

AN ABSTRACT OF THE THESIS OF

Peeradet Tongumpai for the degree of Doctor of Philosophy
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(Coffea arabica L.) Fruits.

Abstract approved:


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Greenhouse and field grown coffee plants were used to study the synchronization of flowering and fruiting. The purpose of these studies was to develop methods of improving the selective harvesting of mature coffee fruits by machine. The studies were divided into 4 parts: 1) Synchronization of flowering, 2) Synchronization of fruiting, 3) Determination of fruit detachment and fruit removal force (FRF), and 4) Alteration of the FRF of ripe fruits.

Either mist irrigation or water deficit stress followed by sufficient watering were effective in inducing flowering. Gibberellic acid treatment was less effective.

Gibberellic acid applied to field grown coffee plants with fruit at different ages up to 40 days after anthesis advanced fruit maturity but did not synchronize fruit maturation. Ethephon sprayed on fruits at 230 days after

anthesis resulted in uniform ripening of fruit on greenhouse grown coffee plants.

The mechanism of abscission of green and ripe fruits was studied. No abscission layer was found at the sites of detachment in either green or ripe fruits. The detachment of ripe fruits occurred at random, by the breaking of parenchymatous pericarp tissue immediately above the pedicel. In contrast, the detachment of green fruits occurred at random along the pedicel.

The FRF of green fruits was greater than the FRF of ripe fruits. The FRF decreased as fruits matured in both the ethephon and control treatments.

Indices of fruit maturation were correlated with FRF. Exocarp color change, expressed quantitatively as the degree of lightness (L) and hue angle (θ), was correlated with the FRF. The soluble solids content of the pericarp tissue decreased as fruit matured. There was a strong inverse correlation between FRF and soluble solids content of the pericarp. Further reduction in the FRF of ripe fruits was achieved by mist irrigation. The FRF of green fruits was not affected by the mist irrigation treatment.

These studies provide strong evidence for the selective harvesting of mature coffee fruits by machine by integration of synchronized flowering and fruiting, and the reduction in FRF of ripe fruits.

Strategies for Machine Harvesting of Mature Coffee
(Coffea arabica L.) Fruits

by

Peeradet Tongumpai

A THESIS

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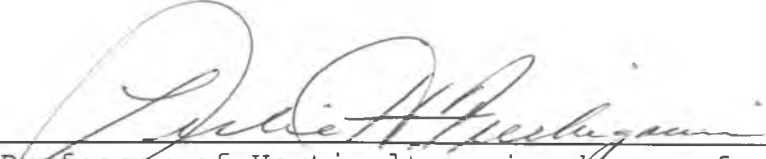
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Chairman of Department of Horticulture

Dean of Graduate School

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Table of Contents

Chapter 1. Introduction.....	1
Chapter 2. Review of Literatures.....	8
Flowering.....	8
Fruit Development.....	12
Machine Harvesting.....	21
Chapter 3. Effect of Mist Irrigation and Desiccation on Anthesis of Coffee Flower Bud.....	23
Abstract.....	23
Introduction.....	25
Materials and Methods.....	28
Results.....	31
Discussion.....	35
References.....	44
Chapter 4. The Effect of Gibberellic Acid on Fruit Growth and Maturation of Coffee.....	47
References.....	55
Chapter 5. Anatomical Study on the Fruit Separation of Unripe and Ripe Coffee Fruits.....	56
Abstract.....	56
Introduction.....	57
Materials and Methods.....	60
Results.....	61
Discussion.....	63
References.....	72
Chapter 6. Effect of Ethephon on Fruit Color Change and Fruit Removal Force.....	74
References.....	83

Table of Contents, Continued

Chapter 7. Harvest Indices for Machine Harvesting of Coffee Fruits.....	85
Abstract.....	85
Introduction.....	86
Materials and Methods.....	89
Results.....	93
Discussion.....	97
References.....	112
 Chapter 8. Osmotic Movement of Water into Pericarp Tissue of Coffee Fruits and Fruit Removal Force.....	115
Abstract.....	115
Introduction.....	116
Materials and Methods.....	120
Results.....	125
Discussion.....	131
References.....	145
 Concluding Remarks.....	147
 Bibliography.....	149
 Appendices	
A. Correlation between Soluble Solids Content of Pericarp and FRF of Ripe Coffee Fruits	160
B. Correlation between Leaf Water Potential and FRF of Green and Ripe Coffee Fruits.....	162
C. Effect of NAA on Fruit Removal Force of Coffee.....	164
D. Effect of Shoot Tip Removal on Flowering of Coffee (<u>Coffea arabica</u> L. 'Guatemalan').....	166

List of Figures

3.1	Flower water content as affected by irrigation, desiccation, and GA treatment.....	41
3.2	Length of coffee flowers after drip irrigation, mist irrigation, desiccation and GA treatments.....	42
3.3	Weight change of coffee flower after drip irrigation, mist irrigation, desiccation and GA treatments.....	42
3.4	Change in osmotic potential of coffee flowers after drip irrigation, mist irrigation, desiccation and GA treatments.....	43
4.1	Coffee fruit diameter following gibberellic acid application after anthesis.....	53
4.2	Percent ripe fruits of coffee harvested after 100 ppm GA application.....	54
5.1	Diagram of fruit detachment sites.....	66
5.2	Pericarp tissues of ripe (top) and green (bottom) fruits adjacent to pedicel (magnification 40 X).....	67
5.3	The separation of pericarp from the pedicel of ripe fruit.....	68
5.4	Cross-section of pedicel tissue of ripe (top) and green (bottom) fruits (magnification 40 X)...	69
5.5	Pedicel tissues of green fruit between the pedicel and peduncle (magnification 40 X).....	70
5.6	Part of peduncle and pedicel (top) tissues separated from the pedicel tissues (bottom) following physical detachment of green fruits (magnification 40 X).....	71

List of Figures, Continued

6.1	Effect of 200 ppm ethephon on fruit color change as determined by the change of hue angle (θ), where $\theta = \tan^{-1} b/a$	81
6.2	Effect of 200 ppm ethephon on FRF at different fruit age.....	81
6.3	Effect of 200 ppm ethephon on the soluble solids content of the pericarp at different fruit age.....	82
6.4	Seed dry weight at different fruit age after 200 ppm ethephon application.....	82
7.1	Change in fruit color (determined with a Chromameter) with fruit age.....	102
7.2	Changes of FRF and soluble solids content of the pericarp with fruit maturation.....	103
7.3	Relationship between FRF and soluble solids content of pericarp.....	104
7.4	Relationship between FRF and fruit color, L and hue angle (determined by a Chromameter).....	105
7.5	Correlation between percent water absorption of pericarp (Y_1) with fruit age, $Y_1 = -1704+11.4X-0.04X^2$ ($R^2=0.86$) and percent RWC (Y_2) with fruit age, $Y_2 = 1114-6.86X+0.01X^2$ ($R^2=0.64$).....	106
7.6	Relationship between RWC, water absorption capacity and soluble solids content of pericarp tissue.....	107
7.7	Relationship of dry matter content (%) of pericarp and fruit age, $Y = -5.3+0.1X$ ($R^2=0.92$).....	108

List of Figures, Continued

7.8	The relations of water potential (Y_1), osmotic potential (Y_2), and turgor pressure with fruit age ($Y_1 = 46.6 - 0.33X + 0.0005X^2$; $R^2 = 0.60$ and $Y_2 = 43.6 - 0.30X + 0.0005X^2$; $R^2 = 0.66$).....	109
7.9	Relationship between soluble solids content of pericarp and water potential (Y_1) and osmotic potential (Y_2).....	110
7.10	Change of seed dry weight with fruit age Each point is the mean of 12 fruits from 4 plants.....	111
8.1	Percent water absorption of ripe coffee fruit through exocarp via osmosis after immersion in water.....	135
8.2	Effect of mist irrigation duration on the FRF of coffee fruits at 270 and 305 days after anthesis.....	136
8.3	Effect of overhead mist irrigation on percent of fruit crack per coffee tree.....	137
8.4	Relationship between FRF and percent cracking of fruits per tree due to mist irrigation.....	137
8.5	Water saturation deficit of pericarp tissue after different periods of mist irrigation.....	138
8.6	Changes of pericarp water content in coffee fruits after different periods of mist irrigation.....	138
8.7	Effect of mist irrigation on the pericarp water relation in 305 days old coffee fruits.....	139
8.8	Relationship between FRF and pericarp turgor pressure in coffee fruits.....	139

List of Figures, Continued

8.9	Changes in soluble solids content of coffee pericarp tissue after different periods of mist irrigation.....	140
8.10	Reduction of tetrazolium chloride (TTC) as determined by the absorbance of formazan at 495 nm per g of coffee pericarp tissue fresh weight after different periods of irrigation.....	141
8.11	Electrolyte leakage of coffee pericarp tissue at different maturity as determined by soluble solids content.....	142
8.12	Changes in FRF of coffee fruits at different ages before and after 24 hr mist irrigation.....	143
8.13	Pericarp water absorption capacity of coffee fruits at different ages before and after 24 hr mist irrigation.....	143
8.14	Changes in WSD of coffee fruits at different ages before and after 24 hr mist irrigation.....	144
8.15	Changes in pericarp water content of coffee fruits at different ages before and after 24 hr mist irrigation.....	144
A1	Correlation between FRF and soluble solids content of field grown coffee fruits ($r = 0.68$).....	161
B1	Correlation between leaf water potential and the FRF of ripe and green fruits.....	163
C1	Effect of NAA at 12.5 to 50 ppm on the FRF of green and ripe coffee fruits.....	165
C2	Effect of NAA at 50 to 200 ppm on the FRF of green and ripe coffee fruits.....	165

List of Tables

- 3.1. Effect of irrigation and GA treatments on flower opening of coffee.....40
- 4.1. Mean fruit size (diameter and fresh weight) and harvesting date of coffee as affected by gibberellin treatments at various stage of fruit development.....51
- 4.2. Average seed size (dry weight and diameter) and seed number per fruit as affected by gibberellin treatments at various stage of fruit development.....52
- 5.1. Fruit removal force of green and ripe fruits and percent detachment at four sites between the fruit and stem tissues.....66
- D1. Effect of shoot tip removal in 1990 on number of flowers and fruits per branch of coffee.....167

Strategies for Machine Harvesting
of Mature Coffee (Coffea arabica L.) Fruits

Chapter 1
Introduction

Machine harvesting of coffee beans is not widely practice due to uneven maturation of coffee fruits. Hand-harvesting of coffee is the normal practice in most production areas, including Hawaii, because uneven fruit maturation makes mechanical harvest impracticed. A major concern affecting large scale coffee production is the high cost of labor that is required to selectively harvest mature beans. Twenty years ago the labor for harvesting accounted for over 70% of the total labor input for coffee production (Monroe and Wang, 1968).

In Hawaii ripening and harvesting of coffee fruits extends over a 5 months period from August to December. Because of the extended ripening period, most of the coffee in Hawaii is hand-harvested. To be competitive with other coffee producing areas of the world, producers of coffee in Hawaii must selectively machine harvest only the mature coffee. To accomplish this, one strategy that has been proposed is to synchronize flowering (Crisosto et al. 1992; Schuch, 1990). It is well known that a period of water stress followed by sufficient watering results in

the breaking of flower bud dormancy and enhancement of flowering (Alvim, 1973; Astegiano et al., 1988; Browning, 1975a; Browning, 1975b, Browning, 1977; Crisosto et al., 1992; Magalhaes and Angelocci, 1976). Unfortunately, controlling water deficit stress in the field is not practical due to unpredictable rainfall. In addition, synchronous flowering does not cause uniform fruit ripening. Schuch et al. (1990) found that although 81% of the flowers reached anthesis about 10 days after a major rain (9 cm/day) in March but only 54% of fruits ripened and were harvestable in October, and the rest ripened in November to December. Although synchronous fruit maturation is not obtained by synchronization of flowering such practices may still be important for concentrating fruit maturation. Concentrating fruit maturation is important for reducing the costs of machine harvesting (Winston et al., 1992).

Attempts have been made to induce a synchronous fruit ripening by chemical means, such as gibberellin (Schuch et al., 1990) and ethephon (Browning and Cannell, 1970; Opile and Browning, 1975; Oyebade, 1976; Sondahl et al., 1974; Winston et al., 1992). Ethephon treatments as high as 1400 ppm failed to concentrate the harvesting of mature fruits from multiple flowerings (Opile and Browning, 1975). Others have reported the beneficial effect of ethephon on

concentrating fruit ripening (Browning and Cannell, 1970; Sondahl et al., 1974; Winston et al., 1992).

Some authors reported that ethephon treatment enhanced abscission of ripe coffee fruits (Opile and Browning, 1975; Oyebade, 1976). If this is a real and reproducible response it would greatly enhance the feasibility of machine harvesting, however, more research is needed to verify the effect of ethephon on bean quality and fruit abscission.

Another strategy for machine harvesting of mature coffee is to improve the selectivity of machine harvest of ripe fruits. This alternative may be necessary to augment the problem of achieving synchronous fruit maturation in coffee. Fruit removal force (FRF) and weight (W) ratio (FRF/W ratio) has been used to indicate the ease of fruit detachment of tree crops (Wang, 1965; Wang and Shellenberger, 1967). The FRF of green and ripe fruits of different coffee cultivars was found to vary (Crisosto and Nagao, 1991). The FRF of green fruits is always greater than the FRF of ripe fruits. A large difference in FRF between green and ripe fruits should favor the selective harvesting of mature fruit by mechanical means. To alter the FRF of coffee fruits a better understanding of the process of coffee fruit separation from the stem is necessary.

Coffee plants lack the ability to shed "excess" immature fruits (Cannell, 1985). Ripe coffee fruits also do not abscise naturally and the reason for this is not clear (Opile and Browning, 1975). It is possible that the separation of coffee fruits is mechanical and not physiological by way of an abscission layer as in the separation of the guayule (Parthenium argentatum) leaf (Addicott, 1945). The separation of ripe coffee fruits occurred only when an external force was applied.

The objective of this project was to develop an effective method for machine harvesting of mature coffee fruits. To achieve this objective, the following strategies were studied:

1. Synchronization of flowering.
2. Synchronization of fruit maturation.
3. Determination of fruit detachment and FRF.
4. Altering the FRF of ripe fruit.

Synchronization of flowering

Synchronization of flowering is a prerequisite for concentrating fruit maturation. Water deficit stress followed by irrigation is able to induce uniform flowering of coffee by breaking flower bud dormancy (Alvim, 1960; Crisosto et al., 1992; Mes, 1957; Schuch, 1990; van der Veen, 1968) but this practice is not practical under field

conditions due to unpredictable rainfall. Gibberellin (GA) applied at the 4 mm dormant bud stage of coffee could partially overcome bud dormancy and promote more synchronous flowering (Schuch, 1990).

The poorly developed xylem connection in the peduncle of dormant flower buds reduces water movement into the floral tissues and inhibits normal metabolic activities (Gopal and Vesudeva, 1973; Mes, 1957). The breaking of flower bud dormancy is accompanied by a rapid influx of water into the flower bud. The uptake of water is thought to be involved in the breaking of dormancy (Astegiano et al., 1988; Magalhaes and Angelocci, 1976).

Synchronization of fruit development

Coffee fruits remain at the "pin-head" stage for 6 or 8 weeks after the flowers are fertilized (Wormer, 1964). The duration of this "pin-head" stage can vary with the climate. Opile (1979) called this a dormant period associated with high levels of abscisic acid (ABA) and low levels of GA. The difference in time of fruit dormancy breaking may be the cause of nonsynchronous fruit maturation. GA treatment may overcome fruit dormancy and induce uniform fruit maturation. Ethephon can induce uniform coffee fruit ripening (Browning and Cannell, 1970;

Crisosto and Grantz 1990; Oyebade 1976; Sondahl et al., 1974; Winston et al., 1992).

Determination of fruit detachment and FRF

The mechanism of green and ripe coffee fruit detachment has not been reported previously. An understanding of this would be useful for studies on selective machine harvesting of mature coffee fruits. Anatomical studies of the pericarp and pedicel tissues at the sites of fruit separation following fruit removal by applied force were studied.

The FRF of green and ripe coffee fruits were studied and related to the change in exocarp color, and the pericarp soluble solid content and water relations. These indices may be useful for determining the proper timing for machine harvesting.

Lowering the FRF of ripe fruit

Reduction of the FRF of ripe fruits may be accomplished by weakening of the pericarp cells. The fruits of plant species bearing soft, fleshy pericarp tissue are subject to skin-cracking disorders caused by high internal turgor pressures generated during rainfall (Considine and Brown, 1986). Cherry (Davenport et al.,

1972), apple (Byers et al., 1990), and grape (Considine and Kriedemann, 1972) fruits can absorb water through the exocarp by osmosis. Cracking of these fruits following rainfall are common. It is possible that ripe coffee fruits may behave in the same manner. If cell rupturing of the pericarp tissue does occur following periods of watering this should reduce the FRF. In contrast to the other fruits, cracking of the coffee pericarp tissue may be advantageous because the pericarp tissue is discarded during processing of coffee beans. Experiments were designed to determine the uptake of water through the exocarp tissue of different age coffee fruits. The effect of water uptake was related to the water relations of the pericarp tissue, fruit cracking and FRF.

Chapter 2

Review of Literature

Flowering

Flower bud induction

Coffee (Coffea arabica L.) is a typical short-day plant. The critical daylength is reported to be between 13 to 14 hr (Piringer and Borthwick, 1955) and has been confirmed by Schuch et al. (1990a). In the region near the equator where the plant is continuously exposed to inductive photoperiods, flower bud differentiation occurs throughout the year (Alvim, 1973). Mature plants are less sensitive to daylength than juvenile seedlings (Cannell, 1972). Flower initiation was also stimulated at day/night temperature of 23/17 °C, and inhibited at high (30°C) and low (17°C) temperature (Mes 1957). Wormer and Gituanja (1970a) found a good correlation between starch level of individual branches and the percentage of nodes with at least one flower buds.

Flower bud development

Flower bud differentiation is induced by short-day photoperiod, whereas flower bud opening (anthesis) is

associated with rainfall following a dry period (Alvim, 1973). After flower buds reach a length of 4-5 mm, they remain dormant until stimulated into flowering by water following a period of water stress. The xylem connection in the peduncle of dormant flower buds is poorly developed (Mes, 1957). During dormancy, water uptake into the flower bud is restricted causing lower water potential in the flower buds compared to the leaf (Magalhaes and Angelocci, 1976; Schuch, 1990). However, during this period, the pathway of water from the leaf to the flower bud is thought to be mostly symplastic (Astegiano et al., 1988). The availability of water enables the xylem vessels to develop rapidly in the peduncle and causes the corolla to expand rapidly, and the flowers open simultaneously 8 to 12 days after heavy rainfall ("blossom shower") (Wrigley, 1988). The water content of flower buds at dormancy is 65.8%. During the enlargement of the flower buds after "blossom showers", the water content gradually increases, reaching 85.5% at anthesis (Gopal et al., 1974).

A drought period followed by rain seems to be instrumental in the breaking of flower bud dormancy (Alvim, 1960; Mes, 1957; Schuch, 1990; van der Veen, 1968). Magalhaes and Angelocci (1976) concluded that the process of breaking of flower bud dormancy of coffee was associated with rapid changes in water balance between flower buds with the subtending leaves. The sudden

reversal of the negative water potential gradient established between flower buds and subtending leaves causes an influx of water into the buds due to the rapid increase in leaf water potential (Magalhaes and Angelocci, 1976). The rapid uptake of water by the flower buds is indicative of dormancy breaking (Astegiano et al., 1988).

In the expansion of rose petals, the increase in soluble sugar content promoted an influx of water to drive cell expansion (Ho and Nichols, 1977). The increase in soluble sugar content of the outer whorl of the rose petal coincided with the time when the rate of petal expansion increased (Evans and Reid, 1988). The soluble sugar content is derived largely from starch hydrolysis, and that a major function of low-molecular-weight carbohydrates is to lower osmotic potential in petal cells. Astegiano et al. (1988) found that the mobilization of water and calcium to coffee flower buds was greater in plants that had experienced a drought period. This may be an indicator that an osmotic adjustment of the flower buds occurred due to water deficit stress. Osmotic adjustment is the active accumulation of solutes in the plant tissues resulting in the lowering of osmotic potential (Zhang and Archbold, 1993a). Osmotic adjustment may be advantageous in maintaining turgor during drying conditions by lowering total tissue water potential (Cortes and Sinclair, 1987). This phenomenon was found in many plants as in the leaves

of Fragaria chiloensis (Zhang and Archbold, 1993a), soybean leaf (Cortes and Sinclair, 1987), magnolia and sweetgum leaves (Augé and Stodola, 1989).

Exogenous applications of gibberellins (GA) could replace the stimulus induced by water stress for overcoming dormancy of coffee flower buds (Alvim, 1958; Browning, 1975; Schuch et al., 1990b; van der Veen, 1968). Browning (1977) reported that the exogenous GA could replace the stimulus generated by irrigation and rainfall even when it was applied without water contact directly to the buds. Furthermore, increased amounts of extractable GA are produced rapidly in buds after irrigation or rainfall accompanied by a temperature reduction (Browning, 1973). Abscisic acid (ABA) prevented the flower bud from developing to anthesis. The effect of ABA gradually decreased as the length of drought increased. Exogenous ABA did not prevent the response of exogenous GA on overcoming coffee flower bud dormancy (Browning, 1972).

Extensive reviews on flower induction, flower development and flower bud dormancy can be found elsewhere (Alvim, 1985; Barros et al., 1978; Browning, 1977; Cannell, 1985).

Fruit Development

Stage of development

The coffee fruit is a drupe which normally contains two seeds but occasionally more. It is commonly referred to as a cherry or berry (Wrigley, 1988). Fruit growth of coffee follows a sigmoid curve, which plateaus as a result of retarded growth rate after the fruit has reached about half of its mature size (Clowes and Allison, 1982). For the first 6 or 8 weeks after the flowers are fertilized, cell division occurs in the ovary but the fruits remain at the "pin-head" stage, increasing little either in size or weight (Wormer, 1964). The duration of this "pin-head" stage can vary with the climate. Opile (1979) called this a dormant period associated with high levels of abscisic acid (ABA) and low levels of gibberellin (GA). Following this lag period the fruit enters the rapid growth phase which lasts about 10 to 17 weeks after flowering. At the end of this period of rapid fruit expansion, the parchment is fully formed and the integument, which forms the silverskin, stops growing (Wormer, 1964). The endosperm remains small until 11 or 12 weeks after flowering, and thereafter suppresses and replaces the integument. About 19 weeks after flowering the endosperm completely fills the cavity. The endosperm continues to increase greatly in

dry weight up to about 30 weeks after flowering, when the beans reach their full dry weight. The fruits ripen at 30 to 35 weeks after flowering. During the seven weeks' ripening period the dry weight of the pericarp increased by 106 percent while the dry weight of the whole fruit increased by 49 percent (Wormer, 1964).

Fruit ripening

On a single plant, the fruits from flowers opening on the same date ripen over a period of at least six weeks (Clowes, 1977). During ripening the fruit expands and pericarp increases greatly in dry weight, the exocarp turns red, and the mesocarp softens and becomes mucilaginous (Cannell, 1971). The seeds attain their final dry weight when berries are still green. During ripening of the berry, which is in fact ripening of the pericarp, the fresh weight of the seeds drop slightly due to loss of water (Wormer, 1964). Fruit color change, from green to red or yellow (depending on cultivars), is associated with fruit ripening. The red pigments of coffee fruits are anthocyanins which are located in the epidermal layer of the fruit. The yellow fruits contain leucoanthocyanins and flavonols and lacks anthocyanins (Mazza and Miniati, 1993). Fruit skin color can be used as an index for

harvesting of many fruit crops as in sweet cherries (Drake et al, 1982) and grape (Watada and Abbott, 1975).

