UNIVERSITI TEKNOLOGI MARA

PROTEOMIC AND ENZYMATIC STUDIES IN RELATION TO REGENERATION CAPACITY OF *BOESENBERGIA ROTUNDA* CELL SUSPENSION CULTURE

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Thesis submitted in fulfillment
of the requirements for the degree of
Master of Science
(By Research)

Faculty of Plantation and Agrotechnology

January 2018

ABSTRACT

Gradual loss of cell proliferation and regeneration capacity in prolonged cultures of cell suspension continues to be a significant limitation, particularly those with economic interest and related studies. In this study, biochemical changes and proteins associated with cell proliferation and regeneration capacity from *Boesenbergia rotunda* cell suspension culture were investigated. The results showed that the optimum settle cell volume (SCV) of 6 and 9 month-old cell suspension proliferation were decreased of 42.17 % and 53.78 % respectively when compared to 0 month-old cells. In general, shoots of 4 ± 2 cm were formed after 7 months of culture. Regeneration response was recorded at 66.5 %, 56.6 %, and 47.4 % of 0, 6 and 9 month-old cells, respectively. Differences between 6 and 0 month-old cells for superoxide dismutase (SOD), hydrogen peroxide (H_2O_2), catalase (CAT) and ascorbate peroxidase (APX) were 14.40 Unit SOD/mg protein, 5.27 µmol H_2O_2/mg protein, 74.38 µmol H_2O_2/mg protein, and 2.23 µmol AsA/mg protein, respectively. While the activities of SOD, H_2O_2 and APX decreased from 6 to 9 month-old cells, CAT activity was found to increase. We applied a gel-based proteomic technique to analyze protein changes for 0, 6 and 9 month-old cells. Protein extraction protocol for *B.* rotunda cell suspension culture were optimized by comparing several extraction protocol by which TCA-acetone in combination with DTT showed the best result. A total of 13 protein spots showed significant differential expression and 8 protein spots were successfully identified. They were classified as protein synthesis, energy metabolism, defense and stress responses, and catabolic proteins.

ACKNOWLEDGEMENT

Alhamdulillah. First and foremost, I would like to thank Allah for His blessing that enable me to accomplish this study successfully. This project is the result of the dedicated effort of many individuals, several of whom deserve special mention:

First of foremost, I would like to express my greatest gratitude and appreciation to my supervisors, Dr. Nor Azma Yusuf and my co-supervisor Professor Dr. Norzulaani Khalid for the supervision, guidance and constructive recommendations throughout the project.

Special thanks to Dr. Tan Boon Chin for his advice, guidance and helping me to get essential scientific facts besides guiding me through the laboratory technique and analysis.

To my family I unreservedly express my deepest and most profound appreciation: my parents Aziz Abu Bakar and Kamariah Khalid, my siblings Aiman Ariffudin Aziz and Eman Najua Aziz, the individual important to me Rizal, Norshaida, Ropelis, Siti Wilis, and Norazua for the steadfast support, patience and understanding.

Not forgetting also many thanks to CEBAR laboratory assistants and staff members of University of Malaya. My appreciation to the Institute of Biological Sciences PBRL assistant science officer Pn Azlina, fellow research students Ain, Gayatri, Nurul, Tan, Wani, Fatin, Zakuan Wan Sin, Fong, Diyana, As and William. Special thanks to Sher Ming and Wendy for their guidance in plant tissue culture and enzyme assay. Special thanks also go to my brothers, Nabeel Ata and Nazrin Abd Aziz for always being there when and wherever needed.

To all of these people mentioned and unmentioned, I am deeply grateful for their help and support.

TABLE OF CONTENTS

		Page
CONFRIMATION BY PANEL OF EXAMINERS		ii
AUTHOR'S DECLARATION		iii
ABSTRACT		iv
ACKNOWLEDGMENT		v
TABLE OF CONTENTS		vi
LIST OF TABLES		xi
LIST OF FIGURES		xiii
LIST OF ABBREVIATIONS		xv
CHAPTER ONE : INTRODUCTION		
1.1	General Introduction	1
1.2	Scope of Research	3
1.3	Objective of Study	3
1.4	Limitations of Study	4
1.5	Hypothesis	4
1.6	Significance of Study	4
CHAPTER TWO : LITERATURE REVIEW		
2.1	Zingiberaceae Species	5
2.1.1	*Boesenbergia* Species in Malaysia	7
2.2	*Boesenbergia rotunda*	8
2.2.1	Origin and Taxonomy of *Boesenbergia rotunda*	8
2.2.2	Morphological and Botanical Description of *Boesenbergia rotunda*	10
2.2.3	Nutritional Composition and Medicinal Properties of *Boesenbergia rotunda*	11
2.2.4	Conventional Propagation of *Boesenbergia rotunda*	15
2.3	Plant Tissue Culture	15
2.3.1	Establishment of Surface Sterilization	16

2.3.2 Callus Culture 17
2.3.3 Suspension Culture 18
2.3.4 Prolong Cell Culture 20
2.4 Reactive Oxygen Species (ROS) and Antioxidants In Plant 21
2.4.1 Reactive Oxygen Species 21
2.4.2 Enzymatic Antioxidant 22
2.4.2.1 Superoxide Dismutase 23
2.4.2.2 Ascorbate Peroxidase 24
2.4.2.3 Catalase 25
2.4.2.4 Glutathione Reductase 25
2.4.2.5 Guaiacol Peroxidase 26
2.4.3 Non-enzymatic Antioxidant 26
2.5 Proteomic Studies in Plant 27
2.5.1 Gel Base Protein Separation 27
2.5.1.1 One-dimensional Gel Electrophoresis (1-DE) 28
2.5.1.2 Two-dimensional Gel Electrophoresis (2-DE) 28
2.5.2 Gel-free Protein Separation 30
2.5.2.1 Gel-free Label Based 30
2.5.2.2 Gel-free Non-label Based 32
2.5.3 Protein Extraction 34
2.5.3.1 Trichloroacetic Acid Acetone Precipitation Extraction Method (TCA-acetone) 35
2.5.3.2 Phenol Extraction Method 35
2.5.3.3 Trichloroacetic Acid Acetone Precipitation-Phenol Extraction Method (TCA-acetone Phenol) 36
2.5.3.4 Total Protein and Precipitation Extraction Method 37
2.5.4 Protein Separation and Staining Procedure 37
2.5.5 Mass Spectrometry 38
2.5.6 Application of Protein Profiling for Cell Culture Studies 39
2.5.6.1 Somatic Embryogenesis 40
2.5.6.2 Differentially Expressed Protein Induced by Elicitor 41
2.5.6.3 Regeneration Response from Short-term and Long-term Cell Suspension Culture 41