This is to certify that the

dissertation entitled

EFFECTS OF FRUIT, SEED AND SHOOT DEVELOPMENT
ON FLOWER INDUCTION IN APPLE

presented by

JOHN CALVIN NEILSEN

has been accepted towards fulfillment
of the requirements for

Ph.D. degree in Horticulture

Major professor

Date May 8, 1998

EFFECTS OF FRUIT, SEED AND SHOOT DEVELOPMENT ON FLOWER INDUCTION IN APPLE

BY

JOHN CALVIN NEILSEN

DISSERTATION

Submitted to
Michigan State University
in partial fulfillment of the requirements
for the degree of

DOCTOR OF PHILOSOPHY

Department of Horticulture

1998

ABSTRACT

EFFECTS OF FRUIT, SEED AND SHOOT DEVELOPMENT ON FLOWER INDUCTION IN APPLE

By

John Calvin Neilsen

Studies were conducted to determine the effects of seeds, shoot length, crop density, and gibberellin application on flower induction in apple. Spurs of 'Spencer Seedless' were defruited at intervals during fruit development in 1988-1991. Fruit mass and seed number were recorded for each spur. Flower induction was inhibited in spurs bearing fruits containing more than five seeds, the effect increasing as time of defruiting was delayed. When fruits were left until harvest in 1994 and 1996, flowering decreased linearly with an increase in seed number per spur, but response varied with bourse shoot length. All shoots less than 2 mm long failed to flower. In spurs bearing seeded fruits, flowering increased with shoot length, and most shoots > 10 mm long flowered. Neither fruit mass per spur nor crop density affected flowering in the absence of seeds. In spurs bearing fruits containing five or more seeds, high crop density appeared to inhibit flowering, but fruit mass per spur had little effect. In 'Paulared' flowering was not inhibited if trees were entirely defruited within 60 days after full bloom, but declined rapidly as fruit removal was delayed further. In 1996, fruits were removed 24 days after full bloom, leaving either 1 or 2 fruits per bearing terminal; half of the trees were girdled to stimulate flowering. Flowering in 1997 was variable, with more flowering than expected for trees bearing moderate to heavy crops. No effect of bourse shoot length on flowering was detected, and non-bearing shoots flowered only slightly more than did bourse shoots on terminals that bore one or two fruits. In the absence of girdling, percentage flowering decreased as crop density increased. To determine if substitution of gibberellins for seeds would inhibit flowering, fruits of several seeded and seedless cultivars were cut transversely 30 days after full bloom, and the seeds removed and replaced with lanolin pastes containing gibberellins A_3 or $A_{4/7}$. Percentage flowering in the controls (fruits intact) was relatively high, and neither seed removal nor gibberellin treatment had appreciable effects, regardless of cultivar. Sprays of $GA_{4/7}$ were applied to whole trees of 'Gala' apple to determine if flowering could be inhibited in lateral buds. Treatments applied 3 July inhibited flowering more than did later applications, but the variability was high, resulting in nonsignificant effects of concentration. These experiments demonstrate that, in 'Spencer Seedless', seeds rather than fruits are responsible for inhibition of flowering and that the effects of seeds declines as bourse shoot length increases. These relationships could not be confirmed in the seeded cultivar 'Paulared' as response was too variable. Substitution of lanolin pastes containing gibberellins for seeds did not inhibit flowering, suggesting that the role that seeds play in biennial bearing interacts with other growth factors, including shoot growth and cropload.

To my parents, Calvin E. Neilsen and Sarah C. Neilsen,

champions of agriculture, who planted the seed,

and my eternal companion Kelly A. Neilsen,

whose faith in me nurtured it to fruition.

ACKNOWLEDGMENTS

I'm grateful to Dr. Frank G. Dennis Jr. for serving as my major professor and chair of my advisory committee. He persevered beyond my expectations. I also appreciate the service of Drs. Martin J. Bukovac, James A. Flore, Kenneth L. Poff and Jan A. D. Zeevaart as members of my advisory committee for their advice and encouragement.