Anthocyanins in the pericarp of muscadine grapes increased with fruit maturity (Flora, 1978). Soluble solid content of grape was well correlated with anthocyanin content (Watada and Abbott, 1975). This close association between soluble solid content and anthocyanin content may be due to the increased concentration of both with ripening.

Ethephon has been used to enhance fruit ripening. Oyebade (1976) found that a single application of ethephon at 200 ppm and above showed a marked influence on ripening when applied to mature green berries. Within the first six days after chemical application, ripening was visually noticeable. A similar experiment was done by Winston et al. (1992) who sprayed coffee plants with ethephon at various concentrations ranging from 125 to 2000 ppm. They found that higher rates increased the number of ripe fruits, but those concentrations of more than 500 ppm caused unacceptable leaf abscission. The rate of 250 ppm was marginally acceptable in terms of leaf abscission. Ethephon had a less pronounced effect on ripening when applied immediately before the ripening process began. Crisosto and Grantz (1990) reported ethephon at 100 ppm was also effective in inducing uniform ripening when applied at the onset of ripening. In one experiment,

however, ethephon as high as 1400 ppm failed to concentrate the harvesting of trees bearing a crop from multiple flowerings, although ripening was still brought forward (Opile and Browning, 1975). Other reports support the successful use of ethephon in enhancing fruit ripening (Browning and Cannell, 1970; Sondahl et al., 1974).

Crisosto and Grantz (1990) reported that cupping tests (coffee tasting) did not reveal any quality differences between ethephon-treated and untreated coffee beans. In contrast, Sondahl et al. (1974) found a decrease in cupping quality if the ethephon treatment was done too early. Late treatment, after fruit ripening, resulted in good cup quality. These observations suggest that ethephon can be used to obtain more uniform fruit maturation without reducing the quality of coffee, providing application is made after the fruits begin to ripen.

Fruit drop

The coffee plant is unusual among woody perennials in that it lacks the ability to shed excess immature fruits (Cannell, 1985). Ripe coffee fruits also do not abscise naturally (Opile and Browning, 1975).

Ethephon is an effective abscission inducing chemical for many fruit crops such as citrus (Young and Jahn, 1972), cherries (Bukovac et al., 1971), and apple

(Edgerton, 1971). The effect of ethephon on fruit abscission of coffee is controversial. Fruit drop following the ethephon treatments was observed but fallen fruits were generally over-ripe and abscised easier because of their softer pericarp (Browning and Cannell, 1970). It was concluded that ethephon increased the fall of coffee berries partly by increasing berry ripeness. This conclusion was confirmed by Oyebade (1976). On the contrary, Winston et al. (1992) reported that ethephon did not cause significant fruit abscission at any concentration from 125 to 2000 ppm.

Browning and Cannell (1970) found no distinct distal abscission layer present in the fruit pedicel of the fallen fruits following ethephon application. However, Opile and Browning (1975) reported distinct distal abscission layers found on the pedicels of many fallen coffee fruits sprayed with both ethephon and triiodobenzoic acid. Unfortunately, they did not present evidence for their claim.

In contrast to fruit drop, NAA treatment has been reported to increase the yield of coffee. This effect may be due to an inhibition of fruit drop (Vasudeva and Venkataramanan, 1981).

Fruit removal force

Fruit removal force (FRF) is the energy required to remove the fruits. It can be measured quantitatively with a pull force gauge. Pressure is applied by pulling at a zero angle parallel to the axis of the fruit, in order to transmit the force equally to all sides of the juncture of the woody peduncle and exocarp of the fruit (Cooper and Henry, 1973). Coffee fruit removal forces are higher than other fruits having about the same weight (Monroe and Wang, 1968). Crisosto and Nagao (1991) compared the FRF of five coffee cultivars. They showed that the FRF of green fruits from 5 cultivars tested were similar and about twice the force required to remove red fruits. They also found that the FRF difference between green and red fruits changed diurnally. There was a larger difference in FRF between green and red fruits removed in the early morning than other times. Ethephon at 50 to 200 ppm reduced the FRF and/or induced fruit abscission of coffee fruits in some cases. Ethephon treatment has also been observed to accelerate FRF reduction in young macadamia fruits (Nagao and Sakai, 1985).

Fruit water relation

Crisosto and Nagao's (1991) report that coffee fruits harvested at 0600 hr gave larger differences in FRF between green and red fruits than other times might be due to changes in plant water relations. Patten et al (1988) studied machine harvesting of blueberries at different times of the day and concluded that during the night and early morning, when plants were at full turgor, fruit removal force was less. No study on fruit water relations of coffee has been reported anywhere.

Total water potential of plant tissues can be partitioned into components mainly osmotic potential, turgor pressure, and matric potential (Turner, 1981). Tissues water potential can be measured with the pressure chamber technique (Turner, 1988). Thermocouple psychrometers are used in measuring tissues water potential and osmotic potential, which then can be used to estimate turgor pressure (Matthews et al., 1987; Turner, 1981; Tyree, 1976).

Soluble solids and osmotic potential of grape fruits are well correlated to each other (Matthews et al., 1987). This relationship was also found in sweet cherries and peach (Andersen and Richardson, 1982). Soluble solid content usually increases with fruit maturity, that is,

the change in osmotic potential follows the same pattern but in opposite direction.

Fruit turgor pressure of tomato (Shackel et al., 1991) and grape (Matthews et al., 1987) were relatively low as compared to the osmotic potential of the same tissues. The turgor of stalk parenchyma of sugarcane was also maintained relatively low during sucrose accumulation (Moore and Cosgrove, 1991). This may be because these tissues have an ability to regulate turgor (Moore and Cosgrove, 1991). They found that the low turgor of stalk parenchyma of sugarcane was maintained by the partitioning of solutes between the cell and wall compartments. The presence of apoplastic solutes was also reported in beet root (Perry et al., 1987; Wyse et al., 1986).

Effect of ethylene on turgor pressure has been reported by Eisinger et al. (1983) who investigated the effect of ethylene on the osmotic potential of pea internode tissue. Ethylene caused a decrease (more negative) in osmotic potential and resultant increase in turgor pressure.

Fruit cracking is a significant problem of reducing fruit quality of many species. The fruits with soft and fleshy pericarp are subject to skin-cracking disorders caused by high internal turgor pressures generated during rainfall (Considine and Brown, 1986). The critical turgor pressure of grape, the pressure which resulted in 50% of

the berries splitting, was approximately 1.5 MPa in grape cultivars sensitive to splitting and 4.0 MPa in resistant cultivars (Considine and Kriedemann, 1972).

Cherries can absorb water through the fruit cuticle (Davenport et al., 1972). Watson et al. (1988) reported that rain causes cherries to absorb moisture, swell and crack, making them unmarketable. Water is absorbed by osmotic pressure through small pits that occur naturally in the exocarp cells. With increasing water uptake, turgor pressure increases and eventually causing the rupture of the cherry exocarp. Under tree irrigation has little effect on splitting and provides supporting evidence that water is absorbed into the cherry directly through the skin and not from the soil. Apple fruits can also absorb water through the skin and may lead to fruit cracks. The primary sites of water uptake were through lenticels and injured areas of the fruit cuticle (Byers et al., 1990). Fruit cracking usually occurs during relatively long rainy periods. Soil water failed to induce fruit cracking. Fruit cracks can occur during high humidity periods only when the relative humidity was near 100% for 6 hours or more (Verner, 1935).

Machine Harvesting

Mitchell (1988) described the methods of hand picking and machine harvest of coffee. In Puerto Rico and Hawaii, tree and branch shakers have been used to harvest ripe fruits onto mesh nets laid on the ground. This method is unsatisfactory resulting in the removal of many green berries. In Brazil, harvesting machines with vibrating fingers that comb through the foliage of the coffee tree either shaking or knocking off the fruits, onto collecting trays are used. Over 95% of the crop is removed by these machines including green fruits.

The concept behind the design of coffee harvesters is based on mechanical vibration. The FRF to weight ratio (FRF/W) have been used as an index of determining the timing of fruit detachment (Wang and Shellenberger, 1967). Wang (1965) concluded that the differences between FRF/W ratio of mature and immature coffee fruits was important for developing indices the selective harvesting of mature fruits. Mechanical harvesting of coffee by vibration is accomplished by increasing the frequency of the applied force and decreasing the amplitude. Selectivity is improved when the shaker frequency is adjusted based on the amount of mature and immature fruits on the tree at time of harvest. Monroe and Wang (1968) suggested that a lower frequency was better for selective harvesting of

ripe fruits when the percentage of ripe fruits on the tree was low. However, they reported that about 15 to 40% of the harvested fruit was immature. Jakeway (1990) evaluated the Korvan harvester (Korvan Industries, Lynden, Washington) for harvesting of seven coffee cultivars. For six of the cultivars the composition of ripe to green fruits was 60% and 14% respectively. One of the cultivars, Catuai, 41% of the fruits harvested was immature. Another problem of harvesting by machine is the amount of unharvested fruits. Approximately 27 to 52% of the fruits was harvested while about 37 to 65% was left unharvested when harvesting was done with machine. Ethephon and other chemicals have also been tried in attempts to promote abscission of ripe berries as an aid to mechanical harvesting, but the results have been disappointing (Mitchell, 1988).

Chapter 3

Effect of Mist Irrigation, Gibberellic Acid and Desiccation on Anthesis of Coffee Flower Bud

Abstract

Container grown coffee (Coffea arabica L. 'Guadalupe') plants with dormant flower buds at the 4-mm stage were subjected to mist irrigation, desiccation and gibberellin (GA) treatments to overcome dormancy. Twenty four hours of mist irrigation effectively induced uniform flower opening within 8-9 days after treatment. Similar results were obtained by first desiccating plants to leaf water potential of -3.6 MPa followed by rewatering to field capacity. All flowers opened synchronously at 9 to 10 days after rewatering. The osmotic potential of desiccated plants remained lower than the controls several days after the rewatering. This phenomenon may be due to osmotic adjustment of the flower buds which regulate rapid water absorption into the buds. Mist irrigation resulted in a rapid uptake of water into the flower buds perhaps by absorption of exogenous water by osmosis. Both treatments achieved the same goal, rapid water absorption and enhanced flower bud opening.

GA treatment was not effective in promoting synchronous flower opening. Most of flower buds were dehydrated and very few flowers developed to anthesis 11 days after treatment. The control plants which were maintained at field capacity produced no open flowers during the time of experiment.

Introduction

Synchronization of flowering is an important strategy for achieving uniform fruit ripening. The nonsynchronous flowering of coffee plants is a result of flower bud dormancy. Alvim (1973) proposed that the flower buds of coffee seem to have two phases of rest: a) a true dormancy which is controlled by growth inhibitor(s); and b) an imposed dormancy or quiescence which may result from inadequate water supply. Moisture stress is necessary to reduce the level of growth inhibitors to release the dormant phase, whereas water uptake is necessary to release the quiescence phase (Alvim, 1973).

The growth of the flower buds stop at the 4-mm stage due to dormancy (Cannell, 1985). Browning (1973) proposed that flower bud dormancy may be a form of true dormancy associated with high endogenous abscisic acid level.

At the 4-mm stage the xylem connections between the flower buds and their parent shoots are poorly developed (Mes, 1957). This was confirmed by dye uptake studies of flower buds (Schuch, 1990) and the movement of ^{45}Ca and tritiated water from subtending leaf to the flower buds (Astegiano et al., 1988) of stress and nonstressed plants.

Numerous attempts have been made to synchronize flower opening. Treatments such as water deficit stress followed by rewatering or in combination with GA spray are

known to be effective in overcoming flower bud dormancy (Alvim, 1958; Alvim, 1973; Browning, 1977; Crisosto, 1990; Schuch et al, 1990). Crisosto (1990) showed that flower opening was stimulated by irrigation after one period of water deficit if predawn leaf water potential declined below -0.8 MPa. Similar results were obtained by Schuch (1990). Plants subjected to leaf water potentials lower than -2.65 MPa reached anthesis within 9 days after rewatering. Under controlled conditions withholding water is an effective means to overcome dormancy, however, controlling water deficit stress to the desired level is nearly impossible in the field because of unpredictable rainfall.

Gibberellin can promote flower bud development toward anthesis (Schuch et al., 1990). Exogenous gibberellin application replaces the stimulus generated by irrigation and rainfall (Browning, 1977). Data from Schuch et al. (1990) showed that GA supplemented the effect of water stress in the regulation of flower opening. Buds that were smaller than 4 mm at the time of treatment did not respond to GA₃ application.

Alvim (1958) showed that submerging the branches in water for 1 hr could induce flower opening but the effect was not as good as the gibberellin treatment. Rapid changes in water balance between flower buds and subtending leaves causes the breaking of bud dormancy has

been proposed by Magalhaes and Angelocci (1976). They concluded that irrigation released flower bud from dormancy in response to a sudden reversal of the negative water potential gradient between flower buds and subtending leaves. Gopal and Vasudeva (1973) proposed that the dormancy of flower buds at the 4-mm stage was caused by the physiologically insufficient water content in plants which ceased the normal metabolic activities of the plant including enlargement of flower buds. From previous work, any treatment that results in the sudden increase of flower bud water uptake could be effective in promoting flower bud growth to anthesis. The objective of this study was to prove that the cause of flower bud dormancy in coffee is due to the inability of the flower bud to acquire water through the xylem system and dormancy could be overcome by water uptake directly through the surface of the flower bud by osmosis.

Materials and Methods

Three year old coffee (Coffea arabica L. 'Guadalupe') plants growing in 4-l plastic containers with pumice, peat, soil and sand (2:1:1:1) as potting media in a greenhouse under day/night temperatures of 25/19 °C and natural photoperiod (Oregon, 44°N latitude) were used for these studies. Plants were approximately 150 cm tall and contained more than a hundred flower buds at the 4-mm dormant stage per plant at the time of treatment. In June 1993 sixteen plants with uniform flower nodes were selected. From each plant fifteen nodes with flowers at the 4-mm stage were tagged for this study. The selected plants were transferred to one bench in the greenhouse and drip irrigated at the rate of 1.5 l/hr/plant continuously for 8 days. The plants were then divided into the following 4 groups of four plants each: Group 1, watering was withheld for 6 days to induce water stress (not more than -3.0 MPa). Groups 2, 3 and 4 plants were kept under the drip irrigation to prevent water stress. At this point, the leaf water potential of all plants were measured at 1000 hr with the pressure chamber (PMS, Corvallis, Oregon). Two terminal shoots with 4 leaves from the middle part of the trees were used for leaf water potential measurements. The stressed plants (Group 1) were then rewatered and resumed drip irrigation treatment.

Group 2: Fourteen days after drip irrigation, four plants were transferred to a mist chamber for the mist application experiment. The plants were held under the mist generated by a fog generator (Tobacco Conditioner Model 35, Agritech, Sanford, North Carolina) which emitted a fine spray of water continuously at the rate of 100 l/hr for 24 hr to keep all above ground plant parts in contact with water. After mist application the plants were placed under drip irrigation. Group 3: Four plants were treated with gibberellic acid by spraying the above ground plant parts to run off with 100 ppm GA₃ (ProGibb, Abbott Laboratories, North Chicago, Illinois) with a hand sprayer. After treatment these plants were placed under drip irrigation. Group 4: Four plants was maintained continuously under drip irrigation throughout the study. Flower number at anthesis were counted daily for 14 days.

Two flowers per tree were tagged and their growth measured once every 3 days. The floral length was measured from flower tip to the base of corolla tube, including the ovary, with a micrometer and vernier caliper. The average flower weight was determined by weighing 10 flowers from each plant and dividing the total weight by 10. The osmotic potential of the flowers was measured at 3 day intervals by placing the flower buds in a disposable centrifuge tube with a lid, freezing the samples at -80°C, thawing the samples at room temperature, centrifuging at

5000 rpm for 5 min, and determining the osmotic potential. Eight μ l of the sap was transferring with a micropipet to a small paper disk in the osmometer (Wescor 5100C, Wescor, Logan, Utah). Flowers from each tree were collected at 3 day intervals, weighed and dried for 24 hr at 80°C. Percent water content of the flowers was calculated by the following formula:

$$\text{Percent water content} = \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100$$

Data were analyzed with SAS by GLM procedure when all four treatments were available and means comparison were done by Duncan's Multiple Range Test.

Results

At the beginning of the experiment the leaf water potential was -0.54 ± 0.03 MPa. On the last day of the desiccation treatment, the leaf water potential was -3.6 ± 0.2 MPa. After 24 hr of rewatering to saturation the leaf water potential of these plants returned to normal (-0.47 ± 0.21 MPa).

The leaf water potential following 24 hours of mist irrigation increased to -0.17 ± 0.01 MPa. Twenty four hours after resuming drip irrigation the leaf water potential decreased to -0.63 ± 0.50 MPa. The leaf water potential of the control and GA treated plants was -0.53 ± 0.26 MPa throughout the study.

Flower opening

None of the flower buds from the control or GA treated plants developed to anthesis within 9 days (Table 3.1). In contrast, plants subjected to either desiccation or mist irrigation treatments produced uniform flowering to anthesis within 9 days after treatment. Desiccation stimulated opening of 80% and 100% of the flower buds by the 9th and 10th days respectively. Mist irrigation treatment stimulated 72% and 100% floral bud opening 8 and 9 days after treatment respectively (table 3.1).

Majority of the flower buds of the GA treated plants desiccated and abscised. Few flowers developed to anthesis (0.43 flowers per node) 11 days after treatment.

Flower water content

The water content of the floral buds at the start of the study was $62.9 \pm 0.3\%$ (Figure 3.1). After desiccation of plants to -3.6 MPa the flower water content decreased to $48.6 \pm 0.7\%$. Rewatering of the desiccated plants for 24 hr increased the water content of the flower buds to $68.5 \pm 0.9\%$. The flower water content of the control was $65.4 \pm 0.8\%$.

Twenty four hours of mist irrigation increased the flower water content to $68.1 \pm 0.4\%$, similar to the rewatered desiccation treatment. The water content of flower buds of the control plants remained unchanged throughout the 9 days treatment period. In contrast, the flower water content of the desiccation and mist treatments continued to increase similarly and parallel to each other throughout the test period (Figure 3.1). At anthesis, the flower water content of the desiccation and mist treatments were $83.6 \pm 0.3\%$. The flower water content of the GA treated plants increased slightly throughout the treatment period and significantly less than the desiccated and mist treatments (Figure 3.1).

Flower growth

The growth (length and weight) of the flowers in the mist irrigation and desiccation treatments increased similarly and parallel from the time of treatment to anthesis (figure 3.2 and 3.3). The growth of the flowers was correlated with the increased water content ($r = 0.88$). The length and weight of the flowers in the GA treatment increased slowly as compared with the desiccation and mist treatments. Only 2 out of ten of the tagged flowers developed to anthesis. No visible growth of flowers was observed in the control treatment.

Osmotic potential

The osmotic potential of coffee flowers prior to the treatments was -1.09 ± 0.01 MPa (Figure 3.4). Following desiccation, the flower osmotic potential decreased to -3.12 ± 0.15 MPa. The osmotic potential increased rapidly after rewatering but remained lower than the initial value and the control 4 days after rewatering. At full bloom, the osmotic potential of the flowers decreased.

Mist irrigation increased the osmotic potential of the flowers immediately after treatment. Four days after treatment, the osmotic potential returned to normal (-1.09 ± 0.03 MPa) and then followed the same pattern as

the desiccation treatment. The control plants had a constant osmotic potential throughout the 9 days study period. The flower osmotic potential of the GA treated plants increased slightly on the 9th day after treatment.

Discussion

Desiccation treatment prior to rewatering by drip irrigation induced flower opening 9-10 days after treatment. This confirms previous work that a period of water stress followed by watering at the 4-mm stage of floral development induces flower opening (Alvim, 1960; Astegiano et al., 1988; Browning, 1977; Crisosto et al., 1992; Schuch 1990).

The mechanism of how water stress followed by rewatering induces flower opening is not clear. Browning (1973) found the gibberellin content in flower buds to increase rapidly as the result of watering after a period of drought. Magalhaes and Angelocci (1976) proposed that the rapid changes in water balance between flower buds and subtending leaves caused the breaking of bud dormancy. They concluded that irrigation released flower buds from dormancy in response to a sudden reversal of the negative water potential gradient between flower buds and subtending leaves. Gopal and Vasudeva (1973) proposed that flower bud dormancy at the 4-mm stage is caused by insufficient water content which reduces normal metabolic activities of the plant including enlargement of flower buds. Astegiano et al. (1988) reported that the mobilization of water and calcium to buds was greater in plants that had experienced a drought period.

Our results showed desiccation to decrease the osmotic potential of flowers, and rewatering caused the rapid increase of osmotic potential. As the water content in the flowers decreased from 62.9 to 48.6% due to desiccation, the solute concentration increased. After 24 hr of rewatering, the water content of the flower buds increased and the osmotic potential increased due to the dilution effect. The osmotic potential of the flower after 24 hr of rewatering did not increase above the control . This suggests that the low osmotic potential of the flowers at the end of the desiccation period was not solely due to the concentration of solutes due to decreased water content. The osmotic adjustment of the flower buds due to water deficit stress was reported to be important in many plant species (Hsiao, 1973; Morgan, 1984; Zhang and Archbold, 1993a). The lowered osmotic potential may be the result of starch to sugar conversion as reported in some species e.g. strawberry (Zhang and Archbold, 1993b). During the expansion and opening of the rose flower an increase in the soluble sugar content of the floral tissue was observed by Evans and Reid (1988). Ho and Nichols (1977) suggested that the change in soluble sugar content was largely due to starch hydrolysis. This finding was supported by Hammond (1982), who suggested that rose flower opening is accompanied by a concomitant increase in amylase activity. Gopal et al. (1975) studied

the changes in carbohydrates status of coffee flower buds and found that total sugars increased while the starch content decreased 2 days after rainfall.

One day after rewatering, the flower bud water content gradually increased and seemed to be associated with flower growth toward anthesis. The rapid water uptake by flower buds within 24 hr after rewatering was demonstrated by Schuch (1990). Schuch suggested that desiccation induced opening of the xylem connections between flower buds and the parent branches allowing the passage of water to the developing flower buds. Dye uptake increased rapidly from day 3 to anthesis indicating the establishment of water conducting elements from the pedicel into the flower. The pattern of water movement in flower buds was described by Gopal et al. (1974). The low osmotic potential of the stressed flower bud may be a reason for the increased movement of water into the flower. Water stress may also trigger xylem differentiation. The confined development of the xylem opening and the lowered osmotic potential of the flower tissues may both play roles in water movement into the flower bud.

Mist irrigation enhanced flowering similar to the desiccation treatment. All of the flowers developed to anthesis within 9 days after misting. This proves that water stress is not necessary to overcome dormancy of

coffee flower buds. Gopal and Vasudeva's (1973) hypothesized that the low water content of the flower bud was insufficient for the normal metabolic activity for flower bud enlargement. That hypothesis is consistent with the result of our study that the direct increase in water content alone was capable of overcoming dormancy and promoting flower development. Our results supports the conclusion of Mes (1957) that any treatment that reduced the water tension inside the flower buds would release dormancy and regulate flower opening. Mist irrigation allowed the direct contact of flowers with water. Alvim (1958) reported that flower buds can absorb exogenous water directly. Twenty four hours of mist irrigation was sufficient for inducing complete flowering. This result may explain the effect of heavy rainfall on flower opening as observed in the field by some investigators (Alvim, 1958; Schuch, 1990; van de Veen, 1968). The osmotic potential of the flower buds immediately after mist irrigation decreased while the water content increased (Figure 3.1 and 3.4). On day 7, at the candle stage of development, the osmotic potential of both desiccation and mist irrigation treatments increased to the maximum and the water content was about 85%. The osmotic potential was lowered again at anthesis probably due to the contamination of flower nectar since the whole flower was measured.

