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GROWTH, DEVELOPMENT AND QUALITY OF WAX APPLE (*SYZYGIUM SAMARANGENSE* [BLUME] MERRILL & L. M. PERRY) FRUITS AS INFLUENCE BY SELECTED HORTICULTURAL TECHNIQUES

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GROWTH, DEVELOPMENT AND QUALITY OF WAX APPLE (*SYZYGIUM SAMARANGENSE* [BLUME] MERRILL & L. M. PERRY) FRUITS AS INFLUENCED BY SELECTED HORTICULTURAL TECHNIQUES

ABSTRACT

Plant growth regulators, regulating chemicals and growth manipulating techniques are important tools in horticulture. These techniques can regulate various physiological and biochemical aspects of growth and development, and improve growth, yield and nutritional quality of fruits. Currently, there is a limited report if any, describing how plant growth regulators (PGRs) affect the fruit quality of wax apple, an increasingly popular fruit in Asia. The present study was carried out to investigate the effects of plant growth regulators, gibberellic acid (GA_3) at 20, 50 and 100 mg/L, 2, 4-dichlorophenoxy acetic acid (2,4-D) and naphthalene acetic acid (NAA) at 5, 10 and 20 mg/L, girdling practices (50%, 100%, I, C and V-shaped) and hydrogen peroxide (H_2O_2) at 5, 20 and 50 mM on the growth, development and quality of the wax apple (*Syzygium samarangense*) var. *Jambu Air Madu* fruits Various physiological, biochemical and phytochemical parameters were monitored during three seasons of fruit growth between December, 2008 to May, 2011 at MARDI, Jalan Kebun, Klang, and a farm in Banting. The results showed that the application of GA_3 enhanced fruit growth, increased fruit length, diameter and color development, fruit number, weight and yield. GA_3 also increased the number of buds and fruit set and reduced bud and fruit drop. With regard to fruit quality, the application of GA_3 increased fruit juice, biomass, total soluble solids (TSS), glucose, fructose, sucrose, total sugar, sugar acid ratio, soluble protein and total flavonoids content in the fruits. In addition, anthocyanin content, total phenol, ascorbic acid and antioxidant activity was higher in GA_3-treated fruits. Less titrable acidity was also recorded in treated fruits. Treatment with 2, 4-D enhanced bud development, fruit growth, fruit length and diameter,

peel color development and increased fruit set, yields, and fruit drymatter. The treatment also reduced bud and fruit drop. Chlorophyll fluorescence, photosynthetic yield, stomatal conductance and chlorophyll readings were also high in 2, 4-D-treated branches. Fruit quality parameters such as TSS, total sugar and anthocyanin content were also high in the treated fruits. NAA treatments increased bud number and total number of fruits, enhanced faster fruit growth, fruit size and red color development. Furthermore, NAA treatment increased photosynthetic quantum yield, chlorophyll content, stomatal conductivity, fruit set, fruit weight and total yield in addition to decreasing fruit drop. With regard to fruit quality, NAA treatment increased TSS, fruit juice, total sugar, phenol, flavonoid contents and higher antioxidant activity in the fruits, in addition to a higher anthocyanin, protein content and phenylalanine ammonia lyase (PAL) activity. Girdling significantly increased fruit growth and development, and color development. Apart from improving leaf physiological characteristics, girdling also increased juice K^+ content, TSS, total sugars, total phenolics, flavonoids and anthocyanin content. A higher antioxidant activity in the fruit was also observed. Leaf chlorophyll content and dry matter, total soluble solids and total sugar contents of fruits were significantly increased in wax apple after treatment with H_2O_2. The application of H_2O_2 significantly reduced bud drop and enhanced fruit growth, resulting in larger fruit size, increased fruit set, fruit number, fruit biomass and yield. H_2O_2 treatment increased the K^+, anthocyanin, carotenoid, flavonoid, phenolic and soluble protein content. Higher PAL and antioxidant activities were measured in the treated fruits. There was a positive correlation between peel color and TSS and anthocyanin, stomatal conductance with photosynthesis and yield, phenols and flavonoids with antioxidant activity, PAL activity and anthocyanin formation. Finally, net photosynthesis, stomatal conductance and transpiration, leaf chlorophyll and dry matter content in the leaves of the

wax apple were significantly increased after GA_3 and H_2O_2 treatments. Sucrose phosphate synthase (SPS) activity and rubisco expression was also high in GA_3 treatments. In addition, a higher soluble protein content, PAL and anthocyanin formation were recorded in the treated fruits. From this study, it can be concluded that spraying PGRs (50 mg/L GA_3, 10 mg/L NAA or 2, 4-D), 20 mM H_2O_2 and C-shaped girdling application results in better yield and quality of wax apple fruits under field conditions.

PERTUMBUHAN, PERKEMBANGAN DAN KUALITI BUAH WAX APPLE (*SYZYGIUM SAMARANGENSE* [BLUME] MERRILL& L. M. PERRY) YANG DIPENGARUHI OLEH TEKNIK-TEKNIK HORTIKULTUR TERPILIH

ABSTRAK Dan pengaruh mereka

Pengawalaturan pertumbuhan pokok (Plant growth regulators, PGRs), bahan kimia yang mengawalatur dan memanipulasi teknik pertumbuhan adalah kaedah yang penting dalam sistem hortikultur. Teknik ini boleh mengawal pelbagai aspek fisiologi dan biokimia pertumbuhan dan meningkatkan kualiti pertumbuhan, hasil dan pemakanan buah-buahan. Pada masa ini, terdapat laporan terhad yang menjelaskan bagaimana pengawalaturan ini menjejaskan kualiti kutikel buah epal yang semakin popular di Asia. Oleh itu, kajian ini telah dijalankan untuk menyiasat kesan pengawalaturan pertumbuhan tumbuh-tumbuhan, asid gibberellic (GA_3) pada 20, 50 dan 100 mg / L, 2, 4-dichlorophenoxy asetik asid (2,4-D) dan asid asetik naftalena (NAA) pada 5, 10 dan 20 mg / L, kaedah girdling (50%, 100%, I C dan berbentuk V,) dan hidrogen peroksida (H_2O_2) pada 5, 20 dan 50 mM pada pertumbuhan dan kualiti kutikel pada buah jambu air madu (*Syzygium samarangense*). Pelbagai parameter fisiologi, biokimia dan fitokimia dipantau sepanjang tiga musim pertumbuhan buah-buahan antara Oktober 2008 hingga Ogos, 2011 di MARDI, Jalan Kebun, Klang, dan sebuah ladang di Banting. Hasil kajian menunjukkan bahawa penggunaan GA_3 meningkatkan pertumbuhan buah-buahan, panjang buah-buahan, garis pusat dan perkembangan warna, bilangan buah-buahan, berat dan hasil buah-buahan. GA_3 juga meningkatkan bilangan tunas dan set buah-buahan dan mengurangkan keguguran putik dan buah-buahan. Dengan mengambil kira kualiti buah-buahan, penggunaan GA_3 meningkatkan jus buah-buahan, biomass, total soluble solids (TSS), glukosa, fruktosa, sukrosa, jumlah nisbah gula acid, gula, protein larut dan jumlah kandungan flavonoid dalam buah-buahan. Di samping itu, kandungan antosianin, jumlah fenol, asid askorbik

dan aktiviti antioksidan juga meningkat dalam buah-buahan yang dirawat dengan GA_3. Pengurangan keasidan titrable juga direkodkan dalam buah-buahan yang dirawat.

