University of Alberta

The Role of Plant Hormones in Saskatoon Fruit Development

by

Roisin Carrie McGarry



A thesis submitted to the Faculty of Graduate Studies and Research in partial fulfillment of the requirements for the degree of Master of Science

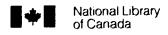
in

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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Research for acceptance, a thesis entitled The Role of Plant Hormones in Saskatoon Fruit Development submitted by Roisin Carrie McGarry in partial fulfillment of the requirements for the degree of Master of Science in Horticulture.

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ABSTRACT

The focus of this research was to compare saskatoon (Amelanchier alnifolia Nutt.) fruit growth among cultivars, and determine the effects of gibberellins and ethephon on reducing seed number and enhancing uniform fruit ripening, respectively. Saskatoon fruit growth patterns, similar among cultivars, exhibited three phases of development: Stage I (from compressed buds at 18 heat units, to 300 heat units) is characterized by rapid mesocarp cell division and elongation; Stage II (300 to 600 heat units) is characterized by steady fruit growth and rapid seed development; Stage III (600 heat units to harvest maturity) is characterized by continued mesocarp cell elongation, resulting in exponential fruit growth. In general, gibberellins did not affect saskatoon fruit set, but significantly decreased the seed content per fruit. Ethephon significantly enhanced saskatoon fruit ripening without deleteriously affecting fruit quality. The results from this research suggest that gibberellins and ethephon can improve saskatoon fruit quality and yield.

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LIST OF ABBREVIATIONS

ANOVA analysis of variance

BA N-(phenylmethyl)-1H-purine 6 amine

GA gibberellin

GA₃ gibberellic acid

 $GA_{4/7}$ gibberellins GA_4 and GA_7

GDD growing degree days

HU heat unit

LSD least significant difference test

SSC soluble solids concentration

TA titratable acidity

 T_{base} base temperature

T_{max} daily maximum temperature

T_{min} daily minimum temperature

1. INTRODUCTION

1.1 Development of the Saskatoon as a Fruit Crop

The saskatoon, Canada's native prairie fruit, has recently gained popularity as a cultivated fruit crop. Saskatoon fruit production is rapidly increasing to meet growing consumer demand (St. Pierre, 1992a). In 1995, over 400 ha of cultivated saskatoon orchards existed in Alberta, with 160 - 180 ha producing fruit (Hausher, 1996, personal communication). Estimated yields from mature saskatoon orchards approximate 3300 kg/ha with potential returns ranging from \$9500 - \$15,000/ha (Hausher, 1996, personal communication). Saskatoon fruit processing, a viable and expanding industry, has generated a wide range of value-added products, including jams, jellies, sauces, pie fillings, chocolates, wines and liquors, and flavor concentrates. This research has addressed two areas for improving saskatoon fruit quality and yield: reducing seed number per fruit and increasing uniform fruit ripening.

1.2 Taxonomic Identification

Saskatoon shrubs are members of the sub-family Maloideae (Campbell et ai, 1991), family Rosaceae, order Rosales, sub-class Rosidae, class Magnoliopsida, and division Magnoliophyta (Cronquist, 1988). After initial classification as *Aronia alnifolia* and *Pyrus alnifolia*, the saskatoon shrub was identified by Thomas Nuttal in 1834 with the current binomial, *Amelanchier alnifolia*. This species, native to western North America, includes the majority of current cultivars, such as cv.'s Thiessen, Northline, Smoky, Regent, and Pembina (St. P erre, 1992b). Other saskatoon species used as horticultural crops include *Amelanchier sanguinea* (cv. Parkhill) and *A. canadensis* (cv. Success; St. Pierre, 1992b).

The sub-family Maloideae includes pome fruits, such as apple, pear, quince, and saskatoon. Pomes are fleshy false fruits, so named as the origin of the fruit wall, comprised of a cartilaginous or membranous endocarp (Sterling, 1965), fleshy mesocarp, and exocarp, arises from an inferior ovary and accessory tissues (Douglas, 1944).

1.3 Shoot Structure

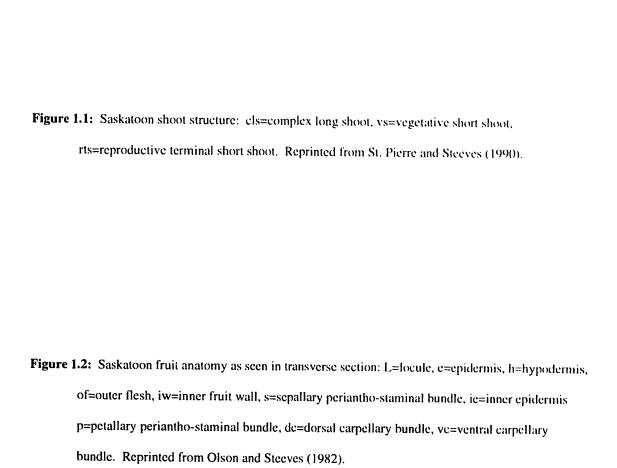
Shoot structure in *Amelanchier alnifolia* is dimorphic (St. Pierre and Steeves, 1990). Each ramet, an aerial and branched shoot, is composed of determinate long and snort shoots. (St. Pierre and Steeves, 1990). Raceme-like inflorescences, developed from initials formed in the previous season (Steeves and Steeves, 1990), are borne on short shoots (Fig. 1.1; St. Pierre and Steeves, 1990).

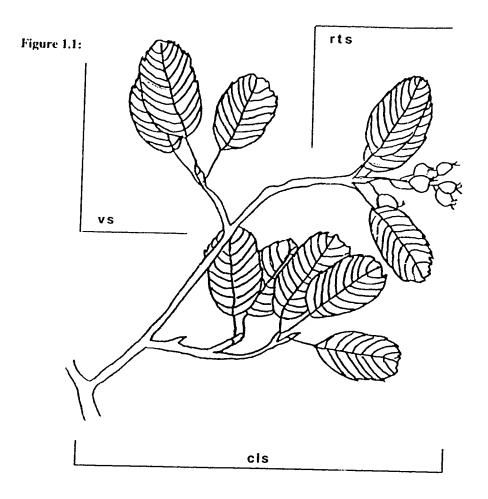
1.4 Fruit Set

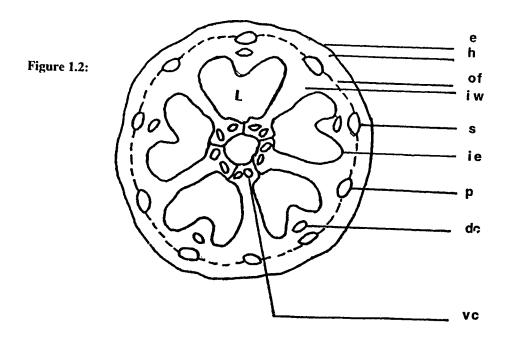
Saskatoon flowers are actinomorphic, pentamerous, and perfect, and can be set by insect-mediated self- or cross-pollination and non-insect mediated autogamy (Olson, 1984). Fruit set among natural populations of saskatoon has been variable (55-98%) despite consistently heavy annual blooms (St. Pierre, 1989; Steeves et al, 1979). St. Pierre (1989) observed that only 19% of flowers from natural stands attained full fruit maturity. Steeves et al (1979) reported that although 74.4% of flowers from natural saskatoon stands were successfully pollinated and initially set fruit, only 33.1% of those blossoms yielded fruits at harvest maturity. These observations suggest that the majority of fruit loss could be attributed to post-pollination events. St. Pierre (1989) identified that 27% of fruit loss resulted from frost injury, 54% from pest infestation, and the remaining 19% of the total fruit lost was not damaged.

1.5 Fruit Morphology and Development

Using fruits from natural stands, Olson and Steeves (1982) described saskatoon fruit morphology and development. The developing saskatoon fruit wall is a highly vascularized structure (Olson and Steeves, 1982). Ten periantho-staminal bundles, alternating as sepallary or petallary, leave the stele of the pedicel to vascularize the outer fruit wall (Fig. 1.2). The sepallary bundles branch to form five centripetally located dorsal carpellary bundles. Interior to these bundles are the ten ventral carpellary bundles. Apart from the vasculature, the saskatoon fruit wall is comprised of the outer epidermis, hypodermis, outer flesh, inner fruit wall, and inner epidermis. A thin cuticle covers the tangential walls of the vacuolated, rectangular cells comprising the exterior uniseriate epidermis. As the diameter of the wild fruit increased during development, the cells of the epidermis increased 1.9 times in diameter from bloom to maturity, and







the cuticle thickened. The next region, the biseriate hypodermis, also contained vacuolated, rectangular-shaped cells which increased 4.2 times in diameter from flower bloom to fruit maturity. The third component, the outer flesh, lies inside the hypodermis and extends to the ring of periantho-stammal bundles. Cell diameter in the outer flesh increased 4.6 times from bloom to maturity, and intercellular spaces became increasingly prevalent. Unlike the apple fruit, no core line extends between the periantho-staminal bundles to clearly separate the inner and outer fruit flesh (Olson and Steeves, 1982). The inner fruit wall lies between the periantho-staminal bundles and the inner epidermis. Cells of the inner fruit wall enlarged 4.5 times in diameter from bloom to maturity, and aerenchyma and brachysclereids became prevalent during development. The uniscriate inner epidermis, lining the gynoecial locules, divided anticlinally, and cell diameter increased 1.6 times from bloom to maturity. Olson and Steeves (1982) attributed saskatoon fruit growth to be primarily the result of sizable increases in the number and size of cells within the outer flesh and inner fruit wall (mesocarp).

1.6 Fruit Ripening in Amelanchier

In general, saskatoon fruits do not ripen uniformly on the shrub (Green and Mazza, 1986). Within a single harvest, Rogiers and Knowles (1995) have identified nine saskatoon fruit maturity stages, grouped according to fruit color and size. Mechanisms of fruit ripening asynchrony in *Amelanchier arborea* was examined by Gorchov (1985). Observations made during the growing season indicated that non-uniform fruit ripening was not due to flowering asynchrony in this specie, as 95% of flowers opened at one time. However, 98% of fruit ripening asynchrony in *A. arborea* was due to variable fruit developmental time, a factor highly correlated with seed number per fruit. Variable seed number accounted for 42-53% of fruit ripening asynchrony in *A. arborea*. Gorchov (1985) concluded that fruits with more seeds ripened faster.

1.7 Seed Development in Amelanchier

Although saskatoon fruit can potentially yield ten seeds per fruit, this is rarely achieved. St. Pierre and Steeves (1990) observed that 86% of wild fruits sampled contained one to three viable seeds; in all, 73-80% of the ovules aborted. Low seed to ovule ratios are not unusual in angiosperms (Bawa and Webb,

1984; Garwood and Horvitz, 1985; Gorchov, 1985; Gorchov and Estabrook, 1987), and can result from the intensity of sexual selection (Bawa and Webb, 1984), pollen scarcity, failed entargement of all ovules, or insufficient numbers of pollen tubes reaching the ovules for double entilization (Garwood and Horvitz, 1985). Gorchov and Estabrook (1987) tested the effects of pollen and resource limitations on seed development in *Amelanchier arborea*, a fruit with variable seed number at maturity. Supplementing pollination and altering resources by defoliating, girdling, and fruit thinning, did not affect seed number in *A. arborea* fruits (Gorchov and Estabrook, 1987). Seed development in *A. arborea* was examined with respect to carpel-dependent (both ovules within a carpel develop similarly) and carpel-independent (each ovule within a carpel develops independently) processes. Since 32% of all carpels contained both a developed and aborted seed (42% and 26% of carpels contained two seeds and zero developed seeds, respectively), the authors surmised that seed development in *A. arborea* was not exclusively governed by carpel-dependent or carpel-independent processes (Gorchov and Estabrook, 1987). Seed development in *A. arborea* was explained using a mixed model whom some carpels failed entirely while the ovules in remaining carpels were equally likely to develop (Gorchov and Estabrook, 1987).

1.8 The Role of Gibberellins in Fruit Set and Parthenocarpy

Maximizing yields of salable saskatoon fruits requires improved fruit set. Among exogenous hormone treatments, gibberellins (GAs) are the most active chemicals available to increase the set of open-pollinated flowers (Dennis, 1986). However, the effects of GAs on fruit set in apples have not been consistent (Dennis, 1986). GAs applied to uninjured apple blossoms have been shown to increase (Edgerton, 1981), decrease (Edgerton, 1981; Taylor, 1975; Wertheim, 1982), or not affect fruit set (Dennis et al., 1983; Edgerton, 1981; Stembridge and Morrell, 1972; Wertheim, 1982). Luckwill and Silva (1979) illustrated the ambiguous role of GAs on fruit set in cv. Golden Delicious apples. They found that GA₃ applied at 500 ppm during full bloom and again 14 days later significantly reduced fruit set (56% of control) while GA₃ applied at full bloom and 39 days later did not significantly affect fruit set (95% of control). Such conflicting responses have been attributed to variations in environmental conditions, plant health, time of GA application, GA concentration, and type of GA applied (Greene, 1989).

High concentrations of GAs can stimulate vegetative growth (c. 173) or affect the return bloom of fruit crops. GAs can promote flowering in Chrysanthemum and conifers (Pharis and King, 1985) while inhibiting bloom in apples and trees that form flower initials the previous year (Luckwill and Silva, 1979). Luckwill and Silva (1979) observed that GA3 applied at 500 ppm to cy. Golden Deheious apples significantly reduced the return bloom in the year following treatment. They found that Go copiled at full bloom and again 14 or 39 days later significantly reduced the number of returning blossoms (31% of control and 84% of control, respectively). Luckwill and Silva (1979) concluded that GA₃ inhibited floral induction on cv. Golden Delicious spurs. In contrast, Looney et al (1985) found that GA4 was not inhibitory to apple tree flowering. Prior to GA₄ application, spur-type cv. Golden Delicious apples exhibiting prominent biennial bearing were first thinned by removing the fruits from 80-90% of flowering spurs in order to ensure some degree of return bloom in the following "off" year. Although the return bloom in the subsequent year was low for all trees, GA4 treatments applied at 3, 30, 300 µg/spur and GA4+zeatin (at 30 µg each) increased the proportion of flowering spurs 2.2, 2.4, 5.8, and 3.3 times, respectively. Treatments applied 4.5 weeks after full bloom returned more blossoms the following year (3.8 times) compared to treatments applied 7 weeks after full bloom (3.0 times). Looney et al (1985) concluded that GA₄ could play a promotive role in apple tree flowering. The potential success of GAs on flowering among fruit crops rests upon selecting the most effective GA, incorporating synergists or antagonists, employing multiple applications at reduced rates, and by optimizing the method and timing of application (Dennis, 1973; Looney and Pharis, 1986).