I gratefully acknowledge the assistance of Drs. Martin J. Bukovac and Jerome Hull Jr. with experiments reported in Section 4 and Abbott Laboratories for the gift of Provide® and GA_{4+7}.

I gratefully acknowledge the assistance of Dr. Jun Ban, Mario Mandujano, Dr. John Everard, Adam J. Neilsen and many other colleagues for assistance in data collection and discussion of the results. I also acknowledge my friends and family who provided financial and moral support to make it possible for me to pursue this goal.

Finally, I express deep love and appreciation to my dear wife Kelly A. Neilsen and our children Adam, Maren, Anna, Camille, and Sarah who have endured many deprivations during the many years I've pursued education.

TABLE OF CONTENTS

LIST OF TABLES x

LIST OF FIGURES xiii

LITERATURE REVIEW 1

- Economic importance of flower formation 1
- Growth and flowering habit 2
- Time of induction 4
- Factors influencing flower induction 5
  - Genotype and juvenility 5
  - Environmental and cultural factors 5
    - Shoot length and leaf area 6
  - Leaf/fruit ratio 8
  - Seeds. 9
- Possible reasons for effects of seeds 11
  - Competition for carbohydrate 11

Competition for water 12

Competition for "florigen" 12

Seed-produced hormones 12

Summary 15

Literature Cited 15

SECTION 1. EFFECTS OF SEED NUMBER, FRUIT REMOVAL, BOURSE SHOOT LENGTH AND CROP DENSITY ON FLOWERING IN 'SPENCER SEEDLESS' APPLE. 21

Abstract 21

Introduction 22

Materials and Methods 23

Plant material 23

Pollination and fruit removal 23

Effects of fruit removal, seed number, bourse shoot length and fruit mass on flower induction 24

Effects of seed number, crop density and bourse shoot length 24

Results 25

Effects of fruit removal, seeds, bourse shoot length and fruit mass on flower induction 25

Effects of seed number, crop density and bourse shoot length 28

Discussion 37

Literature Cited 42

SECTION 2. EFFECTS OF FRUIT REMOVAL, CROP DENSITY, AND TRUNK GIRDLING ON FLOWER FORMATION IN 'PAULARED' APPLE: GENERAL VS. LOCAL EFFECTS OF FRUITS. 45

Abstract 45

Introduction 47

Materials and Methods 48

Expt. 1. Effects of time of fruit removal on flower initiation 48

Expt. 2. Effects of crop density, fruit number per spur, and girdling on flower initiation 48

Results 50

Expt. 1. Effects of time of fruit removal on flower initiation 50

Expt. 2. Effects of crop density, fruit number per spur, and girdling on flower initiation 50

Discussion 57

Literature Cited 61

SECTION 3. EFFECTS OF DEFRUITING, SEED REMOVAL AND REPLACEMENT OF SEEDS WITH GIBBERELLINS ON FLOWER FORMATION IN APPLE. 63

Abstract 63

Introduction 64

Materials and Methods 65

Expt. 1. Seeded cultivars 65

Expt. 2. Seedless cultivars 66

Results 66

Expt. 1. Seeded cultivars 66

Expt. 2. Seedless cultivars 69

Discussion 69

Literature Cited 74

SECTION 4. EFFECT OF GIBBERELLIN SPRAYS ON FLOWER FORMATION IN LATERAL BUDS OF 'GALA' APPLE. 76

Abstract 76

Introduction 77

Materials and Methods 78

Results 79

Discussion 83

Literature Cited 91

LIST OF REFERENCES 100

LIST OF TABLES

SECTION 2. 45

Table 1. Effect of time of defruiting entire 'Paulared' apple trees in 1991 and 1992 on mean shoot length (± standard error) in the same growing season and number of lateral flowers per shoot the following year. 52