Rawatan dengan 2, 4-D menggalakkan pertumbuhan tunas dan buah, panjang buah-buahan dan garis pusat, pertumbuhan warna buah ranum dan meningkatkan set, hasil, dan berat bersih buah-buahan. Ia juga mengurangkan keguguran putik dan buah-buahan. Pendarfluor Klorofil, hasil fotosintesis, kealiran stomatal dan bacaan klorofil juga tinggi pada dahan yang dirawat dengan 2, 4-D. Parameter kualiti buah-buahan seperti gula TSS, jumlah dan kandungan antosianin juga tinggi dalam buah-buahan yang dirawat. Rawatan NAA meningkatkan bilangan putik dan jumlah buah-buahan, menggalakkan pertumbuhan buah-buahan, saiz buah dan perkembangan warna merah dengan lebih cepat. Tambahan pula, rawatan NAA meningkatkan jumlah hasil fotosintesis, kandungan klorofil, kekonduksian stomatal, set buah-buahan, berat buah-buahan dan hasil jumlah di samping mengurangkan keguguran buah-buahan. Dengan mengambil kira kualiti buah-buahan, rawatan NAA meningkatkan TSS, jus buah-buahan, gula jumlah, fenol, kandungan flavonoid dan aktiviti antioksidan dalam buah-buahan, sebagai tambahan kepada aktiviti antosianin, kandungan protein dan PAL yang lebih tinggi. Kaedah *girdling* meningkatkan pertumbuhan dan perkembangan warna buah-buahan. Tambahan pula, ciri-ciri fisiologi daun juga bertambah baik dengan kaedah *girdling*. Kaedah ini juga meningkatkan kandungan K^+, TSS, jumlah gula, phenolic jumlah, flavonoid dan kandungan antosianin. Aktiviti antioksidan yang lebih tinggi dalam buah juga direkodkan. Kandungan klorofil dan berat bersih daun, jumlah pepejal larut dan jumlah kandungan gula buah-buahan meningkat dengan ketara dalam lapisan kutikel buah epal ber lilin var. Jambu madu) selepas dirawat dengan H_2O_2. Penggunaan H_2O_2 mengurangkan putik drop dan menggalakkan pertumbuhan buah-buahan, menghasilkan saiz buah yang lebih besar, meningkatkan set, bilangan, biomas dan

hasil buah-buahan. Rawatan H_2O_2 meningkatkan kandungan K +, antosianin, protein karotenoid, flavonoid, fenolik dan protein larut. Kandungan lyase ammonia Phenylalanine (PAL) dan aktiviti-aktiviti antioksidan yang lebih tinggi didapati dalam buah-buahan yang dirawat. Terdapat korelasi positif antara warna buah ranum, TSS dan antosianin, kealiran stomatal dengan hasil fotosintesis; fenol dan flavonoid dengan aktiviti antioksidan, aktiviti PAL dan pembentukan antosianin. Kesimpulannya, hasil bersih fotosintesis, kealiran stomatal dan transpirasi, klorofil daun dan kandungan berat bersih dalam daun epal ber lilin meningkat dengan ketara selepas rawatan GA_3 dan H_2O_2. Aktiviti sukrosa fosfat synthase (SPS) juga tinggi dalam buah-buahan yang dirawat dengan GA_3 dan H_2O_2. Di samping itu, kandungan protein larut, PAL dan pembentukan antosianin yang lebih tinggi dicatatkan dalam buah-buahan yang dirawat. Daripada kajian ini, dapat disimpulkan bahawa penyemburan PGRs (50 mg/L GA_3, 10 mg/L NAA atau 2, 4-D), 20 mM H_2O_2 dan kaedah C-shaped girdling memberikan hasil dan kualiti lapisan kutikel buah epal yang lebih baik menurut syarat-syarat yang ditetapkan.

DEDICATION

This dissertation is dedicated to my late father Khandaker Siddiqur Rahman and my family. Their love, support, and encouragement throughout this academic research journey meant everything to me.

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TABLE OF CONTNETS

Page

ORIGINAL LITERARY WORK DECLARATION…….....i

ABSTRACT……...ii

ABSTRAK……...v

DEDICATION……..viii

ACKOWLEDGEMENTS…….ix

TABLE OF CONTENTS……...x

LIST OF TABLES……xvii

LIST OF FIGURES……..xix

ABREVIATIONS……..……xxv

APPENDICES…….xxvii

CHAPTER 1: INTRODUCTION ……1

CHAPTER 2: LITERATURE REVIEW

2 .1 The botany of *Syzygium samaragense* ……...9

2.2 Ecology and distribution of *Syzygium spp*…….13

2.3 Fruit growth, development and harvest ……...15

2.4 Growth and ripening of wax apple……....16

2. 5 Functional uses of *Syzygium spp*……...18

2.6 Physicochemical changes in fruits during ripening……..21

2.7 Postharvest changes in non-climacteric fruits……..22

2.7.1 Tissue firmness……..22

2.7.2 pH…….24

2.7.3 Total soluble solids (TSS)……25

2.7.4 Titrable acidity content……..27

2.7.5 Total sugar content……....28

2.8 The use of selected horticultural techniques improve to fruit growth, development and quality……30

2.9 The effect of gibberellic acid on growth and development……...30

2.10 Gibberellic acid on fruit quality……35

2.11 NAA and 2, 4-D on growth and development……..38

2.12 Auxin (NAA and 2, 4-D) on fruit quality……..42

2.13 Effects of girdling on growth and development ……...44

2.14 Effects of girdling on leaf and fruit quality……...47

2.15 Influence of hydrogen peroxide on fruit growth and development……..48

2.16 Influence of hydrogen peroxide on fruit quality……...50
2.17 Plant growth regulators on color development and anthocyanin formation…….....52
2.18 The effects of plant growth regulators on phenylalanine ammonia lyase (PAL) enzyme activity……. ….54
2.19 The effects of gibberellic acid on antioxidant content, phenols and flavonoids of treated plants……..57
2.19.1 Antioxidant assays……57
2.19.2 The DPPH assay…….....58
2.19.3 The ABTS assay……...61
2.19.4 Antioxidant activity in fruits…….....62
2.19.5 Phenolic content…….....65
2.19.6 Flavonoid content……..69
2.20 Growth regulators effects on photosynthesis……..72
2.21 The effects of gibberellic acid on protein and rbcA gene of treated plants……..75

CHAPTER 3 MATERIALS AND METHODS

3.1 a. Experimental site……81
3.1 b. Experimental design…….....81
3.2.1 Experiment 1.The effect of GA_3 treatments on fruit growth, development and quality of wax apple
(a) Plant materials …….....85
(b) Treatment application……...85
(c) Measurements of physiological parameter
(i) Number of buds……...85
(ii) Bud drop (%)……...86
(iii) Fruit set (%)…….....86
(iv) Fruit drop (%)…….....86
(v) Fruit growth…….....87
(vi) Fruit weight (average)…….....87
(vii) Peel color (%)…….....87
(viii) Yield and fruit dry biomass…….....87
(ix) Leaf characteristics (Leaf length, wide, fresh weight and drymatter content)……87
(d) Measurements of biochemical parameter
(i) Determination of chlorophyll a, b and carotenoid Content.....88
(ii) Protein determination by Bradford's method……89
(iii) Juice content……..89
(iv) Fresh biomass and dry matter……....90
(v) Total soluble solids (TSS) and pH……90
(vi) Titratable acidity and TSS/TA ratio……..90
(vii) Total ascorbic acid content assay……..90
(viii) Measurement of the K^+ content in fruit juice……91

(xi) Determination of total sugars……91

(x) Glucose, fructose and inverted sugar……. ……..92
(xi) The total phenolic content (TPC)…….....92
(xii) Total flavonoid content (TFC)…….....92
(xiii) Antioxidant assays…….....93
(xiv) Total anthocyanin content…….....95

