

WAN ZALHA BINTI WAN SEMBOK

REGULATION OF FRUIT COLOUR DEVELOPMENT,
QUALITY AND STORAGE LIFE OF 'CRIPPS PINK'
APPLE WITH DEFICIT IRRIGATION AND PLANT
BIOREGULATORS

CURTIN UNIVERSITY OF TECHNOLOGY
BENTLEY, WESTERN AUSTRALIA

0130073273
School of Agriculture and Environment

**Regulation of Fruit Colour Development, Quality and Storage Life
of 'Cripps Pink' Apple with Deficit Irrigation and
Plant Bioregulators**

Wan Zaliha Binti Wan Sembok

**This thesis is presented for the Degree of
Doctor of Philosophy
of
Curtin University of Technology**

November 2009

THESIS

Declaration

To the best of my knowledge and belief this thesis contains no material previously published by any other person except where due the acknowledgement has been made. This thesis contains no material which has been accepted for the award of any other degree or diploma in any university.

Signature: _____

Date: _____

Dedication

To

My mother (*Maznah Hassan*)

&

My father (*Late Wan Sembok Wan Ali*)

**'A constant source of inspiration during the entire period of my PhD study and
throughout my life'**

Acknowledgements

In the name of *Allah*, the most Beneficent and the Merciful.

I am grateful to *Allah* Almighty for *His* help and blessings that enable me to complete this research work and the thesis. Special thanks to my supervisor Professor Dr. Zora Singh for his creative comments and guidance throughout my research and the thesis completion process. I extend my thanks to Associate Supervisor, Dr Ritu Gupta for her help and suggestion during my thesis completion. Thanks to Dr. Ewald E. Swinny, Chemist and Research officer, Chemistry Centre, Western Australia for his help and guidance in biochemical analysis. My special thanks to Associate Professor M. Hossein Behboudian, Institute of Natural Resources, Massey University, New Zealand for his help and suggestion in irrigation experiments.

I would like to express my deep appreciation to University Malaysia Terengganu, Malaysia for granting study leave and awarding me a scholarship and research grant for my PhD programme. My sincere thanks to G. Casotti & Company, Karragullen, Perth Hills and Giumelli & Sons, Carmel, Western Australia for generously providing the 'Cripps Pink' apple trees and fruit required for this research. I would also like to thank Nufarm Australia Ltd., Kwinana, Western Australia for providing Regalis (Prohexadione-calcium) as a gift sample for my experiments.

My special thanks to Ms. Susan M. Petersen, Laboratory Manager, Curtin Horticulture Research Laboratory for her technical support and assistance during my laboratory work and also to my lab-mates, Minh Tam Pham, Maria Fransisca Sumual, Sukhvinder Pal Singh, Dr. Khuyen Dang and Dr. Harsimranjit Singh for their help, cooperation, discussion and encouragement during the course of my studies. I am thankful to my friends, Dr. Nolila, Dr. Marina, Rosazalin, Hannerita and others for their support and motivation.

I owe a heartfelt debt of gratitude to my mother (Maznah Hassan) for being my pillar of strength during the highs and lows of this course and also for her endless love, tireless support and prayers for my success. I would also like to express my deep appreciation to my siblings; Wan Ahmad, Wan Ismail, Wan Zubaidah, Wan Zaharah,

Wan Junaidah and Wan Fatimah Suraya for their constant encouragement, motivation and their prayers for my success.

Abstract

Poor and erratic fruit colour development in 'Cripps Pink' apple causes serious economic losses to the growers and/or exporters of Western Australia and other parts of the world. Many internal and external factors such as genetic, light, temperature, irrigation, application of chemicals and also soil and tree factors affect the biosynthesis of anthocyanins consequently fruit colour. Some of the past approaches followed to improve fruit skin colour resulted in limited outcomes. The aim of my research was to evaluate the effects of water saving strategies and newly developed plant bioregulators in improving fruit colour development without adversely affecting fruit size and quality of 'Cripps Pink' apple at harvest, following cold and controlled atmosphere (CA) storage. I also investigated the individual polyphenolics profiles, their identification and confirmation in the skin of this apple cultivar. Nine polyphenolic compounds (cyanidin 3-*O*-galactoside, chlorogenic acid, phloridzin, quercetin 3-*O*-rutinoside, quercetin 3-*O*-galactoside, quercetin 3-*O*-glucoside, quercetin 3-*O*-xyloside, quercetin 3-*O*-arabinoside and quercetin 3-*O*-rhamnoside) in the fruit skin of 'Cripps Pink' apple were identified, quantified and re-confirmed using high performance liquid chromatography-electrospray ionization mass spectrometry (HPLC-ESI-MS). Increased concentration of cyanidin 3-*O*-galactoside in 'Cripps Pink' apple skin coincided with the increase in total anthocyanins concentrations.