GA treatment was only partially effective in overcoming dormancy. The flowering of the well-watered (drip irrigated) GA treatment was poor and not synchronous as reported elsewhere (Alvim, 1958; Schuch et al. 1990). GA treatment had no effect on osmotic potential during the first 6 days. Flower bud size, weight, and water content of the GA treatment increased slightly.

Table 3.1. Effect of irrigation and GA treatments on
flower opening of coffee.

Treatment	% flower at anthesis			%flowering ^{1/}
	time after treatment			
	day 8	day 9	day 10	
Control	0	0	0	0
Desiccation	0	79.6	20.3	100
Misting	72.4	27.6	0	100
GA	0	0	0	0

1/ percent flowering referred to percent of selected nodes
with flower at anthesis.

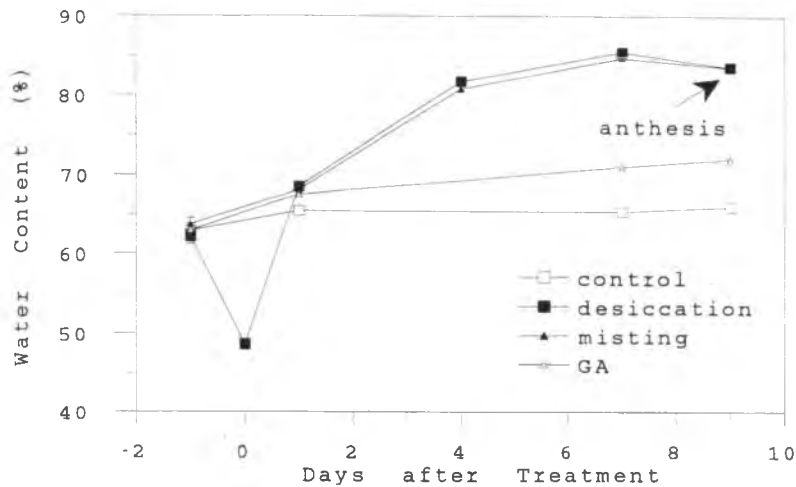


Figure 3.1. Flower water content as affected by irrigation, desiccation, and GA treatment.

Explanation:

day -1 = One day before any treatments.

day 0 = a) Plants desiccated for 8 days to -3.6 MPa and resumed normal drip irrigation.

b) Begin mist irrigation treatment.

c) GA applied.

day 1 = a) One day after rewatering of the desiccation treatment.

b) Completion of mist irrigation (24 hr treatment) and resumption of drip irrigation.

day 9 = Time of anthesis (desiccation and mist irrigation treatment).

Control plants treated with drip irrigation throughout study.

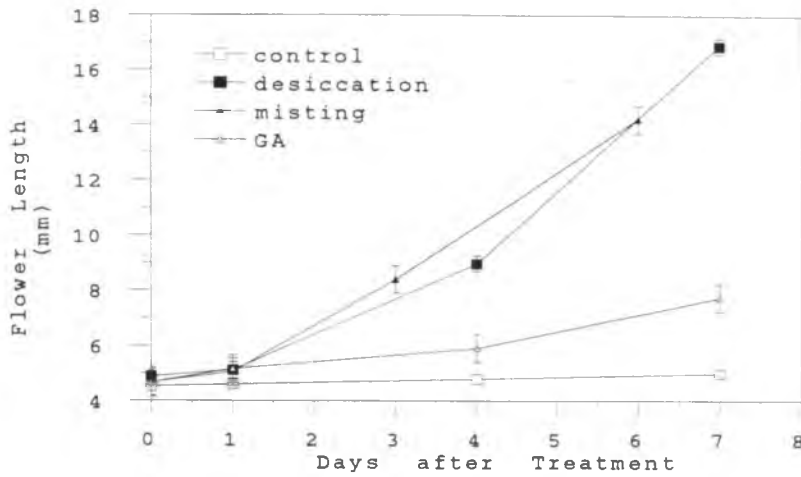


Figure 3.2. Length of coffee flowers after drip irrigation, mist irrigation, desiccation and GA treatments.

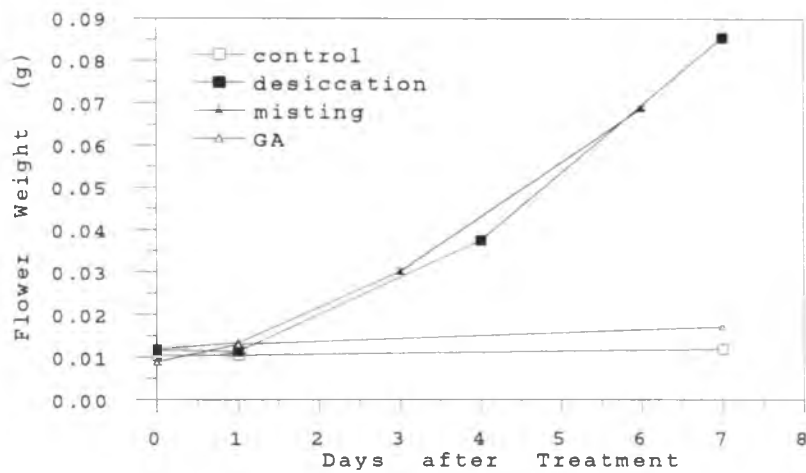


Figure 3.3. Weight change of coffee flower after drip irrigation, mist irrigation, desiccation and GA treatments.

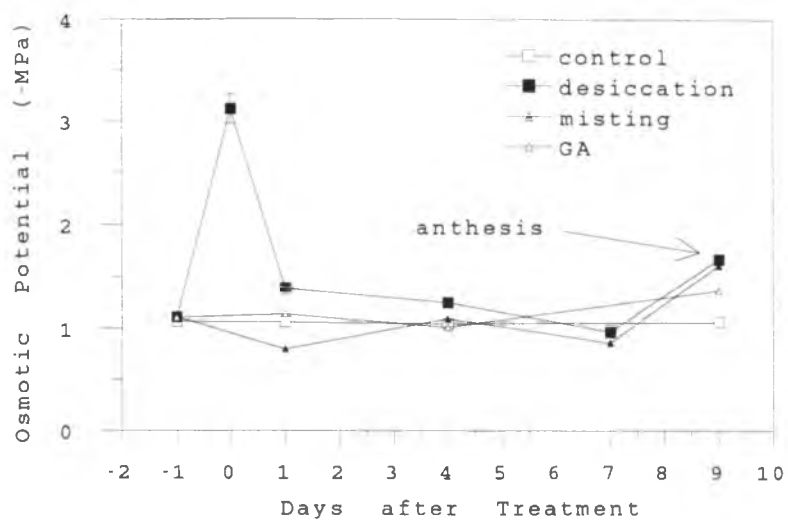


Figure 3.4. Change in osmotic potential of coffee flowers after drip irrigation, mist irrigation, desiccation and GA treatments.

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Chapter 4

The Effect of Gibberellic Acid on Fruit Growth and Maturation of Coffee

Synchronization of flowering in coffee (Coffea arabica L.) has not resulted in synchronous fruit maturation. The fruits from flowering on a given date ripen over a period of at least six weeks (Clowes, 1977). Similar results were reported by Schuch et al. (1990). Asynchronous fruit maturation may be caused by variable rates of fruit growth and development. The coffee fruit remains in the "pin-head" stage, with no visible growth, for approximately 6-8 weeks after anthesis (Cannell, 1974; Clowes and Allison, 1982; Wormer, 1964). These fruits are thought to be dormant due to high levels of endogenous abscisic acid and low levels of active gibberellins (GA) (Opile, 1979). Gibberellin has been reported to be effective in breaking dormancy of plant parts including coffee flower buds (Browning, 1977; Schuch et al., 1990; van de Veen, 1968; Wormer and Gituanja, 1970). Schuch et al. (1990) reported a carry over effect of gibberellin on fruit maturation when it was applied at the flower bud stages.

Fruit growth of coffee follows a sigmoid curve, which is interrupted by retarded growth rate after the fruit has reached about half size (Clowes and Allison, 1982). For

the first 6 or 8 weeks after the flowers are fertilized, cell division occur in the ovary but the fruits remain at the "pin-head" stage, increasing little either in size or weight (Wormer, 1964). The duration of this "pin-head" stage can vary with the climate. Opile (1979) called this a dormant period associated with high levels of abscisic acid (ABA) and low levels of gibberellin (GA). It is possible that gibberellin may overcome coffee fruit dormancy in the early stages. Overcoming fruit dormancy may result in enhancement of fruit growth and development and possible synchronization of fruit maturation. The objective of this study was to determine the effects of GA treatments immediately after anthesis on overcoming the lag ("dormant") phase of fruit development and its subsequent effect on fruit maturation.

Nine uniform 3 year old coffee plants c.v. Yellow Catura growing in the field at Mountain View, Hawaii were used for this study. About ten branches from each tree with flowers at anthesis (full bloom) were tagged on May 17, 1990. About five flowers from each node were marked with waterproof ink to identify the date of anthesis. The experiment included 6 treatments (100 ppm GA + 1% Tween 20 applied 11, 18, 26, 32, and 40 days after anthesis on individual branches, one branch per plant; and control 1% Tween 20) randomized in a complete block design with 9 blocks (plants). Fruit diameter at the center of the fruit

was measured weekly with a micrometer (for the first 10 weeks when the fruits were very small) and a vernier caliper thereafter. Fruits were harvested at maturity (completely yellow pericarp color). Fruits were weighed individually after harvest. The pericarp was then removed and the seeds oven dried at 70°C for 48 hr. The parchment was then removed from the seed and the dry weight of the seed determined. Seed size, both diameter and length, was measured with a vernier caliper.

After first week after anthesis, the fruits were 2 mm in width. This stage is called the "pinhead" stage (Cannell, 1974). Gibberellic acid treatment at all stages of fruit development caused an increase in fruit size within 1 week (Figure 4.1). The diameter of the treated fruits remained greater than the control up to 74 to 80 days after anthesis. Thereafter, their diameter was comparable to the control.

Fruits treated with gibberellic acid 11 and 18 days after anthesis matured significantly earlier than the control (Figure 4.2, Table 4.1). Gibberellic acid fruit application at later stages, i.e. 26 to 40 days after full bloom, did not enhance ripening significantly, however, there was a tendency for earlier ripening due to the GA treatment generally (Figure 4.2, Table 4.1). Fruits treated with GA could be harvested about 10 days earlier than the control. Although GA treatment was effective in

enhancing earlier fruit maturation, the timing of fruit ripening within treatments was still variable. The two earliest GA application treatments did result in more synchronous fruit ripening than the other treatments (30 versus 50 days), however, although this may be desirable, for machine harvesting a closer concentration of fruit maturation would be necessary.

In general, cell division of the ovary tissue is completed before anthesis. The rate of cell enlargement and elongation was enhanced by the gibberellin treatment. However, although the fruits reached their final size earlier following GA treatment the final size and weight of the untreated fruits were eventually the same (Table 4.1 and 4.2). Seed weight, seed diameter and seed length were not affected by the gibberellin treatments. The numbers of seeds per fruit were not significantly different among the treatments (Table 4.2).

Table 4.1. Mean fruit size (diameter and fresh weight) and harvesting date of coffee as affected by gibberellin treatments at various stage of fruit development.

Treatment	Fruit weight (g)	Fruit diameter (mm)	Harvest date ^{1/}
control	1.84	10.64	239.2 a
GA 11 days AFB ^{2/}	1.72	11.01	228.9 d
GA 18 days AFB	1.66	10.63	229.6 cd
GA 26 days AFB	1.80	10.87	235.7 ab
GA 32 days AFB	1.80	10.90	235.1 abc
GA 40 days AFB	1.65	10.53	233.4 bcd

1/ Mean separation by Duncan Multiple Range Test at $p = 0.01$

2/ AFB = after full bloom

Table 4.2. Average seed size (dry weight and diameter) and seed number per fruit as affected by gibberellin treatments at various stage of fruit development.

Treatment	Seed weight (g)	Seed diameter (mm)	Seed length (mm)	Seeds per fruit
control	0.225	7.30	10.83	1.47
GA 11 days AFB	0.230	7.39	10.86	1.63
GA 18 days AFB	0.221	7.14	10.53	1.61
GA 26 days AFB	0.194	7.13	10.43	1.78
GA 32 days AFB	0.233	7.33	10.74	1.62
GA 40 days AFB	0.220	7.42	10.59	1.63

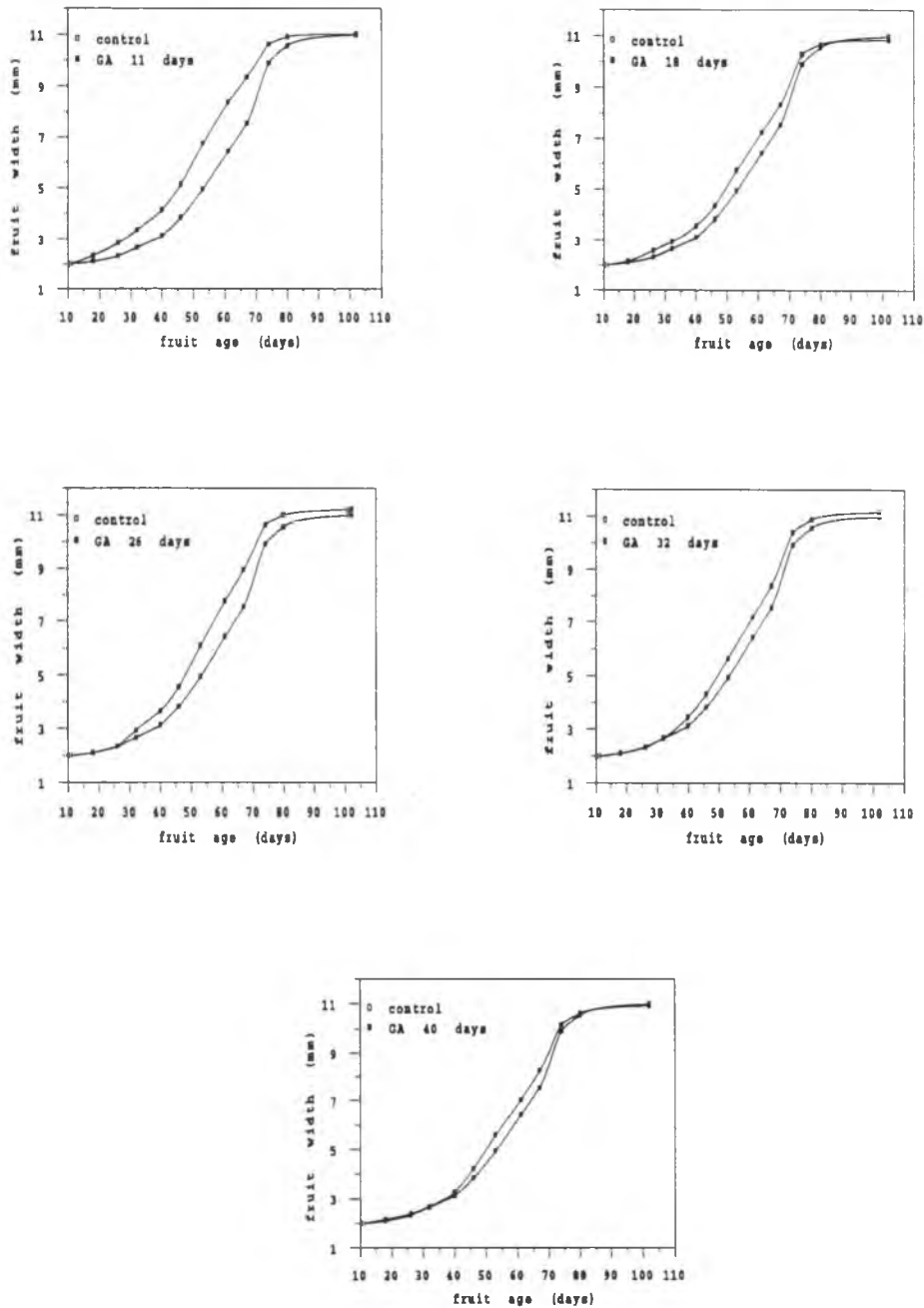


Figure 4.1. Coffee fruit diameter following gibberellic acid application after anthesis.

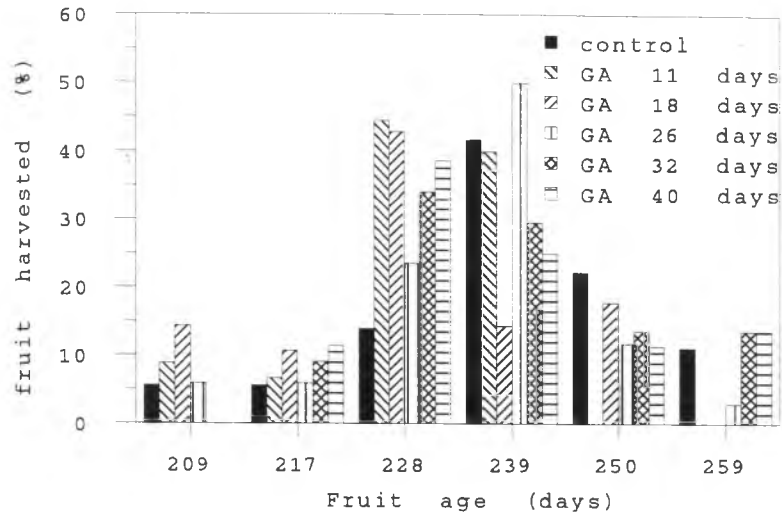


Figure 4.2. Percent ripe fruits of coffee harvested after 100 ppm GA application.

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Chapter 5
Anatomical Study on Fruit Separation of Unripe
and Ripe Coffee Fruits

Abstract

The fruit removal force (FRF) for detaching unripe (green) and ripe coffee (*Coffea arabica* L. 'Guadalupe') fruits from the stem by force pulling in parallel direction to the fruit axis were measured. The FRF to detach ripe fruits was significantly lower than the FRF for unripe fruits. The separation of green fruits was primarily at the junction between peduncle and stem (55%) and the junction between pedicel and peduncle (40%). Only a few green fruits (5%) detached at the junction between the pedicel and pericarp tissues. The separation of ripe fruit from the stem occurred at the junction between pedicel and pericarp tissues. None of ripe and unripe fruits abscised naturally in the greenhouse. Ripe fruits remained attached as long as no external force was applied even after the fruits dried completely. No abscission layer was found in the area where fruits detached. The separation of fruits may be entirely mechanical with cells breaking by the applied force.

Introduction

Coffee fruits at the same age (same day after anthesis) do not necessarily ripen at the same time. Schuch et al. (1990) found that most of the flowers (81%) reached anthesis about 10 days after a major rain (9 cm/day) in March but only 54% of fruits were harvestable in October. Fruits continued to ripen throughout November and December. Attempts to synchronize fruit development by chemical treatment at the early stages of fruit development were unsuccessful (Tongumpai, 1993, chapter 4). These studies suggest that synchronization of flowering or fruit development may not achieve synchronous fruit ripening.

The concept of harvesting coffee by machine is based on mechanical vibration and shaking of the limbs and fruits. The FRF and fruit weight are important factors used to indicate the ease of fruit separation (Wang and Shellenberger, 1967). The FRF of green (immature) fruit is always higher than that of the mature fruit. The differences in FRF of green and ripe fruits are cultivar dependent (Crisosto and Nagao, 1991). The selectivity of the machine to harvest ripe fruits only is dependent on the difference in FRF between ripe and green fruits. The machine's selectivity increases as the difference in FRF of ripe and unripe fruits increases. To alter the FRF of

coffee fruits to facilitate machine harvesting, a better understanding of the mechanism of fruit abscission is needed.

When ripe fruits are harvested in the field either by machine or hand-picked, the fruits were removed without the pedicel attached. The separation usually occurred at the junction between the pedicel and pericarp tissue. This may be due to an abscission layer, if present, in the area between the pedicel and pericarp. The detachment of the green fruit is different. Most of the harvested green fruits have the pedicel attached. These observations suggest the mechanism of fruit separation of green and ripe fruits is different.

The coffee plant is unusual among woody perennials due to its inability to shed "excess" immature fruits (Cannell, 1985). Ripe coffee fruits also do not abscise naturally and the reason for this is not clear (Opile and Browning, 1975). The fruits remain on the tree even after the fruits wilt and dry. The dried fruit can remain intact indefinitely unless removed mechanically, i.e. by hand or shaking. These observations question the presence of an abscission zone in the coffee fruit.

The abscission of reproductive structures in plants is characterized by a definite zone located either at the base of an organ or associated plant parts (Simons, 1973). The process of detachment of plant organs has been defined

by Esau (1960) as the abscission zone which is the zone at the base of leaves, fruits or other plant parts containing an abscission layer and the protective layer. The abscission layer is defined as a distinct layer of cells, the disjunction or breakdown of which separates a plant part from the plant.

Abscission is one mechanism of shedding plant parts (Addicott and Lyon, 1973). The process of abscission involves a group of biochemical steps that leads to the dissolution of cell walls in an abscission zone. Another mechanism of shedding involves the nonphysiological separation of plant parts. Addicott and Lyon (1973) stated that the variations in the pattern of shedding are based on morphologically and physiologically differences. Morphologically, some organs have a well-defined abscission zone while others have none; and physiologically, some organs can abscise with great rapidity while others lack any physiological mechanism to assist shedding.

The objective of this study was to determine the abscission mechanism of immature and mature coffee fruits. This study was a part of another project on developing procedures for the discriminate harvesting of mature coffee fruits by machine.

Materials and Methods

Six 3 year old coffee (Coffea arabica L. 'Guadalupe') plants produced in a greenhouse at Corvallis, Oregon were used in these studies.

Fruit removal force (FRF) of both unripe (green) and ripe fruits was measured with a hand-held digital force gauge (Shimpo FG-5.0, Shimpo America, Lincolnwood, Illinois) by placing the modified hook of the gauge around the pedicel at the base of the fruit and pulling the gauge parallel to the fruit axis. Between 10 to 20 ripe and green fruits of each tree were sampled. The FRF was read in pounds and converted to Newton by multiplying by 4.5. The point of separation for each fruit was recorded.

Anatomical studies of tissues at the separation area of both green and ripe fruits were done. Sections of pericarp, pedicel and peduncle tissues from green and ripe fruits were fixed in FAA solution (formalin: acetic acid: 50% alcohol 1:1:18). The tissues were dehydrated with a series of tertiary butyl alcohol and infiltrated in paraffin. Longitudinal sections (10 μ m thick) of the tissues were sliced with a microtome. Cross sections of pedicel tissues were also made to compare the development of the xylem of green and ripe fruits. The sections were stained with safranin and fast green as described by Johansen (1968) and observed under the light microscope.

Results

Fruit removal force and points of detachment

FRF of green fruits was significantly greater than the FRF of ripe fruits. The average FRF of green fruits was 9.05 ± 0.28 N while the FRF of ripe fruits was 3.86 ± 0.30 N. All ripe fruits (100%) detached from the tree at the junction between the pedicel and basal end of the pericarp tissue (Table 5.1 and Figure 5.1). This type of detachment was not observed in green fruits. The separation of green fruits occurred at various points along the pedicel. About half of the green fruits detached from the tree at the junction between the peduncle and stem (point D) and 40% separated between the pedicel and peduncle junction (point C). Only 5% of green fruits detached by breaking along the pedicel (group B) and none detached in the same manner as ripe fruits at the pedicel and pericarp junction (point A) (Table 5.1 and Figure 5.1).