3.2.2 Experiment 2. Effects of 2, 4-Dichlorophenoxy acetic acid (2, 4-D) on growth and development of wax apple……95
(a) Plant materials…….....96
(b) Treatment application……96
(C) Measurements fruit growth parameters
(i) Number of bud, bud drop, fruit set, fruit drop, average fruit weight and peel color……...96
(ii) Chlorophyll content (SPAD value)……..96
(iii) Chlorophyll fluorescence……..96
(iv) Firmness of fruits……...97
(v) Stomatal conductance…….....97
(d) Measurements fruit biochemical parameters…….....97

3.2.3 Experiment 3. Effects of naphthalene acetic acid (NAA) on growth, development and quality of wax apple
(a) Plant material……..99
(b) Treatment application……..99
(c) Measurement of fruit growth and development characteristics……..99
(d) Determination of nutritional quality parameter……...99
(e) Phenylalanine ammonia lyase (PAL) enzyme assay……...99

3.2.4 Experiment 4. Effect of different type of girdling on growth, development and quality of wax apple
(a) Plant material……..100
(b) Treatment application……..101
(c) Measurements of physiological characteristics
i. Inflorescence development……..101
ii. Fruit retention (%)……...101
iii. Fruit growth and yield contributing characteristics……103
(iv) Fruit size……103
(d) Determination of nutritional quality parameters……...103

3.2.5 Experiment 5.The influence of hydrogen peroxide on growth, biochemical and phytochemical properties of wax apple

(a) Plant material……103
(b) Treatment application……104
(c) Fruit growth and developmental characteristics……...104

(d) Measurement of photosynthetical characters (net photosynthetic rate, transpiration and stomatal conductance)……..104
(e) Determination of fruit biochemical and phytochemical Characteristics……105
(f) Phenylalanine ammonia lyase (PAL) activity……105

3.2.6 Experiment 6. Effect of GA_3 on selected physiological process of wax apple (*Syzygium samarangense*)

(a) Plant material……...106
(b) Treatment application……...106
(c) Measurement of physiological parameter……....106
(i) Photosynthetical characteristics……...106
(ii) Sucrose phosphate synthase (SPS) activity……....106
(iii) Phenylalanine ammonia lyase (PAL) activity and anthocyanin content……107

3.3 Statistical analysis……...108

.

CHAPTER 4 RESULTS

4.1 Effects of gibberellic acid (GA_3) on growth, development and biochemical properties of wax apple fruits.

4.1.1 Number of buds and bud drop……109
4.1.2 Fruit set and fruit drop……109
4.1.3 Fruit growth (length and diameter)……....111
4.1.4 Gibberellic acid on color development..……....111
4.1.5 Yield and average fruit weight……...115
4.1.6 Gibberellic acid on leaf size, weight and quantum yield……....115
4.1.7 Chlorophyll content in leaf and fruit……117
4.1.8 Gibberellic acid on soluble leaf protein…….…....117
4.1.9 Fruit juice …….....117
4.1.10 pH of fruit juice……....119
4.1.11 K^+ content and total soluble solids (TSS)…….....119
4.1.12 Total soluble sugar……....119
4.1.13 Total flavonoids, total phenols, carotenoid and anthocyanin content……..121
4.1.14 Glucose, fructose and sucrose content……..122
4.1.15 Titrable acidity and TSS and Acidity ratio……...122
4.1.16 Ascorbic acid……124
4.1.17 Color versus TSS content of fruit and total phenolics versus antioxidant activity……....124

4.2 Effects of 2, 4-D on growth and development of wax apple fruits

4.2.1 Bud development, bud drop and fruit set……....126
4.2.2 Fruit drop and average fruit weight……126
4.2.3 Leaf dry matter and chlorophyll content……128
4.2.4 Fruit growth (length and diameter)……..128
4.2.5 Chlorophyll fluorescence and photosynthetic yield……131
4.2.6 Stomatal conductance and fruit yield……..131
4.2.7 Total soluble solids (TSS) and peel color development…..133
4.2.8 Color at 7^{th} week, pulp firmness and anthocyanin content……133
4.2.9 Total sugar content……..136

4.3 Effects of NAA on growth, development and quality of wax apple fruit.

4.3.1 Total number of buds, fruit set, and fruit drop…….137
4.3.2 Leaf chlorophyll content……137
4.3.3 Fruit growth (length and diameter)…… ……137
4.3.4 NAA on color development……141
4.3.5 Total number of fruits/branch……141
4.3.6 Chlorophyll fluorescence, photosynthetic yield, stomatal conductance and dry matter content……143
4.3.7 Correlation between stomatal conductance and yield…....145
4.3.8 TSS content and correlation between TSS and peel color in fruit…….....145
4.3.9 Total sugar content…….……147
4.3.10 K^+ content of fruit…….…..147
4.3.11 Total phenolic and flavonoid content in fruit…….……147
4.3.12 Antioxidant activity and correlation between total phenol and antioxidant activity…….….149
4.3.13 NAA on leaf soluble protein…….….149
4.3.14 Anthocyanin content in fruit……..149
4.3.15 NAA on PAL activity……151

4.4 Effect of girdling on growth, development and quality of wax apple

4.4.1 Bud number……....153
4.4.2 Inflorescence development and fruit retention……..153
4.4.3 Bud drop and fruit set percentage……..153
4.4.4 Fruit drop percentage……154
4.4.5 Fruit growth (length and diameter)……....154
4.4.6 Fruit size (length and diameter) and L/D ratio……...157
4.4.7 Fruit yield (kg) and dry matter……...157

4.4.8 Average fruit weight and number of fruits……...157
4.4.9 Peel color……....160
4.4.10 Effect on leaf chlorophyll content……163
4.4.11 Leaf size, chlorophyll and quantum yield……163
4.4.12 Fruit juice, fruit biomass and pH content…….164
4.4.13 Potassium and TSS content ……166
4.4.14 Total sugars content…….166
4.4.15 Total phenolics and flavonoids ……...168
4.4.16 Correlation between antioxidant activity and total flavonoid content ……168

4.5 Influence on hydrogen peroxide on growth, yield, biochemical and phytochemical properties of wax apple

4.5.1 Leaf drymatter and chlorophyll content.....171
4.5.2 Photosynthesis, stomatal conductance and leaf transpiration..... 171
4.5.3 Bud drop, fruit set and fruit drop percentage.....176
4.5.4 Fruit growth (length and diameter)..... 177
4.5.5 Color development……...177
4.5.6 Fruit yield (kg), drymatter content and total number of fruits.....181
4.5.7 TSS content of fruits.....183
4.5.8 Total sugar.....183
4.5.9 K^+ content.....183
4.5.10 Total phenolic and flavonoid content.....185
4.5.11 Antioxidant activity in the fruit.....185
4.5.12 Correlation between flavonoid content and antioxidant activity.....187
4.5.13 Carotenoid contents.....187
4.5.14 PAL activity and anthocyanin formation……..…187
4.5.15 Soluble sugars, fruit protein and endogenous hydrogen peroxide concentration.....189

4.6 Effect of GA_3 on selected physiological processes of wax apple

4.6.1 Gibberellic acid on PAL enzyme activity.……193
4.6. 2 Gibberellic acid on anthocyanin content……...195
4.6.3 Correlation between anthocyanin and PAL activity…….195
4.6.4 Gibberellic acid on photosynthetical characteristics..... 198
4.6.5 Gibberellic acid on sucrose phosphate synthase

(SPS) activity.....202

CHAPTER 5 DISCUSSION

5.1 Effects of Gibberellic acid ……204
5.2 Effects of 2, 4-Dichlorophenoxy acetic acid……..208
5.3 Effects of Naphthalene acetic acid ……....213
5.4 Girdling effects……...218
5.5 Hydrogen peroxide effects…….225
5.6 Gibberellic acid on plant physiology...…… ……..……230