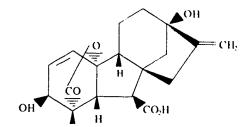
Extensive research in horticulture has been directed at promoting parthenocarpic, or "seedless", fruit development. Natural parthenocarpy arises from unsuccessful pollination, as in bananas (Israeli and Lahav, 1986), failed fertilization, as seen in some blueberries (Eck, 1986), or by embryo abortion, as with grapes (Lavee and Nir, 1986). Exogenous GAs can promote parthenocarpic fruit development from unpollinated flowers by substituting for seed-produced GAs, or inducing production of other growth substances, at a critical stage of development (Looney and Pharis, 1986; Pharis and King, 1985). Among pollinated flowers, exogenous GAs can reduce pollen germination (Crane, 1964) or inhibit embryogenesis (Pharis and King, 1985) to yield parthenocarpic fruits. At present, only GA₃, GA₄₇₇, and GA₄₇₇+BA [N-(phenylmethyl)-1H-purine 6-amine] are available for commercial use, by the trade names "Progibb",

"Provide", and "Promalin", respectively (Fig. 1.3a-d). Although GA₄ and GA₇, the predominant and native hormones in apple seeds (Dennis and Nitsch, 1966), are generally more active than GA₃ at inducing parthenocarpy in apples (Dennis, 1986), all commercial formulas have been successful in reducing apple seediness (Bangerth, 1976; Bukovac, 1963; Greene, 1984; McLaughlin and Greene, 1984; Taylor, 1975). Since the saskatoon and apple fruits are taxonomically similar, GA treatments may improve saskatoon fruit set and reduce seed number.

1.9 The Role of Ethylene in Fruit Ripening

Fruit ripening encompasses many processes, transforming the texture, flavor, fragrance, and appearance of a developing fruit. Fruits have been traditionally classified as climacteric or non-climacteric depending upon their ripening behavior (McGlasson, 1985). Climacteric fruits, such as saskatoon (Rogiers and Knowles, 1996), are characterized by large increases in respiration and ethylene (C₂H₄) production accompanying the biochemical changes taking place within the ripening fruit (McGlasson, 1985). In contrast, non-climacteric fruits do not exhibit changes in respiration or ethylene production associated with fruit development (McGlasson, 1985). Ethylene promotes fruit ripening by activating transcription of many genes (Gillaspy et al, 1993) which control the climacteric rise in respiration and biochemical changes within the fruit (Oeller et al, 1991). Exogenous ethylene can elicit a full ripening response in climacteric fruits if applied at a receptive time of fruit development (McGlasson, 1985). However, if ethylene is applied too carly during fruit growth, no response is evoked (McGlasson, 1985). Developmental sensitivity has been demonstrated in tomato fruits, where ethylene applied prior to the mature green stage of fruit development did not advance fruit ripening (Varga and Bruinsma, 1986).

An ethylene-releasing agent, ethephon (2-chloroethylphosphonic acid), has been widely used in crop management to stimulate fruit ripening. Ethylene, phosphate and chlorine ions are released through the nucleophilic attack on the phosphonate dianion of ethephon by a water molecule at pH>4 (Yang, 1969). Ethephon has been employed to hasten post-harvest fruit ripening (jujube: Abbas et al, 1994; papaya: Bel et al, 1992; peach: Dekazos, 1985), and to enhance uniform ripening in planta for efficient mechanical



CO
$$H$$
 CO_2H

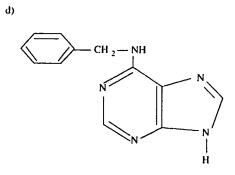


Figure 1.3: The chemical structures of: a) GA₃; b) GA₄; c) GA₇; d) BA

harvesting (peppers: Batal and Granberry, 1982; coffee: Browning and Cannell, 1970; Winston et al, 1992; tomato: Dostal and Wilcox, 1971; English walnut: Martin, 1971; cherry: Olien and Bukovac, 1978; Sims et al, 1974; and olives: Tous et al, 1995). As mechanical harvesting becomes more popular in saskatoon orchards, ethephon could be an effective agent to promote uniform ripening, thereby aiding in saskatoon crop management.

1.10 Research Objectives

- 1) Prior to implementing orchard management strategies for improving saskatoon fruit quality, cultivar-specific growth and development patterns in saskatoon fruit first need to be established. The first objective was to compare fruit growth patterns and mesocarp development among large, medium, and small fruited saskatoon cultivars; to investigate the role of seed number on final fruit size; and to examine the relationship between mesocarp cell number and size, and final fruit size among these cultivars.
- 2) Increasing saskatoon fruit set can improve fruit yields, and reducing seed number per fruit can improve the quality and texture of many value-added saskatoon products. Gibberellins have been employed with other fruit crops to improve fruit set and/or stimulate parthenocarpic fruit development. The second objective was to test the creeks of gibberellins on fruit set and seed number in saskatoon.
- 3) Mechanically harvesting saskatoon fruits is preferable to hand-harvesting, a laborious and costly procedure. However, reductions in salable fruit yields can occur with once-over mechanical harvesting due to non-uniform ripening of saskatoon fruits. The third objective was to examine the effects of ethephon on saskatoon fruit ripening.

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2. SASKATOON FRUIT DEVELOPMENT

2.1 Introduction

The hardy saskatoon shrub, Amelanchier alnifolia (Nutt.), has considerable potential for cultivation across western Canada, particularly with its natural adaptation to the harsh prairie climate. With the rapid increase in popularity of this native fruit, growing numbers of saskatoon orchards are currently being developed across the Canadian prairies to meet consumer demand (St. Pierre, 1992). Comparisons of fruit growth patterns among cultivars can be particularly insightful for many horticultural procedures, such as selecting superior genotypes, establishing breeding programs, and implementing orchard management strategies. However, no thorough studies comparing flower and fruit development among cultivated varieties of saskatoon have been reported.

The saskatoon fruit, a pentalocular pome, has the potential to produce ten seeds though this is rarely achieved. St. Pierre and Steeves (1990) observed saskatoon fruits from natural stands to contain one to five seeds, with 80% of fruits containing only one to three seeds. Since fruit size is related to seed number in many species (Dennis, 1984), the number of seeds per saskatoon fruit may be a significant factor contributing to the fruit size variation observed among cultivars. The objectives of this study were to compare fruit developmental patterns among large, medium, and small truited saskatoon cultivars, and to examine the role of seed number on final fruit size among these cultivars.

2.2 Materials and Methods

Field growth studies were conducted during two seasons (1995) at the Alberta Crop Diversification Centre North in Edmonton, Alberta, Canada. Six-year old saskatoon shrubs of cv.'s Thiessen, Northline, Smoky, and Regent were selected following a split-split-plot design, with cultivar as the main plot, year of study as the split-plot, and sampling date as the split-split-plot. Main plots were arranged in randomized blocks with three replications for cv. Thiessen, and two replications for each remaining cultivar.

Two flowers/fruits were randomly harvested from each of ten randomly selected inflorescences/infructescences per shrub. Tissues were collected weekly from 50-70% flower bloom (May 13, 1994 and May 23, 1995) to fully mature ripe fruit (July 15, 1994 and July 18, 1995), and harvested onto ice. From fruit set (June 17, 1994 and June 13, 1995) to maturity, an additional ten fruits were similarly harvested for seed analysis.

Flower/fruit pedicels were trimmed to a 2 mm length, and, using calipers, pedicel diameters were measured at the pedicel-flower/fruit abscission zone and fruit diameters at the widest region of the fruit perpendicular to the pedicel-calyx axis. Flower/fruit diameters were measured at seven sampling dates each year (June 3 to July 15, 1994; June 7 to July 18, 1995). Fresh flower/fruits weights, measured at nine sampling dates each year (May 13 to July 15, 1994; May 23 to July 18, 1995), were determined gravimetrically. After drying for one week at 60°C, dry flower/fruit weights, measured at nine sampling dates each year (May 13 to July 15, 1994; May 23 to July 18, 1995), were determined gravimetrically. Seed analysis required dissection of seeds from fruits, scoring of seed number, and gravimetrically determining the fresh weight per seed. Plump seeds containing fully formed cotyledons and embryo axis were scored as "fully developed" seeds.

Sampling dates in each year were expressed as heat units (HU) with the following equation:

$$HU = [(T_{max} + T_{min})/2] - T_{base}$$
 (Equation 2.1)

where T_{max} = daily maximum temperature (0 C), T_{min} = daily minimum temperature (1 C), and T_{base} = base temperature of 4.4 0 C. Negative HUs were assigned a value of zero. Cumulative HUs (from April 1) were expressed as growing degree days (GDD) as follows:

$$GDD = \sum_{1}^{n} HU$$
 (Equation 2.2)

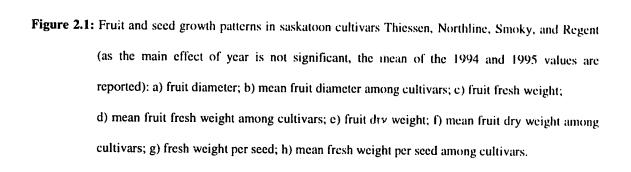
where n = days. For statistical analysis, the General Linear Model of SAS 6.10 program (SAS Institute Inc., Cary, NC 27513, USA) was used.

2.3 Results

Patterns of fruit development among cultivars were compared using growing degree days to standardize time to crop development, enabling the trends observed in this study to be adapted to varying seasonal conditions.

The seasonal fruit diameter, fresh weight, and dry weight growth patterns exhibited similarly shaped curves for all saskatoon cultivars studied. In general, cv. Thiessen was observed to have the largest fruit throughout the season, followed by cv.'s Northline, Smoky, and Regent, in descending order (Fig. 2.1a,c,e). The average trends in saskatoon fruit development among cultivars are illustrated in Fig. 2.1 (b,d,f). The fruit and seed growth patterns did not differ significantly between years of study (ANOVA, P>0. 0), therefore, the data represents the means of the 1994 and 1995 values (Fig. 2.1). Seed fresh weight increased linearly up to 670 GDD in all cultivars studied (Fig. 2.1g,h; ANOVA, P<0.05). The most rapid increase in the rate of seed fresh weight growth (from 440 to 670 GDD) was observed in cv. Northline $(2.55 \times 10^{-2} \text{ mg/GDD}, R^2 = 0.979)$, followed by cv.'s Smoky $(1.46 \times 10^{-2} \text{ mg/GDD}, R^2 = 0.948)$, Thiessen $(1.54 \times 10^{-2} \text{ mg/GDD}, R^2 = 0.993)$, and Regent $(1.00 \times 10^{-2} \text{ mg/GDD}, R^2 = 0.935)$. In general, the pattern of saskatoon fruit development, using the fruit fresh weight data, exhibits three phases of fruit growth. Stage I of fruit development (50-70% bloom at 150 GDD, to 300 GDD) is characterized by an initial rate of growth of 0.361mg/GDD which increases to 0.831 mg/GDD between 225 to 300 GDD. In Stage II (300 to 600 GDD), the growth rate slows to 0.490 mg/GDD. At this time, rapid seed development is occurring (Fig. 2.1g,h). Stage III, the final phase of fruit growth, is characterized by exponential growth from 600 GDD to harvest maturity (fruit fresh weight increases 1.98 mg/GDD between 590 to 670 GDD, and 8.55 mg/GDD between 670 to 750 GDD).

Pedicel diameters (measured one week prior to harvest maturity) differed significantly among cultivars (ANOVA, P<0.01), with pedicels from fruits of cv. Thiessen having larger diameters than those from fruits of cv.'s Northline, Smoky, and Regent (Table 2.1; LSD, P<0.01). In general, pedicel diameters increased linearly with increasing fruit fresh and dry weights (Fig. 2.2a), fruit diameter (Fig. 2.2b), and number of fully developed seeds present per fruit (Fig. 2.2c) at P<0.05, 0.05, 0.001, respectively.



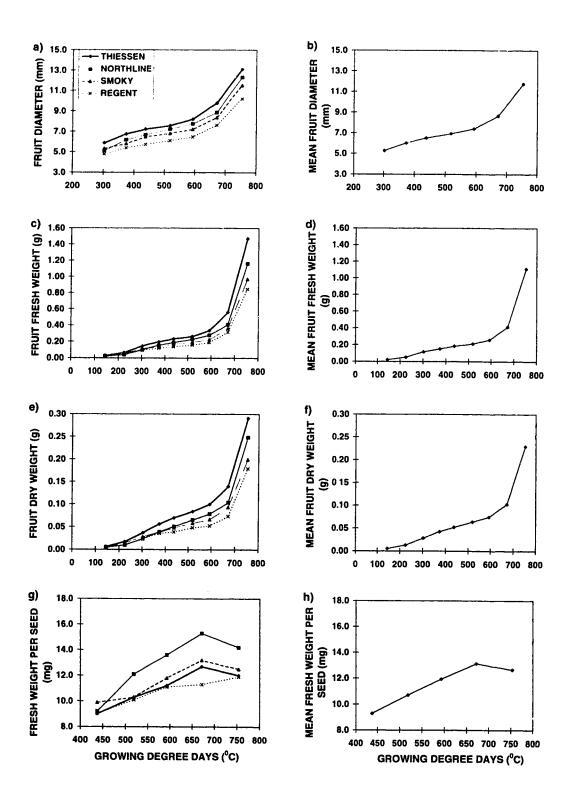
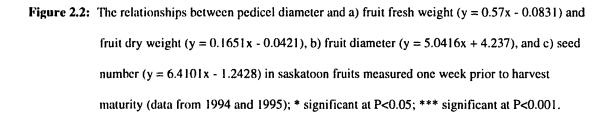
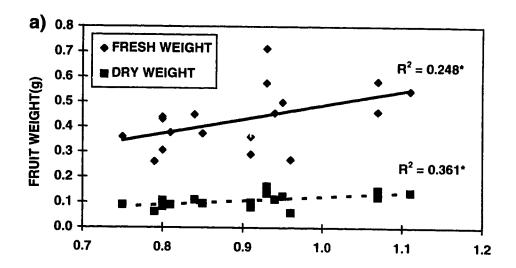


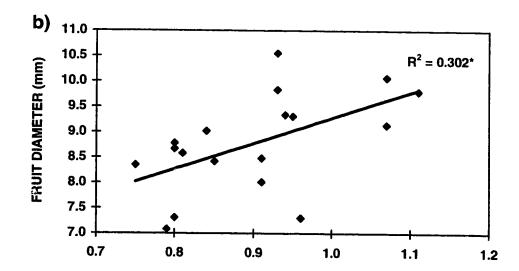
Table 2.1: Pedicel diameters of four saskatoon cultivars measured one week prior to harvest maturity.

