four transgenic plants that were qualified using GUS staining assay, and they have been transplanted into soil for T2 seeds.

In the present thesis, the revised version of literature review, which has been examined by Dr. Andrew Jacobs, Dr. Oliver Cotsaftis, and Prof. John Randles on March, is present in the first part. The second part is the revised manuscript of the *TdDHN8/WCOR410* promoter analysis according to the format of *The Plant Journal*. The final version of my master thesis was revised based on the critical suggestions by Prof. Peter Langridge, Dr. Oliver Cotsaftis, and Dr. Bujun Shi. Although some big progress was made in last few months, yet we recognize that more hard work is still needed to address the problem of the big variation of transformation efficiency in cell suspension cultures via particle bombardment, and extend our findings in the thesis in the next few months. At the end of my master study, I thank those lovely persons who help me for the master research in plant genomics center. The big gratitude are also given to our program coordinator Dr. Amanda Able for her assistance during my two-year master study in The University of Adelaide, and my supervisors Dr. Sergiy Lopato and Dr. Serik Eliby for their kindness and patience in my master research project.

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