CHAPTER 6 CONCLUSIONS…….....235

FUTURE STUDY…….....242

PUBLICATIONS…….....243

REFERENCES……245

LIST OF TABLES Page

Table 4.1 : Effects of different treatments of GA_3 on number of buds, bud drop, fruit set and pre harvest fruit dropping and yield of wax apple fruit……110

Table 4.2 : The effects of GA_3 on leaf size, weight and Chlorophyll fluorescence of wax apple…….....118

Table 4.3 : Effects of different treatments of GA_3 on leaf chlorophyll, fruit juice and pH of wax apple fruits……120

Table 4.4 : Effects of different treatments of GA_3 on TSS (% Brix), total sugar, flavonoids and total phenols, anthocyanin and carotenoids content in wax apple fruits…….120

Table 4.5 : Effects of different treatments of GA_3 on glucose, fructose, sucrose, TA, TSS/TA and Vit-C content of wax apple fruits…… …….123

Table 4.6 : Effects of different treatments of 2, 4-D on number of buds, bud drop, fruit set, pre harvest fruit drop and yield of wax apple……...127

Table 4.7 : Effects of different treatments of 2, 4-D on colour development, anthocyanin and total sugar content of wax apple fruit……...135

Table 4.8 : Effects of NAA treatment on bud number, fruit set, fruit drop, yield, dry matter and chlorophyll content of wax apple……..…138

Table 4.9 : Effects of different treatments of NAA on chlorophyll fluorescence, photosynthetic yield, stomatal conductance, leaf DM and TSS (°Brix) of wax apple……...144

Table 4.10 : Effects of different treatments of NAA on K^+, total phenol, total flavonoid, antioxidant activity and anthocyanin content of wax apple…..148

Table 4.11 : Effect of different types of girdling on bud number, bud drop, fruit set, fruit drop and yield per branch in wax apple trees…….155

Table 4.12 : Effects of different types of girdling on inflorescence development, fruit retention, fruit size, leaf and fruit drymatter of wax apple fruits……159

Table 4.13 : The effects of different types of girdling on leaf chlorophyll content and leaf size of wax apple……..165

Table 4.14 : Effects of different types of girdling on juice content, dry matter, K^+ and TSS (°Brix) content of wax apple……167

Table 4.15 : Effects of different types of girdling on total sugar, total phenol, total flavonoid and DPPH concentrations of wax apple fruits…….169

Table 4.16 : Effects of H_2O_2 treatment on leaf dry weight, chlorophyll content, bud drop, fruit set and fruit drop in *Syzygium samarangense* based on harvest values for all seasons.....172

Table 4.17 : Effects of H_2O_2 treatment on yield, fruit dry matter, K+ content and phenolic content in *Syzygium samarangense* based on harvest values for all seasons..... 182

Table 4.18 : Effects of H_2O_2 treatments on the number of fruits and their juice content in *Syzygium samarangense* for three different harvest seasons..... 184

Table 4.19 : Effects of H_2O_2 treatments on total soluble solids (TSS) and total sugar content in *Syzygium samarangense* for three different harvest seasons.....184

Table 4.20 : Effects of different H_2O_2treatments on K^+, total phenol, total flavonoid, TEAC and DPPH concentrations of wax apple at harvest for the three growing seasons.....186

Table 4.21 : Effects of H_2O_2 treatments on the number of fruits and their juice content in *Syzygium samarangense* for three different harvest seasons.....191

LIST OF FIGURES **Page**

Fig. 1.1 : Flow chart of research activity of this study from plant selection to data analysis and report writing…….....8

Fig. 2.1 : Leaf, flower bud, flower and different part of flower, floral diagram and fruit of wax apple (*Syzygium samarangense*) (Source: Nakasone and Paul, 1998)……..11

Fig. 2.2 : Blooming flower of wax apple (*Syzygium samarangense*)…….....12

Fig. 2.3 : Different stages of fruit growth and development of wax apple (*Syzygium samarangense*)…… …….....12

Fig. 2.4 : Location of wax apple cultivation native and exotic range…….....14

Fig. 2.5 : Ripening fruits of wax apple (*S. samarangense*)……...17

Fig. 2.6 : Peel color of wax apple (*Syzygium samarangense*)……...17

Fig. 2.7a : Pulp color of wax apple (*Syzygium samarangense*)……19

Fig. 2.7b : Fruit size and fruit diameter of distal and proximal end of *S. samarangense*……19

Fig. 2.8 : (A): Fruit, (B): Fresh biomass, (C): Juice and (D): Seed of wax apple…...23

Fig. 2.9 : Growth regulators gibberellic acid and auxin (NAA and 2, 4-D)…….....31

Fig. 2.10 : Growth promoting chemical hydrogen peroxide……..31

Fig. 2.11 : Outline of the phenylpropanoid biosynthetic pathway…….....55

Fig. 2. 12 : Diphenylpicrylhydrazine (free radical) and Diphenylpicrylhydrazine (non radical)……59

Fig. 2.13 : The neutralizing reactions free radical by ascorbic acid……..60

Fig. 2.14 : Oxidation of ABTS to $ABTS^{\bullet+}$ radical…….....63

.

Fig. 2.15 : Phenolic compounds…….....66

Fig. 2.16 : Flavonoids include: Flavone, Flavanone, Catechin, Anthocyanin, Flavanonol Flavonol……..70

Fig. 3.1 : Google map showing experimental location in Klang and Banting, Selangor Malaysia…….....82

Fig. 3.2 : Experimental field in (1) MARDI station, Jalan Kebun, Klang and (2a, b, c) Olak Lempit, Banting, Selangor……...83

Fig. 3.3 : Experimental design and treatment setting…….....84

Fig. 3.4 : Protected spray of growth regulators and H_2O_2……...98

Fig. 3.5 : Different girdling techniques were performed in this study. A: Control, B: I25%, C: 50 %, D: 100 %, E: C-shaped, and F: V- shaped girdling.....102

Fig. 4.1 : Fruit growth (length) as influenced by different treatments of GA_3 110

Fig. 4.2 : Fruit growth (diameter) as affected by different treatments of GA_3 112

Fig. 4.3 : Fruit growth of wax apple at one week after anthesis as affected by GA_3 treatments,(A) = control, (B) = 20 mg/L, (C) = 50 mg/L and (D) = 100 mg/L GA_3.....112

Fig. 4.4a : The effects of GA_3 treatments on peel color (%) of wax apple fruits after anthesis (n = 6).....113

Fig. 4.4b : Fruit size and color development at three week after anthesis. (A) = Untreated control, (B) = 20 mg/L, (C) = 50 mg/L and (D) = 100 mg/L GA_3 treatments.....113

Fig. 4.4c : Fruit color development at four week after anthesis. (A) = Untreated control, (B) = 20 mg/L, (C) = 50 mg/L and (D) = 100 mg/L GA_3 treatments.....114

Fig. 4.5a : Fruit size as affected by GA_3 treatments. (A) = Untreated control, (B) = 20 mg/L, (C) = 50 mg/L and (D) = 100 mg/L GA_3.....114

Fig. 4.5b : Fruit size as affected by GA_3 treatments. (A) = Untreated control, (B) = 20 mg/L, (C) = 50 mg/L and (D) = 100 mg/L GA_3.....116

Fig. 4.5c : Fruit size as affected by GA_3 treatments. (A) = Untreated control, (B) = 20 mg/L, (C) = 50 mg/L and (D) = 100 mg/L GA_3.....116

Fig. 4.6 : The effects of GA_3 treatments on soluble protein content (mg/g FW) of wax apple leaves.....118

Fig. 4.7 : Correlation between peel color and TSS content of wax apple fruits affected by 50 mg/L GA_3 treatment.....125

Fig. 4.8 : Regression analysis of antioxidant capacity (DPPH measurements) and total phenols of wax apple fruits..... ...125