Table 2. Main effects of girdling, crop density and number of fruits per spur in 1996 on flowering of 'Paulared' apple in 1997. Percentages calculated from 21 to 46 bourse shoots per tree for each category. Three replicate trees were used for each treatment in a split-plot design. 53

Table 3. Effect of medium (M) and high (H) crop density on flower induction of non-bearing shoots and spurs bearing 1 or 2 fruits on individual, non-girdled 'Paulared' apple trees. 57

SECTION 3. 63

Table 1. Harvest data for five 'Golden Delicious' apple trees used in Expt. 1. Clarksville Horticulture Experiment Station, 1996 - 1997. 67

Table 2. Effects of defruiting, fruit cutting, seed removal, and replacement of seeds

with lanolin pastes on flowering of 'Golden Delicious' apple. Clarksville Horticulture Experiment Station, 1996 - 1997. Treatments were applied to 5 trees on 26 June 1996. 67

Table 3. Effects of defruiting, fruit cutting, seed removal, and replacement of seeds with lanolin pastes on flowering of 'Jonagold' apple. Horticulture Teaching and Research Center, 1996 - 1997. Treatments were applied to 6 trees on 29 June 1996. 68

Table 4. Effects of defruiting and fruit cutting on flowering of 'Delicious' apple. Clarksville Horticulture Experiment Station, 1996 - 1997. Treatments were applied to 7 trees on 30 June 1996. 69

Table 5. Effects of seed number and of applications of lanolin pastes to locules of cut fruits of 'Spencer Seedless' at the Horticulture Teaching and Research Center, 1996 - 1997. Treatments were applied to fruits on one tree on 29 June 1996. 70

Table 6. Effects of defruiting and applications of lanolin pastes to locules of cut fruits of 'Ohio No. 3' apple at the Horticulture Teaching and Research Center, 1996 - 1997. Treatments were applied to fruits on 2 trees on 25 June 1996. 71

SECTION 4. 76

Table 1. Effect of $GA_{4/7}$ sprays applied 3 July 1996 to trees with varying crop density on bloom rating and percentage of lateral buds flowering in 1997 in 'Gala' apple at Clarksville Horticulture Experiment Station, Clarksville, Mich. All values are

means for 5 trees. 80

Table 2. Effect of timing and concentration of ProVide® treatments in 1996 on crop density and bloom rating in 1997 in 'Gala' apple at Clarksville Horticulture Experiment Station, Clarksville, Michigan. Five trees per treatment. 81

Table 3. Effect of timing and concentration of ProVide® treatments in 1996 on flowering of terminal and of lateral buds in 1997 in 'Gala' apple at Clarksville Horticulture Experiment Station, Clarksville, Michigan. Analysis of variance based on transformed (arcsin square-root) data. 82

LIST OF FIGURES

SECTION 1. 21

Figure 1. Effects of seeds and time of fruit removal the previous year on flowering of terminal buds of 'Spencer Seedless' apple bourse shoots. Each point represents from 6 to 45 bourse shoots. Year indicates time of fruit removal. 26

Figure 2. Effects of shoot length and seed number the previous year on flowering of terminal buds of bourse shoots of 'Spencer Seedless' apple. All fruits were harvested at maturity. Each point represents one bourse shoot. 27

Figure 3. Relationship between total seed number and total weight of fruit per spur at harvest in 1989, 1990, 1991, and 1992 for 'Spencer Seedless' apple spurs that were flowering or vegetative the following season. All fruits were harvested at maturity. Only fruits from hand-pollinated branches were weighed in 1990. Each point represents one bourse shoot. 29