Natural abscission of both ripe and green fruits was not found in the greenhouse throughout the period of study. Ripe fruits remained on the tree even when the fruits wilted and dried by aging. The dried fruits remained attached to the branches without dropping unless removed by an external force.

Detachment of ripe fruits

Longitudinal section of ripe coffee fruits at the junction between the pedicel and basal end of the pericarp revealed no abscission layer present in the area where the fruit separation occurred (Figure 5.2). Fruit detachment occurred by random breaking of parenchymatous tissue of the pericarp at about 1-2 mm below the pedicel and pericarp junction. After fruit detachment, parts of pericarp tissue remains attached to the pedicel (Figure 5.3). Cross section of the pedicel indicates a well developed xylem in the pedicel of ripe fruits (Figure 5.4).

Detachment of green fruits

Longitudinal sections of the pedicel and peduncle of green fruits showed no abscission layers along either the pedicel or peduncle tissue at any point where the fruits can possibly abscise. Fruit separation occurs most likely at the weakest point along the pedicel and peduncle tissue (Figure 5.5). The separation occurs at random along the pedicel (Figure 5.6). Cells of the green pericarp tissue below the pedicel are relatively small when compared to those of the ripe fruit (Figure 5.2).

Discussion

The FRF of green fruits were always greater than the FRF of ripe fruits. This finding confirmed the work of Crisosto and Nagao (1991). The location of the break point between the stem and the fruit of green and ripe fruits was different indicating the mechanism of fruit detachment for green and ripe fruits to be different.

The detachment of green fruits along the pedicel occurs primarily between either the pedicel and peduncle or peduncle and stem. Anatomical studies of this area and the pedicel did not reveal any abscission layer. Fruit separation at these points by physically pulling the fruits with the FRF gauge suggests those area to be the weak points between the stem and the fruit tissues. Occasional breakage along the pedicel of green fruits occurred at random.

In ripe fruits the point of detachment, by physical pulling of the fruits with the FRF gauge, was between the pedicel and base of the pericarp tissues. Anatomical observations of this area did not reveal the presence of an abscission zone. Comparison of the area between the pericarp and pedicel tissues of green and ripe fruits showed the parenchymatous cells of ripe fruits to be larger than those of the green fruits. The larger cells of the ripe fruits were weaker than the smaller cells,

therefore, when physical force was applied to ripe fruits the fruits detached at the point of the larger cells. Microscopic studies of the detached area revealed remnants of broken pericarp cells still attached to the pedicel and a tear in the pericarp tissue at the original point of attachment.

The lack of a definitive abscission zone at the point of detachment in green and ripe coffee fruits explains the reason why coffee fruits do not abscise naturally. Others have also observed that the arabica coffee fruit do not abscise naturally (Cannell, 1985; Opile and Browning, 1975), however, the reason for this phenomenon was not clear (Opile and Browning, 1975). Under field and greenhouse conditions the fruits of coffee did not abscise if no external force was applied. The fruits remained attached to the stem even after the fruits turned black and dried. Separation of fruits from the stem is completely mechanical.

Another reason for the difference in detachment point of ripe and green fruits may be due to the differences in the development of the xylem tissues. The pedicel of ripe fruits has well-developed xylem vessels while those of green fruit is not well-developed. This may explain why the break point of ripe fruits is between the pedicel and pericarp tissue and not along the pedicel tissue. In contrast, the poorly developed xylem vessels of the

pedicel of green fruits may explain why the break point of green fruits was along the pedicel rather than between pedicel and pericarp.

The lack of abscission zone between the fruit and stem tissues of green and ripe fruits of coffee presents a challenge for developing strategies for machine harvesting of ripe coffee fruits. Magnifying the difference between the FRF of green and ripe fruits would improve the discriminate harvesting of ripe fruits. Companion studies to either increase the FRF of green fruits by strengthening the pedicel tissues, or decrease the FRF of ripe fruits by weakening of the pericarp tissue has been tested. Experiments to increase the strength of the pedicel tissue of green fruits has not been successful to date (Tongumpai, 1993). Studies to weaken the pericarp tissue by hydration and fruit cracking under controlled greenhouse tests has been successful (Tongumpai, 1993). Field experiments are needed to verify the success of these procedures for discriminating the machine harvest of ripe coffee fruits only.

Table 5.1. Fruit removal force of green and ripe fruits and percent detachment at four sites between the fruit and stem tissues.

	FRF(N) 1/	Site of Detachment ^{2/}			
		A	B	C	D
Green	9.05 a	0	5	40	55
Ripe	3.86 b	100	0	0	0

1/ means separation by t-test, $p = 0.01$

2/ points of detachment (see Figure 5.1)

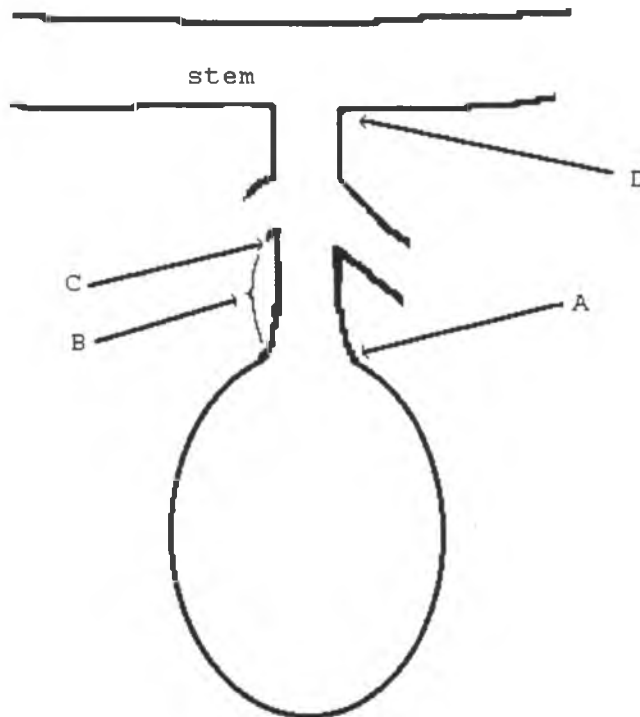


Figure 5.1. Diagram of fruit detachment sites.

- A = pedicel and pericarp junction
- B = along the pedicel but not at any junction
- C = pedicel and peduncle junction
- D = peduncle and stem junction

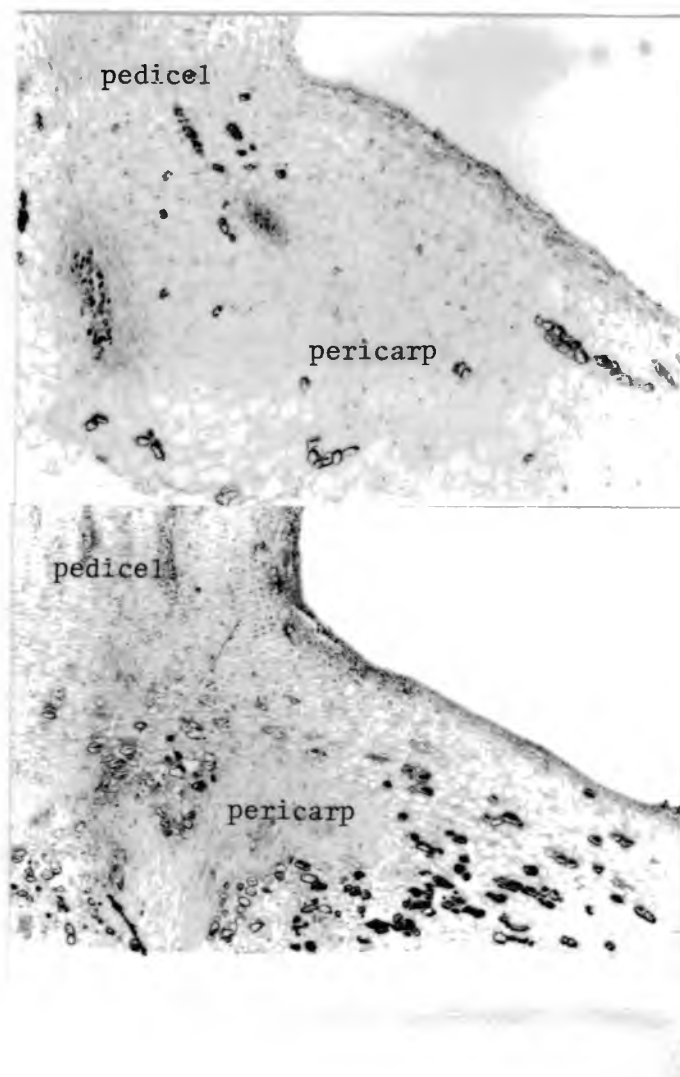


Figure 5.2. Pericarp tissues of ripe (top) and green (bottom) fruits adjacent to pedicel (magnification 40 X).

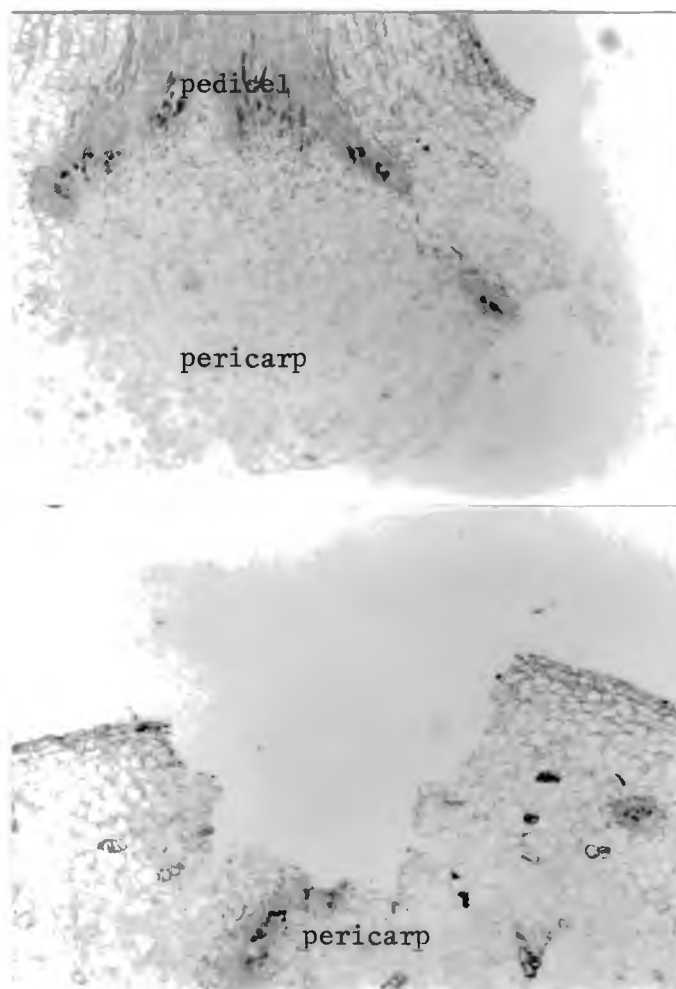


Figure 5.3. The separation of pericarp from the pedicel of ripe fruit. Remnant of the pericarp tissue still attached to the pedicel (top) and the hole in the pericarp (bottom) caused by physical detachment of fruit. Cell breakage occurred randomly in the separation area (magnification 40 X).

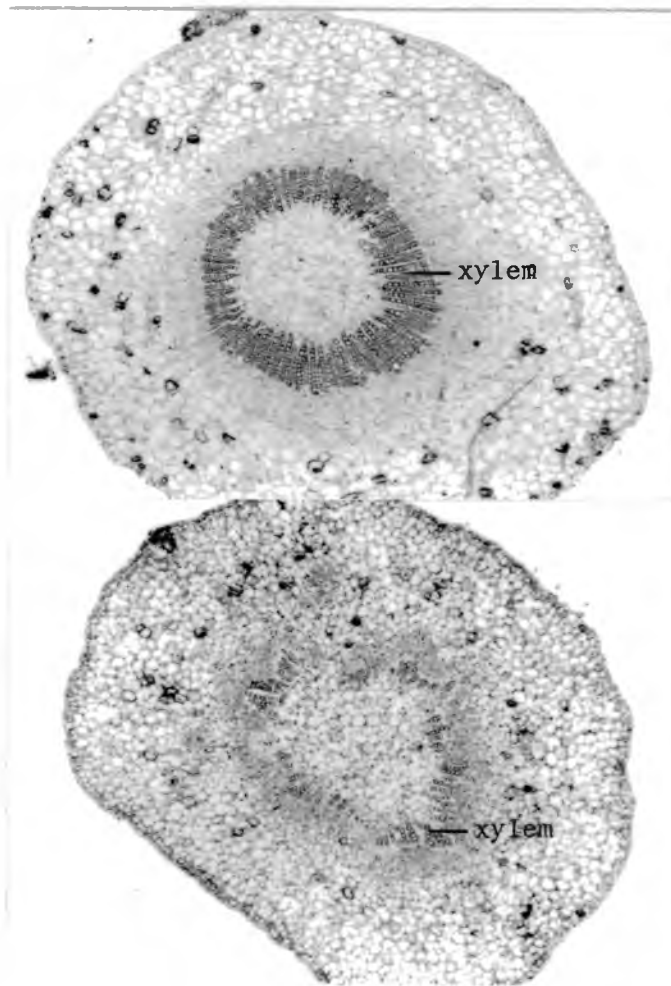


Figure 5.4. Cross-section of pedicel tissue of ripe (top) and green (bottom) fruits (magnification 40 X).

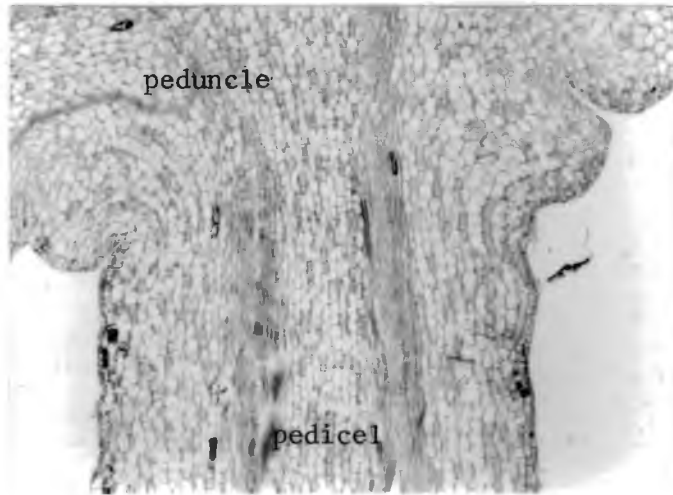


Figure 5.5. Pedicel tissues of green fruit between the pedicel and peduncle (magnification 40 X).

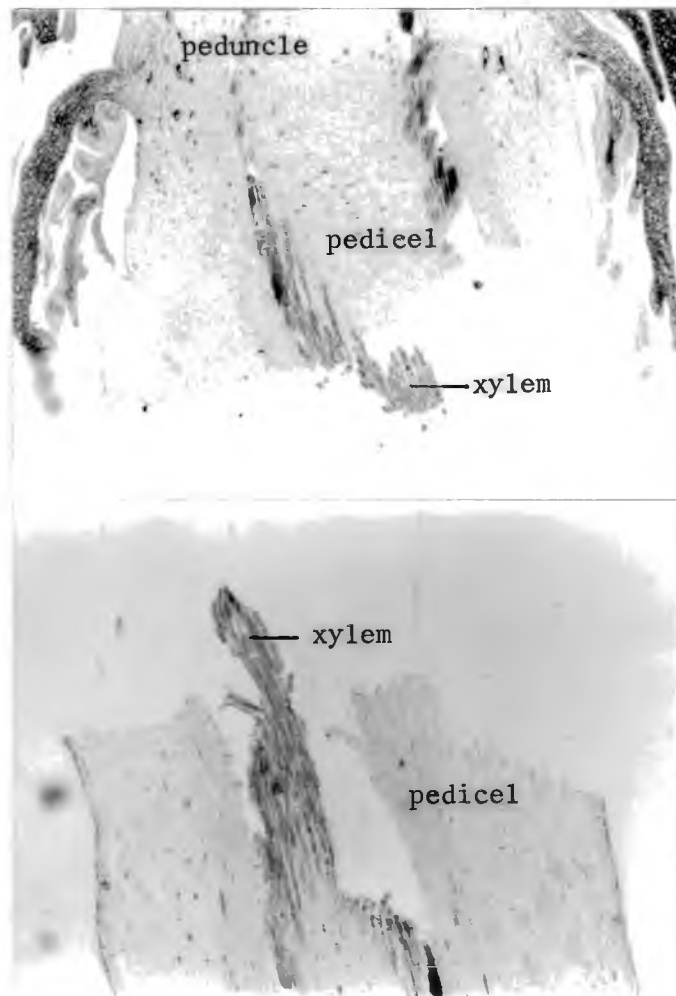


Figure 5.6. Part of peduncle and pedicel (top) tissues separated from the pedicel tissues (bottom) following physical detachment of green fruits (magnification 40 X).

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Chapter 6

The Effect of Ethephon on Fruit Ripening and Fruit Removal Force

Coffee (Coffea arabica L.) fruits that flowered on the same date do not necessarily ripen at the same time. These fruits ripen over a period of at least six weeks (Clowes, 1977b). Schuch et al. (1990) found that most flowers (81%) reached anthesis about 10 days after a major rain (9 cm/day) in March but only 54% of fruits were harvestable in October. Fruits were harvested from October to December. This implies that the synchronization of flowering does not lead to synchronous fruit ripening. Schuch et al. (1990) also reported that gibberellic acid (GA₃) significantly enhanced earlier anthesis of coffee flower buds and more uniform fruit ripening. GA₃ application to fruits from 11 to 40 days after anthesis did not completely synchronize ripening (Tongumpai, 1993, chapter 4).

Ethephon can enhance fruit ripening of many species including apple, cherries, tomato, blueberry (Thomas, 1982). Ethephon also enhances fruit ripening of coffee (Browning and Cannell, 1970; Crisosto and Grantz, 1989; Opile and Browning, 1975; Oyebade, 1976; Sondahl et al., 1974; Winston et al., 1992). Ethephon at 500 to 2000 ppm applied to the fruits accelerated maturation 15 days after

application (Sondahl et al., 1974). Oyebade (1976) found that a single dose of ethephon at 200 ppm and above increased ripening when applied to mature green fruits. The ripening was noticeable within six days after chemical application. Opile and Browning (1975) failed to synchronize fruit ripening with ethephon at 1400 ppm when applied to trees bearing crop from multiple flowering. High concentration of ethephon caused excessive leaf abscission (Winston et al., 1992). A rate of 250 ppm was marginally acceptable in terms of leaf abscission. Lower rates (i.e. 100 ppm) also caused 80% leaf abscission as reported by Crisosto and Grantz (1989).

The effect of ethephon on coffee fruit abscission has been reported by many (Browning and Cannell, 1970; Crisosto and Grantz, 1989; Opile and Browning, 1975; Oyebade, 1976). Winston et al. (1992) reported that ethephon at 125 to 2000 ppm did not cause significant fruit abscission. Browning and Cannell (1970) reported that ethephon treatment to fruiting coffee plants caused about 20% fruit drop compared with 7% from the untreated trees following vigorous shaking. Many of the fruits that fell were over-ripe and may have fallen because of their softer pericarp. This finding was confirmed by Oyebade (1976). Crisosto and Grantz (1989) reported that ethephon treatment caused 25% fruit removal force (FRF) reduction.

Unfortunately, in these studies the stage of fruit development at time of harvesting was not mentioned.

These studies suggested that ethephon may be useful in machine harvesting of coffee by inducing fruit ripening and lowering the FRF. Further studies on the effect of ethephon on quantitative color changes and the reduction in FRF at different times after application might be useful. The objective of this study was to determine the effect of ethephon on quantitative change in fruit color when applied to mature green fruits. The effect of ethephon on the change of FRF was also determined.

Coffee trees cv. Guadeloupe were grown in 4-l plastic containers in a mixture of pumice, peat, soil and sand (2:1:1:1) for 3 years in a greenhouse under day/night temperatures of 25/19 °C and natural photoperiod (Oregon, 44°N latitude). After the plants had flower buds at the 4 mm stage water deficit stress induced flower opening (Schuch, 1990). After anthesis in July 1992 the flowers were tagged and date recorded. The age of the fruit was determined after anthesis. Six plants induced to flower on the same date were used for the ethephon study. Three plants were sprayed with ethephon at 200 ppm and 0.1% Tween 20 to run off when the fruits were at 230 days old. Five fruits from each tree were picked every 15 days starting from the date of application. All data were analyzed with SAS by TTEST procedure. The FRF was measured

with the digital force gauge (Shimpo FG-5.0, Shimpo America, Lincolnwood, Illinois). The fruit was detached from the branch by placing the modified hook of the force gauge to the collar of the fruit and pulling in the direction parallel to the fruit axis.

The exocarp color of the harvested fruit was measured with a chromameter (Minolta CR-200b, Minolta Camera, Osaka, Japan) where the degree of lightness (L), redness (a^*) and yellowness (b^*) were determined. Hue was determined by the hue angle (θ) which equals to $\tan^{-1}b^*/a^*$ (Sapers et al., 1983). A high positive value of θ indicates a yellow to orange (with high degree of yellow) color and a low value indicates a red color (with less yellow). A negative θ value indicates a green color.

The juice of ripe fruit was extracted by squeezing the pericarp tissue with the hand. The juice of the pericarp of green fruits was extracted by freezing the tissue at -80°C in sealed centrifuge tubes and thawing at room temperature and centrifuging at 5000 rpm. The soluble solids of the juice was measured with a handheld refractometer.

The seeds were removed from the fruit and dried in the oven at 80°C for 48 hr for dry weight measurement.

Ethephon enhanced fruit ripening of 230 days old mature green fruit. The hue of the treated fruits were orange (high positive value) and red (low positive value

of hue) 15 and 30 days after treatment respectively and maintained their color throughout the study (305 days old after anthesis) (Figure 6.1). The hue of the control fruits remained negative (green color) 15 days after treatment. The majority of the control fruits were still green 260 days after anthesis (30 days after start of experiment). The variation in hue angle of the control fruits was quite large as compared with the ethephon treated fruits. The control fruits turned to the orange color (red with more yellow) 275 days after anthesis and became completely red after 290 days old. In contrast to the ethephon treatment the control fruits were 30 days later in acquiring the red color. High variation in hue of the control fruits was found. Ethephon has been reported to regulate color change of many fruits such as apple (Looney, 1983), blueberry (Thomas, 1982) and coffee (Bartholomew and Criley, 1983). This experiment confirmed earlier reports that ethephon can enhance color change in coffee fruits. Fruits ripened sooner and more uniformly with ethephon treatment. The pattern of fruit color change of both control and ethephon treatment was similar.

The FRF of fruits treated with ethephon was lowered throughout the test period (Figure 6.2). The difference in FRF between the controls and ethephon treatments was significant 15 days after the treatment ($p < 0.01$) and continued for 60 days after treatment. Thereafter the

difference became less obvious ($p < 0.05$). No significant difference in FRF between control and ethephon treatment was found in 305 days old fruits. A high correlation between fruit age and FRF of both treatment was found ($r = -0.85$, $p < 0.01$). Fruit abscission following the ethephon treatment was observed by others (Browning and Cannell, 1970; Opile and Browning, 1975; Oyebade, 1976). Most of these reports stated that the fallen fruits were overripe and had softer pericarp. No fruit abscission was found in this experiment. Winston et al. (1992) found that ethephon at 125 to 2000 ppm did not cause significant fruit abscission. In a companion, no abscission layer was found at the breakage point of either green or ripe fruits (Tongumpai, 1993, Chapter 5). Physical force was necessary to remove the fruits from the stem.