Fig. 4.9 : Fruit growth (length) as influenced by different treatments of 2, 4-D (n=6).....129

Fig. 4.10 : Fruit growth (diameter) as affected by different treatments of 2, 4-D (n=6)..... 129

Fig. 4.11a : The effect of 2, 4-D treatments on fruit growth and color development of wax apple. (A) = control, (B) = 5mg/L, (C) = 10 mg/L and (D) = 20 mg/L 2, 4- D.....130

Fig. 4.11b : The effect of 2, 4-D treatments on fruit size of wax apple. (A) = control, (B) = 5mg/L, (C) = 10 mg/L and (D) = 20 mg/L 2, 4-D.....130

Fig. 4.11c : The effect of 2, 4-D treatments on fruit size of wax apple. (A) = control, (B) = 5mg/L, (C) = 10 mg/L and (D) = 20 mg/L 2, 4-D.....132

Fig. 4.12 : Chlorophyll fluorescence intensity as affected by 2, 4-D treatments. F_0 = Lower fluorescence, Fm= Higher fluorescence and Fv= Relative variable fluorescence.....132

Fig. 4.13 : Photosynthetic or quantum yield (Fv/Fm) as affected by treatments of 2, 4- D.....134

Fig. 4.14 : Stomatal conductance (mmol m^{-2} s^{-1}) and yield (Kg) as affected by 2, 4-D treatments.....134

Fig. 4.15 : TSS (°Brix) and peel color (%) as affected by 2, 4-D treatments in wax apple fruits.....135

Fig. 4. 16 : The effect of different treatments of NAA on fruit growth (length) of wax apple..... 138

Fig. 4.17 : Fruit size at two week after anthesis affected by NAA treatments. (A) = control, (B) = 5 mg/L, (C) = 10 mg/L and (D) = 20 mg/L NAA.....139

Fig.4.18 : Final fruit diameter (cm) and fruit growth at 2^{nd} week as affected by different treatments of NAA.....139

Fig. 4.19a : Fruit size affected by NAA treatments. (A) = control, (B) = 5 mg/L, (C) = 10 mg/L and (D) = 20 mg/L NAA.....140

Fig. 4.19b : Fruit size affected by NAA treatments. (A) = control, (B) = 5 mg/L, (C) = 10 mg/L and (D) = 20 mg/L NAA.....140

Fig. 4.20 : The effects of different treatments of NAA on color-cover (%) of wax apple after anthesis.....142

Fig. 4.21 : Fruit color at four week after anthesis**.** (A) = control, (B) = 5 mg/L, (C) = 10 mg/L and (D) = 20 mg/L NAA.....142

Fig. 4.22 : The effects of NAA treatments on total number of fruits of wax apple throughout the three growing season.....144

Fig. 4.23 : Correlation between stomatal conductance and yield of wax apple fruits affected by NAA treatments.....146

Fig. 4.24 : Correlation between peel color (%) and TSS (°Brix) of 10 mg/L NAA treated wax apple fruits.....146

Fig. 4.25 : Fluctuation of total sugar content during the three growing seasons in wax apple fruit treated with NAA.....148

Fig. 4.26 : Correlation between total phenols and antioxidant capacity via TEAC methods in 10 mg/L NAA treated wax apple fruits.....150

Fig. 4.27 : The effects of different treatments of NAA on soluble protein content (mg/g FW) in wax apple leaves.....150

Fig. 4.28 : The effects of NAA treatments on anthocyanin content (mg/L) in wax apple fruit.....152

Fig. 4.29 : PAL activity (nmol- cinnamic acid min^{-1} mg $protein^{-1}$) at 15 min of observation as affected by different treatment of NAA in wax apple fruit.....152

Fig. 4.30: Fruit growth (length) as affected by different girdling treatments.....156

Fig. 4.31 : Fruit growth and color development as affected by different types of girdling. A= control, B= I-shaped, C= 50%, D= 100%, E = C-shaped and F= shaped girdling.....156

Fig. 4.32 : Fruit growth (diameter) as affected by different girdling treatments.....158

Fig. 4.33 a : Fruit size as affected by different girdling treatments**.** A= control, B= I-shaped, C= 50%, D= 100%, E = C-shaped and F= shaped girdling.....158

Fig. 4.33 b : Fruit size and color as affected by different girdling treatments**.** A= control, B= I-shaped, C= 50%, D= 100%, E = C-shaped and F = shaped girdling.....159

Fig. 4.34 : Mean fruit weight (g) as affected by different girdling techniques. Bars indicate ± S.E. (n=6).....161

Fig. 4.35 : Total number of fruits as affected by different types of girdling techniques.....161

Fig. 4.36 : The effect of different treatments of girdling on peel color (%) development of wax apple after anthesis.....162

Fig. 4.37 : SPAD value readings of leaves as affected by different types of girdling practices (n = 12).....162

Fig. 4.38 : Quantum yield (Fv/Fm) as affected by different treatments of girdling.....165

Fig. 4.39 : Correlation between total flavonoids and antioxidant activity via TEAC methods in the C-shaped girdled fruit.....169

Fig. 4.40 : Effects of hydrogen peroxide treatments on net photosynthesis of wax apple trees under field condition.....174

Fig. 4.41 : Effects of hydrogen peroxide treatments on stomatal conductance of wax apple leaves under field condition.....174

Fig. 4.42 : Effects of hydrogen peroxide treatments on transpiration of wax apple leaves under field condition.....175

Fig. 4.43 : Relationship between stomatal conductance and transpiration of 20 mM H_2O_2 treated wax apple trees.....175

Fig. 4.44 : Fruit growth (length) as affected by different treatments of H_2O_2.....178

Fig. 4.45 : Fruit growth (diameter) as affected by different treatments of H_2O_2.....178

Fig. 4.46 : Fruit growth and color development at 4^{th} week after anthesis. A= control, B= 5 mM H_2O_2, C= 20 mM H_2O_2 and D= 50 mM H_2O_2.....179

Fig. 4.47 : The effect of different treatments of H_2O_2 on peel color (%) development of wax apple fruit after anthesis.....179

Fig. 4.48 : Fruit size as affected by H_2O_2 treatments. A= control, B= 5 mM H_2O_2, C= 20 mM H_2O_2 and D= 50 mM H_2O_2.....180

Fig. 4.49 : Correlation between total flavonoids and antioxidant activity via DPPH methods in the 20 mM H_2O_2 treated wax apple fruits.....188

Fig. 4.50 : Correlation between total phenols and antioxidant activity via TEAC methods in the 20 mM H_2O_2 treated wax apple fruits..... .188

Fig. 4.51 : PAL activity (nmol- cinnamic acid min^{-1} mg $protein^{-1}$) in wax apple fruit as affected by different treatment of H_2O_2.....190

Fig. 4.52a : Fruit size of wax apple as affected by GA_3, NAA, 2, 4-D and H_2O_2 treatments.....190

Fig. 4.52b : Fruit quality as affected by GA_3, NAA, 2, 4-D, girdling and H_2O_2 treatments. A= control, B= 50 mg/L GA_3, C= 10 mg/L NAA, D= 10 mg/L 2, 4-D, E= C-shaped girdling, and F= 20 mM H_2O_2.....191

Fig. 4.53 : PAL enzyme activity (nmol- cinnamic acid min^{-1} mg $protein^{-1}$) at 1 min of observation with GA_3 treatments..... ...194

Fig. 4.54 : PAL enzyme activity (nmol- cinnamic acid min^{-1} mg $protein^{-1}$) as affected by different treatment of GA_3 in wax apple fruit..... 194