Water saving strategies, regulated deficit irrigation (RDI) and withholding irrigation (WHI), have been carried out for two seasons (2005-06 and 2006-07, and 2006-07 and 2007-08, respectively) in a commercial apple orchard. The treatment (75% RDI applied for 72 days, commencing on 135 days after full bloom (DAFB) and WHI for 20 to 30 days, commencing on 135 and 145 DAFB) increased red skin colour, concentration of total anthocyanins and polyphenolic compounds such as cyanidin 3-*O*-galactoside and quercetin glycosides. These treatments also improved fruit firmness and soluble solids concentration (SSC) of 'Cripps Pink' apple at harvest without adversely affecting postharvest quality in cold and controlled atmosphere (CA) storage, and also saved the irrigation water. To the best of my knowledge, this may be the first report on the effects of water-deficit on accumulation of flavonoids and other phenolic compounds in red-skinned apple particularly 'Cripps Pink'

cultivar and also its impact on postharvest storage performance in CA storage. Soil-plant water relations such as volumetric soil water content, stomatal conductance, leaf water potential and stem water potential was pronounced with the application of these water saving strategies applied in the middle of stage II of fruit development of 'Cripps Pink' apple. The sparse leaf abscission due to water-deficit has improved light penetration, consequently improved red skin colouration through increased accumulation of anthocyanins particularly cyanidin 3-*O*-galactoside. This highlighted the importance of water stress and light in regulating colour and biosynthesis of anthocyanins.

Newly developed plant growth regulator, Prohexadione-calcium (ProCa) improved fruit colour development of this apple cultivar by manipulating the light interception into the tree canopy and onto the fruit through reduction of vegetative growth. The reduction of shoot length was pronounced with three spray applications of ProCa (500 mg·L⁻¹) on 3, 33 and 63 DAFB or two sprays of ProCa (500 mg·L⁻¹) on 2 and 32 DAFB in combination with summer pruning (SP). The above mentioned treatments increased concentration of anthocyanins, cyanidin 3-*O*-galactoside, and all individual quercetin glycosides (quercetin 3-*O*-rutinoside, quercetin 3-*O*-galactoside, quercetin 3-*O*-glucoside, quercetin 3-*O*-xyloside, quercetin 3-*O*-arabinoside and quercetin 3-*O*-rhamnoside) and also maintained other fruit quality attributes such as fruit firmness and SSC of this apple cultivar.

Lysophosphatidylethanolamine (LPE) spray, 125 mg·L⁻¹ (at two and four weeks prior to anticipated commercial harvest) or 250 mg·L⁻¹ (at four weeks before harvest) appeared to be promising in improving fruit colour development, accumulation of anthocyanins and polyphenolic compounds (cyanidin 3-*O*-galactoside, quercetin glycosides and also individual quercetin glycosides such as quercetin 3-*O*-xyloside, quercetin 3-*O*-arabinoside and quercetin 3-*O*-rhamnoside), and other fruit quality attributes of 'Cripps Pink' apple. However, the mode of action of LPE in improving red colour in apple skin is possibly associated with enhanced ethylene production.

In conclusion, fruit colour development of 'Cripps Pink' apple can be improved by applications of water saving techniques in the middle of stage II of fruit development such as 75% RDI for 72 days commencing on 135 DAFB or WHI for 20 (135-155

DAFB) to 30 (145-175 DAFB) days, and also newly developed plant bioregulators such as ProCa (three spray applications of ProCa ($500 \text{ mg}\cdot\text{L}^{-1}$) on 3, 33 and 63 DAFB or two sprays of ProCa ($500 \text{ mg}\cdot\text{L}^{-1}$) on 2 and 32 DAFB in combination with SP) or LPE (two spray applications ($125 \text{ mg}\cdot\text{L}^{-1}$) at two and four weeks prior to anticipated commercial harvest or single spray ($250 \text{ mg}\cdot\text{L}^{-1}$) at four weeks before harvest) without adversely affecting other fruit quality attributes.