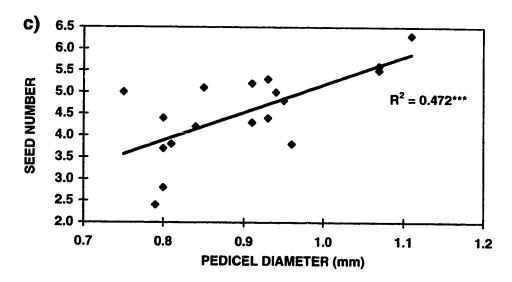
1.01	ER (mm)
1.01	a
0.88	b
0.83	b
0.84	b
	0.83

Mean separation among cultivars for pedicel diameter (ab) by LSD, P<0.01.







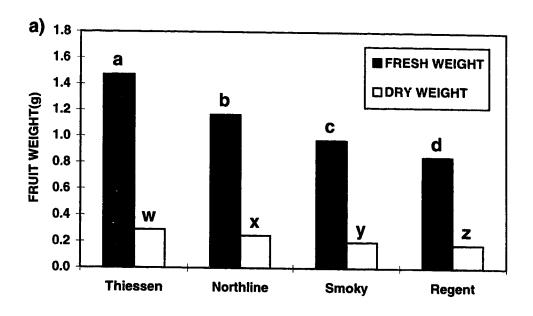


At harvest maturity (July 15, 1994 and July 18, 1995), fruit diameters and fruit fresh and dry weights varied significantly among cultivars (ANOVA, P<0.01). Fruit fresh and dry weights and diameters of cv. Thiessen were the largest, followed by cv.'s Northline, Smoky, and Regent, in descending order (Fig. 2.3a,b). The number of fully developed seeds per fruit at harvest maturity also varied significantly among cultivars (ANOVA, P<0.05), with cv. Thiessen fruit containing significantly more seeds per fruit than the other cultivars (Table 2.2; LSD, P<0.01). The fresh weight per seed varied significantly among cultivars (ANOVA, P<0.01; Table 2.2), with cv. Northline exhibiting the greatest fresh weight per seed (LSD, P<0.01). The total seed fresh weight per fruit varied significantly among cultivars (ANOVA, P<0.05), with the fresh weight of seeds per fruit from cv.'s Thiessen and Northline significantly greater than those from cv.'s Regent and Smoky (Table 2.2; LSD, P<0.01). At maturity, seed number correlated linearly with fruit fresh and dry weights (Fig. 2.4a), and fruit diameter (Fig. 2.4b) at P<0.001. Seed fresh weight per fruit correlated linearly with fruit diameter (Fig. 2.5a), and fruit fresh and dry weights (Fig. 2.5b) at P<0.001.

2.4 Discussion

In general, the pattern of saskatoon fruit development is characterized by a relatively steady rate of growth with a single period of exponential growth occurring later in development. If saskatoons remain on the shrub past harvest maturity, fruit growth slows and eventually ceases (Rogiers and Knowles, in review). Pome fruits, such as apple and pear, typically exhibit sigmoid growth curves with a single period of exponential fruit growth (Westwood, 1988). In contrast, berries and drupes typically exhibit double sigmoid growth curves, where a phase of minimal fruit development separates two periods of exponential fruit growth (Coombe, 1976; Monselise, 1986).

Patterns of saskatoon fruit development can be used to optimize orchard management strategies. Orchard irrigation can be scheduled at the onset of the exponential phase of saskatoon fruit growth (Stage III) to ensure and/or enhance fruit fresh weight gain. Pesticides should be applied during the phase of minimal fruit growth (Stage II) as to maximize the resources available to the remaining fruits. Since saskatoon from attain harvest maturity during the period of very rapid fruit development, immediate post-harvest fruit care should be provided.



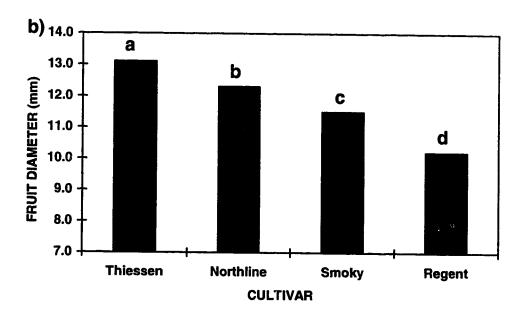


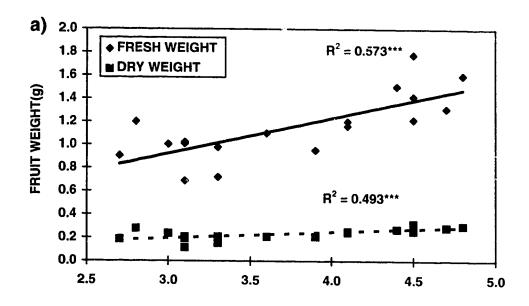
Figure 2.3: a) Fruit fresh and dry weights and b) fruit diameters of four saskatoon cultivars at harvest maturity. Mean separation within cultivar for fresh weight (abcd), dry weight (wxyz), and diameter (abcd) by LSD, P<0.05.

Table 2.2: The number of seeds per fruit, fresh weight per seed, and total seed fresh weight per fruit among saskatoon cultivars measured at harvest maturity *.

CULTIVAR	NUMBER OF SEEDS	FRESH WEIGHT	TOTAL SEED FRESH WEIGHT
	PER FRUIT	PER SEED (mg)	PER FRUIT (mg)
Thiessen	4.57 a	11.9 b	54.6 a
Northline	3.65 b	14.2 a	51.5 a
Smoky	3.20 b	12.5 b	40.1 b
Regent	3.18 b	11.9 b	37.7 b

Mean separation within columns (ab) by LSD, P<0.01.

* Only fully developed seeds per fruit were analyzed.



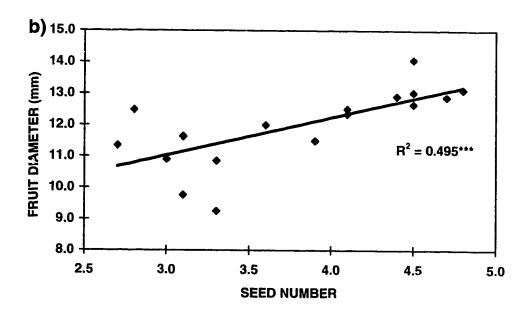
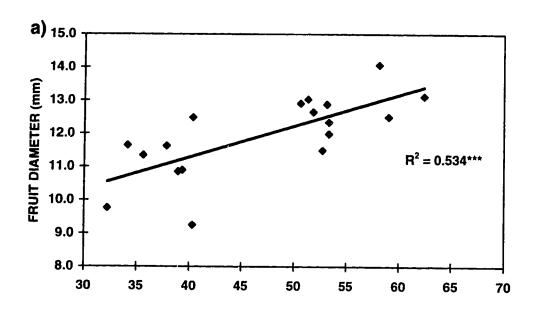


Figure 2.4: The relationship between seed number per fruit and a) fruit fresh weight (y = 0.307x + 0.0022) and fruit dry weight (y = 0.0541x + 0.0332), b) fruit diameters (y = 1.2017x + 7.4279) in saskatoon fruits at harvest maturity (data from 1994 and 1995); *** Significant at P<0.001.



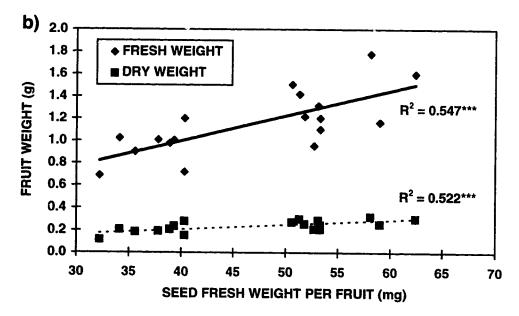


Figure 2.5: The relationship between seed fresh weight per fruit and

a) fruit diameter (y = 0.0941x + 7.5213), b) fruit fresh weight (y = 0.0226x + 0.0934) and fruit dry weight (y = 0.0042x + 0.0394) in saskatoon fruits at harvest maturity (data from 1994 and 1995); *** significant at P<0.001.

Sustained fruit growth is often correlated to the number of developed seeds per fruit (Vaccinium corymbosum: Gorchov, 1985; Brewer and Dobson, 1969; Eaton, 1967; tomato: Dempsey and Boynton, 1965; and Amelanchier arborea: Gorchov, 1985); seed number can also affect final fruit size (Dennis, 1984). In mature saskatoons fruits, pedicel and fruit size increased linearly with the number of developed seeds, suggesting that saskatoon seeds influence growth of the surrounding fruit tissues. Seed number distinguished ev. Thiessen (4.57 seeds per fruit), the cultivar producing the largest fruits, from the remaining cultivars (3.34 seeds per fruit). Thiessen fruits also had larger pedicels compared to the other cultivars studied. Immature seeds are known to contain high concentrations of hormones (Coombe, 1976). It is possible that saskatoon seeds may release hormones which stimulate vascularization within the pedicel. Subsequently, larger fruit size in cv. Thiessen may result from seed-derived hormones enhancing pedicel vascularization which, in turn, can increase the delivery of photosynthates to the developing fruits. The cultivar producing the penultimate sized fruits, cv. Northline, had the largest fresh weight per seed compared to the other cultivars studied. Seed fresh weight per fruit differentiated the larger from smaller cultivars (cv.'s Thiesen and Northline vs. cv.'s Smoky and Regent). The results of this research indicate that fruit size in saskatoon is related to seed number per fruit and the total seed fresh weight per fruit. Furthermore, these data suggest that seed number is more critical than seed size in determining the largest fruit size in the saskatoon cultivars studied.

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3. MESOCARP DEVELOPMENT IN SASKATOON FRUITS

3.1 Introduction

Canada's native saskatoon shrub (*Amelanchier alnifolia* Nutt.) has considerable potential as a cultivated horticultural crop (St. Pierre, 1992). Comparisons of saskatoon fruit growth among cultivars can offer insight into many horticultural procedures, including breeding and orchard management, but such research has been limited (Chapter 2). Fruit size among saskatoon cv.'s Thiessen, Northline, Smoky, and Regent varies consistently throughout the season to yield significantly different sized fruits at harvest maturity (cv. Thiessen > Northline > Smoky > Regent; Chapter 2).

Fruit size is largely a function of cell division in the early stages of fruit growth, and cell enlargement in the final stages of fruit growth (Smith, 1950; Westwood et al, 1967). Using saskatoon fruits from natural stands, Olson and Steeves (1982) observed changes occurring within the outer and inner epidermis, hypodermis, and the outer and inner flesh (mesocarp) of the saskatoon fruit wall during development. Cells of the uniseriate outer and inner epidermis exhibited only minor cellular expansion during fruit growth while cells of the biseriate hypodermis expanded considerably (4.2 times). The outer flesh of the saskatoon fruit increased from 4-7 layers at anthesis to 10-16 layers at maturity, with cells expanding 4.6 times in diameter. The inner fruit flesh expanded from 10-13 layers at anthesis to 13-17 layers at maturity, with cells increasing 4.5 times in diameter during this period. Olson and Steeves (1982) concluded that fruit enlargement in wild saskatoons occurred as a result of cell division and expansion predominantly within the outer and inner flesh.

Characteristic varietal size among apple cultivars results primarily from cell multiplication after pollination (Smith, 1950). Similarly, peach fruit size differences within and among cultivars have been attributed to differences in mesocarp cell number rather than cell size (Bradley, 1959; Scorza et al, 1991). Although the size of saskatoon fruits at harvest maturity differs significantly among cultivars (Chapter 2), the roles of cell division and elongation upon final fruit size have not been determined. The objective of this study was to determine if fruit size differences among saskatoon cultivars result from differences in mesocarp cell number or cell size.

3.2 Materials and Methods

Tissues for histological studies were harvested during the 1994 growing season at the Alberta Crop Diversification Centre North in Edmonton, Alberta, Canada. Six-year old saskatoon shrubs of cv.'s Thiessen, Northline, Smoky, and Regent were selected following a split-split-plot design with cultivar as the main plot, sampling date as the split-plot (compressed bud, full bloom, and four post-bloom stages), and fruits as the spit-split-plot (4 fruits per replication). Main plots were arranged in randomized blocks with three replications for cv. Thiessen, and two replications for each remaining cultivar.

Two compressed buds/flowers/fruits were randomly harvested from each of ten randomly selected inflorescences/infructescences per shrub. Tissues were collected at the compressed bud stage (April 15), full bloom (May 14), and biweckly thereafter (June 1, June 15, June 29) to harvest maturity (July 15), and harvested onto ice. Compressed buds and flowers were vacuum infiltrated overnight in 3.0% glutaraldehyde fixative in 0.1 M phosphate buffer, dehydrated in a graded ethanol series (at 30 minute intervals for each 15% increment in ethanol) followed by two changes in propylene oxide, and infiltrated with Spurr's resin (1969). Using a glass knife, buds and flowers were sectioned 1 μm thick using the Reichert "Om U 2" Ultramicrotome (Reichert, Vienna, Austria), and stained with 0.5% toluidine blue-O in 0.1% sodium carbonate (pH 11.1). Post-bloom tissues were vacuum aspirated overnight in formalin-acetic acid fixative, dehydrated in a graded ethanol/tertiary butyl alcohol series (at 2 hour intervals for each increment), embedded in Paraplast Plus paraffin (Oxford Labware, St. Louis, MO), sectioned 10 μm (June 1 samples), 15 μm (June 15 samples), and 20 μm (June 29 and July 15 samples) thick with a Leitz rotary microtome, and stained with 2% safranin (O'Brien and McCully, 1981).