Fig. 4.55 : PAL enzyme activity (nmol- cinnamic acid min^{-1} mg $protein^{-1}$)

at different maturity stages of GA_3- treated wax apple fruit.....196

Fig. 4.56 : PAL enzyme activity (nmol- cinnamic acid min^{-1} mg $protein^{-1}$) as affected by different maturity stages of wax apple fruits.....196

Fig. 4.57 : The effect of different treatment of GA_3 on anthocyanin content (mg/L) in wax apple fruit.....197

Fig. 4.58 : The effect of stages of fruit development on anthocyanin content (mg/L) of wax apple fruit.....197

Fig. 4.59 : Correlation between PAL activity and anthocyanin content in 50 mg/L GA_3 treated wax apple fruit.....199

Fig. 4.60 : Effects of GA_3 treatments on net photosynthesis of wax apple trees under field condition..... ...199

Fig. 4.61 : Effects of GA_3 treatments on stomatal conductance of wax apple leaves under field condition.....200

Fig. 4.62 : Effects of GA_3 treatments on transpiration of wax apple leaves under field condition.....200

Fig. 4.63 : Relationship between stomatal conductance and photosynthesis of 50 mg/L GA_3 treated wax apple trees.....201

Fig. 4.64 : Relationship between transpiration and photosynthesis of 50 mg/L GA_3 treated wax apple trees.....203

Fig. 4.65 : Effects of GA_3 treatment on SPS activity of wax apple leaves.....203

Fig. 6.1 : Interaction among the parameters studied.....240

ABBREVIATIONS

2, 4-D	2, 4- Dichlorophenoxy acetic acid
ABTS	2, 2 -azino-di-[3-ethylbenzthiazoline sulphonate]
AEAC	ascorbic acid equivalent to antioxidant capacity
ANOVA	analysis of variance
ATP	adenosine triphosphate
C/N	carbon nitrogen ratio
CE	catechin equivalents
DM	dry matter
DMRT	Duncan's Multiple Range Test
DNA	deoxyribonucleic acid
DPPH	1, 1-diphenyl-2-picrylhydrazyl hydrazyl
EDTA	ethylene di-amine tetra acetic acid
g	gram
GA_3	gibberellic acid
GAE	gallic acid equivalents
H_2O_2	hydrogen peroxide
K^+	potassium
KOH	potassium hydroxide
L	liter
LSD	least significant difference
MARDI	Malaysian Agricultural Research and Development Institute
mg	milligram
ml	milliliter
mM	milli mole
MOPS	morpholine propane sulfonic acid

ABBREVIATIONS (CONT.)

MSTAT-C	Michigan State University Statistical Program-C
NAA	naphthalene acetic acid
OR	over ripe
PAL	phenylalanine ammonia lyase
PAR	photosynthetic active radiation
PGRs	plant growth regulators
ppm	parts per million
PSII	photosystem II
rbcA	rubisco 1, 6 bi phosphate
RCBD	randomized complete block design
ROS	reactive oxygen species
rpm	revolutions per minute
RubisCoase	ribulose 1, 5-bisphosphate carboxylase/oxygenase
SPAD	soil plant analysis development
SPS	sucrose phosphate synthase
TA	titrable acidity
TAC	total antioxidant capacity
TAL	tyrosine ammonia lyase
TEAC	trolox equivalent to antioxidant capacity
TFC	total flavonoid content
TPC	total phenolic content
TSS	total Soluble Solids
UDP	uridine diphosphate
µg	microgram

APPENDICES

Appendix 1 Physico-chemical characteristics of soils of two orchards located at MARDI, Jalan Kebun, Klang and Banting, Selangor

Experimental field	pH value	Moister (%)	Carbon (%)	Organic matter (%)	N (%)	Clay (%)	Silt (%)	Sand (%)	CEC
MARDI	4.8	80	34.2	59.3	3.1	44.6	11.7	43.7	145.0
Banting	4.63	195%	-	70	4.67	38	27	35	--

Source: Ismail *et al.* (1994)

Appendix 2 Climatology information of two orchards located at MARDI, Jalan Kebun, Klang and Banting, Selangor (2008-2011)

Average	Temperature (°C)			Rainfall (mm)	Relative humidity (%)
	Min	Max	Average		
Monthly	23	37	33	225	84%

Source: Malaysian Metrological Department

Appendix 3 Food value of *S. samarangense* (wax apple)

Food value per 100 g of edible portion

Moisture	91.40-92.96 g
Protein	0.50 g
Sugar	6.56 g
Iron	0.001 g
Ash	0.21-0.27 g
Calcium	0.01 g
Phosphorus	0.03 g
Sulphuric Acid	0.17%
Citric Acid	0.15%

Source: Morton (1987)

Appendix 4 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by plant growth regulator treatments

Variation sources	df	Mean square (MS)						
		Bud number	Bud drop	Fruit set	Fruit drop	Yield	Fruit weight	Biomass
Rep	5	147.14**	12.00^{ns}	17.83^{ns}	15.40^{ns}	0.002^{ns}	48.19^{ns}	2.90**
GA_3	3	67.04^{ns}	59.11^{ns}	1905.36**	337.5**	0.458**	537.43**	60.50**
Error	15	41.80	29.24	40.48	20.86	0.008	68.13	0.396
Rep	5	119.60**	22.87^{ns}	11.58^{ns}	80.04^{ns}	0.026^{ns}	14.61^{ns}	0.093^{ns}
NAA	3	129.50**	554.20**	393.26**	41.37^{ns}	0.045*	117.91**	3.77**
Error	15	9.60	36.02	21.10	52.84	0.012	10.01	0.30
Rep	5	76.00*	70.98*	61.46^{ns}	6.74^{ns}	0.019^{ns}	18.64**	0.047^{ns}
2, 4-D	3	18.00^{ns}	216.17**	50.67^{ns}	553.04**	0.024*	19.18**	0.283**
Error	15	17.47	19.63	29.60	49.67	0.007	2.95	0.038

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 5 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by girdling and H_2O_2 treatments

Variation sources	df	Mean square (MS)						
		Bud number	Bud drop	Fruit set	Fruit drop	Yield	Fruit wt	Fruit DM
Rep	5	49.69^{ns}	6.06^{ns}	7.71^{ns}	51.77^{ns}	0.020**	11.54^{ns}	0.084
Girdling	5	15.76^{ns}	494.06**	568.97**	83.64^{ns}	0.174**	21.21^{ns}	0.635*
Error	25	37.93	11.61	58.64	45.19	0.002	13.59	0.24
Rep	5	27.28^{ns}	19.26^{ns}	51.84^{ns}	49.90^{ns}	0.003^{ns}	52.14^{ns}	0.003^{ns}
H_2O_2	3	88.38*	66.56^{ns}	139.71^{ns}	198.00**	0.094**	207.56**	0.782**
Error	15	22.74	39.42	56.04	20.16	0.006	25.87	0.018

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 6 Analysis of variance (ANOVA) for fruit growth, color development and chlorophyll content of wax apple affected by GA_3 treatments

Variation sources	df	Mean square (MS)						
		Length 3rd	Length 7th	Diameter 3rd	Diameter 7th	Color dev.	Chlo a	Chlo b
Rep	5	0.048^{ns}	0.041^{ns}	0.119^{ns}	0.094^{ns}	60.40^{ns}	0.65^{ns}	0.214^{ns}
GA_3	3	8.49**	4.58**	2.89**	0.362**	5516.89**	0.827^{ns}	0.385^{ns}
Error	15	0.06	0.095	0.043	0.015	26.57	0.364	0.193

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 7 Analysis of variance (ANOVA) for fruit growth, color development and pulp firmness of wax apple affected by 2, 4-D treatments