Figure 4. The relationship between seed number per spur and percentage of 'Spencer Seedless' apple bourse buds that flowered the following growing season in 1994 ($y = 96.1 - 15.2x$, $R^2 = 0.97$) and 1996 ($y = 88.2 - 19.6x$, $R^2 = 0.96$). Seed number classes are based on observation of apparent break points and sample

size. 30

Figure 5. The relationship between seed number and shoot length of bourse shoots of 'Spencer Seedless' apple in 1994 and 1996 vs. percentage of bourse buds that flowered the following growing season. Seed number and shoot length classes were based on observation of apparent break points and sample size. 31

Figure 6. Distribution of bearing spurs of 'Spencer Seedless' apple by total fruit mass borne per spur in 1994 and 1996. No spurs bore less than 40 g. of fruit. 40 g = 40 to 59 g; 60 g = 60 to 79 g, etc. 33

Figure 7. Effect of total 'Spencer Seedless' apple fruit mass (g) borne per spur on percentage of spurs that flowered the following year, in 1994 and 1996. Spurs bearing seedless and seeded fruits were ranked by total fruit mass (20 g increments). Classes were combined when necessary to give sufficient numbers to determine *P* = 0.95 confidence limits. Number of spurs per class varied from 49 to 337, as required to calculate confidence limits for binomial data. Classes represent spurs with fruit mass from the mass given to the next class above for a given treatment, except for the lowest class, which represents all fruits less than the value given. Insufficient spurs bore seedless fruit in 1996 to classify, so mean fruit mass of all seedless spurs is reported. 34

Figure 8. The relationship between seed number per spur in 'Spencer Seedless' apple and percentage of bourse buds that flowered the following growing season on spurs bearing less than or greater than the median fruit mass. Seed number classes are based on observation of apparent break points and sample size. .. 35

Figure 9. The relationship between seed number and crop density per cm^2 branch cross-sectional area in 'Spencer Seedless' apple in 1994 vs. percentage of bourse buds that flowered the following growing season. Seed number and crop density classes are based on observation of apparent break points and sample size. . . 36

SECTION 2. 45

Figure 1. Effect of time of defruiting entire ‘Paulared’ apple trees in 1991 and 1992 on percentage of terminal buds of fruiting spurs flowering the following year. Shoots with dead terminal buds were excluded. Bars represent the standard errors of the means for three replicate trees. 51

Figure 2. Effects of crop density and number of fruits borne on a spur on the average number of lateral flowers per bourse shoot. 54

Figure 3. Effect of crop density on percentage of terminals flowering (90 shoots per tree) on girdled ($y = 98.9 - 1.5x$, $R^2 = 0.093$, $P = 0.34$) and nongirdled ($y = 105.7 - 7.0x$, $R^2 = 0.56$, $P = 0.000056$)‘Paulared’ trees. 56

SECTION 4. 76

Figure 1. Crop density on ‘Gala’ apple trees used (A) and effects of concentration and time of application of gibberellin in the form of ProVide® in 1996 on bloom rating in 1997 (B). Each point is the mean for five replicate trees. 84

Figure 2. Effects of concentration and time of application of gibberellin in the form of ProVide® on percentage terminal (A) and lateral (B) buds flowering the

following year on ‘Gala’ apple. Each point is the mean for five replicate trees. 85

Figure 3. Relationship ($y = 8.32 - 0.38x$, $R^2 = 0.38$, $P < 0.0001$) between return bloom rating in 1997 and crop density in 1996 in ‘Gala’ apple over three treatment dates and three concentrations of ProVide® at Clarksville Horticulture Experiment Station. 86

Figure 4. Relationship between crop density in 1996 in ‘Gala’ apple and percentage of lateral nodes flowering in 1997 over three treatment dates for ProVide® concentrations from 150 to 600 $mg \cdot L^{-1}$ at Clarksville Horticulture Experiment Station. 87