Four tissue sections per bud/flower/fruit (four buds/flowers/fruits per shrub per sampling date) were viewed through a compound microscope at 10X magnification, and the image was relayed through a video camera (Solid State 4 Color Video Camera, Hitachi VK-C350, Hitachi Ltd., Japan) to an attached Macintosh IIfx computer monitor. Four regions of mesocarp tissue (avoiding vascular bundles) were selected within each section, and the sample area, the number of cells within the sample, and the area per cell were determined using image analysis software NIH 1.5 (National Institute of Health, USA). The total amount of mesocarp tissue present in bud and flower sections was determined by computer analysis. The

total amount of mesocarp tissue present in fruit sections was calculated using the equation for the area of a circle

area =
$$\pi$$
 (diameter/2)² (Equation 3.1)

with diameters measured using an ocular micrometer. The total number of mesocarp cells per bud/flower/fruit section was estimated using the following ratio:

Sampling dates in each year were expressed as heat units (HU) with the following equation:

$$HU = [(T_{max} + T_{min})/2] - T_{base}$$
 (Equation 3.3)

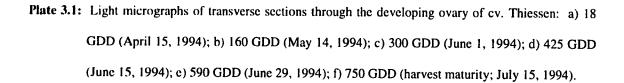
where T_{max} = daily maximum temperature (0 C), T_{min} = daily minimum temperature (0 C), and T_{base} = base temperature of 4.4 0 C. Negative HU were assigned a value of zero. Cumulative HU (from April 1) were expressed as growing degree days (GDD) as follows:

$$GDD = \sum_{1}^{n} HU$$
 (Equation 3.4)

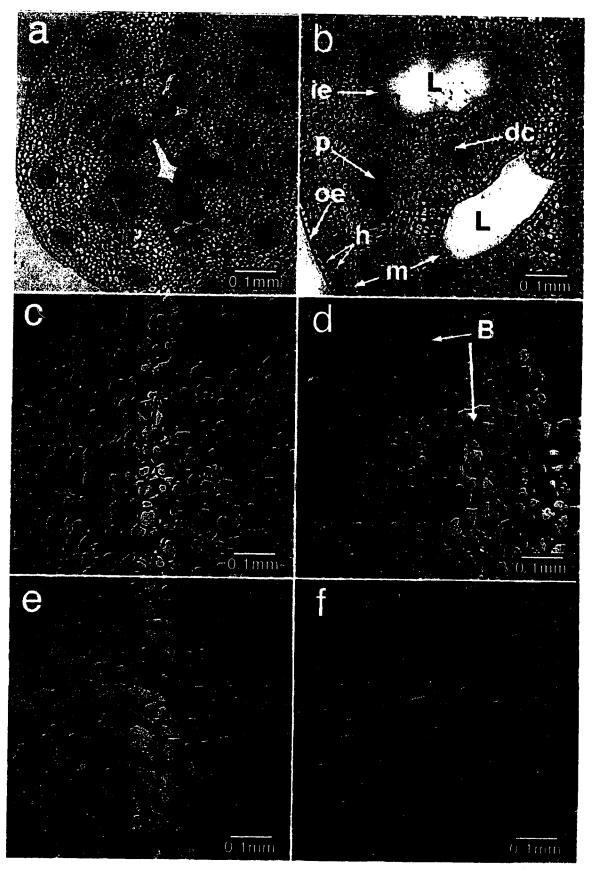
where n = days. For statistical analysis, the General Linear Model of SAS 6.10 program (SAS Institute Inc., Cary, NC 27513, USA) was used.

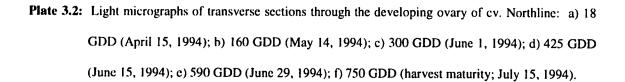
3.3 Results

Mesocarp development of cv.'s Thiessen, Northline, Smoky, and Regent fruits is illustrated in Plates 3.1 - 3.4. During early flower development (compressed buds, 18 GDD, to 160 GDD), mesocarp cells within each cultivar are similar in size and shape (Plates 3.1-3.4, a,b). By 300 GDD, each component of the fruit wall becomes more distinct: cells of the outer and inner epidermis are more compressed; cells within the hypodermis are more rectangular in shape; and cell size and shape within the mesocarp is less uniform (Plates 3.1-3.4, c). By 425 GDD, brachysclereids and aerenchyma are apparent (Plates 3.1-3.4, d), and most abundant in cv. Thiessen fruits, infrequent in cv.'s Northline and Smoky fruits, and absent from cv. Regent fruits. From 425 GDD to harvest maturity (750 GDD), aerenchyma become more prevalent within the mesocarp of all cultivars studied (Plates 3.1-3.4, d,e,f).



L=locule; ie=inner epidermis; p=periantho-staminal bundle; oe=outer epidermis; h=hypodermis; m=mesocarp; dc=dorsal carpellary bundle; B=brachysclereid





c=carpel; o=ovule; oe=outer epidermis; h=hypodermis; ie=inner epidermis; m=mesocarp; p=periantho-staminal bundle

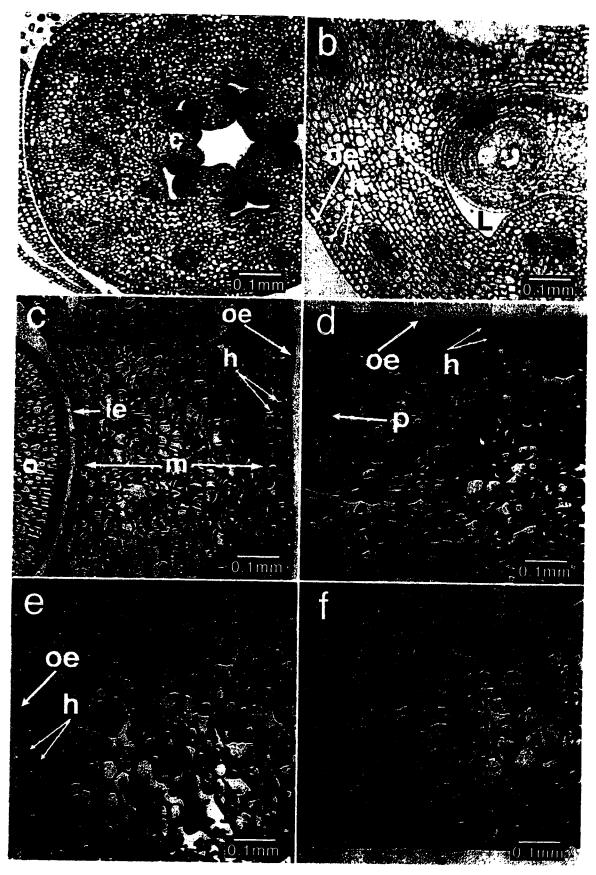
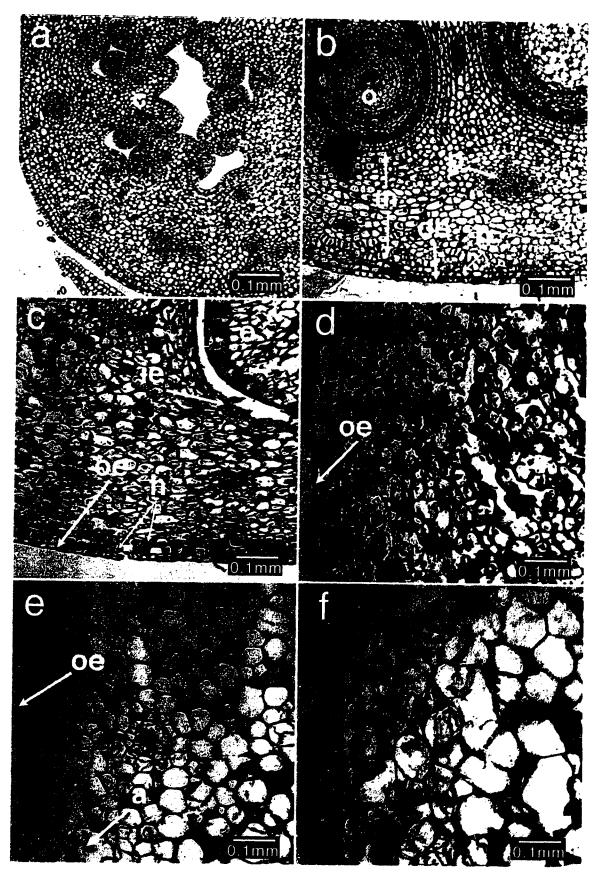


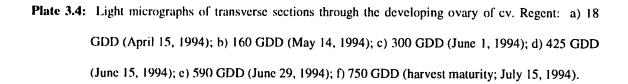
Plate 3.3: Light micrographs of transverse sections through the developing ovary of cv. Smoky: a) 18

GDD (April 15, 1994); b) 160 GDD (May 14, 1994); c) 300 GDD (June 1, 1994); d) 425 GDD

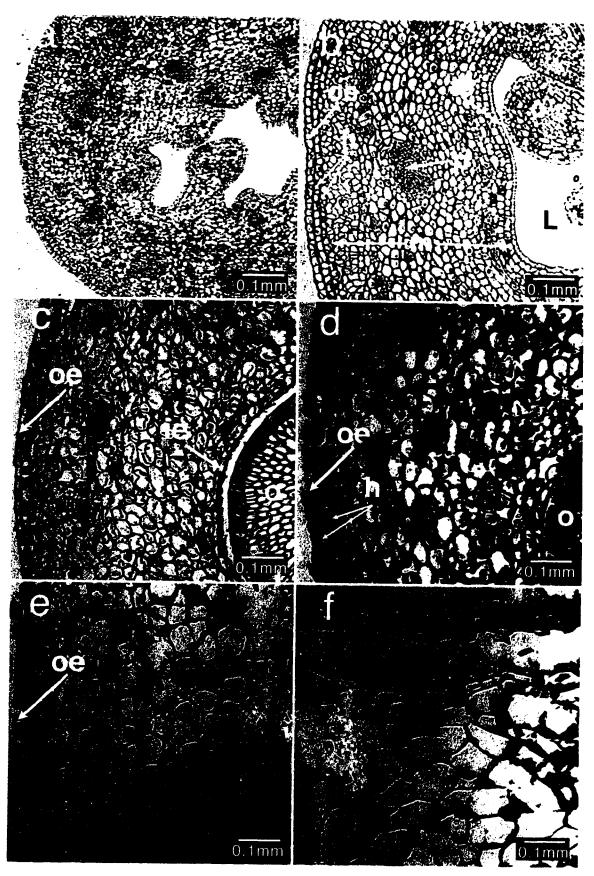
(June 15, 1994); c) 590 GDD (June 29, 1994); f) 750 GDD (harvest maturity; July 15, 1994).

c=carpel; o=ovule; m=mesocarp; p=periantho-staminal bundle; oe=outer epidermis; h=hypodermis; ie=inner epidermis; a=aerenchyma





o=ovule; oe=outer epidermis; h=hypodermis; ie=inner epidermis; p=periantho-staminal bundle; m=mesocarp; L=locule



The number of mesocarp cells present per fruit cross-section was significantly different among saskatoon cultivars (ANOVA, P<0.05), with significantly fewer mesocarp cells present in cv. Regent fruits than the other cultivars (Fig. 3.1; LSD, P<0.05). In cv's Thiessen, Northline, and Smoky, the number of cells present per cross-section increased by an average of 4.33 cells/GDD from 18 to 160 GDD, and then rapidly increased by 80.5 cells/GDD until 300 GDD (Fig. 3.1). After 300 GDD, cell number in fruits from cv.'s Thiessen, Northline, and Smoky remained relatively constant (Fig. 3.1). In cv. Regent fruits, cell number per cross-section initially increased by 2.58 cells/GDD up to 160 GDD, then by 11.5 cells/GDD between 160 to 300 GDD, and then rapidly increased by 80.0 cells/GDD between 300 to 425 GDD (Fig. 3.1). The size of mesocarp cells did not vary among cultivars (ANOVA, P>0.10). In general, the size of mesocarp cells initially increased by 0.370 μm²/GDD up to 160 GDD, and then increased by 0.996 μm²/GDD up to 300 GDD. Mesocarp cells continued to expand at a reduced rate, increasing 0.210 μm²/GDD from 300 GDD to harvest maturity (750 GDD; Fig. 3.2 a,b).

3.4 Discussion

The previous growth study among saskatoon fruit cultivars established that cv. Thiessen has the largest fruits (fruit diameter and fruit fresh/dry weights) throughout the season and at harvest maturity, followed by cv.'s Northline, Smoky, and Regent, in descending order (Chapter 2). In general, the pattern of saskatoon fruit development is characterized by a relatively steady increase in fruit diameter, fruit fresh weight, and fruit dry weight with a single period of exponential growth occurring later in development (Chapter 2). The patterns of mesocarp development with respect to cell number and size were similar among the larger fruited saskatoon cultivars (cv.'s Thiessen, Northline, and Smoky). Cell division in cv.'s Thiessen, Northline and Smoky was most active during Stage I (18 to 300 GDD), and minimal during Stages II (300 to 600 GDD) and III (600 GDD to harvest maturity at 760 GDD) of saskatoon fruit growth (Fig. 3.1; stages defined in Chapter 2). Cell division in cv. Regent fruits occurred later in the growing season, continuing past Stage I to 425 GDD. Variable mesocarp cell counts were obtained from 425 GDD to harvest maturity within each cultivar; however, the general trend suggests that cell division was minimal during this time. The pattern of cell division within the mesocarp of saskatoon fruits was similar to that of

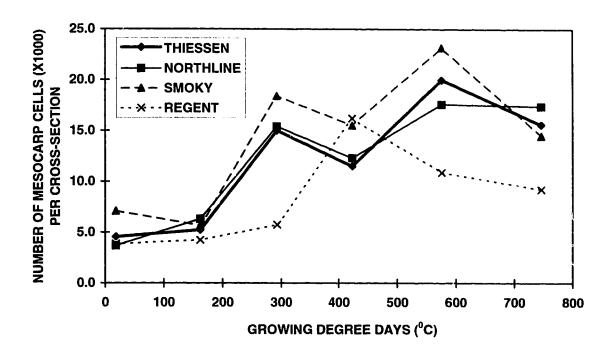


Figure 3.1: Changes in mesocarp cell number (x1000) (per cross-section) during fruit development in saskatoon cv.'s Thiessen, Northline, Smoky, and Regent.

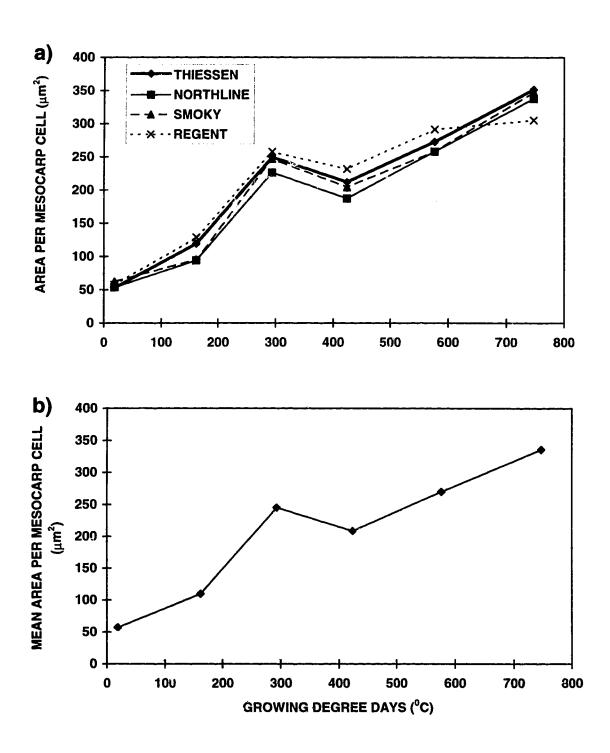


Figure 3.2: Changes in mesocarp cell size (μm²) during saskatoon fruit development: a) in cv.'s Thiessen,

Northline, Smoky, and Regent; b) mean mesocarp cell size among cultivars.

other fruits, where cell number increases rapidly during early fruit development and then decreases or ceases in the later stages of fruit growth (apple Blanpied and Wilde, 1968; Dennis, 1986; Tukey and Young, 1942; apricot: Jackson and Coombe, 1966; kiwi: Hopping, 1986). The pattern of cell elongation within the mesocarp of saskatoon fruits was also similar to that of other fruits, where cell size increases most actively during Stage I of fruit growth, and thereafter continues to expand at a reduced rate (apricot: Jackson and Coombe, 1976; avocado: Schroeder, 1953; kiwi: Hopping, 1986). A slight decrease in cell size was apparent at 425 GDD in all saskatoon cultivars studied, suggesting that some external event, such as temperature or drought stress, may have reduced cell size at this time. In general, saskatoon fruit growth can be attributed to cell division and cell elongation within the mesocarp during Stage I, and continued cell elongation during Stages II and II.