The soluble solids content of the pericarp was related to fruit maturity ($r = 0.88$, $p < 0.01$). The soluble solids content of the ethephon treated fruits was higher than the control fruits 15 to 30 days after treatment ($p < 0.01$). This coincided with changes in the exocarp color. A high negative correlation between soluble solids content and degree of fruit yellowness (b-value) was found ($r = -0.92$, $p < 0.01$) (data not shown). No difference in soluble solids content of the ethephon and control fruits was found in the 275 days old fruits (Figure 6.3). Soluble solids content was also negatively

correlated with FRF ($r = -0.91$, $p < 0.01$). This study confirms other studies that fruit color and soluble solids content of pericarp can be used as indices of fruit maturity and FRF (Tongumpai, 1993, chapter 7).

Seed dry weight increased as fruit maturity increased ($r = 0.78$, $p < 0.01$) (Figure 6.4). Ethephon did not affect seed dry weight up to 45 days after treatment. Thereafter the seed dry weight of the ethephon treatment was lower than the control ($p = 0.05$). Winston et al. (1992) suggested that ethephon treatment had no effect on seed weight when applied at physiological seed maturity. Clowes (1977a) suggested that ethephon should only be applied when the seeds had completed expansion and were in the filling and ripening stage of development as described by Wormer (1964). Early ethephon application causes uneven and premature ripening of fruit and lowers the quality of the seeds.

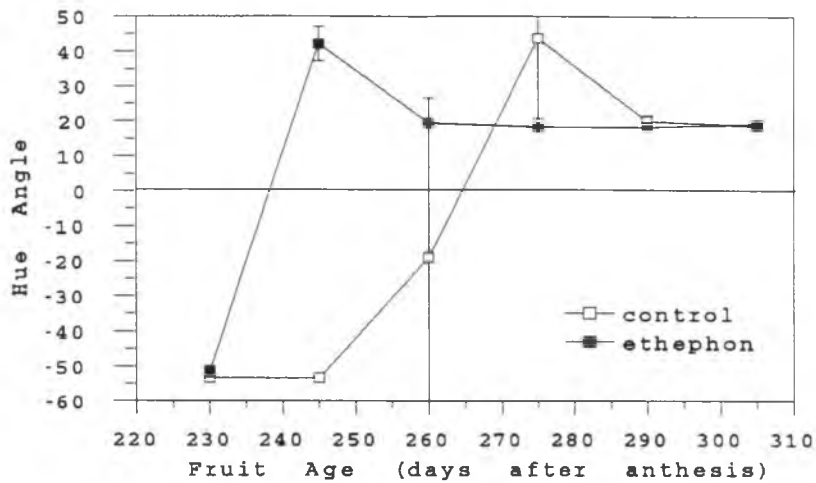


Figure 6.1. Effect of 200 ppm ethephon on fruit color change as determined by the change of hue angle (θ), where $\theta = \tan^{-1} b/a$.

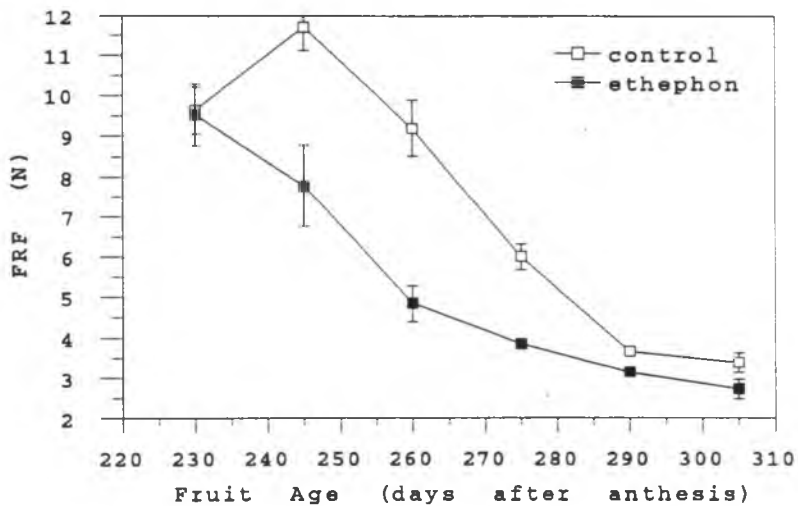


Figure 6.2. Effect of 200 ppm ethephon on FRF at different fruit age.

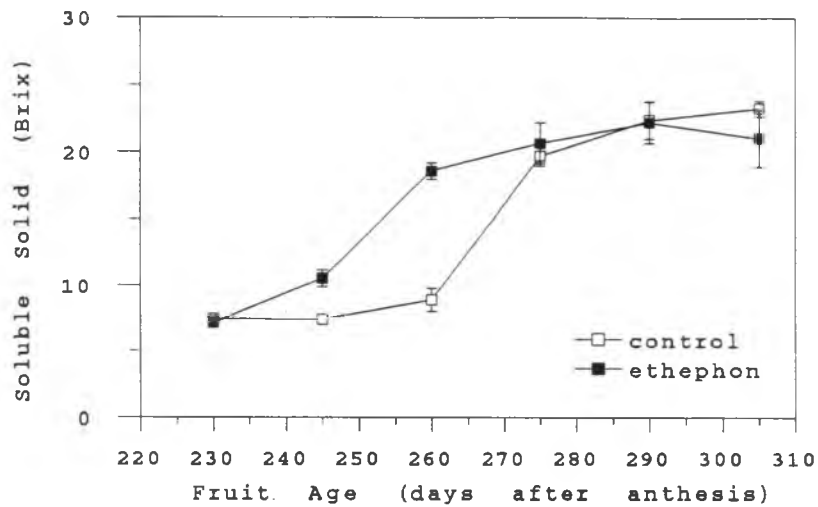


Figure 6.3. Effect of 200 ppm ethephon on the soluble solids content of the pericarp at different fruit age.

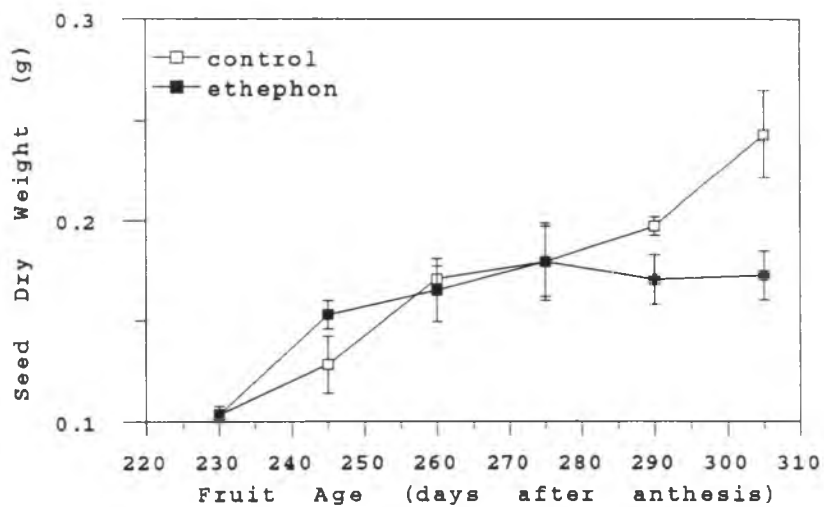


Figure 6.4. Seed dry weight at different fruit age after 200 ppm ethephon application.

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Chapter 7

Harvest Indices for Machine Harvesting of Coffee

(Coffea arabica L. 'Guadalupe') FruitsAbstract

Several indices of fruit maturity were related to the FRF of Coffea arabica L. 'Guadalupe' fruits. Green fruit FRF remained constant at 8.9 ± 0.3 N regardless of age and decreased as the fruit turned red. The FRF of red fruits continued to decrease with increasing fruit age. Percent seed dry weight of ripe fruits did not change with fruit age.

Fruit color expressed as the degree of lightness (L) and hue, correlated well with the FRF. High negative correlation between FRF and soluble solids content of the pericarp was found. Pericarp water potential and osmotic potential significantly decreased with fruit age. These parameters were correlated with the soluble solids content of the pericarp. Turgor pressure of fruit remained constant throughout the ripening period. These studies suggest the possible use of color and soluble solids content as harvest indices for machine harvesting of coffee fruits.

Introduction

Harvesting machines for coffee fruits have been developed and are being used commercially. These machines have vibrating fingers that reach through the foliage and branches of the plant (Mitchell, 1988). Over 95% of the crop is removed indiscriminately by the machines. Wang (1965) found that the ratio of FRF and fruit weight decreased as fruits matured, indicating that some degree of selectivity is possible. Monroe and Wang (1968) stated that the selectivity could be improved by adjusting the frequency of the vibrating fingers . A lower frequency is better when the percent of ripe fruit on the tree at harvest time is low.

The difference between fruit removal force (FRF) of green and ripe fruit is more important than the absolute magnitude of either. Crisosto and Nagao (1991) found differences in FRF among different cultivars of coffee. They suggested consideration of these differences when selecting coffee cultivars suited for machine harvest. The FRF of green fruit was always greater than the ripe fruit regardless of cultivars.

The quality of coffee is related to fruit maturity. Fully red fruits have the best quality (Mitchell, 1988). The quality of under-ripe (green) and over-ripe (black) fruits is reduced. Thus, in order to obtain the highest

quality of coffee only fully ripe fruits should be harvested. To achieve this goal by machine harvesting, the FRF between unripe and fully ripe fruits must be great enough for the machine to discriminate between them.

Wormer (1964) found that coffee beans attained their full dry weight 30 weeks after anthesis or about 6 weeks before picking. The fruit begins to change color after the 30th week. The color changes from green to yellow to red. Fruits can be harvested at any time during this ripening period without much effect on quality as long as the seed can be separated from the pericarp by pulping process (David Roche, personal communication). In Hawaii, ripe fruits can retain their quality on the tree for more than 3 weeks. One of the first symptoms of fruit maturation is color change. Color change has been used successfully as an index for harvesting fruits, i.e. sweet cherries (Drake et al., 1982). Watada and Abbott (1975) found a good correlation between soluble solids content and anthocyanin content in grape berry. This close association was anticipated due to the increased concentration of both with fruit ripening.

Anthocyanin is the main pigment of coffee (Mazza and Miniati, 1993). As the fruits ripen, as in the case of tomato fruits, the accumulation of simple sugars in the pericarp increases (Picha, 1987). With increased fruit ripening, tissue softening occurs followed by fruit drop

and senescence (Ahrens and Huber, 1990; Kojima et al., 1991).

In coffee, there is no abscission layer in the fruit pedicel. The fruits detach from the pedicel by randomly breaking at the junction of the pedicel and pericarp tissue (Tongumpai, 1993). Due to the absence of an abscission zone I have hypothesized that the reduction in FRF was due to either fruit tissue softening as fruits aged or the increase in turgor pressure as the sugar content increased during ripening. The objective of this study was to determine the changes of fruit properties during fruit maturation and their relationship to changes in FRF.

Materials and Methods

Eight potted coffee plants (Coffea arabica L. 'Guadalupe') produced in the greenhouse at Corvallis, Oregon (44° N latitude) were used in this study. Each flower cluster was tagged with a small paper label fastened with a string at anthesis. After 260 days, the FRF of five fruits from each tree was measured by pulling the fruit parallel to the fruit axis with a digital force gauge (Shimpo FG-5.0, Shimpo America Corp., Lincolnwood, Illinois) until the fruits detached from the pedicel as described by Cooper and Henry (1973). The force required to detach the fruits from the pedicel was recorded as the fruit removal force (FRF) in Newtons. All measurements were done at 10 day intervals until the fruits were 330 days after anthesis.

Fruit color was measured with a chroma meter (Minolta CR-200b, Minolta Camera, Osaka, Japan) and reported according to the Judd-Hunter system (Francis, 1970). The L value represents the degree of lightness or darkness. The negative value of a* indicates the greenness while the positive value represents the red color. The b* value indicates the blueness (negative b*) and yellowness (positive b*). Hue angle (θ), which reflects the shade of color, is calculated as θ equals to $\tan^{-1} b^*/a^*$ (Sapers

et al., 1983). Exocarp color was presented as L and Hue as suggested by Francis (1980).

The wet-loop thermocouple psychrometer was used to measure the fruit water potential and osmotic potential of two fruits from each of four plants. The proximal end of each fruit was cut into a 1-cm disk and placed in a psychrometer cup. The procedures of measurement were described by Matthews et al. (1987). Pericarp turgor pressure was estimated by the difference between pericarp water potential and osmotic potential (Turner, 1981).

The juice from the pericarp was extracted by hand for soluble solid measurements. Soluble solids content of pericarp tissue was measured with a hand-held refractometer (Fisher) and expressed as degree Brix.

The fresh and dry weights of seed of 3 fruits from each of 4 plants were studied. The seeds were dried in an oven at 80°C for 48 hr. The percent seed dry weight was calculated by the following equation:

$$\text{Percent dry weight} = 100 - \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100$$

Four plants were used for the water content and water absorption study. Five fruits from each tree were harvested at 10 day interval. Two fruits each were used to determine fruit weight at full turgor and relative water content. A fruit disk was sliced from the proximal end of each fruit to approximately 1 cm diameter, weighed and

floated in distilled water for 24 hr. The fruit disk was weighed and dried in the oven at 80° C for 48 hr and the dry weights determined. The initial fresh weight (FW), full turgid weight (TW), and dry weight (DW) were used to calculate the relative water content (RWC) of pericarp tissue as described by Turner (1981):

$$\text{RWC} = \frac{(\text{FW} - \text{DW})}{(\text{TW} - \text{DW})} \times 100$$

Tissue water absorption capacity after 24 hours was determined by the following:

$$\text{water absorption capacity} = \frac{(\text{TW} - \text{FW})}{\text{FW}} \times 100$$

Three fruit disks obtained from separate fruits from each of four trees were used for pericarp dry matter content determination. The fruit disks were dried in the oven at 80° C for 48 hr. The pericarp dry matter content was determined on a fresh weight basis as follows:

$$\text{dry matter content} = 100 - \left(\frac{\text{FW} - \text{DW}}{\text{FW}} \right) \times 100$$

Correlations between fruit removal force, fruit color, soluble solids content, and fruit water relation were studied. All data were analyzed with SAS by GLM procedure.

Another set of twelve coffee plants with green fruits (unspecified ages) was used for determining FRF of immature fruits. FRF of twenty fruits from each tree were measured with the hand-held digital force gauge as

described above. The means and standard error of FRF of green fruits was determined.

Results

Fruit color change

Exocarp color of green fruits as determined by the degree of lightness (L) and hue (θ) remained constant until ripening occurred (data not shown). When the fruit turned red the hue angle became positive. As fruit maturation (X) increased the red color became darker as indicated by the decrease in L-value (Y_1), where $Y_1 = 246.2 - 1.3X + 0.002X^2$; $p < 0.01$ $R^2 = 0.71$ (Figure 7.1). The color change from orange to red and deep red with fruit maturation was indicated by the change in hue angle (Y_2) with fruit age (X), where $Y_2 = 217.2 - 1.3X + 0.002X^2$; $p < 0.01$, $R^2 = 0.42$.

Ripe fruits remained on the tree, under greenhouse conditions, for over 2 months before turning black and dehydrating. Fruit color correlated well with the FRF (Figure 7.4). The L-value and hue angle decreased as the FRF decreased (Figure 7.4). The relations between L-value (Y_1), hue angle (Y_2) and FRF (X) were $Y_1 = 40.1 + 0.23X^2$ ($p < 0.01$, $R^2 = 0.77$), and $Y_2 = 21.5 - 1.9X + 0.3X^2$ ($p < 0.01$, $R^2 = 0.53$).

Fruit removal force (FRF)

The FRF of green fruits remained constant at 8.9 ± 0.32 N regardless of age. When the fruits began to turn red at 250 days after anthesis the FRF decreased significantly as fruits matured (become redder). The FRF 330 days after anthesis decreased by more than twice that of the fruit at 260 days (Figure 7.2). The relation of FRF with fruit age was $Y = 74.2 - 0.43X + 0.00064X^2$ ($p < 0.001$, $R^2 = 0.74$), where $Y = \text{FRF}$ and $X = \text{fruit age}$.

Pericarp soluble solids content

The soluble solids content of the pericarp significantly increased as fruit age increased. The change of soluble solids content with age was curvilinear (Figure 7.2). A good correlation between soluble solids content and fruit age was found, $Y = -246.4 + 1.8X + 0.003X^2$ ($p < 0.01$, $R^2 = 0.78$), where $Y = \text{soluble solids content (Brix)}$ and $X = \text{fruit age (days)}$. A linear relationship between FRF and SS content was found (Figure 7.3), $Y = 12.0 - 0.3X$ ($p < 0.001$, $R^2 = 0.67$), where $Y = \text{FRF}$ and $X = \text{soluble solids content of the pericarp tissue}$.

Pericarp water absorption and relative water content

The correlation between pericarp water absorption capacity and fruit age was highly significant, $Y = -1704 + 11.4X - 0.2X^2$ ($p < 0.001$, $R^2 = 0.86$), where Y = water absorption capacity and X = fruit age (Figure 7.5). The correlation between RWC and fruit age was similarly high but the inverse of the above relationship [$Y = 1114 - 6.9X + 0.1X^2$ ($p < 0.001$, $R^2 = 0.64$), where Y = RWC and X = fruit age] (Figure 7.5).

A high correlation between RWC and soluble solids content in the pericarp tissue was found, $Y = 127.1 - 2.57X$ ($p < 0.001$, $R^2 = 0.84$), where Y = RWC and X = soluble solids content (Figure 7.6). A high correlation between water absorption capacity of pericarp tissue and soluble solids content was also found, $Y = -70.3 + 4.7X$ ($p < 0.001$, $R^2 = 0.89$), where Y = water absorption capacity and X = soluble solids content (Figure 7.6).

Percent dry matter content of the pericarp (Y) gradually increased with fruit age (X) ($Y = -5.3 + 0.1X$ ($p < 0.01$, $R^2 = 0.92$)) (Figure 7.7), however, no correlation between percent dry matter and soluble solids content was found.

Pericarp water relation

Pericarp water potential (Y_1) and the osmotic potential (Y_2) significantly decreased with fruit age (X) ($Y_1 = 46.6 - 0.3X + 0.0005X^2$; $p < 0.01$, $R^2 = 0.60$, and $Y_2 = 43.7 - 0.3X + 0.0005X^2$; $p < 0.01$, $R^2 = 0.66$ respectively) (Figure 7.8). The estimated turgor pressure of the pericarp tissue remained constant regardless of fruit age.

A high correlation between the osmotic potential and soluble solids content of the pericarp was found ($r = 0.86$). Osmotic potential can be estimated by $Y_2 = -0.13X$ ($p < 0.001$, $R^2 = 0.75$) where X = soluble solids content (Brix). Soluble solids content can also be used to estimate fruit water potential by $Y_1 = -0.11X$ ($p < 0.001$, $R^2 = 0.58$) (Figure 7.9).

Seed dry weight

Seed dry weight remained constant between 260 and 330 days after anthesis. The average seed dry weight throughout the study was 0.45 ± 0.02 g/seed (Figure 7.10).

Discussion

FRF of the green fruit, younger than 250 days after anthesis, remained constant at 8.9 ± 0.1 N regardless of age. FRF significantly decreased as fruit age increased after the development of the red coloration. The difference in FRF of green and ripe fruit increased with maturation. The difference in FRF between the green and ripe fruits may be important for machine harvesting of mature fruits. Under greenhouse conditions ripe fruit could be maintained on the tree for over 2 months. The prolonged period of ripening without fruit drop would allow a greater proportion of the fruits to ripen and permit a greater percentage of ripe fruits to be machine harvested at reduced FRF.

The reduction in FRF with fruit maturation may be the consequence of changes in the pericarp tissue. The decrease in FRF was not due to turgor pressure changes of the pericarp tissue. The pericarp turgor pressure remained constant throughout fruit maturation. The maintenance of low pericarp turgor pressure throughout the fruit aging was also observed in grape (Matthews et al., 1987). The reduction in FRF may be due to fruit softening. During tomato fruit ripening, the fruit firmness decreased inversely with the increase in polygalacturonase enzyme

activity (Ahrens and Huber, 1990; Crookes and Grierson, 1983).

Percent seed dry weight remained constant throughout the ripening process. This may be because there was no significant change in seed content after the fruit turned red. This confirmed the finding of Wormer (1964) who stated that the beans attained their final dry weight when the berry was still green.

Soluble solids content of fruits may be used to indicate fruit maturity. Horton (1992) related fruit firmness to soluble solids content and fruit ripening of peach. Both fruit firmness and soluble solids content changed with maturity stages. The same is also true for coffee. The soluble solids content of coffee pericarp increased with fruit maturation. A high correlation between pericarp soluble solids content and FRF was found suggesting that pericarp soluble solids content may be used as an index of maturity for machine harvesting.

Fruit color was also a good indicator of fruit maturity. The correlation between fruit color (degree of lightness and hue), fruit age, and FRF was significantly high. In contrast to soluble solid measurements, the quantitative measurement of color in the field is not practical. The reduction in L-value as measured with the chromameter indicated that as the fruit matured, the color

became darker. Riper fruits acquired a deeper red color as compared with the less mature fruits.

Pericarp water potential decreased with fruit maturity. This change corresponded to changes in pericarp osmotic potential and can be estimated by the pericarp soluble solids content. A high negative correlation between osmotic potential and the refractive index (expressed as soluble solids content in term of °Brix) was found ($r = -0.74$). This may be due to the solute content of ripening fruit which is largely simple sugars as in tomato (Picha, 1987). The refractive index, therefore, can be used to estimate the osmotic potential of the pericarp as in the case of grape (Matthews et al., 1987).

The osmotic potential of the pericarp tissue was as low as -3.6 MPa and water potential was -3.3 MPa for the 310 days old fruit. Similar results were obtained on other fruits, e.g. grape (Considine and Brown, 1986; Matthews et al., 1987). In ripe grape fruits the osmotic potential was -3.0 to -4.0 MPa while the turgor pressure was as low as 0.1 to 0.4 MPa (Matthews et al., 1987). The turgor pressure of tomato fruit was also much less than expected, based on tissue osmotic potential (Shackel et al., 1991). These low turgor values may indicate the presence of apoplastic solutes. The turgor pressure of coffee pericarp was as low as 0.1 to 0.4 MPa while the osmotic potential was below -2.2 MPa. This was probably due to apoplastic

solutes as reported in other species, e.g. sugarcane parenchyma of the storage stalk (Moore and Cosgrove, 1991), sugar beet root (Perry et al., 1987; Wyse et al., 1986) and probably tomato fruit (Shackel et al., 1991).

The RWC of the pericarp decreased as fruit aged. This suggested that the pericarp water absorption capacity increased as fruits aged. This change corresponded to the changes in water potential and osmotic potential of the pericarp tissue. To absorb water, fruits with low RWC must be in direct contact with free water or be in a high humidity environment. In these situations fruits absorb water readily and if enough water is absorbed fruit cracking may occur due to excessive turgor pressure buildup. Fruit cracking caused by high internal turgor pressures generated during rainfall or irrigation occurs in many plant species with soft, fleshy pericarp (Abbott et al. 1986; Considine and Brown, 1986; Considine and Kriedemann, 1971). In coffee, controlled induction of fruit cracking may be used as a strategy for discriminate machine harvesting of mature coffee fruits. The large differences of osmotic potential and soluble solids content of ripe and green fruits causes cracking of ripe fruit only following rainfall and irrigation. Cracking of the pericarp tissue reduces the FRF. Because the coffee pericarp tissue is not important and discarded, induction of pericarp cracking should be an acceptable method of

harvesting ripe fruits without affecting coffee bean quality.