Variation sources	df	Mean square (MS)						
		Length 3rd	Length 7th	Diameter 3rd	Diameter 7th	Color	SPAD	P. firmness
Rep	5	0.120	0.10*	0.078^{ns}	0.035^{ns}	5.60^{ns}	36.76^{ns}	0.039^{ns}
2, 4-D	3	0.83**	0.49**	0.698**	0.580**	932.6**	390.8**	0.347**
Error	15	0.12	0.012	0.039	0.053	26.71	11.56	0.034

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 8 Analysis of variance (ANOVA) for fruit growth, color development and SPAD value of wax apple affected by NAA treatments

Variation sources	df	Mean square (MS)						
		Length 3rd	Length 7th	Diameter 3rd	Diameter 7th	Color 21	Color 27	SPAD
Rep	5	0.284**	0.059ns	0.053ns	0.21ns	2.96ns	20.96	17.18ns
NAA	3	10.02**	2.624**	0.828**	1.236**	314.2**	809.05	330.76**
Error	15	0.029	0.051	0.021	0.109	17.45	20.25	29.01

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 9 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by girdling and H_2O_2 treatments

Variation sources	df	Mean square (MS)						
		Length 3rd	Length 7th	Diameter 3rd	Diameter 7th	Color dev.	Chlo	Leaf DM
Rep	5	0.152^{ns}	0.072^{ns}	0.129 ^{ns}	0.117^{ns}	13.44^{ns}	24.27^{ns}	0.029^{ns}
Girdling	5	2.87**	1.878**	1.262**	0.671**	2573.78**	351.83**	0.591**
Error	25	0.182	0.059	0.039	0.015	10.67	23.02	0.086
Rep	5	0.019^{ns}	0.057^{ns}	0.027^{ns}	0.030^{ns}	20.20 ^{ns}	50.75^{ns}	0.006^{ns}
H_2O_2	3	2.371**	3.364**	0.493**	0.788**	895**	230.79**	0.069*
Error	15	0.034	0.046	0.029	0.022	10.00	16.62	0.015

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 10 Analysis of variance (ANOVA) for biochemical properties of wax apple affected by plant growth regulator treatments

Variation sources	df	Mean square (MS)						
		K^+	TSS	Total sugar	Flavonoid	Phenol	Anthocyanin	Carotenoid
Rep	5	1.27^{ns}	0.39^{ns}	0.063^{ns}	4.54^{ns}	11177^{ns}	0.9973^{ns}	0.091^{ns}
GA_3	3	256.22*	46.02**	13.87**	312.67**	198959**	11.09**	35.295**
Error	15	66.82	1.58	0.234	4.47	7243	0.07	0.356
Rep	5	190.5^{ns}	0.164^{ns}	0.71^{ns}	3.38^{ns}	932.62^{ns}	0.085^{ns}	--
NAA	3	5210.83**	29. 65**	14.92**	355.74**	150394**	12.38**	--
Error	15	60.23	0.58	0.38	5.06	3854.4	0.244	--
Rep	5	6.16^{ns}	0.97^{ns}	0.113^{ns}	22.23^{ns}	436.72^{ns}	0.044^{ns}	--
2, 4-D	3	864.68**	23.97**	9.42**	272.86**	135452**	2.009**	--
Error	15	76.69	1.98	0.37	6.81	352.6	0.006	--

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 11 Analysis of variance (ANOVA) for biochemical properties of wax apple affected by girdling treatments

Variation sources	df	Mean square (MS)						
		F. biomass	K^+	TSS	Total sugar	Flavonoid	Phenol	Anthocyanin
Rep	5	0.97^{ns}	42.378^{ns}	0.315^{ns}	0.184^{ns}	67.91^{ns}	75953^{**}	0.31^{*}
Girdling	5	63.85^{**}	56.97^{**}	42.32^{**}	9.284^{**}	746.06^{**}	33784^{*}	5.923^{**}
Error	25	0.528	79.07	1.21	0.477	39.03	10816	0.092

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 12 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by H_2O_2 application

Variation sources	df	Number of fruits (3 yrs) Mean square (MS)			Fruit Juice (3 yrs)		
		1^{st}	2^{nd}	3^{rd}	1^{st}	2^{nd}	3^{rd}
Rep	5	2.60^{ns}	1.00^{ns}	2.60^{ns}	56.80^{ns}	51.70^{ns}	7.10^{ns}
H_2O_2	3	60.00^{**}	22.00^{**}	29.50^{**}	217.50^{**}	325.50^{**}	244.00^{**}
Error	15	3.40	3.00	3.26	32.66	18.63	14.43

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 13 Analysis of variance (ANOVA) for biochemical characteristics of wax apple affected by H_2O_2 application

Variation sources	df	Mean square (MS)						
		K^+	TSS	Total sugar	Flavonoid	Phenol	Anthocyanin	Carotenoid
Rep	5	52.80^{ns}	0.031^{ns}	0.237^{ns}	2.65^{ns}	11608^{ns}	0.75^{ns}	0.27^{ns}
H_2O_2	3	309.53^{*}	23.68^{**}	5.43^{**}	68.84^{**}	250568^{**}	10.88^{**}	20.96^{**}
Error	15	49.80	0.519	0.28	11.09	14994	0.48	0.102

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 14 Analysis of variance (ANOVA) for biochemical properties of wax apple affected by GA_3 application

Variation sources	df	Mean square (MS)						
		Total chlorophyll	Fruit chlorophyll	Fruit juice	pH value	DPPH (mg/100g)	TEAC (mg/100g)	TSS/Acidity ratio
Rep	5	0.938ns	0.008ns	147.77**	0.217ns	1.483ns	0.045ns	0.699ns
GA_3	3	1.893^{*}	0.199^{**}	161.37^{**}	0.143^{ns}	10.79^{**}	0.105^{*}	76.096
Error	15	0.39	0.005	32.44	0.072	1.19	0.032	0.274

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 15 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by NAA

Variation sources	df	Mean square (MS)							
		Fruit number			Chlorophyll fluorescence			Quantum yield	Stomatal conductance
		1^{st}	2^{nd}	3^{rd}	F0	Fm	Fv		
Rep	5	6.46^{**}	7.26^{ns}	10.44^{ns}	1085^{ns}	32223^{ns}	16714^{ns}	0.001^{ns}	20826^{**}
NAA	3	25.5^{**}	52.17^{**}	17.37^{**}	46824^{**}	5732125^{**}	5061301^{**}	0.054^{**}	147850^{**}
Error	15	1.09	5.80	0.84	3298.26	136919	91148	0.003	2861

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 16 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by girdling treatments

Variation sources	df	Mean square (MS)							
		Fruit number			Chlorophyll fluorescence			Quantum yield	TEAC assay
		1st	2nd	3rd	F0	Fm	Fv		
Rep	5	4.21*	8.82ns	0.96ns	6502ns	23087ns	38579n	0.005*	0.041ns
Gird.	5	37.89**	69.86**	42.58**	90934**	6662630**	5388686 **	0.031**	0.274**
Error	25	1.56	4.12	3.12	2902	37534	24073	0.001	0.012

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 17 Analysis of variance (ANOVA) for biochemical characteristics of wax apple affected by GA_3 treatments

Variation sources	df	Mean square (MS)					
		Glucose	Fructose	Sucrose	Titrable acidity	Vitamin-C	PAL (stage)
Rep	5	0.511ns	0.062ns	0.03ns	0.007**	0.078ns	535.82**
GA_3	3	7.99**	8.39**	4.39*	0.008**	2.27**	7793.38**
Error	15	0.33	0.089	0.04	0.001	0.146	51.09

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 18 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by girdling treatments

Variation sources	df	Mean square (MS)							
		Inflo. dev.	Fruit Ret.	Fruit length	Fruit diam.	L/D ratio	Mean fruit weight 1st season	2nd season	3rd season
Rep	5	4.84ns	15.27ns	0.972ns	0.087ns	0.004 ns	1.329ns	5.98ns	28.56**
Girdling	5	98.24**	224.2**	1.509*	0.568*	0.006*	210.02**	226.64**	185.63**
Error	25	6.52	11.90	0.45	0.24	0.003	10.85	9.71	7.66