Cv. Regent fruits contained ancantly fewer mesocarp cells than the cultivars producing larger sized fruits (cv.'s Thiessen, Northline and Smoky). This suggests that a minimum number of mesocarp cells may be required in order to attain the larger fruit sizes observed in cv.'s Thiessen, Northline, and Smoky. Studies with other fruit crops have similarly reported that fruit size differences among cultivars result from distinctions in mesocarp cell number (peach: Scorza et al, 1991; apple: Smith, 1950; avocado: Schroeder, 1953; and apricot: Jackson and Coombe, 1966); generally, these differences are established during very early fruit development (Bergh, 1985; Smith, 1950). Since mesocarp cell number did not vary significantly among the larger fruited saskatoon cultivars (Thiessen, Northline, and Smoky), factors external to these cellular processes, such as the density of bloom and fruit set (Westwood et al, 1967) may have affected fruit size. Furthermore, in Chapter 2, it was established that seed number, and seed fresh weight per fruit significantly affect fruit size among the saskatoon cultivars studied. Therefore, these studies indicate that mesocarp cell number, seed number, and seed fresh weight per fruit contribute significantly to the observed fruit size differences among saskatoon cv.'s Thiessen, Northline, Smoky, and Regent.

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4. THE EFFECTS OF GIBBERELLINS ON FRUIT SET AND SEED NUMBER IN SASKATOON

4.1 Introduction

Saskatoon fruit production among natural stands has been unreliable despite consistently heavy annual blooms (St. Pierre, 1989). St. Pierre (1989) observed that only 19% of flowers attained full fruit maturity. Low fruit yields were not due to unsuccessful pollination (Steeves et al, 1979). Steeves et al (1979) observed that 74.4% of flowers from natural stands were successfully pollinated and initially set fruit. However, only 33.1% of those blossoms yielded fruits at harvest maturity (Steeves et al, 1979). These observation suggest that post-pollination events reduce the set of wild saskatoon flowers.

Fruit set can be affected by numerous conditions, cultural practices, and chemical treatments applied prior to, during, and shortly after anthesis (Dennis, 1986). Gibberellins (GAs) are among the most active chemicals applied to increase the set of open-pollinated flowers (Dennis, 1986). Additionally, GAs applied to unpollinated (Barker and Collins, 1965; Looney and Pharis, 1986; Pharis and King, 1985) and pollinated (Crane, 1964; Pharis and King, 1985) flowers can promote parthenocarpic fruit development, as demonstrated in apple, blueberry, and tomato (Naylar, 1984). Each saskatoon fruit, a pentalocular pome, is capable of producing ten seeds (St. Pierre and Steeves, 1990); however, mature saskatoons usually contain 3-5 seeds per fruit at harvest maturity (Chapter 2). Reducing the seed content in saskatoon fruits could improve the quality of many value-added products, including saskatoon jams, pie filling, and raisins. The objective of this study was to test the effectiveness of GAs in promoting fruit set and reducing seed number in saskatoon fruits.

4.2 Materials and Methods

4.2.1 Field Experiments

Field experiments were conducted in two seasons (1994 and 1995) at the Alberta Crop Diversification Centre North in Edmonton, Alberta, Canada. Six-year old trees of cv.'s Smoky and

Northline were selected following a split-plot design with application time (3 stages of flower development) as the main plot, and treatment (3 chemicals at 2 concentrations) as the split plot. Main plots were arranged in randomized blocks with three replicates. Applications were timed weekly to coincide with 50-70% bloom [May 12, 1994 (19.5°C) for cv. Smoky, and May 13, 1994 (18.5°C) for cv. Northline; and May 23, 1995 (17.5°C) for cv.'s Smoky and Northline], full bloom [May 19, 1994 (14.0°C); and May 30, 1995 (28.5°C)], and petal fall [May 25, 1994 (26.0°C) for cv. Northline, and May 26, 1994 (24.8°C) for cv. Smoky; and June 7, 1995 (17.4°C) for cv.'s Smoky and Northline]. Treatments (in 0.2% Tween 20) included GA₃ (4% w/w), GA_{4/7} (2.0% w/w), and GA_{4/7}+BA (1.8% w/w each) (Abbott Laboratories, Chemical and Agricultural Products Division, North Chicago, IL 60064), at 100 and 200 ppm, and a control solution of 0.2% Tween 20. Treatments were randomly assigned to branches 37 cm long, bearing three inflorescences. Treatments were applied to run-off, using screens surrounding each branch to prevent drift during application.

Treated fruits were collected onto ice at harvest maturity (July 20, 1994; July 26, 1995), and a subsample of ten fully mature (purple-blue colored) fruits was analyzed from each treatment replicate. Fruit diameters were measured using calipers at the widest region perpendicular to the pedicel-calyx axis, and the fresh weight per fruit was determined gravimetrically. Seed analysis involved dissection of seeds from fruits, scoring seed number, gravimetrically determining the fresh weight per seed, and, after drying at 60°C for one week, gravimetrically determining the dry weight per seed. Plump seeds containing fully formed cotyledons and embryo axis were scored as "fully developed" seeds. Fruit set was calculated as the percentage of fruits harvested compared to the number of flowers present at or prior to treatment. Return bloom was calculated as a percentage of flowers present in the year after treatment compared to the number of flowers present at or prior to treatment.

At the Alberta Crop Diversification Centre North in 1995, emasculated flowers of cv. Smoky were treated with GAs who a randomized complete block design with three replicates and five treatments. GA₃, GA₄₇, GA₄₇ + 32 (100 ppm in 0.2% Tween 20) and a control solution of 0.2% Tween 20 were randomly assigned to branches bearing three emasculated inflorescences. The inflorescences were emasculated at petal elongation (Olson, 1984), treatments were applied to run-off, and the treated

inflorescences were immediately covered with pollination bags. Analyses followed as per the previous field trials.

4.2.2 Greenhouse Experiment

Saskatoon plants (three-year old cv. Pembina) grown in one gallon size pots were removed from 4°C storage on March 8, 1995, and placed in a greenhouse maintained at 21°C with a 16 hour photoperiod following a randomized complete block design (four replicates and nine treatments). The sib-pollinated plants were treated at 50-70% bloom with GA₃, GA_{4/7}, and GA_{4/7}+BA (in 0.2% Tween 20) at 100 and 200 ppm, and a control solution of 0.2% Tween 20. To observe fruit development on self-fertilized plants, inflorescences were covered with pollination bags prior to bloom. Analyses followed as per the experiments.

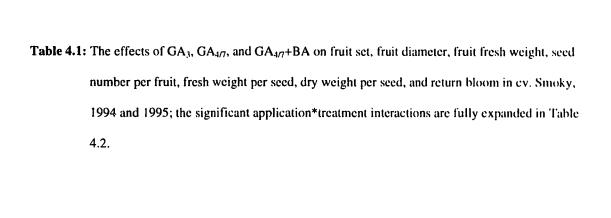
All statistical analyses were completed using the General Linear Model of SAS 6.10 program (SAS Institute Inc., Cary, NC 27513, USA).

4.3 Results

4.3.1 Field Experiments

Fruit set in cv. Smoky in 1994 was significantly different among treatments, with a significant application by treatment interaction (Table 4.1; ANOVA, P<0.05). At 50-70% bloom, fruit set was significantly reduced by 100 ppm GA_{4/7} and GA_{4/7}+BA; at full bloom, 200 ppm GA₃ and 100 ppm GA_{4/7}+BA significantly reduced fruit set; at petal fall, GA₃ (100 and 200 ppm) and 200 ppm GA_{4/7} significantly reduced fruit set (Table 4.2; LSD, P<0.05). The GA treatments did not affect fruit set in cv. Smoky in 1995 (Table 4.1; ANOVA, P>0.10), or cv. Northline in 1994 or 1995 (Table 4.3; ANOVA, P>0.10).

Seed number per fruit in cv. Smoky in 1995 was significantly different among treatments, with a significant application by treatment interaction (Table 4.1, ANOVA, P<0.05). At 50-70% bloom, seed number was significantly reduced by 100 ppm GA_{4/1}, at full bloom by 100 ppm GA₃, and at petal fall by 100 and 200 ppm GA₃ (Table 4.2; LSD, P<0.05). Seed number per fruit in cv. Smoky in 1994 was not affected by GA treatments (ANOVA, P>0.05). In cv. Northline in 1994, seed number was significantly



		% FRUIT	FRUIT	FRUIT FRESH	SEED NUMBER	FRESH WEIGHT PER	DRY WEIGHT PER	% RETURN
		SET	(mm)	WEIGHT (g)	PER FRUIT	SEED (mg)	SEED (mg)	BLOOM
1994	application							
	50-70% sloom	53.4	11.9	0.892	2.47 b	11.2	6.56	354
	fuli bloom	56.2	12.6	1.01	2.94 a	11.6	6.79	210
	petal fall	61.2	12.2	0.928	2.62 b	11.2	6.56	166
	treatment)	3
	control	6.89	12.1	0.973	3.02	11.8	6.82	116
	100 ppen GA ₃	42.8	12.5	0.987	2.39	11.3	9:99	127
	200 ppm GA ₃	31.7	12.2	0.926	2.58	10.8	6.54	061
	100 ppm GA40	62.3	12.4	0.941	2.90	11.5	6.71	326
	NO ppm GAM	63.6	12.6	0.951	2.32	11.4	6.76	151
	(W ppm GA47+BA	58.0	12.0	0.931	2.77	11.6	6.84	347
	200 ppm GA ₄₇₇ +BA	71.2	11.8	0.892	2.75	10.9	6.22	146
	application*treatment	*	su	su	ns	ns	ns.	SII
	co-efficient of	35.6	6.31	13.0	19.4	10.9	7.38	127.8
	variation (%)	;						
1995	application							
	50-70% bloom	71.4 b	12.8	1.11	3.51	10.9 b	6.38	20.4
	full bloom	89.1 a	12.9	1.28	3.88	11.5 ab	09.9	25.8
	petal fall	81.6 a	12.6	1.31	3.66	11.9 a	6.85	18.7
	treatment							
	control	65.7	12.3 b	1.15	3.99	12.0	6.87	37.5
	100 ppm GA ₃	80.1	13.2 a	1.31	3.32	11.6	6.72	12.6
	200 ppm GA ₃	83.9	13.1 a	1.30	3.46	10.8	6.49	9.51
	100 ppm GA ₄₇	6.98	12.8 ab	1.20	3.64	11.2	6.34	25.0
	200 ppm GA ₄₇	84.2	13.2 a	1.29	3.78	11.4	99.6	22.1
	100 ppm GA4π+BA	81.6	12.4 b	1.22	3.76	11.5	89.9	20.2
	200 ppm GA ₄₇₇ +BA	82.3	12.5 ab	1.17	3.86	11.5	6.52	24.6
	application*treatment	us	Sh	su	*	ns	ns	SH.
	co-efficient of	14.0	5.56	11.4	12.3	7.03	5.88	142.0
	variation (%)							

Mean separation within columns (ab) by LSD, P<0.05; * indicates a significant application by treatment interaction (ANOVA, P<0.05; refer to Table 4.2); ns indicates no significant application by treatment interaction (ANOVA, P>0.05).

Table 4.2: — se effects of GA₃, GA₄₇₇, and GA₄₇₇+BA applied at 50-70% bloom, full bloom, and petal fall, on fruit set (1994) and seed number (1995) in ev. Smoky.

APPLICATION	TREATMENT	% FRUIT SET (1994)	SEED NUMBER PER FRUIT (1995)
50-70% bloom	control	40.6 b	3.87 a
	100 pg	27.3 c	3.73 ab
	200 pr	36.5 b	3.70 ab
	100 ppi. 7	74.9 a	2.98 в
	200 ppm GA _{4/7}	61.2 ab	3.47 ab
	100 ppm GA _{4/7} +BA	79.2 a	3.37 ab
	200 ppm GA _{4/7} +BA	54.4 abc	3.47 ab
full bloom	control	76.4 ab	4.15 a
	100 ppm GA ₃	66.9 ab	3.23 b
	200 ppm GA ₃	34.7 c	3.77 ab
	100 ppm GA _{4/7}	48.9 b	4.30 a
	200 ppm GA _{4/7}	82.3 a	3.87 ab
	100 ppm GA _{4/7} +BA	29.0 с	4.00 a
	200 ppm GA _{4/7} +BA	54.7 ab	3.87 ab
petal fall	control	89.8 ab	3.97 a
	100 ppm GA ₃	34.0 c	3.00 b
	200 ppm GA ₃	23.8 d	2.90 b
	100 ppm GA _{4/7}	63.1 bc	3.63 a
	200 ppm GA _{4/7}	47.3 cd	4.00 a
	100 ppm GA _{4/7} +BA	65.8 bc	3.90 a
	200 ppm GA _{4/7} +BA	100 a	4.23 a

Mean separation within application time (abcu) by LSD, P<0.05.

Table 4.3: The effects of GA₃, GA_{4/7}, and GA_{4/7}+BA on fruit set, fruit diameter, fruit fresh weight, seed number per fruit, fresh weight per seed, dry weight per seed, and return bloom in ev. Northline, 1994 and 1995.