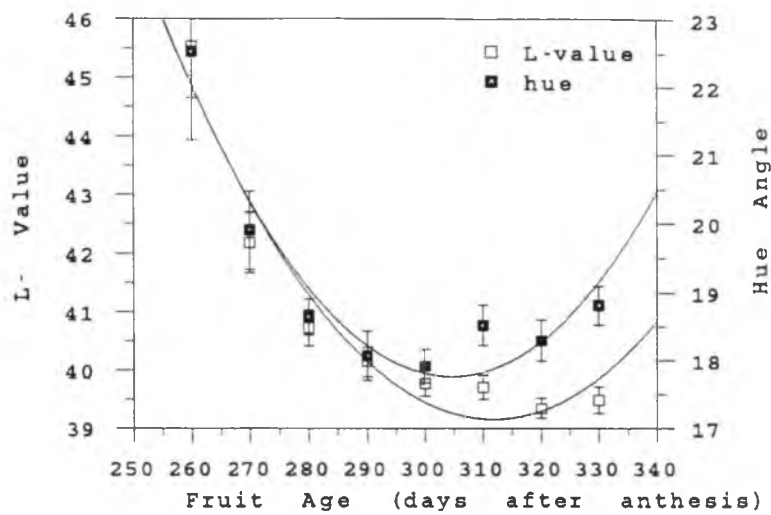


Figure 7.1. Change in fruit color (determined with a Chromameter) with fruit age. Each point is the mean of 40 fruits from 8 plants.

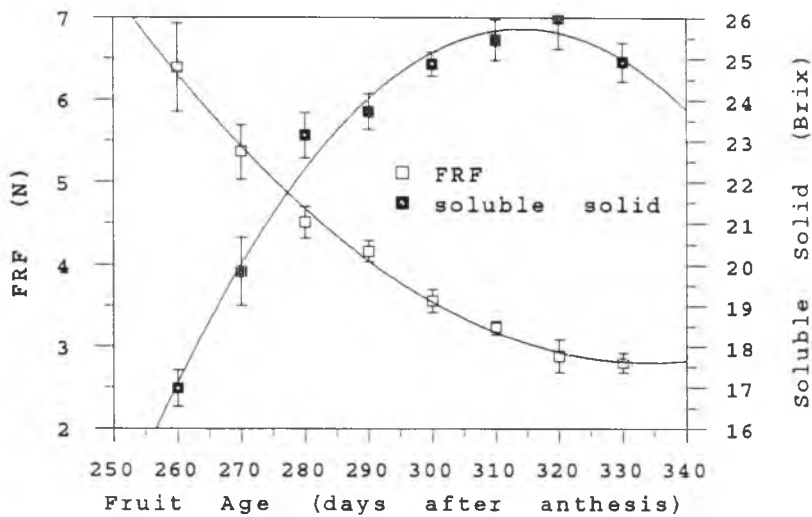


Figure 7.2. Changes of FRF and soluble solids content of the pericarp with fruit maturation. FRF (Y_1) decreased as fruit age increased, $Y_1 = 74.2 - 0.43X + 0.0006X^2$; $R^2 = 0.74$. Soluble solids content of the pericarp (Y_2) changes inversely with increasing fruit maturity, $Y_2 = -246.4 + 1.8X + 0.003X^2$; $R^2 = 0.78$. Each point represented 40 measurements.

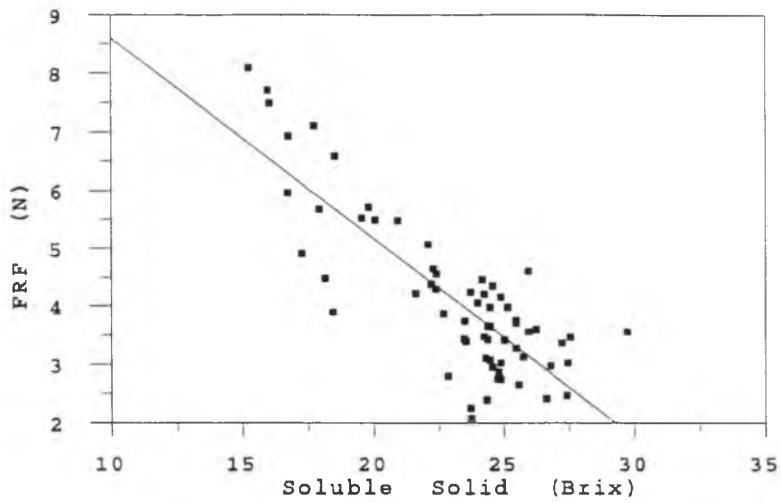


Figure 7.3. Relationship between FRF and soluble solids content of pericarp. FRF decreased as soluble solids content in pericarp increased, $Y = 12 - 0.3X$; $R^2 = 0.67$. Each point is the mean of 5 fruits.

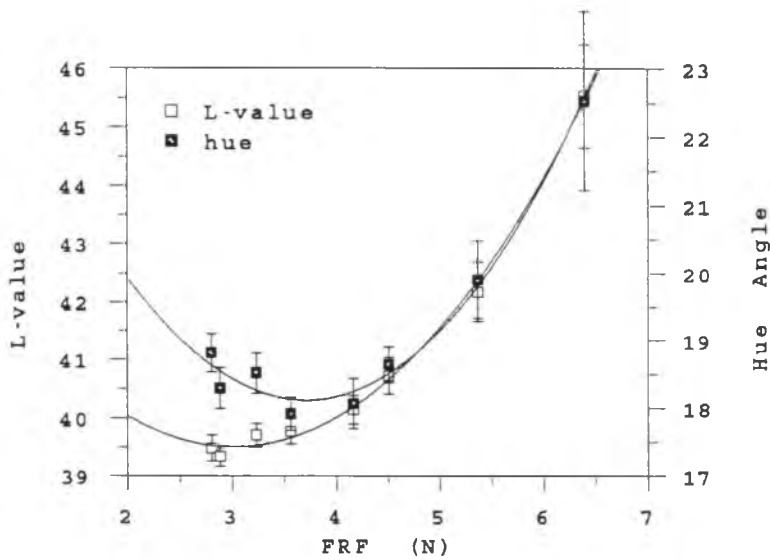


Figure 7.4. Relationship between FRF and fruit color, L and hue angle (determined by a Chromameter). Each point is the mean of 40 fruits from 8 plants.

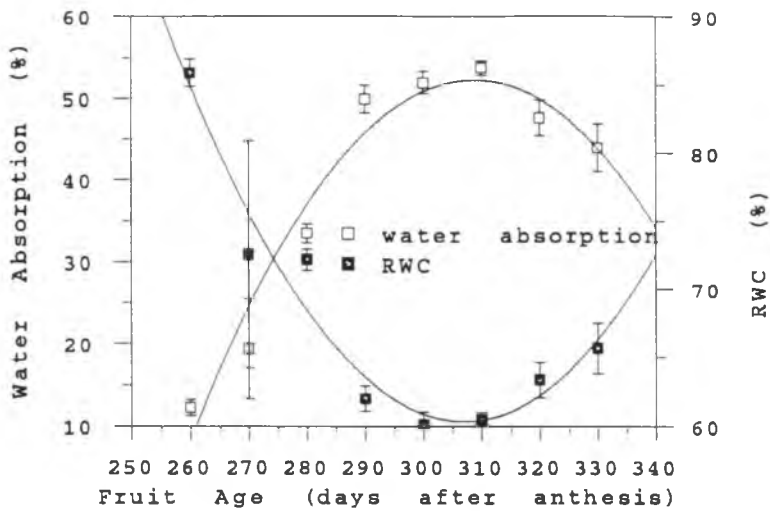


Figure 7.5. Correlation between percent water absorption of pericarp (Y_1) with fruit age, $Y_1 = -1704 + 11.4X - 0.04X^2$ ($R^2=0.86$) and percent RWC (Y_2) with fruit age, $Y_2 = 1114 - 6.86X + 0.01X^2$ ($R^2=0.64$). Each point is the mean of 12 fruits from 4 plants.

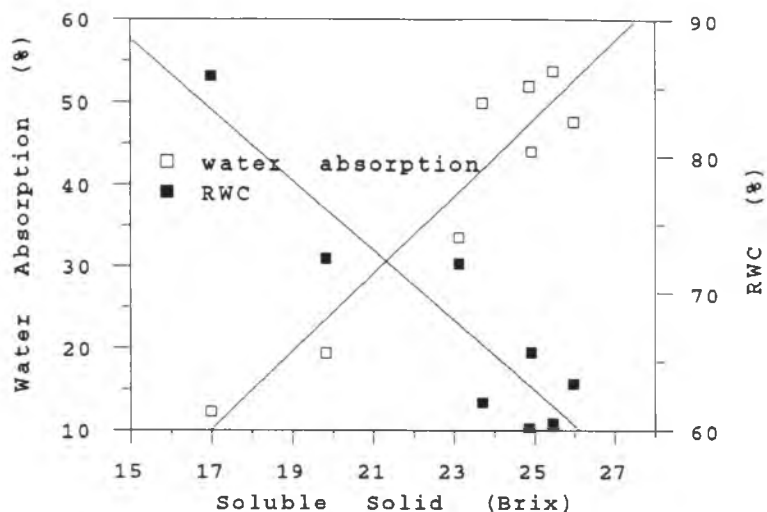


Figure 7.6. Relationship between RWC, water absorption capacity and soluble solids content of pericarp tissue. RWC decreased as soluble solids content increased ($Y = 127.1 - 2.57X$; $R^2 = 0.84$) while water absorption capacity increased as soluble solid increased ($Y = -70.3 + 4.7X$; $R^2 = 0.89$). Each point is the mean of 12 fruits from 4 plants.

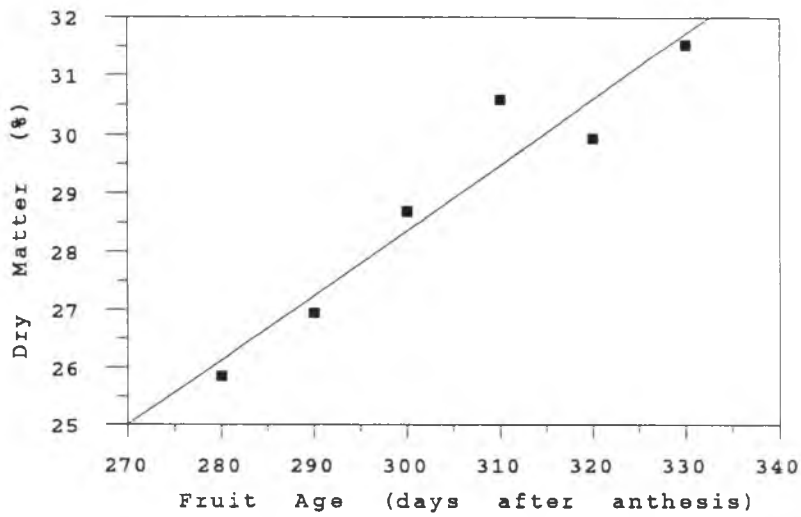


Figure 7.7. Relationship of dry matter content (%) of pericarp and fruit age, $Y = -5.3 + 0.1X$ ($R^2 = 0.92$). Each point is the mean of 8 fruits from 4 plants.

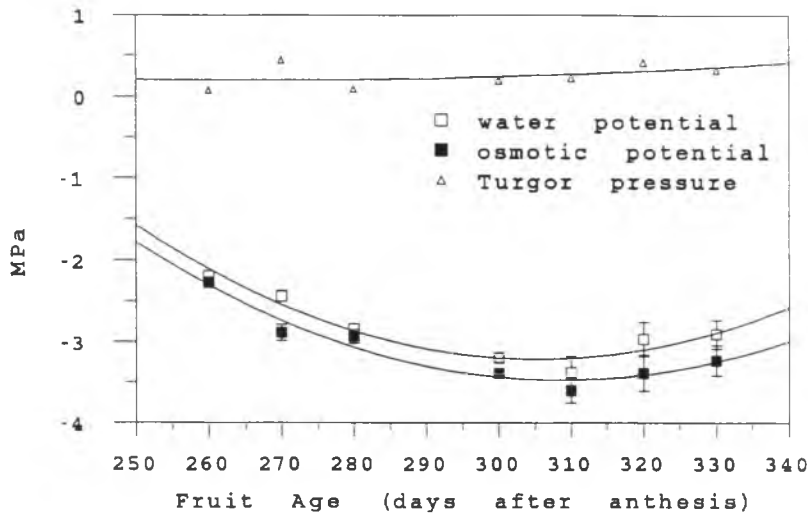


Figure 7.8. The relations of water potential (Y_1), osmotic potential (Y_2), and turgor pressure with fruit age ($Y_1 = 46.6 - 0.33X + 0.0005X^2$; $R^2 = 0.60$ and $Y_2 = 43.6 - 0.30X + 0.0005X^2$; $R^2 = 0.66$). Each point is the mean of 8 fruits from 4 plants.

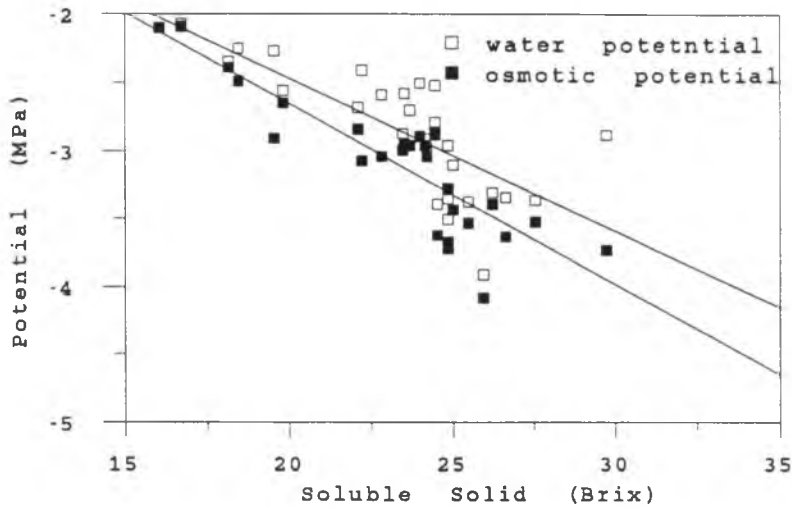


Figure 7.9. Relationship between soluble solids content of pericarp and water potential (Y_1) and osmotic potential (Y_2). Each point represented 2 measurements.

$$Y_1 = -0.12X; R^2 = 0.58$$

$$Y_2 = -0.13X; R^2 = 0.74$$

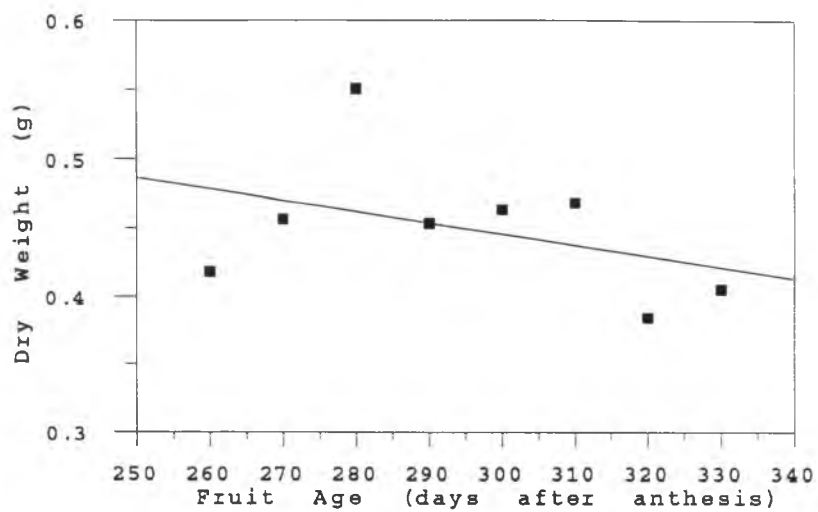


Figure 7.10. Change of seed dry weight with fruit age
Each point is the mean of 12 fruits from 4
plants.

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Chapter 8

Osmotic Movement of Water into Pericarp Tissue of Coffee
Fruits and Fruit Removal ForceAbstract

The movement of water into the pericarp tissue of coffee (Coffea arabica L. 'Guadalupe') fruit and its effect on fruit removal force was studied. Mist irrigation was applied to coffee plants with 270 and 305 days old fruits. Water relations and fruit removal force (FRF) of mature coffee fruits were studied after 24 to 72 hr of continuous irrigation. The FRF of ripe coffee fruit decreased as the duration of mist irrigation increased from 24 to 72 hr. Percent fruit cracking and pericarp water content increased with increasing irrigation. The viability of the pericarp tissue decreased linearly as the duration of irrigation increased from 24 to 72 hr. The turgor pressure of the pericarp increased with increasing irrigation. The increase in turgor and cracking of the pericarp tissue may be the cause of the lowering FRF following irrigation. Without mist irrigation, the FRF of ripe fruits decreased with increasing maturity. After mist irrigation the FRF of ripe fruits at all ages was reduced significantly. Mist irrigation had no effect on the FRF of green fruits.

Introduction

Coffee fruits do not generally ripen at once (Goto and Fukunaga, 1956). Fruits at several stages of development may be found on any one tree. This creates a serious problem for machine harvest.

The FRF of coffee is high compared to other fruits having about the same weight (Wang, 1965). Since coffee fruits do not ripen uniformly, fruit removal equipment must be designed to be selective in harvesting the mature fruits only (Monroe and Wang, 1968). The selectivity of the harvester can be improved by adjusting the frequency of the shaker to accommodate the FRF of mature and immature fruits. It is possible to harvest ripe fruit selectively by adjustment of the vibration amplitude and frequency (Willson, 1985). Unfortunately, even with the adjustment, the amount of immature fruits harvested was still high, ranging from 15 to 40%, depending on condition at harvest time. Since machine harvesting of coffee is based on mechanical vibration, the ratio of the FRF to fruit weight ratio (FRF/W) has been used to determine the ease of detachment (Wang and Shellenberger, 1967). Crisosto and Nagao (1991) found that the FRF of different cultivars of coffee differed and the FRF of green fruits was always higher than the ripe fruits.

The mechanism of fruit detachment of green and ripe fruits is different. Green fruits can be detached at any point along the pedicel or at the junction between the pedicel and main stem (Tongumpai, 1993). The FRF of green fruits does not vary. This is not the case for the ripe fruits. Ripe fruits always detach at the junction between the pedicel and pericarp with parts of pericarp tissue attached to the pedicel. The detachment was due to random breakage of the pericarp tissue, rather than breakage at a specific abscission layer (Tongumpai, 1993).

Reduction of the FRF of ripe fruits may be accomplished by weakening of the pericarp cells (Tongumpai, 1993). The difference in FRF between green and ripe fruits increases with increasing fruit maturity. Therefore, delaying harvesting of ripe fruits as long as possible should improve the discriminate harvesting of ripe fruits by machine. Procedures of increasing the difference in FRF between green and ripe fruits should favor the selective machine harvesting of ripe fruits.

The FRF of ripe coffee berries under water stress increased linearly as the xylem water potential decreased. In contrast, no change in FRF of green fruits occurred with changes in xylem water potential (Tongumpai, 1993). Crisosto and Nagao (1991) reported that the FRF of mature fruits increased during the day and was lowest at early morning. This may imply that the FRF of ripe fruits

increases as the fruit turgor pressure decreases due to water stress. In other word, one possible way to reduce the FRF of the ripe fruit is to increase fruit turgor pressure.

The fruits of many plant species bearing soft, fleshy pericarp are subject to skin-cracking disorders caused by high internal turgor pressures generated during rainfall (Considine and Brown, 1986). Cherry fruit cracking is a major problem caused by excessive water uptake through the exocarp during rainy periods just before harvest. Water is absorbed into the fruit by osmosis and the amount of uptake is due to the osmotic pressure of the pericarp tissue. It was thought that water was absorbed through the fruit cuticle (Davenport et al., 1972). As more water is taken into the cherry, turgor pressure of the cells increases and eventually resulting in cell rupture of the exocarp (Watson et al., 1988). Apple fruits can also absorb water through the skin leading to fruit cracking. The primary sites of water uptake were lenticels and injured areas of the fruit cuticle (Byers et al., 1990). Fruit cracking usually occurs only during relatively long rainy periods. Soil water failed to induce fruit cracking. Fruit cracking occurs during periods of heavy rain or high humidity when the relative humidity was near 100% for 6 hr or more (Verner, 1935).

Coffee fruits have high soluble solids content in the pericarp tissue and the content increase with maturity (Tongumpai, 1993). This results in a reduction of the osmotic potential of the fruit which then favors the absorption of exogenous water by osmosis. This may increase the turgor pressure of the pericarp tissue following either long rainy period or overhead irrigation. The high turgor pressure buildup may result in cell breakage, and eventually fruit cracking. The increase in either cell turgor or fruit cracking should weaken the pericarp tissue resulting in lowering of the FRF. Fruit cracking of coffee will not reduce the quality of the coffee bean (seed), and normally is practiced during processing to remove the bean from the pericarp tissue in the pulping process.

The objective of this study was to determine the effects of overhead irrigation on the water relations of the pericarp tissue and on the FRF of coffee fruits.

Materials and Methods

Coffee (Coffea arabica L. 'Guadalupe') seedling were grown in 4 l plastic pots in a mixture of pumice, peat, soil and sand (2:1:1:1). The plants were grown for 3 years in a greenhouse at day/night temperatures of 25/19°C and natural photoperiod (Oregon, 44°N latitude). During July 1992 the flowers at anthesis were tagged. The age of each fruit was determined following anthesis.

Ten ripe coffee fruits at various ages were harvested with pedicel intact to study the fruit water absorption through the exocarp by osmosis. The cut surface of the pedicel was sealed with paraffin. Fruits were partially immersed in distilled water with pedicel exposed to the air. Change of fruit water content was determined by fruit weight change up to 48 hours after immersion.

Experiment 1: Duration of mist irrigation

Two sets of four coffee plants each with 270 and 305 days old fruits were placed in the mist chamber. The mist was generated by a fog generator (Tobacco Conditioner Model 35, Agritech, Sanford, North Carolina) emitting fine droplets of water at the rate of 100 l per hour. During the mist treatment the fruit surfaces were always in contact with water. The plants were exposed to either 0,

24, 48, or 72 hours of mist. Fruit removal force was measured at the end of each irrigation period with a force gauge (Shimpo MF 2.0, Shimpo America, Lincolnwood, Illinois) by inserting the modified hook over the pedicel and pulling the fruit parallel to the fruit axis. The force required to detach the fruit was converted to Newtons (N) by multiplying the value in pounds by 4.5.

Pericarp water absorption capacity was measured by slicing a 1 cm diameter disk from the proximal end of the fruit and placing the disk in distilled water for 24 hr. The difference between the initial (FW) and turgid (TW) weight (after hydration) was used to calculate the percent water absorption capacity:

$$\text{Percent water absorption} = \frac{\text{TW} - \text{FW}}{\text{FW}} \times 100$$

Water saturation deficit (WSD) was calculated by the following equation:

$$\text{WSD} = \frac{(\text{TW} - \text{FW})}{(\text{TW} - \text{DW})} \times 100$$

where DW is the dry weight of the full turgid fruit disk after drying in a convection oven at 80°C for 24 hr. The pericarp water potential and osmotic potential of the 305 day old fruits were also determined with a wet loop thermocouple psychrometer. The procedures of measurement were described by Matthews et al. (1987). Pericarp turgor pressure was estimated by the difference between pericarp water potential and osmotic potential (Turner, 1981).