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 19 Analysis of variance (ANOVA) for physicochemical properties of wax apple affected by girdling treatment

Variation sources	df	Mean square (MS)						
		Leaf chlorophyll			Fruit	Total sugar		
		a	*b*	*a+b*	juice	1st season	2nd season	3rd season
Rep	5	0.32^{ns}	0.323^{ns}	0.336^{ns}	79.73^{**}	1.33^{ns}	0.19^{ns}	0.495^{ns}
Girdling	5	2.052^{*}	1.047^{*}	6.32^{**}	97.80^{**}	4.65^{**}	9.34^{**}	7.00^{**}
Error	25	0.436	0.159	0.156	4.61	0.45	0.55	0.43

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 20 Analysis of variance (ANOVA) for leaf protein and PAL activity of wax apple fruits affected by GA_3 treatments

Variation sources	df	Mean square (MS)						
		Protein (leaf)	PAL (fruit)	PAL 0 min	PAL 30 min	PAL 45 min	PAL fruit stage 30 min	PAL fruit stage 45 min
Rep	5	0.099^{ns}	0.082^{ns}	1.790^{ns}	1.064^{ns}	1.319^{ns}	15.92^{ns}	2.17^{ns}
GA_3	3	0.674^{**}	0.516^{**}	65.39^{**}	327.20^{**}	392.58^{**}	105.59^{**}	63.69^{**}
Error	15	0.075	0.073	0.70	5.19	5.23	1.30	0.547

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 21 Analysis of variance (ANOVA) for physicochemical characteristics of wax apple affected by NAA and GA_3 treatments

Variation sources	df	NAA	Mean square (MS)			GA_3		
		Protein (leaf)	PAL (fruit)	PAL 0 min	PAL 30 min	Leaf length	Leaf weight	Quantum yield
Rep	5	0.112^{ns}	0.074^{ns}	0.315^{ns}	0.215^{ns}	1.47^{ns}	0.028^{ns}	0.004^{**}
NAA	3	1.90^{**}	0.540^{**}	40.52^{**}	347.23^{**}	10.14^{**}	1.423^{**}	0.004^{**}
Error	15	0.107	0.037	1.37	2.73	0.21	0.022	0.001

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 22 Analysis of variance (ANOVA) for physicochemical characteristics of wax apple affected by H_2O_2 application

Variation sources	df	Mean square (MS)						
		Fruit protein	Glucose (%)	Fructose (%)	Sucrose (%)	H_2O_2 PAL	Photosynthesis 800 PAR	2000 PAR
Rep	5	0.039^{ns}	0.702^{ns}	0.156^{ns}	0.043^{ns}	1.198^{ns}	4.201^{ns}	24.38^{ns}
H_2O_2	3	0.686^{**}	5.45^{**}	5.40^{**}	4.02^{**}	363.45^{**}	1164.05^{**}	854.58^{**}
Error	15	0.032	0.67	0.08	0.026	2.71	4.22	8.33

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 23 Analysis of Variance (ANOVA) for physiological characteristics of wax apple affected by H_2O_2 and GA_3 application

Variation sources	df	Mean square (MS)					
		Stomatal conductance		Transpiration		Endogenous	SPS activity
		800 PAR	2000 PAR	800 PAR	2000 PAR	H_2O_2	GA_3
Rep	5	0.000^{ns}	0.000^{ns}	0.083^{**}	0.003^{ns}	0.001^{ns}	1.87^{ns}
H_2O_2	3	0.004^{**}	0.003^{**}	0.283^{**}	5.613^{**}	0.343^{**}	113.20^{**}
Error	15	0.0001	0.0001	0.002	0.022	0.004	1.63

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 24 Analysis of variance (ANOVA) for physiological characteristics of wax apple affected by GA_3 treatment

Variation sources	df	Mean square (MS) Photosynthesis 400 PAR	Photosynthesis 2000 PAR	Stomatal conductance 400 PAR	Stomatal conductance 800PAR	Transpiration 400 PAR	Transpiration 2000PAR	Fruit protein
Rep	5	17.71^{*}	9.64^{ns}	0.000^{ns}	0.000^{ns}	0.001^{ns}	0.011^{ns}	0.082^{ns}
GA_3	3	1945.9^{**}	1555.93^{**}	0.003^{**}	0.001^{**}	0.203^{**}	0.912^{**}	0.516^{**}
Error	15	4.31	20.29	0.000	0.000	0.002	0.016	0.073

*significant different at $p<0.05$ ** significant difference at $p<0.01$ *ns:* no significant
PAR: Photosynthetic Active Radiation

Appendix 25 Analysis of variance (ANOVA) for biochemical characteristics of wax apple affected by H_2O_2 application

Variation sources	df	1st season Mean square (MS) leaf DM	SPAD	Bud drop	Fruit set	Fruit drop	Yield	Dry matter
Rep	5	0.011^{ns}	28.70^{*}	39.50^{**}	9.24^{**}	27.70^{*}	0.007^{*}	0.089^{ns}
H_2O_2	3	0.139^{**}	188.0^{**}	157.5^{**}	37.04^{**}	185.5^{**}	0.021^{**}	0.758^{**}
Error	15	0.009	8.033	2.70	0.442	6.5	0.001	0. 074
		3rd season						
Rep	5	0.019^{**}	12.80^{ns}	66.40^{**}	7.90^{ns}	17.30^{ns}	0.005^{*}	0.003^{ns}
H_2O_2	3	0.204^{**}	292.0^{**}	409.5^{**}	193.5^{**}	92.00^{**}	0.015^{**}	0.368^{**}
Error	15	0.001	17.86	10.00	8.83	7.57	0.001	0.002

*significant different at $p<0.05$ ** significant difference at $p<0.01$ *ns:* no significant

Appendix 26 Analysis of variance (ANOVA) for biochemical characteristics of wax apple affected by H_2O_2 application

Variation sources	df	Mean square (MS) TSS	K^+	Phenol	Flavonoid	TEAC	DPPH	Anthocyanin
1st season								
Rep	5	0.418^{*}	11.80^{ns}	275.10^{ns}	2.068^{ns}	0.003^{ns}	1.000^{*}	0.358^{*}
H_2O_2	3	4.24^{**}	140.0^{**}	25612^{**}	153.5^{**}	0.032^{ns}	5.560^{**}	2.775^{**}
Error	15	0.087	7.133	494.03	3.12	0.011	0. 692	0.079
3rd season								
Rep	5	0.283^{*}	13.80^{ns}	192.66^{ns}	15.25^{**}	0.056^{ns}	0.149^{ns}	0.313^{ns}
H_2O_2	3	18.29^{**}	138.0^{**}	17717^{**}	126.26^{**}	0.383^{*}	3.80^{**}	3.415^{**}
Error	15	0.067	11.26	245.3	2.30	0.074	0.489	0.110

*significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant

Appendix 27 Analysis of Variance (ANOVA) for physiological characteristics of wax apple affected by H_2O_2 and GA_3 application

Variation sources	df	Mean square (MS) Carotenoid^{1S}	Carotenoid ^{3S}	T. Sugar^{1S}	T. sugar^{3S}	SPS activity
Rep	5	0.225^{ns}	0.703^{**}	0.084^{ns}	0.102^{ns}	0.307^{ns}
H_2O_2	3	6.423^{**}	6.455^{**}	7.462^{**}	4.428^{**}	68.40^{**}
Error	15	0.466	0.367	0.038	0.494	0.634

S= season, *significant different at *p<0.05* ** significant difference at *p<0.01* *ns:* no significant