:		%	FRUIT	FRUIT	SEED	FRESH	DRY WEIGHT	26
		FRUIT SET	DIAMETER (mm)	FRESH WEIGHT (g)	NUMBER PER FRUIT	WEIGHT PER	PER SEED	RETURN BI OOM
1994	application			(G)		(S)	(9,,,)	DECOM
	50-70% bioom	56.2	12.2	1.11	1.26	11.4	7.06	348
	moold llul	48.4	12.7	1.22	2.13	11.9	7 0 7	167
	petal fall	49.5	13.0	1.25	2.51	5	6.48	84.1
	treatment				:)	2	
	control	45.7	12.7	1.28	2.77 a	13.9	7.73	302
	100 ppm GA ₃	48.8	12.5	1.15	1.86 b	10.9	6.23	258
	200 ppm GA ₃	47.4	13.2	1.29	1.94 b	12.2	86.9	<u> </u>
	100 ppm GA _{4/7}	52.0	12.8	1.22	1.82 b	12.5	7.13	091
	200 ppm GA _{4ח}	60.3	12.8	1.21	2.08 b	11.9	6.73	213
	100 ppm GA₄π+BA	59.3	13.1	1.24	1.92 b		29.9	212
	200 ppm GA₁n+BA	46.4	11.1	0.953	1.39 b	8.9	6.59	149
	application*treatment	SH	su	SII	ns	ns	SU	us
	co-efficient of	37.6	12.0	26.7	38.1	29.0	28.0	97.3
	variation (%)							
5661	application							
	50-70% bloom	44.7 b	13.2	1.22	3.18	12.2	7.45	47.9
	full bloom	71.3 a	12.5	1.30	3.52	14.2	7.70	0.79
	petal fall	67.9 a	11.9	1.25	3.38	13.3	7.60	18.3
	treatment							!
	control	63.3	12.5	1.21	3.68	13.0	7.51	73.4
	100 ppm GA3	62.5	12.6	1.25	3.20	12.9	7.24	21.8
	200 ppm GA3	62.8	17.6	1.31	3.26	12.6	7.14	21.7
	100 ppm GA₄♂	64.0	12.4	1.29	3.68	13.9	7.92	25.2
	200 ppm GA₄⊖	62.9	12.7	1.32	3.32	14.2	7.94	62.4
	100 ppm GA _{2.7} +B.A	52.6	13.1	1.42	3.47	12.2	7.80	62.9
	200 ppm GAz+BA	58.0	11.9	966.0	2.89	13.7	7.52	36.5
	application*treatment	211	ns	ns	7.5	115	11.5	SW
	co-efficient of	21.7	8.42	18.4	17.1	181	× 5×	126.43
	variation (%)				•	· · · · · · · · · · · · · · · · · · ·		

Nean separation within columns (ab) by LSD. P<0.05; ns indicates no significant application by treatment interaction (ANOVA. P>0.05).

different among treatments, with all GA treatments significantly reducing the number of seeds per fruit (Table 4.3; LSD, P<0.05). $GA_{4/7}+BA$ slightly reduced the number of seeds per fruit in ev. Northline in 1994 compared to $GA_{4/7}$ (1.66 vs. 1.95; orthogonal contrasts, P<0.01). Seed number per fruit was not affected by treatment in ev. Northline in 1995 (Table 4.3; ANOVA, P>0.10).

Fruit diameter in cv. Smoky fruits in 1995 was significantly different among treatments, with GA₃ (100 and 200 ppm) and GA_{4/7} (200 ppm) significantly increasing fruit diameter (Table 4.1; LSD, P<0.05). Fruit diameter was not affected by treatment in cv. Smoky in 1994 (Table 4.1; ANOVA, P>0.05) or in cv. Northline in 1994 or 1995 (Table 4.3; ANOVA, P>0.05). Fruit fresh weight, and the fresh and dry weights per seed were not affected by GA treatments in cv.'s Smoky (Table 4.1; ANOVA, P>0.05) or Northline (Table 4.3; ANOVA, P>0.05) in 1994 or 1995.

The return bloom was not affected by GA treatments in cv.'s Smoky (Table 4.1; ANOVA, P>0.10) or Northline (Table 4.3; ANOVA, P>0.10); however, extensive variation in flowering was observed among shrubs in both cultivars. Return bloom from the 1994 season was considerably heavier than that from 1995, suggesting a biennial bearing pattern in saskatoon cv.'s Smoky and Northline.

When GA treatments were applied to emasculated flowers of cv. Smoky, no significant differences in fruit set, diameter, weight, or seed number per fruit were detected (Table 4.4; ANOVA, P>0.05); however, extensive variation among GA treatments was observed in all parameters tested (Table 4.4). Emasculated flowers did not set fruit (Table 4.4). The addition of GAs to emasculated flowers tended to improve fruit set, but these fruits did not attain harvest maturity (Table 4.4).

4.3.2 Greenhouse Experiment

In greenhouse grown cv. Pembina plants, fruit set and seed number per fruit were significantly different among treatments (Table 4.5; ANOVA, P<0.05). Seed number per fruit was significantly reduced with 100 ppm GA₃, 200 ppm GA₄₇, and 100 and 200 ppm GA₄₇+BA (Table 4.5; LSD, P<0.05). Self-fertilized cv. Pembina plants also exhibited reduced seed number per fruit (Table 4.5; LSD, P<0.05). Fruit diameter and weight, and the fresh and dry weights per seed were not affected by treatments (Table 4.5; ANOVA, P>0.05).

Table 4.4: The effects of GA₃, GA₄₇, and GA₄₇+BA on emasculated fruit of cv. Smoky, 1995.

TREATMENT	% FRUIT	% MATURE	FRUIT	FRUIT	SEED
	SET	FRUITS *	DIAMETER (mm)	FRESH WEIGHT (g)	NUMBER PER FRUIT
control	6.78	64.6	7.47	9990	1.6
emasculated+no GA	0	0	0	0	0
emasculated+GA3	44.8	14.7	6.07	0.429	0
emasculated+GA _{2G}	18.7	0	0	0	0
emasculated+GA _{1/1} +BA	21.4	0	ت	0	0
co-efficient of variation	104	991	145	148	202
(%)					

* Mature fruits refers to ripe, purple-blue colored fruits. When fruits were harvested, 85.3%, 100%, and 100% of fruits in the emasculated+GA3, emasculated+GA46, and emasculated+GA46+BA, respectively, were at the immature green stage.

Table 4.5: The effects of GA3, GA4n, GA4n+BA, and self-fertilization on fruit set, fruit diameter, fruit fresh weight, seed number per fruit, and seed fresh and dry weight in greenhouse grown cv. Pembina fruits.

TREATMENT	C REUIT	FRUIT	FRUIT FRESH	SEED NUMBER	FRESH	DRV
	SET	DIAMETER	WEIGHT (g)	PER FRUIT	WEIGHT PER	WEIGHT PER
		(mm)			SEED (mg)	SEED (mg)
control	45.8 ab	10.1	0.735	3.33 a	8.75	4.95
100 ppm GA ₃	62.5 a	7.62	0.416	0.985 b	6.78	3.98
200 ppm GA ₃	45.2 ab	89.9	0.406	2.01 ab	5.70	2.68
100 ppm GA ₄₇	43.0 ab	8.83	0.575	2.08 ab	7.35	4.35
200 ppm GA₄⊓	60.0 a	5.53	0.274	0.813 b	4.73	2.98
100 ppm GA477+BA	20.7 bc	6.43	0.367	0.895 b	5.53	3.65
200 ppm GA47+BA	32.8 abcd	7.36	0.384	1.19 b	5.48	3.43
self-fertilized	16.9 bc	4.55	0.304	0.738 b	4.35	2.63
co-efficient of	65.1	49.2	53.7	78.0	49.9	52.8
variation						

Mean separation within columns (abcd) by LSD, P<0.05.

4.4 Discussion

Although gibberellins are among the most active chemicals available to increase fruit set in apples (Dennis, 1986), their effects have been inconsistent (Dennis, 1986) and severely limited in field conditions (Dennis, 1973). GAs can increase (Edgerton, 1981), decrease (Edgerton, 1981; Taylor, 1975; Wertheim, 1982), or not affect (Dennis et al, 1983; Edgerton, 1981; Luckwill and Silva, 1979; Stembridge and Morrell, 1972; Wertheim, 1982) the set of open-pollinated, uninjured apple blossoms. These variations have been previously attributed to fluctuating environmental and plant health conditions (Greene, 1989), as well as the type of GA used (Dennis, 1986), concentration (Dennis, 1986), and time of application (Weaver and McCune, 1960). The effect of GAs on saskatoon fruit set was also variable. In the field experiments conducted, a significant application by treatment interaction was observed on the set of ev. Smoky fruits in 1994; however, the effects of GA₃, GA_{4/1}, and GA_{4/1}+BA were not consistent at each application time (Table 4.2). GA_{4/7} and GA_{4/7}+BA (100 ppm) significantly increased fruit set when applied at 50-70% bloom (Table 4.2), suggesting that these treatments should be applied during early flower development to improve saskatoon fruit set. Weaver and McCune (1960) demonstrated that GA₃ applied at early bloom acted as a pollenicide to reduce the set of Black Corinth grapes. In saskatoon fruits, early bloom applications of 100 ppm GA₃ to cv. Smoky significantly reduced fruit set (to 27.3%; Table 4.2); however, similarly low levels of fruit set were obtained with the full bloom (200 ppm) and petal fall (100 ppm) applications of GA₃ (Table 4.2). Although the time at which GAs were applied did not affect fruit set in cv.'s Smoky and Northline in 1995 (as determined by non-significant application by treatment interactions), the surfactant may have reduced the set of saskatoon flowers in early bloom applications (Tables 4.1 and 4.3). GAs improved the set of emasculated cv. Smoky flowers: however, the proportion of those fruits attaining harvest maturity was dramatically reduced (Table 4.4). These results suggest that although exogenous GAs can set unpollinated saskatoon flowers, the time required for fruit development may be extended, or repeated applications of GAs may be required to obtain mature fruit within the constraints of a growing season.

Exogenous GAs can promote parthenocarpic fruit development of unpollinated flowers by substituting for seed-produced GAs, or GA-induced production of other growth substances by the seed, at a

critical stage of development (Looney and Pharis, 1986; Pharis and King, 1985). Among pollinated flowers, exogenous GAs can reduce pollen germination (Crane, 1964) or inhibit embryogenesis (Pharis and King, 1985) to yield seedless fruits. Previous research with apple fruits has shown that GA₃, GA₄₇₇, and GA₄₇₇+BA can successfully reduce seed number (Bangerth, 1976; Bukovac, 1963; Greene, 1984; Greene et al. 1982; Taylor, 1975). Similarly, GAs were effective at reducing seed number in saskatoon ev.'s Smoky. Northline, and Pembina. However, the reduction in seed number was not consistently repeated in field trials, and more dramatic decreases in seed number were observed among greenhouse grown fruits (0.8-2.0 seeds per GA-treated greenhouse-grown fruit compared to 1.8-4.3 seeds per GA-treated field-grown fruit). These observations suggest that the more controlled greenhouse environment may have better facilitated absorption of the GAs to reduce seed number further. Reducing saskatoon seed number in field experiments requires further experimentation, possibly with higher concentrations of GAs or incorporating different surfactants. Dennis (1973) has suggested that repeater GA applications may influence parthenocarpic fruit development. However, preliminary field experiments employing GA applications at petal fall and again one week later did not affect the number of seeds in saskatoon ev. Pembina fruits (data not shown).

Gibberellins have demonstrated potential for improving fruit set and inducing parthenocarpy in several fruit crops although the results varied depending on the species, cultivar, and type of GA applied. Although GAs did not consistently affect saskatoon fruit set in the cultivars tested, GAs effectively reduced seed number in field experiments, and this effect was greater in the more controlled greenhouse environment. Exogenous GAs have been shown to decrease fruit size (Crane, 1964) and reduce the return bloom (Luckwill and Silva, 1979) in many crops; however, GAs at 100 and 200 ppm did not deleteriously affect fruit size or the return bloom in the saskatoon cultivars tested. Further research will be required to identify the GA concentration, and the type and concentration of surfactant required to obtain maximum seed reduction in the field without negatively affecting fruit set, fruit size, or the return bloom in saskatoon orchards.

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5. THE EFFECTS OF ETHEPHON ON SASKATOON FRUIT RIPENING

5.1 Introduction

As saskatoon orchards expand, mechanically harvesting fruits is rapidly becoming preferred over hand-picking, a costly and laborious process. However, due to the non-uniform ripening pattern exhibited by saskatoon fruits (Green and Mazza, 1986), maximizing the yield of salable fruit is difficult with once-over mechanical harvesting. Ethylene is a naturally occurring ripening hormone in many fruits (McGlasson, 1985). Ethephon, an ethylene-releasing agent, has been used to synchronize ripening in several horticultural crops, including cherry (Olien and Bukovac, 1978), coffee (Browning and Cannell, 1970; Winston et al, 1992), olive (Tous et al, 1995), pepper (Batal and Granberry, 1982; Sims et al., 1974), and tomato (Dostal and Wilcox, 1971). The objective of this experiment was to test the effectiveness of ethephon in stimulating uniform ripening of saskatoon fruits without negatively affecting fruit quality.

5.2 Materials and Methods

Field experiments were conducted during two seasons (1994 and 1995) at the Alberta Crop Diversification Centre North, Edmonton, Alberta, Canada. Six-year-old saskatoon shrubs of cv.'s Northline and Smoky were selected following a split-split-plot design with origin (sucker vs. micropropagated plants) as the main plot, ethephon treatment as the split-plot, and year of treatment as the split-split-plot. Ethephon was applied at 250, 500, and 1000 ppm to cv. Northline, and at 500 and 1000 ppm to cv. Smoky. Main plots were arranged in randomized blocks with three replications.