The numbers of fruit that cracked after each period of irrigation were recorded and expressed as a percentage of total fruits. Soluble solids content of the pericarp was measured with a hand-held refractometer at the end of each mist treatment. Treatments were arranged in a randomized block design with 2 factors (fruit age and duration of mist irrigation). Data were analyzed with SAS by GLM procedure.

Pericarp tissue viability was estimated by the 2,3,5-triphenyl tetrazolium chloride (TTC) reduction and the conductivity tests (Chen et al., 1981; Li et al., 1981; Schaff et al., 1987). The TTC reduction procedure was described in detail by Steponkus and Lanphear (1967). Four plants with 290 days old fruit were placed in the mist chamber for either 0, 24, 48 and 72 hr. Five fruits from each tree were harvested after each irrigation treatment. A 1-cm diameter disk from the proximal end of the pericarp tissue of each fruit was sliced and weighed. The fruit disk was then incubated in the dark at room temperature in 2 ml of tetrazolium solution (0.08% of 2,3,5-triphenyl-tetrazolium chloride in phosphate buffer at pH 7.5) for 24 hr. After incubation the disks were rinsed with distilled water, and the water-insoluble formazan was extracted with 3 ml of 95% ethanol for 24 hr in the dark at room temperature. The absorbance of the formazan derivative was measured at 495 nm with a spectrophotometer (Shimadzu UV

160, Shimadzu Corp., Kyoto, Japan). The data were expressed as absorbance per gram fresh weight of pericarp tissue.

Electrolyte leakage of the pericarp tissue following water absorption was measured with a conductivity meter. The fruits were classified into the following 4 groups based on the soluble solids content in the pericarp: 15, 20, 25 and 30 degree Brix (measured with a hand-held refractometer). Ten fruits from each group were used. A 1-cm fruit disk from the proximal end of the pericarp was incubated at room temperature in stoppered vials with 10 ml of double-distilled water and placed on a shaker. Electrolyte leakage was measured after incubation and at 3 hr intervals, up to 12 hr, then at 12 hr intervals up to 36 hr. The tissues were then killed after 36 hr of incubation in a hot water bath at 80°C for 30 minutes and the total electrolyte leakage measured. The data were expressed as percent electrolyte leakage by subtracting the final conductivity from the initial conductivity divided by the final conductivity and multiplying by 100.

Experiment 2: Fruit maturity and the response to mist irrigation

Three coffee plants with fruits at 280, 290, 305, and 335 days old were used in the study. Plants were placed in

the mist chamber described above for 24 hr to wet the fruit surfaces. The FRF was measured before and after the mist irrigation with a force gauge (Shimpo MF 2.0, Shimpo America, Lincolnwood Illinois). Fruit water absorption capacity and water saturation deficit were calculated as explained in experiment 1.

The effect of mist irrigation on the FRF of green fruits was also studied. Three plants with unripe (green) 180 days old fruits were placed under the mist irrigation for 24 hr. The FRF of 20 fruits from each tree was determined prior to the treatment and immediately after the plants were moved out from the mist chamber. Means comparison was done by t-test procedure.

Results

Water absorption through the exocarp of coffee fruits increased steadily with the duration of immersion up to 36 hr (Figure 8.1). Water absorption after 36 hr of immersion was negligible.

Experiment 1: Duration of mist irrigation

A. Effect of mist irrigation on FRF

The average FRF of the 305 days old fruits was significantly lower than the 270 days old fruits ($p < 0.01$). The initial FRF of the 305 days old treatment before irrigation was 2.9 N. This was 1.7 times lower than the FRF of the 270 days old treatment. Mist irrigation significantly reduced the FRF of fruits of both 270 and 305 days old treatments (Figure 8.2). However, no interaction between fruit age and duration of irrigation was found. Twenty four hours of mist irrigation of the 305 days old fruits significantly reduced the average FRF to 42% as compared to the control. When the duration of mist irrigation was prolonged to 72 hr the significant reduction of FRF was found (68% reduction).

B. Fruit cracking

The percent of cracking in the 305 days old fruits following mist irrigation was significantly greater than the 270 days old fruits ($p < 0.01$) (Figure 8.3). As the duration of mist irrigation increased the percent of fruit cracking increased curvilinearly. The percent of cracking in the 305 days old fruits was approximately 3 times greater than the 270 days old fruits. A significant negative correlation between the percent of fruit cracking and FRF was found ($r = -0.74$) (figure 8.4).

C. Water saturation deficit

Water saturation deficit (WSD) of the 305 days old fruits was significantly greater than the WSD of 270 days old fruits. As the duration of mist irrigation increased the WSD decreased significantly, especially after 48 hr of irrigation. A significant interaction between fruit age and duration of irrigation was found ($p < 0.01$). The change in WSD of the 305 days old fruit following mist irrigation was greater than the WSD of the 270 days old treatment (Figure 8.5).

Percent water content of the pericarp tissue increased significantly with duration of irrigation ($p < 0.01$). The 305 days old fruits had significantly higher

water content than the 270 days old fruits (Figure 8.6). A significant interaction between fruit age and duration of irrigation was found ($p < 0.05$). The 305 days old fruits absorbed more water than the 270 days old fruit. Water content of the 305 days old fruits increased by 6 and 19% of the initial weight within 24 and 72 hr of irrigation, respectively. The water content of the 270 days old fruits increased at a slower rate following irrigation than the 305 days old fruits.

The percent of water content of the pericarp was highly correlated to the percent of fruit cracks ($r = 0.91$, data not shown). With increased water uptake the percentage of fruit cracks increased. A negative correlation between percent water content and FRF was found, i.e. the FRF decreased as the water content increased ($r = -0.75$, data not shown).

D. Fruit water relations

Significant changes in pericarp water and osmotic potential of 305 days old coffee fruits after 48 hr of mist irrigation treatment was found ($p < 0.01$) (Figure 8.7). Osmotic potential was highly correlated with water potential ($r = 0.99$, data not shown). The turgor pressure was significantly higher in all treated fruits as compared with the control ($p < 0.01$). A negative correlation

between FRF and pericarp turgor pressure was found ($r = -0.85$) (figure 8.8).

E. Soluble solids content

The soluble solids content of the pericarp tissue decreased with increasing mist treatment in both 270 and 305 days old fruits (Figure 8.9). High negative correlation between soluble solids content and pericarp osmotic potential and water potential was found ($r = -0.91$ and -0.93 respectively, data not shown). Pericarp water absorption capacity decreased as soluble solids content decreased ($r = 0.91$, data not shown).

F. Pericarp tissue viability

The absorbance of formazan per gram fresh weight of pericarp tissue in the TTC reduction test decreased significantly as fruit age increased (Figure 8.10). The absorbance of formazan of the non irrigated (control) 290 day old fruits was 5.5. After 24, 48, and 72 hr of irrigation treatments the absorbance of formazan decreased by 86, 71, and 56%, respectively, as compared with the initial value (control).

Electrolyte leakage from fruit discs incubated in water reached almost 90% of total electrolytes present in

the tissue after 12 hr of incubation (Figure 8.11). The tissue with the lowest soluble solids content (15 Brix) leaked less electrolytes as compared to others with higher soluble solids content. The total electrolytes of fruits with soluble solids contents of 15 to 25 Brix were similar (223 to 235 mmhos/g fresh weight of pericarp) while those with 30 Brix were greater (361 mmhos/g fresh weight of pericarp).

Experiment 2: The response of different age fruits to mist irrigation

A. Effect of mist irrigation on FRF

Twenty four hours of mist irrigation significantly reduced the FRF of ripe fruits ($p < 0.01$) regardless of fruit age (Figure 8.12). The difference between FRF before and after irrigation did not change with fruit age. The FRF decreased with increasing fruit age both before and after the mist irrigation. The FRF of 335 days old fruits before irrigation was 1.5 times lower than the 280 days old fruits. After 24 hr of irrigation treatment the difference between the two age classes was maintained at 1.5 times.

FRF of green fruits at 180 days old was not affected by mist irrigation. The FRF before and after irrigation treatment were 9.3 ± 0.4 and 10.0 ± 1.0 N. respectively.

B. Fruit water absorption capacity

Older fruits were able to absorb more water than younger fruits (Figure 8.13). Their water absorption capacity decreased drastically after the fruits were subjected to 24-hr mist irrigation ($p < 0.01$) (figure 8.13). There was no interaction between mist irrigation and ripe fruit age on pericarp water absorption capacity. Water saturation deficit (WSD) of older fruits (290 to 335 days old) was higher than the 280 days old fruits. Mist irrigation significantly reduced the WSD (Figure 8.14). However, the percent change of WSD after mist irrigation in fruits of different ages did not change significantly. Percent water content of pericarp tissue increased significantly after mist irrigation ($p < 0.01$). The water content of the pericarp tissue increased with fruit age (Figure 8.15).

Discussion

Mist irrigation significantly reduced the FRF of ripe coffee fruits. The FRF decreased progressively after 24 hr of mist irrigation. The results of this study indicate that the FRF of ripe coffee fruits can be reduced significantly by overhead irrigation.

Coffee fruits were able to absorb water through the epidermis of the pericarp tissue via osmosis as reported for cherries (Davenport et al., 1972; Watson, et al., 1988), grapes (Considine and Kriedemann, 1972) and apples (Byers et al., 1990). Direct contact of water onto the fruit surface by either direct contact of the exocarp with water and mist irrigation led to increases in pericarp water content. The absorption of water in the pericarp tissue was dependent on the duration of irrigation and fruit age (Figure 8.5, 8.14). The greater absorption of water with increasing fruit age was due to the increased soluble solids content, lowering of osmotic potential and water potential of the pericarp tissue (Tongumpai, 1993).

Mist irrigation induced cracking of the pericarp in ripe coffee fruits. The percent of cracking per plant increased with increasing duration of irrigation and age of fruit. The absorption of water and resultant change in turgor pressure of the pericarp was highly correlated with the osmotic potential of the fruit ($r = -0.90$ and 0.71

respectively, data not shown). The osmotic potential of the pericarp tissue decreased with fruit maturation (Tongumpai, 1993). Therefore, as the fruits matured their ability to absorb water increased. The uptake of water by the ripe fruits eventually reached a level that caused rupturing of the cells and pericarp tissue. The rupturing of the cells by water uptake was verified by the tetrazolium and electrolyte leakage studies (Figure 8.10, 8.11).

The electrolyte leakage of discs from different age fruits was a function of the soluble solids content of the pericarp. Almost 90% of the electrolytes leaked from the cells of mature fruits (Figure 8.11) and only about 50% leakage came from the less mature fruits. This suggests that the pericarp of mature fruits imbibed large quantities of water creating excess turgor pressure within the cell to a point that cell rupturing occurred.

The TTC reduction test for cell viability of the pericarp tissue confirmed these studies. Changes in tetrazolium (TTC) reduction is a measure of tissue viability (Chen et al., 1981; Li et al., 1981; Schaff et al., 1987; Steponkus and Lanphear, 1967). The reduction of TTC by the pericarp tissue decreased as the duration of mist irrigation increased (Figure 8.10). The decrease in TTC reduction caused by increased exposure to mist irrigation was likely the result of breakage of the cell

membrane due to excessive turgor pressure. The rupturing of the pericarp cells of mature fruits by excessive increases in turgor pressure following water absorption may explain the fruit cracking phenomenon observed in coffee fruits. When the cells of the pericarp tissue collapse due to rupturing of cell membrane tissue weakening occurs resulting in the decrease of FRF.

An additional observation made which supports cell rupture and death of mature coffee fruits following mist irrigation treatment was the blackening and shrinking of the pericarp tissue of mature fruits a few days after treatment (data not presented). The condition of the green fruits remained unchanged after the irrigation treatment. The appearance of black-shriveled fruits of coffee following periods of heavy rainfall under natural conditions is also commonly observed. This study suggests that the cause of this phenomenon is due to excessive water uptake resulting in cell rupture and death.

Fruit detachment of either green or ripe coffee fruits is not due to the formation of an abscission layer as reported for most other fruits (Tongumpai, 1993, chapter 5). The detachment point of green fruits occur at random between the peduncle and pericarp tissue, while the detachment point of ripe fruits occur between the pericarp and pedicel tissue (Tongumpai, 1993). This explains why the FRF of green fruit was not affected by irrigation

while the FRF of the ripe fruits decreased with fruit maturation and increased duration of irrigation (Figure 8.2, 8.12). Therefore, the reduction of FRF of the ripe fruits results in greater differences between the FRF of green and ripe fruit. The large difference in FRF between green and ripe fruits following overhead irrigation should favor the selective harvesting of mature fruits by machine. The frequency of the shaker or the force needed to detach the ripe fruit following irrigation can be lowered sufficiently to favor harvesting of ripe fruits only. Overhead irrigation of coffee plants prior to harvesting of coffee fruits may be an effective strategy for selective harvesting of mature coffee fruits by machine. Unfortunately, the length of irrigation needed to reduce the FRF may be too long to be practical under field conditions. If this is a problem, additional studies may be required to find methods of reducing the amount of irrigation needed to achieve the same effect. For example, surfactants are known to increase the penetration of water and chemicals through the cuticle of apple leaves (Westwood and Batjer, 1960) and fruits (Byers et al., 1990) and grape berries (Marois et al., 1987). Surfactants may increase the rate of water absorption and reduce FRF in mature coffee fruits with shorter duration of irrigation.

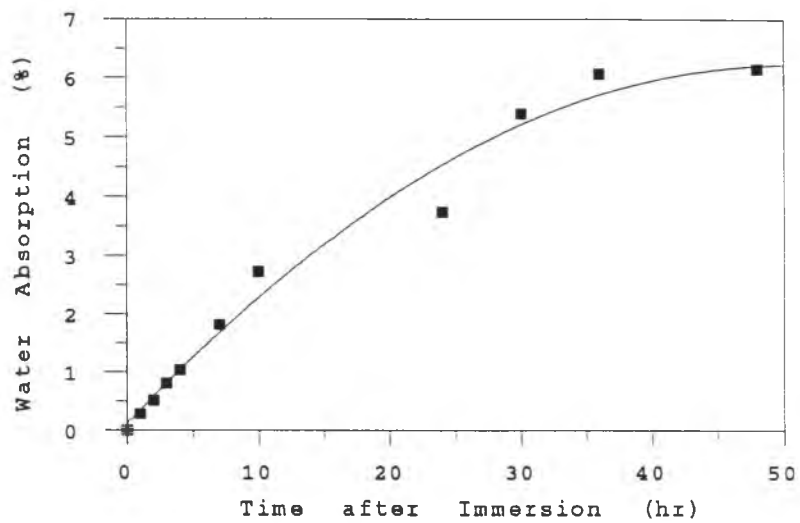


Figure 8.1. Percent water absorption of ripe coffee fruit through exocarp via osmosis after immersion in water.

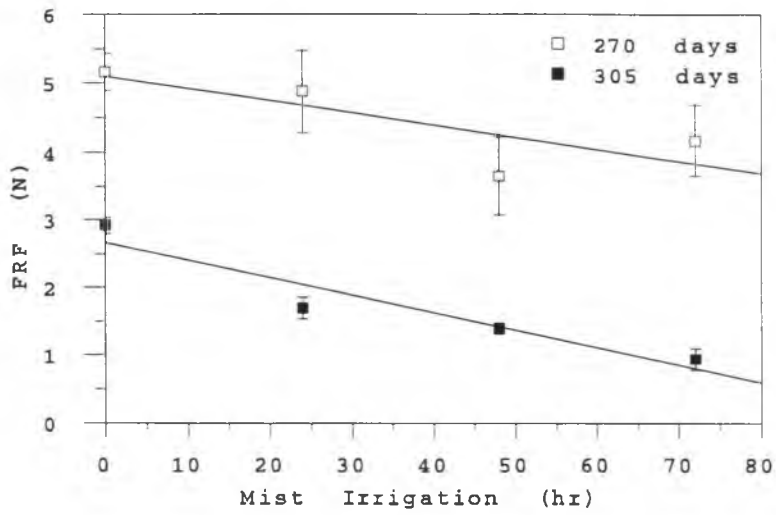


Figure 8.2. Effect of mist irrigation duration on the FRF of coffee fruits at 270 and 305 days after anthesis.

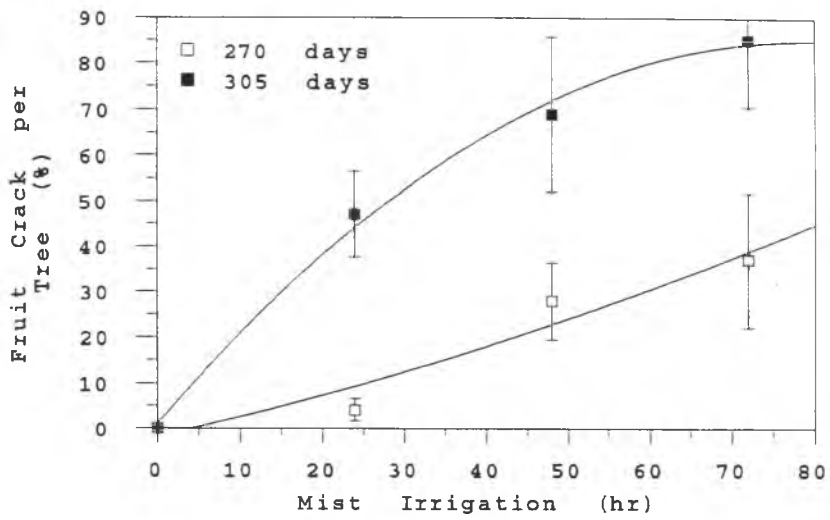


Figure 8.3. Effect of overhead mist irrigation on percent of fruit crack per coffee tree.

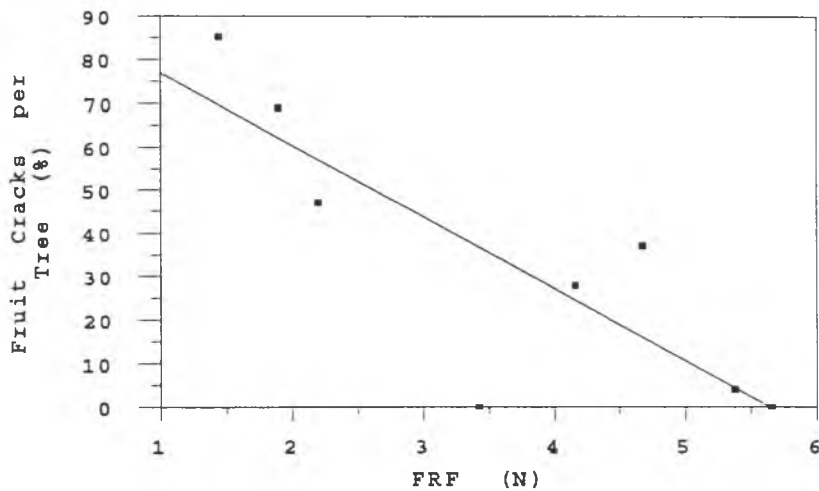


Figure 8.4. Relationship between FRF and percent cracking of fruits per tree due to mist irrigation.

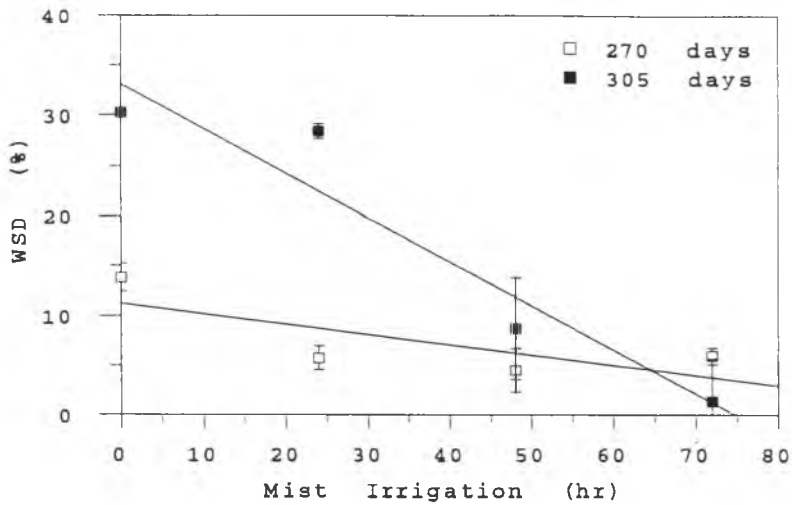


Figure 8.5. Water saturation deficit of pericarp tissue after different periods of mist irrigation.

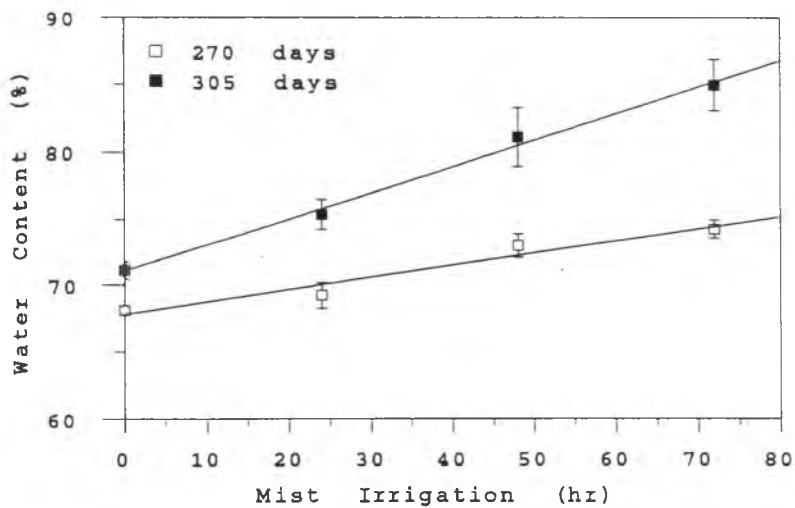


Figure 8.6. Changes of pericarp water content in coffee fruits after different periods of mist irrigation.

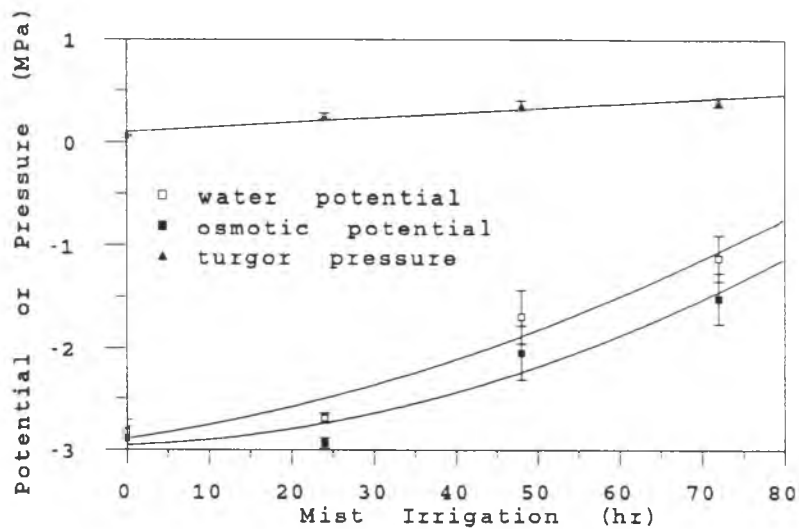


Figure 8.7. Effect of mist irrigation on the pericarp water relation in 305 days old coffee fruits.

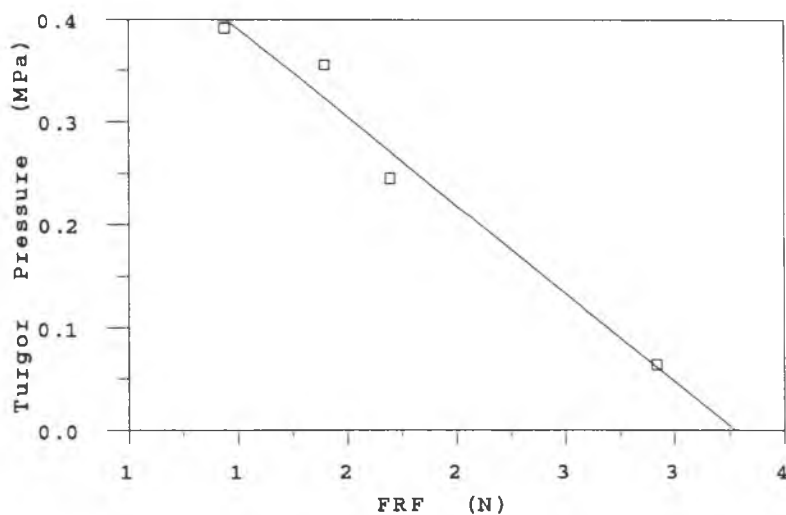


Figure 8.8. Relationship between FRF and pericarp turgor pressure in coffee fruits.

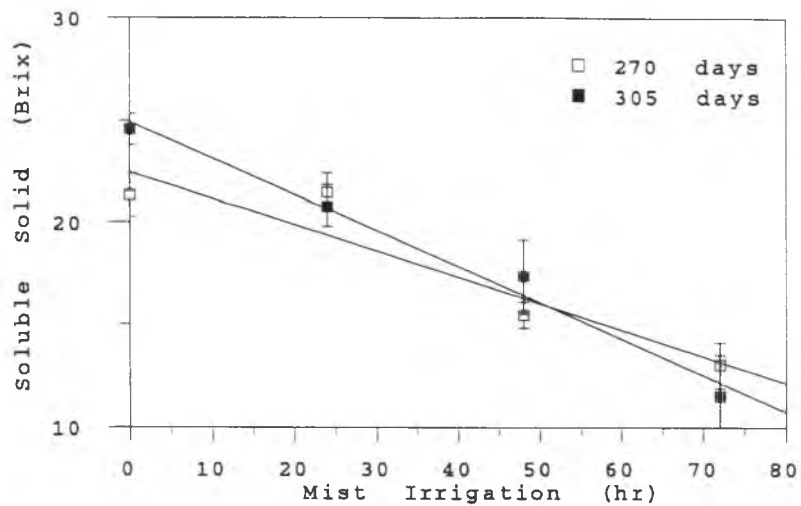


Figure 8.9. Changes in soluble solids content of coffee pericarp tissue after different periods of mist irrigation.

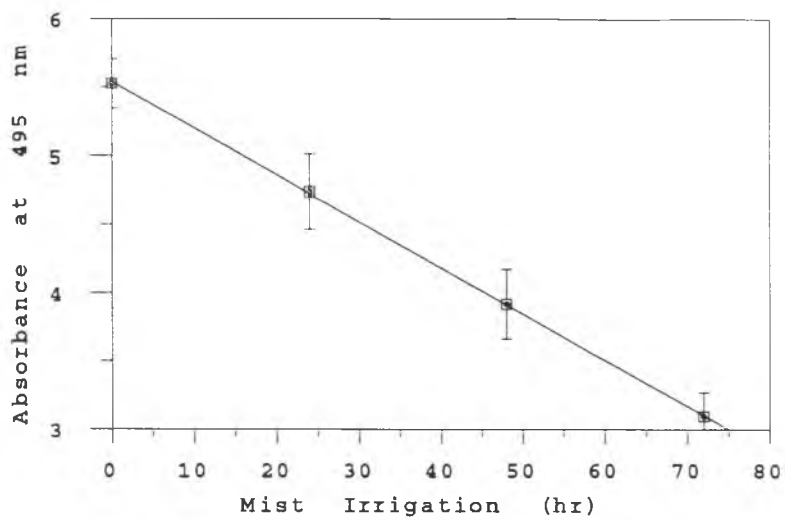


Figure 8.10. Reduction of tetrazolium chloride (TTC) as determined by the absorbance of formazan at 495 nm per g of coffee pericarp tissue fresh weight after different periods of irrigation.

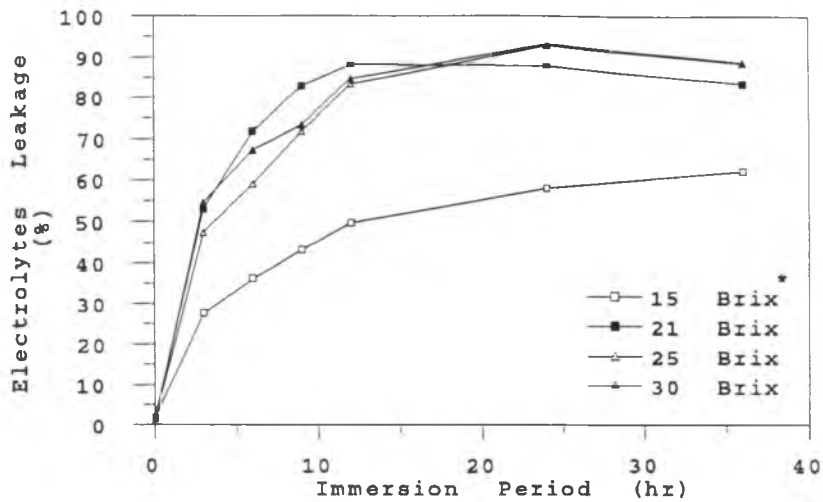


Figure 8.11. Electrolyte leakage of coffee pericarp tissue at different maturity as determined by soluble solids content.

- * 15 Brix = about 250 days old fruit from anthesis
- 21 Brix = about 275 days old fruit from anthesis
- 25 Brix = about 300 days old fruit from anthesis
- 30 Brix = about 330 days old fruit from anthesis

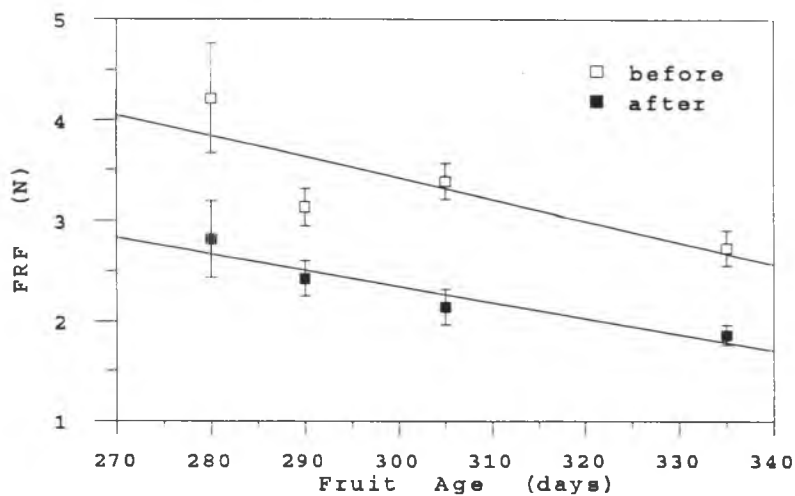


Figure 8.12. Changes in FRF of coffee fruits at different ages before and after 24 hr mist irrigation.

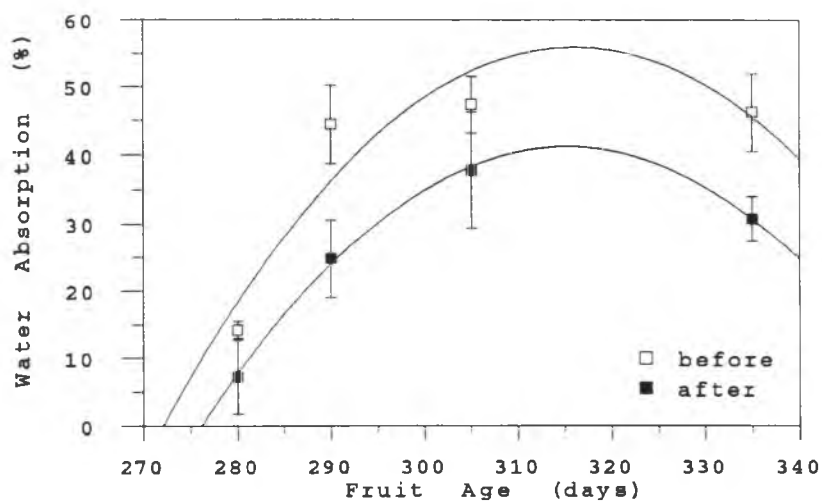


Figure 8.13. Pericarp water absorption capacity of coffee fruits at different ages before and after 24 hr mist irrigation.

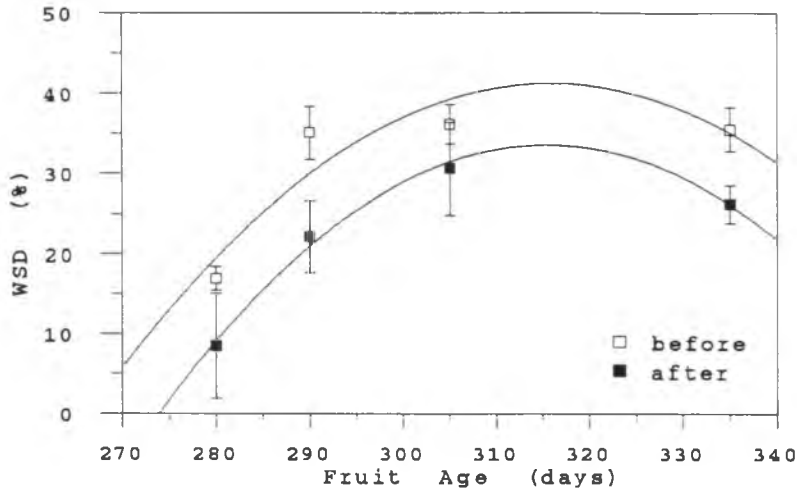


Figure 8.14. Changes in WSD of coffee fruits at different ages before and after 24 hr mist irrigation.

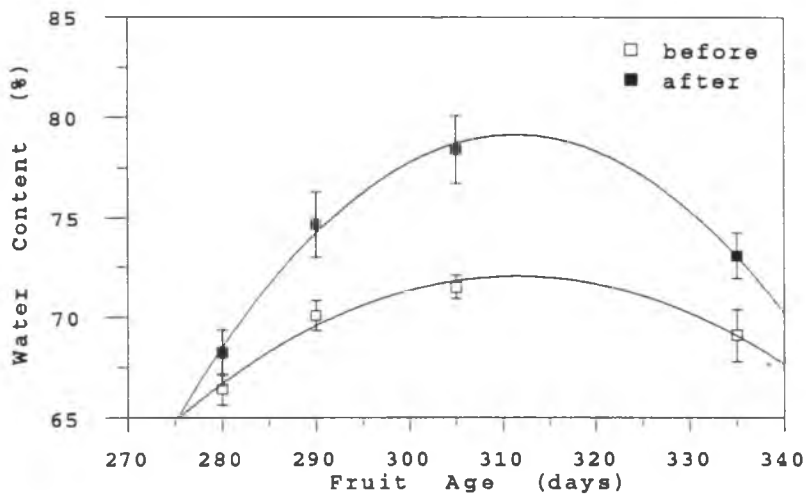


Figure 8.15. Changes in pericarp water content of coffee fruits at different ages before and after 24 hr mist irrigation.

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Concluding Remarks

Strong evidence for the feasibility of machine harvest of mature coffee fruits was found by incorporation of the following studies:

1. Synchronization of flowering.
2. Synchronization of fruit maturation.
3. Alteration of the FRF of ripe fruits.

Synchronization of flowering was achieved either by overhead irrigation treatment or by water deficit stress followed by irrigation to field capacity. This study suggested that water deficit of the flower buds per se was important for overcoming flower bud dormancy. Direct contact of water to the flower buds was important for overcoming dormancy. In commerce, overhead irrigation to overcome dormancy may be manageable and better than previous methods of overcoming dormancy to achieve synchronous flowering. Although synchronization of flowering does not necessarily result in synchronous fruit maturation concentration of flowering does concentrate the period of fruit ripening. Furthermore, this may facilitate the use of chemical-induced fruit ripeners, e.g. ethephon. Ethephon may be able to synchronize fruit ripening of single flowering crops as reputed in this thesis.

When synchronization of flowering and fruit maturation cannot be obtained, an alternative method for

selectively machine harvesting ripe coffee fruits was found. Coffee fruits do not abscise naturally via an abscission zone. Therefore, force is required to detach fruits from the plant. The point of detachment for green and ripe fruits is different. The FRF to detach green fruits is greater than for ripe fruits. Overhead irrigation reduced the FRF of ripe fruits but had no effect on the FRF of green fruits. Lowering the FRF of ripe fruits by overhead irrigation resulted in a greater difference between the FRF of the green and ripe fruits. The large difference in FRF between green and ripe fruits should favor greater discrimination between ripe and unripe fruits by proper adjustment of machines used for harvesting coffee fruits.

Field studies are needed to test the feasibility of synchronizing flowering by overhead irrigation, concentrating fruit ripening by ethephon treatment, and/or discriminating harvest of ripe fruits only by overhead irrigation and proper adjustment of harvest machines. The results reported in this thesis provides good evidence for developing strategies for successfully machine harvesting mature fruits only by either synchronizing flowering and fruiting, and/or by alteration of the FRF of mature fruits.

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Appendices

A. Correlation between Soluble Solids Content of Pericarp
and FRF of Ripe Coffee Fruits

Ten 3 year old coffee (Coffea arabica L. 'Catuai') plants growing in field plots at Pioneer Mill Company at Lahaina, Maui were used. Ten ripe fruits at various ages from each tree were sampled and FRF was measured with a hand-held digital force gauge (Shimpo FG-5.0, Shimpo America Corp., Lincolnwood, Illinois). Soluble solid content of the pericarp tissue of the corresponding fruits was measured with a hand-held refractometer (Fisher Scientific, Santa Clara, California). A negative correlation between FRF and soluble solid content of the pericarp tissue was found (Figure A1). The FRF of ripe fruits decreased with increasing soluble solid content.

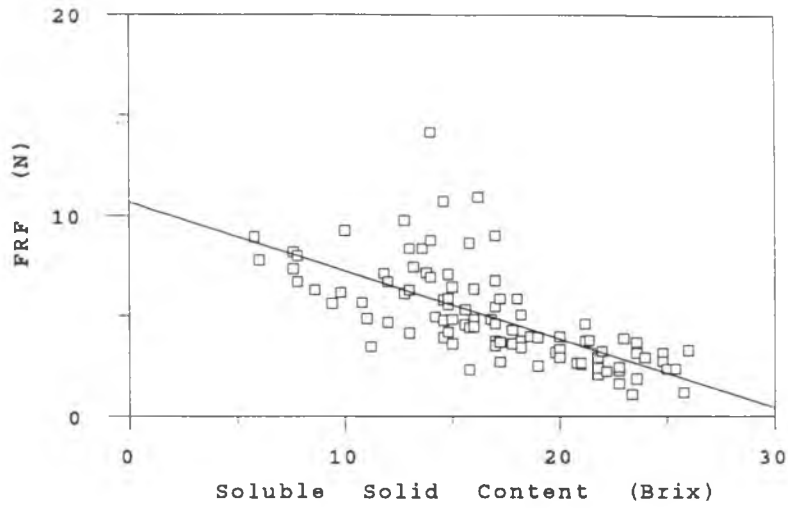


Figure A1. Correlation between FRF and soluble solid content of field grown coffee fruits ($r = 0.68$).

B. Correlation between Leaf Water Potential and FRF of
Green and Ripe Coffee Fruits.

Twenty three 3 year old coffee (Coffea arabica L. 'Catuai') trees growing in field plots at Pioneer Mill Company, Lahaina, Maui were used. Most of these plants were under water deficit stress for more than 1 month. Leaf water potential was measured with a pressure chamber (PMS, Corvallis, Oregon). Ten green and ripe fruits were sampled from each tree and FRF was measured with a digital force gauge (Shimpo FG-5.0, Shimpo America Corp., Lincolnwood, Illinois). A high correlation between leaf water potential and the FRF of ripe fruit was found ($r = 0.96$). There was no correlation between leaf water potential and the FRF of green fruits (Figure B1).

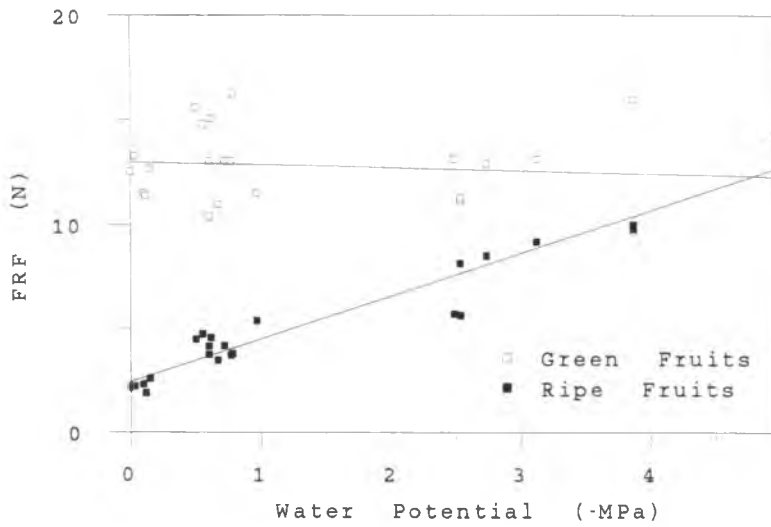


Figure B1. Correlation between leaf water potential and the FRF of ripe and green fruits. Each point represents 10 fruits.

C. Effect of NAA on Fruit Removal Force of Coffee

Twenty 3 years old coffee (Coffea arabica L. 'Catuai'), with both unripe and ripe fruits in the same tree, growing in field plots at Pioneer Mill Company, Lahaina, Maui were used. The whole trees were sprayed with NAA at 12.5, 25 and 50 ppm to run off. The FRF was measured with a handheld digital force gauge (Shimpo FG-5.0, Shimpo America Corp., Lincolnwood, Illinois). The measurement was done at 1, 2, 3, 5 and 8 days after NAA application. Five unripe (green) and fully ripe fruits were sampled for the measurement. The treatments were assigned in randomized block design with 5 blocks. The experiment was repeated by another set of twenty coffee plants and higher concentration of NAA (50, 100 and 200 ppm) was used. The measurement of FRF was done up to 15 days after the NAA application. NAA had no effect on FRF of both green and ripe fruits despite a high concentration was used. The FRF of green fruits was always greater than the FRF of ripe fruits in all treatments (Figure C1, C2).

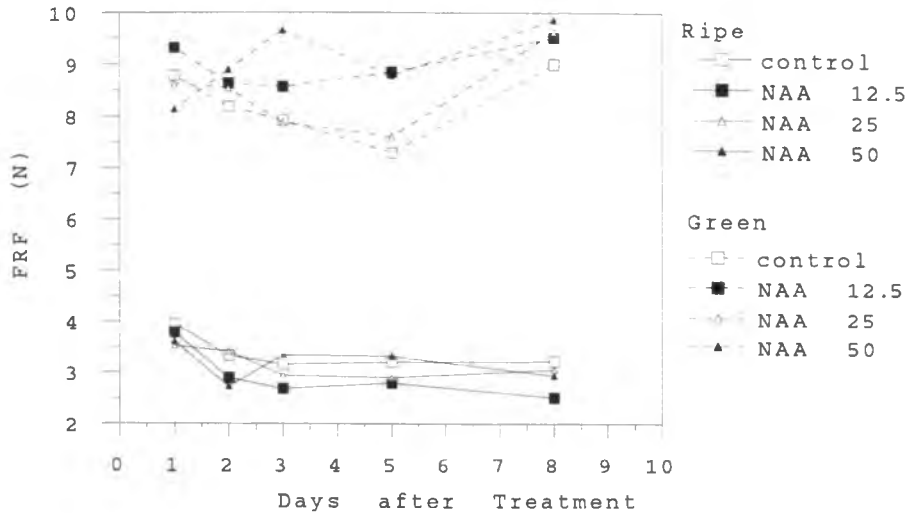


Figure C1. Effect of NAA at 12.5 to 50 ppm on the FRF of green and ripe coffee fruits.

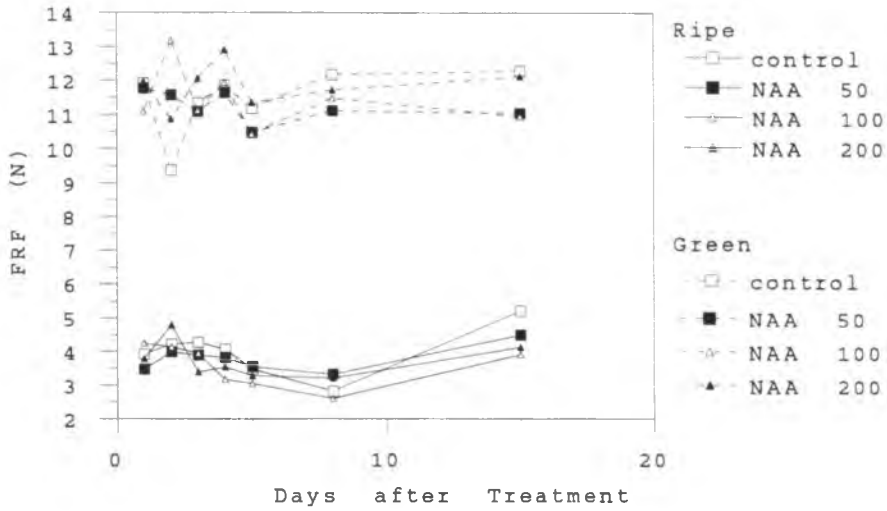


Figure C2. Effect of NAA at 50 to 200 ppm on the FRF of green and ripe coffee fruits.

D. Effect of Shoot Tip Removal on Flowering of Coffee
(Coffea arabica L. 'Guatemalan').

Twenty five 3-year old coffee (Coffea arabica L. 'Guatemalan') growing at Waiakea Agricultural Research Station, Hilo, Hawaii were used. Each tree has two leaders. All shoot tips in one leader of five trees were manually removed in July 1990 while another leader served as control. The same procedure was done in August to november at monthly interval. Number of flowers at anthesis and flowering date of flowers in each branch were recorded at weekly interval started when the first flower opened. Number of vegetative shoots after the treatment was recorded. It was found that shoot tip removal of coffee at anytime between July to November has no effect on flowering. Number of flower at anthesis of each treatment was not significantly different (Table D1). About 50% of flowers reach anthesis on February 5, 1990 in all treatment (data not shown). Shoot tip removal had no effect on number of harvestable fruits (Table D1).

Table D1. Effect of shoot tip removal in 1990 on number of flowers and fruits per branch of coffee.

Time of tip removal	Treatment	# of Flowers per branch	# of Fruits per branch
July	control	227.6	62.9
July	tip removal	151.6	55.9
August	control	217.8	87.6
August	tip removal	190.4	70.2
September	control	349.8	91.3
September	tip removal	309.8	72.9
October	control	387.4	87.5
October	tip removal	403.0	86.5
November	control	477.0	88.0
November	tip removal	540.4	94.7