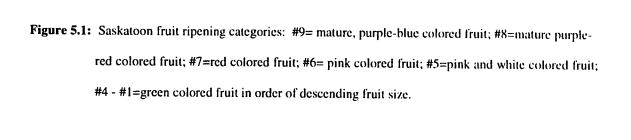
Ethephon solutions were prepared from a stock solution of 480 g/L ethephon (Cerone; Union Carbide, Agricultural Products Co. Inc., USA) diluted with distilled water (pH 5.6) to obtain the desired concentrations, and distilled water was used for the control solutions. Treatments were applied to the entire shrub to run-off when approximately 70% of fruits were red (maturity class #7: Rogiers and Knowles, 1995) [cv. Northline on July 13, 1994 (24.1°C), and July 19 and 21, 1995 (27.3°C and 22.2°C,

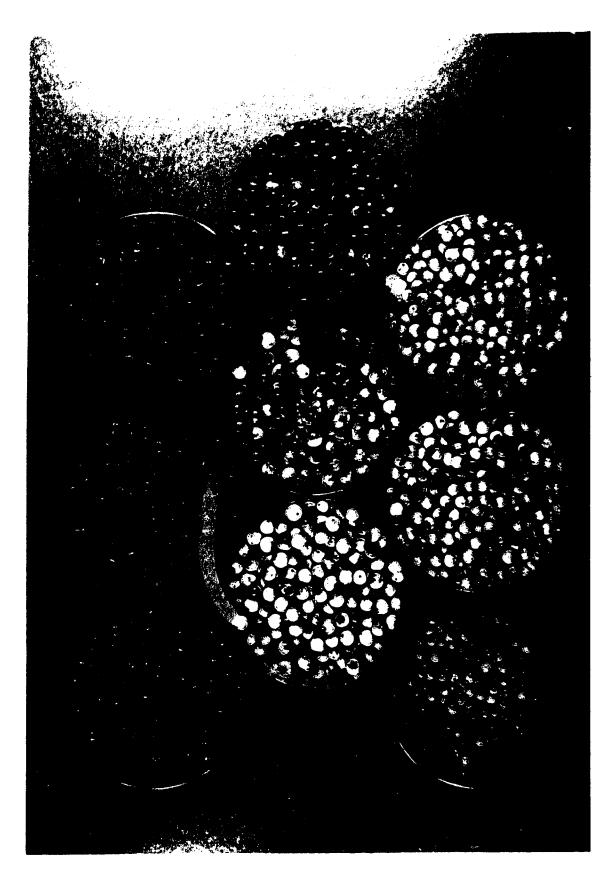
respectively); cv. Smoky on July 13, 1994 (24.1°C) and July 13, 1995 (24.5°C)]. Baffolds were erected between trees to prevent drift during application.

When approximately 70% of the control fruits were purple (maturity class #9: Rogiers and Knowles, 1995) (4-8 days after treatment), fruits were harvested onto ice ev. Northline on July 19, 1994, and July 24 and 25, 1995; cv. Smoky on July 18, 1994, and July 19 and 21, 1995). The total fruit per shrub or a 1 kg sample of harvested fruits (cv. Northline, 1995) was sorted on ice into nine fruit ripening categories grouped according to fruit surface color and size (Rogiers and Knowles, 1995): #9= mature, ripe, purple-blue colored fruits with average diameter of 14 mm; #8= mature, purple-red colored fruits, with average diameter of 14 mm; #7= red colored fruits, with diameter of 13 mm; #6= pink fruits with diameter of 12 mm; #5= pink and white fruits with diameter of 11 mm; #4 to #1= green colored fruit, following in order of descending size (diameters of 11, 10, 9, and 7 mm, respectively; Fig. 5.1). To ensure consistent sorting of fruits into ripening categories, fruit surface color was quantified through the specimen port of a HunterLab Color/Difference Meter D25/L2 (Hunter Laboratory Associates, Inc., Fairfax, Virginia), calibrated according to factory values for the white, black, blue, and pink tiles (Lab values measured with 90° rotations of each fruit filled petri dish x three replications per treatment). The 'L' value indicates the percent lightness, ranging from complete darkness (0% 'L') to pure white light (100% 'L'). The 'a' value indicates the percent red ('a' > 0), green ('a' < 0), or gray ('a' = 0) color present. The 'b' value indicates the percent yellow ('b' > 0), blue ('b' < 0), or gray ('b' = 0) color present. The fresh weight of fruits in each ripening category per shrub was determined gravimetrically. The "ripe fruit" per shrub was comprised of the combined weights of maturity classes #8 and #9 and was expressed as a percentage of the total fruit per shrub. All quality measurements (fruit surface color, fruit firmness, fruit weight, soluble solids, and titratable acids) were determined using fruits from category #9.

Fresh fruit firmness was determined using the 50 kg Kramer shear of the Instron Universal Testing System, model 4201 (Instron Corp., Canton, Mass.) (13 fruits per replication x three replications per treatment).

Soluble solids concentration (SSC), expressed as ⁰ Brix (or percent sucrose equivalents), was determined using the refractometer method (AOAC, 1995b) with the Zeiss Abbe refractometer (Carl Zeiss





Oberkochen/Wurtt, Germany) connected to a water bath (20°C). Frozen fruit (50 g) was blended with 50 mL of distilled water for 3 minutes at high speed in the Waring Blender. The slurry was strained through two layers of cheeseeloth, the liquid was centrifuged at 3500g for 5 minutes, the collected supernatant was centrifuged again at 3500g for 5 minutes, and the final supernatant was filtered through Whatman #4 filter paper. The ⁰Brix were determined from the filtrate (three replications per treatment), and corrected for the initial dilution.

Titratable acidity (TA), expressed as percent malic acid equivalents, was determined using the glass electrode method (AOAC, 1995a) similar to the procedure of Green and Mazza (1986). Frozen fruit (30 g) was blended with 30 mL of distilled water for 3 minutes at high speed in the Waring Blender. An additional 50 mL of distilled water was added, and the slurry was boiled for thirty minutes while replacing water lost to evaporation. The boiled fruit slurry was brought to a final volume of 200 mL, strained through two layers of cheesecloth, centrifuged at 14 000g for 5 minutes, and the supernatant was filtered through Whatman #4 filter paper. Aliquots of filtrate (25 mL x 3 replications) were titrated past the end-point (pH 8.1) with 0.01 M NaOH, and the volume of 0.7 M NaOH required to titrate 100 g of fruit at the end-point was calculated. The amount of malic acid equivalents present were determined by the following equation:

$$g_{\text{malic acid}} = \frac{\text{Volume }_{\text{NaOH}}}{100 \text{ g}} * [\text{NaOH}] * 67 \text{ g/mol}$$
 (Equation 5.1)

where 67 g/mol is the equivalent weight of malic acid. Malic acid equivalents were expressed as grams of acid per 100 g of fruit.

In 1996, the control Northline and Smoky shrubs were harvested to assess fruit bearing patterns in these cultivars. For statistical analysis, the General Linear Model of SAS 6.10 program (SAS Institute Inc., Cary, NC 27513, USA) was used.

5.3 Results

The yield obtained from the control shrubs of cv.'s Northline and Smoky varied significantly among years (ANOVA, P<0.001). In cv. Northline, the total fruit per shrub was significantly less in 1994 and 1996 than in 1995 (Table 5.1, LSD, P<0.05). In cv. Smoky, the fruit yield per shrub was significantly

Table 5.1: Fruit yields from cv.'s Northline and Smoky shrubs from 1994 to 1996.

CULTIVAR	YIELD I	PER SHRUB (k	g/shrub) *
	1994	1995	1996
Northline	2.90 b	7.74 a	3.45 b
Smoky	0.324 z	10.1 x	2.09 y

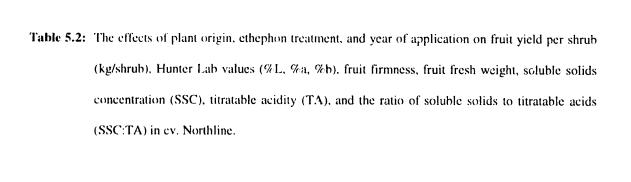
^{*} Yields obtained from control shrubs.

Mean separation within rows (ab)(xyz) by LSD, P<0.05.

different each year, with the least amount of fruit harvested in 1994, and the most fruit collected in 1995 (Table 5.1; LSD, P<0.05). Ethephon treatments did not significantly affect the yield from Northline or Smoky shrubs (Tables 5.2 and 5.3; ANOVA, P>0.10); however, an orthogonal contrast of ethephon treatments vs. the control indicated a small reduction in total fruit per shrub from ethephon-treated shrubs of ev. Northline (19.0% reduction from control; Table 5.2; P<0.01). Due to the minimal fruit yields obtained from ev. Smoky in 1994, the effect of ethephon on synchronizing ripening of ev. Smoky fruits was assessed in 1995 only.

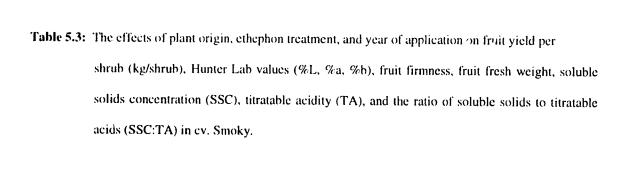
Ethephon treatment significantly affected the percent ripe fruit per shrub in both cultivars (ANOVA, P<0.05). In cv. Northline, the percent ripe fruit per shrub increased linearly with increasing ethephon concentration (Fig. 5.2; R²=0.115, P<0.05), with a maximum increase of 6% attained with 1000 ppm ethephon. The predicted effect of ethephon concentration on percent ripe fruit per shrub based on a 1000 shrub sample size is illustrated in Fig. 5.2 (Sokd and Rolf, 1981). In cv. Smoky, ethephon at 500 and 1000 ppm significantly increased the percent ripe fruit per shrub by 7% and 5%, respectively (Fig. 5.3; LSD, P<0.05).

In cv. Northline, the year of treatment significantly affected fruit color development (Hunter Lab values), soluble solids concentration (35° 2000 cellidity (TA), and the ratio of soluble solids to titratable acids (SSC:TA; Table 5.2; ANOVA 2000 cellidity (TA), and the ratio of soluble solids to titratable acids (SSC:TA; Table 5.2; ANOVA 2000 cellidity (TA), and the ratio of soluble solids to titratable acids (SSC:TA; Table 5.2; ANOVA 2000 cellidity (TA), and the ratio of soluble solids to titratable acids (SSC:TA; Table 5.2; ANOVA, P<0.001), indicating that fruits harvested in 1995 were lighter in color and contained less intense red and yellow pigmentation than fruits from 1994. Values for SSC and SSC:TA were significantly greater while TA values were significantly less in 1994 than in 1995 (ANOVA, P<0.001). The year by origin interaction was significant for SSC, TA, and SSC:TA (Table 5.2; ANOVA, P<0.05). In 1994, the SSC and SSC:TA were greater in fruits from micropropagated derived plants than plants derived from suckers; no difference was observed in 1995 (Table 5.2; LSD, P>0.05). TA did not vary between origins in 1994; however, in 1995, TA was greater in fruit from micropropagated derived plants than plants derived from suckers (Table 5.2; LSD, P<0.05). In cv. Northline, fruit firmness and fresh weight did not differ with respect to shrub origin or year of application (Table 5.2; ANOVA, P>0.10). Ethephon treatment did not affect the Hunter Lab values, firmness, weight, SSC, TA, or SSC:TA of cv. Northline fruits (Table 5.2; ANOVA, P>0.05).



1994	TREATMENT	VIELD	Hunt	Hunter Lab Values	alues		FRUIT	SSC	T.A	SSC:TA
	(wdd)	(kg/shrub)				FIRMNESS (kg/g)	FRESH WEIGHT (g)	(⁰ Brix)	(% malic acid eq.)	
			7 %	% a	9.6 p		•		•	
Micropr spagated	0	1.70	16.7	6.33	1.33	18.4	1.18	16.8	0.361	46.6
	250	1.77	15.7	7.40	1.73	16.9	1.07	15.7	0.373	42.3
	200	1.45	16.1	6.77	1.67	17.6	1.22	16.7	0.399	42.8
	1000	1.57	15.9	7.10	1.43	16.7	1.21	16.7	0.336	48.0
	mean	1.62	19.1	06.90	1.54	17.4	1.17	15.5 a	0.367 c	44.9 a
Sucker	0	4.10	15.9	6.90	1.80	18.2	1.18	15.0	0.388	38.8
	250	1.58	16.1	7.07	1.53	18.0	1.13	15.3	0.369	41.4
	200	2.12	15.6	7.30	1.53	17.8	1.19	15.0	0.404	37.8
	1000	3.59	15.8	7.73	1.47	16.6	1.14	4	0.400	35.2
	теан	2.85	15.9	7.25	1.58	17.6	1.18	14.8 b	0.390 c	38.3 h
	1994 mean	2.23 *	16.0 *	7.08 *	1.56 *	17.5	1.17	15.7 *	6.379 *	41.6 *
1995										
Micropropagated	0	80.9	16.1	1.87	-0.667	17.8	1.06	13.2	0.510	26.1
	250	5.24	18.8	2.43	-1.23	18.7	1.14	14.3	0.597	25.0
	500	7.85	18.5	2.27	-0.967	18.0	1.07	13.8	0.547	24.4
	1000	4.83	19.5	2.17	-1.10	17.7	1.17	13.4	0.514	27.5
	теан	00.9	0.61	2.18	-0.992	18.1	1.11	13.7 c	0.540 a	25.6 c
Sucker	0	9.40	18.9	1.93	-C.767	18.2	1.20	14.0	0.503	28.2
	250	6.85	18.3	1.90	-0.767	17.8	1.15	14.6	0.521	28.0
	200	6.70	18.0	2.23	-1.03	19.0	1.18	14.5	0.493	29.6
	0001	68.9	18.4	2.13	-1:00	18.1	1.19	14.3	0.476	30.2
	mean	7.46	18.4	2.05	-0.892	18.3	1.18	14.3 €	0.498 6	29.0 c
	1995 mean	6.73 *	18.7 *	2.12 *	-0.945	18.2	1.15	14.0 *	0.518 *	27.4 *
					*					

* indicates significant difference between years, by ANOVA. P<0.001; mean separation within origin (abc) by LSD, P<0.05



1994	TREATMENT	YIELD	Hun	Hunter Lab Vaiues	aines	FRUIT	FRUIT	SSC	TA	SSC: TA
	(mdd)	(kg/shrub)				FIRMNESS	FRESH	(⁰ Brix)	(% malic	
		•	T %	% a	% P	(Kg/g)	WEIGHT (g)		acid eq.)	
Micropropagated	0	0.307	17.5	7.13	-0.467	17.9	1.21	17.5	0.193	80.3
	200	0.539	9.91	7.50	-0.900	13.8	1.11	16.8	0.245	68.4
	1000	0.126	18.6	6.87	-1.200		1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	19.3	0.248	72.4
	mean	0.324	17.6	7.17	-0.856	15.7	1.18	17.9	0.236	72.7
Sucker	0	0.341	16.9	7.20	-0.967	18.2	1.18	17.6	0.187	106.5
	200	1.14	16.3	8.23	-0.967	14.0	1.08	15.6	0.214	72.9
	1000	0.603	15.1	7.36	-1.03	14.2	1.08	17.4	0.225	T.T.
	mean	0.695	19.1	2.60	-0.989	14.8	1.11	16.8	0.208	85.7
	1994 MEAN	0.510 *	16.8 *	7.38 *	-0.922 *	15.0	1.13	17.4 *	0.218	81.1 *
1995										
Micropropagated	0	. 91	16.2	2.50	-0.200	12.9	1.09	13.3	0.275	48.7
	200	7.5	16.0	2.33	-0.200	12.7	1.03	13.9	0.274	50.8
	1000	o.52	15.9	2.47	-0.567	14.2	1.02	12.8	0.251	50.8
	mean	8.79	16.0	2.43	-0.322	13.3	1.05	13.3	0.257	50.1
Sucker	0	F. 65	14.7	2.90	-0.733	13.2	1.10	13.5	0.275	48.9
	200	र () ()	15.0	2.43	-0.600	13.4	1.13	13.6	0.246	56.9
	1000	16.2	15.0	3.33	-0.900	12.7	40.1	13.6	0.221	63.6
	mean	96.6	14.9	2.89	-0.744	13.1	1.69	13.6	0.248	56.4
	1995 MEAN	9.37 *	15.5 *	2.66 *	-0.533 *	13.2	1.07	13.5 *	0.258	53.3 *

* indicates significant difference between years, by ANOVA, P<0.001.

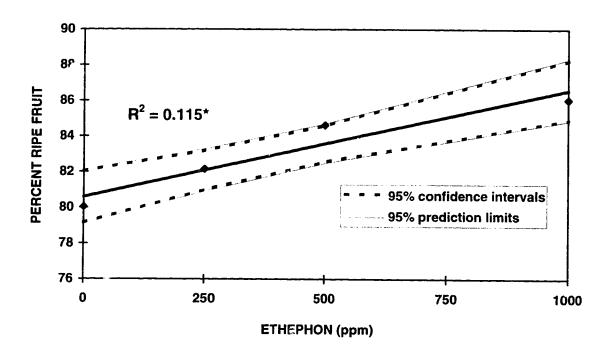


Figure 5.2: The relationship between increasing ethephon concentration and the percent ripe fruit per shrub in saskatoon ev. Northline (mean of 1994 and 1995 values); * significant at P<0.05; dashed lines indicate the upper and lower confidence intervals at P<0.05; thin solid lines indicate the upper and lower prediction limits at P<0.05 using a 1000 shrub sample size.

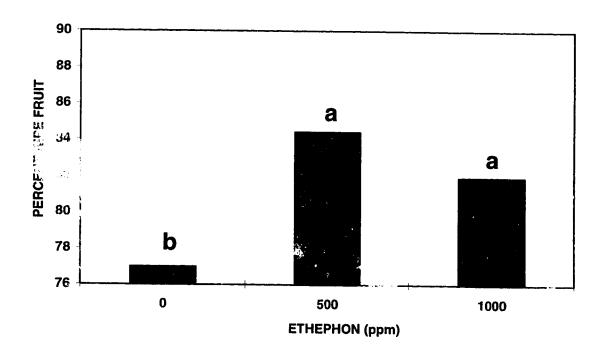


Figure 5.3: The effect of ethephon on the percent ripe fruit per shrub in saskatoon ev. Smoky, 1995; mean separation among reatments (ab) by LSD, P<0.05.

In cv. Smoky, the year of treatment significantly affected Hunter Lab values, SSC, and SSC:TA (Table 5.3; ANOVA, P<0.001). Hunter Lab values were significantly greater in 1994 than in 1995, indicating that fruits harvested in 1994 vere lighter in color yet contained more red and blue pigmentation than fruits from 1995 (ANOVA, P<0.001). Values for SSC and SSC:TA were significantly greater in 1994 than in 1995 (ANOVA, P<0.001). In cv. Smoky, fruit firmness, fresh weight, and TA did not differ with respect to shrub origin or year of application (Table 5.3; ANOVA, P>0.10, 0.10, and 0.05, respectively). Ethephon treatment did not affect the Hunter Lab values, firmness, fresh weight, SSC, TA, or SSC:TA of cv. Smoky fruits (Table 5.3; ANOVA, P>0.05).

5.4 Discussion

Uniform fruit ripening, essential for efficient mechanical harvesting, has been enhanced in many fruit crops with applications of ethephon. Similarly, saskatoon shrubs of cv.'s Northline and Smoky treated with ethephon near harvest maturity yielded greater proportions of ripe fruit per shrub with a single harvest. In general, me increases in ripe fruit were small (6-7%) but significant for both cultivars tested. The success of ethephon in stimulating fruit ripening in field applications can be affected by numerous factors other than the cultivar (Bal et al, 1992; Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987), including the stage of crop maturity (Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987; Winston et al, 1992), the concentration of ethephon (Bal et al, 1992; Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987), the number of ethephon applications (Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987), and the orchard temperature during (Bal et al, 1992) and after ethephon treatment (Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987; Conrad and Sundstrom, 1987; Olien and Bukovac, 1978). Based on these results, applying ethephon to saskatoon shrubs when approximately 70% of fruits are red (maturity class #7) can significantly increase the ripe fruit per shrub. Further experimentation will be required to test the effects of ethephon applied at earlier stages of saskatoon fruit development.

At elevated concentrations, the effectiveness of ethephon as a fruit ripening agent diminishes, and ethephon instead stimulates excessive fruit abscission (Batal and Granberry, 1982; Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987; Cooksey et al., 1994; Knavel and Kemp, 1973), defoliation (Batal and

Granberry, 1982; Cantliffe and Goodwin, 1975; Conrad and Sundstrom, 1987; Cooksey et al, 1994; Martin et al, 1980) and accentuated alternate bearing (Wood, 1989). Such detrimental effects have been observed in pimiento and paprika peppers treated with 1500 to 3000 ppm ethephon (Batal and Granberry, 1982), in Tabasco peppers with 5000 to 15,000 ppm ethephon (Conrad and Sundstrom, 1987), in office with 2250 ppm (Hartmann et al, 1970), and in pecan with 2000 ppm (Martin et al, 1980). In saskatoon ev. Northline, slight reductions in total fruit per shrub were observed among ethephon treated shrubs (Table 5.2; orthogonal contrast, P<0.01), suggesting that ethephon may have stimulated the abscission of fruits (Conrad and Sundstrom, 1987). This trend was not detected in ev. Smoky, suggesting that the saskatoon cultivars tested may differ in sensitivity to ethephon at the time of treatment.

With respect to saskatoon fruit quality, ethephon treatments did not affect the fruit surface color, firmness, fresh weight, SSC, TA, or SSC:TA values in ev.'s Northline or Smoky. This suggests that ethephon applications at concentrations up to 1000 ppm enhance saskatoon fruit ripening without reducing fruit quality. Fruit flavor is attributed to the ratio of soluble solids to titratable acids (Young et al., 1993). The SSC, TA, and SSC:TA values differed between 1994 and 1995, likely reflecting seasonal variations. However, the mean SSC, TA, and SSC:TA values between years (ev. Northline: 14.8° Brix, 0.45% malic acid, 34.7, respectively; ev. Smoky: 15.4° Brix, 0.24% malic acid, 64.3, respectively) were very similar to those values reported by Green and Mazza (1986) (ev. Northline: 16.1° Brix, 0.45% malic acid, 35.5, respectively; ev. Smoky: 16.3° Brix, 0.25% malic acid, 66.2, respectively). In general, these data show that fruits from ev.'s Northline and Smoky contained similar quantities of sugars (SSC). The Eigher SSC:TA from ev. Smoky fruits in 1994 and 1995 was attributed to the reduced amount of acids (TA) present in contrast to ev. Northline fruits.

The total fruit per shrub in cv.'s Northline and Smoky varied significantly among years. In both cultivars, yields alternated in a typical biennial bearing pattern. Analysis of fruit production in subsequent seasons will be required to establish the extent of biennial bearing. Further studies will be necessary to characterize the degree to which alternate bearing may be expressed in other saskatoon cultivars.

Ethephon treatment promoted uniform ripening in saskatoon cv.'s Northline and Smoky. The increase in percent ripe fruit per shrub obtained from each cultivar were small but significant. Ethephon treatment did not adversely affect saskatoon fruit quality, as determined by surface color, fruit firmness,

fresh weight, soluble solids concentration, titratable acidity, or the ratio of soluble solids to titratable acids.

Therefore, ethephon could be a potentially effective ripening agent for saskatoon fruits.

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6. SUMMARY AND CONCLUSIONS

The objective of this research was to examine the effects of plant hormones on saskatoon fruit development. Specifically:

- 1) to obaracterize fruit growth among different sized saskatoon cultivars, and establish the roles of seeds, and mesocarp cell number and size, upon final fruit size among saskatoon fruit cultivars.
- 2) to test the effects of gibberellins on fruit set and seed number in saskatoon.
- 3) to test the effects of ethephon on saskatoon fruit ripening.

The results of this research have established that seasonal fruit growth patterns were similar among the saskatoon cultivars studied (Fig. 2.1a-f). Fruits from cv. Thiessen were the largest throughout the season and at harvest maturity, followed by cv.'s Northline Smoky, and Regent, in descending order (Fig. 2.3). In general, saskatoon fruit development was characted used by a relatively steady rate of fruit growth with a single period of exponential growth occurring later in development. Final fruit size among saskatoon cultivars was significantly affected by seed development. Seed number distinguished cv. Thiessen (4.57 seeds per fruit), the cultivar producing the largest fruits, from the remaining cultivars (3.34 seeds per fruit). The cultivar producing the penultimate sized fruits, cv. Northline, had the largest fresh weight per seed compared to the other cultivars studied. Seed fresh weight per fruit differentiated the larger from smaller cultivars (cv.'s Thiessen and Northline vs. cv.'s Smoky and Regent). These results indicate that fruit size in saskatoon is related to seed number per fruit and the total seed fresh weight per fruit, and seed number is more critical than seed size in discerning the largest fruit size in the saskatoon cultivars studied.

The patterns of mesocarp cell number and size within cv.'s Thiessen, Northline, Smoky, and Regent were used to further characterize saskatoon fruit growth. Mesocarp cell division and clongation occurred most rapidly during Stage I of fruit growth, and cell expansion continued at reduced rates during Stages II and III of saskatoon fruit growth. Mesocarp cell number varied significantly among cultivars, with cv. Regent fruits containing significantly fewer mesocarp cells than the remaining cultivars (Fig. 3.1). Mesocarp cell size did not vary among cultivars (Fig. 3.2). This suggests that the number of mesocarp cells

per fruit is more critical than cell size in determining final fruit size among the saskatoon cultivars examined.

GAs did not consistently affect saskatoon fruit development. Dennis (1973) has advised that the potential of exogenous GAs to improve fruit set and reduce seed number requires selecting the most appropriate GA, optimizing the time and method of application, employing synergists or antagonists with GAs, or repeatedly applying GAs at reduced concentrations. This research has shown that GA₃, GA₄₇, and GA₄₇₇+BA did not consistently affect fruit set or seed number when applied to saskatoon ev.'s Smoky (Table 4.1 and 4.3), Northline (Table 4.2), and Pembina (Table 4.5). The time at which CAs were applied to saskatoon shrubs significantly affected fruit set and seed number; however, these results varied among cultivars and between years of study. For example, early applications of GA₄₇₇ and GA₄₇₇+BA significantly improved fruit set in cv. Smoky in 1994 (Table 4.2) yet did not affect fruit set in 1995 or in cv. Northline. Fruit set in cv.'s Smoky and Northline in 1995 was reduced with early bloom applications (Tables 4.1 and 4.3), suggesting that the surfactant Tween 20 may have reduced saskatoon fruit development when applied at this time. Optimizing the method of GA application to saskatoon plants may require further experimentation. A preliminary study conducted in 1996 tested the effects of non-foliar GA applications upon seed development in greenhouse grown cv. Pembina fruits. Inflorescences at full bloom were infused with GA₃ (10⁵ M) through the peduncle. GA treated fruits did not contain fewer seeds per fruit nor less fresh or dry weight per seed (data not shown), suggesting that delivery of the hormone directly into the vasculature did not necessarily improve GA action. In some fruits, the effectiveness of GAs at reducing seed number can be enhanced when synergists, such as cytokinins or auxins, are employed (Hopping, 1986). In cv. Northline in 1994, GA_{4/7}+BA significantly reduced seed number compared to GA_{4/7}; however, this difference was small (orthogonal contrasts, P<0.01). It would be interesting to test the effects of auxins with GAs on saskatoon seed development using similar foliar applications. In some fruits, repeated GA applications, alone or with synergists/antagonists, can more effectively reduce seed number though often adversely affecting fruit size (Hopping, 1986). A preliminary field experiment conducted in 1995 tested the effects of GA3, GA47, and GA471+BA (100 ppm) applied at petal fall and repeated one week later to saskatoon ev. Pembina shrubs. The repeated application did not significantly affect fruit set, fruit size, seed number per fruit, or seed size (data not shown; ANOVA, P>0.10). It would be interesting to test the effects

of increased GA concentrations on saskatoon fruit set and seed development. Further research will be required to determine the GA concentration, and type and concentration of surfactant that can effectively reduce the seed number of saskatoon fruits in field conditions without negatively affecting fruit set or the return bloom.

Ethephon promoted uniform ripening in saskatoon ev.'s Northline (Fig. 5.2) and Smoky (Fig. 5.3). Ethephon treatments significantly increased the ripe fruit per shrub from a single harvest without compromising fruit quality, as determined according to fruit surface color, fruit weight, firmness, soluble solids concentration, titratable acidity, and the ratio of soluble solids to titratable acids (Table 5.2 and 5.3). Although the increases in the percentages of ripe fruit harvested were small (ev. Northline: 2, 4 and 6%; ev. Smoky: 7 and 5%), they were statistically significant. These small increases could be quite promising for orchardists providing that this treatment is cost-effective. However, applying ethephon at the appropriate concentration and stage of fruit development is critical since elevated concentrations of ethephon can promote fruit abscission and defoliation, and immature fruits may not be sensitive to ethephon, thereby not enhancing fruit ripening. These experiments testing ethephon at concentrations up to 1000 ppm applied to mature, unripe fruits of ev.'s Northline and Smoky have been effective.

In conclusion, saskatoon fruit development can be described by a three phase growth curve, with Stage I characterized by and elongation within the mesocarp, Stage II characterized by seed development, a d by continued mesocarp cell expansion. Seed number is more critical than te edetermining final fruit size; similarly, mesocarp cell number is more critic. nai fruit size among the saskatoon cultivars studied. GA treatments did owever, fruit set in cv. Smoky in 1994 was significantly improved with e of GA_{4/7} and GA_{4/7}+BA. In general, GA treatments reduced the seed content in fruits of cv.'s Smoky, Northline, and Pembina, thereby improving fruit quality for processing. Ethephon treatments enhanced uniform ripening to significantly increase the yield of salable saskatoon fruits obtained with a single harvest. The results from this research suggest that plant hormones can improve saskatoon fruit quality and yield although further research will be needed to optimize the effectiveness of GAs on saskatoon fruit set and seed number, and of ethephon on saskatoon fruit ripening.

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