LITERATURE REVIEW

Economic importance of flower formation

Many fruit and nut trees exhibit cyclic yield variation known as alternate or biennial bearing. This is characterized by a growing season where an abundance of fruits are produced, followed by a growing season where few if any flowers and fruits are produced. This alternate habit can occur among branches of a single tree, among different trees within an orchard, or be synchronized by adverse weather conditions, such as spring freezes, to cause entire orchards and regions to fluctuate widely from year to year. In the "on" years with an excessive crop, fruits are small and may not mature with the quality consumers desire. In the "off" year, what few fruit that are produced are often too large and lack the internal characteristics that allow them to store and maintain good quality through the marketing channels. Thus, alternate bearing poses considerable financial risk for apple growers and creates unstable supply and prices for consumers (Singh, 1948). After decades of work to understand and overcome this problem, irregular cropping is still a major impediment to apple production. While considerable progress has been made through the use of precocious rootstocks and by partial defruiting with chemical thinning agents, understanding of the basic cause(s) of alternate bearing is still limited. Better understanding of the physiology of flowering and fruiting could lead to novel ways to overcome this persistent problem.

Despite the relative complexity of woody plant genomes, Weigel and Nilsson (1995) have successfully transferred the *LFY* gene responsible for precocious flowering from *Arabidopsis* to the woody plant *Populus*, resulting in solitary axillary flowers and an abnormal terminal flower on five-month-old shoots grown in tissue culture. Tobutt (1994) has determined that the apetelous parthenocarpy trait in apple is controlled by a single recessive gene and has combined this with columnar growth habit. The gene for apetelous parthenocarpy may also influence flowering in apple, as seedless apples do not inhibit flowering (Chan and Cain, 1967). Detailed understanding of a plant and its physiology is required to apply the tools of molecular biology in solving production problems. This certainly applies to apple.

Growth and flowering habit

Apple (*Malus domestica* Borkh.) is a woody perennial plant adapted to growth in temperate climates. Thus, in addition to buds formed in the axils of leaves, terminal buds also form as episodic shoot growth ceases, beginning a dormant phase. The shoots grow to various lengths before setting terminal buds. A lateral bud that produces a shoot with less than about six leaves and elongates only 1 to 20 mm is called a spur. These frequently form flower buds and constitute the best form of growth for fruit production. Their growth ceases early in the season, and terminal buds usually begin forming a few days following full bloom. Some lateral and terminal buds produce shoots that have greater than six leaves and lengths from several centimeters to a meter or more. Their growth period can last from just a few days longer than that of spurs to the entire growing season. Thus, terminal buds can form at any time, from a few days after growth starts in spring to late autumn.

The first visible evidence that terminal growth is ceasing is the abscission of the lamina and petiole of the newest emerging leaf; the leaf base forms the first bud scale

(Abbott, 1970). Once terminal buds begin to form, shoot elongation ceases until dormancy is broken, usually by chilling temperatures in temperate climates, but occasionally by summer precipitation, which causes a second growth flush. In spring, buds begin to grow and produce another flush of growth. Much of the new tissue is already differentiated within the bud and the new growth is simply the result of cell expansion, thus the content of these buds is determined during the previous growing season. Apple produces both vegetative buds (containing only shoot primordia) and reproductive buds (containing both an inflorescence and shoot primordia) known as flower, floral or fruit buds.

All terminal and lateral buds have the potential to flower (Zeller, 1960), but the probability of flower buds developing varies greatly with cultivar and growing conditions. This variation leads to cultivars being classified as "spur bearing", "terminal bearing", and "lateral fruiting". These types of fruiting are not exclusive and occur simultaneously to varying degrees in most cultivars. Apple flower buds are mixed buds that produce a terminal inflorescence in the form of a cyme with shoots arising from the axils of leaves attached to the swollen peduncle (bourse). These bourse shoots become the new branch or spur terminals, eventually with their own terminal buds. Usually only one bourse shoot forms, but weak spurs may have none, and two bourse shoots are common on vigorous spurs. Since terminal growth generally ceases after full bloom, and flower induction occurs some time near cessation of terminal growth, flower induction and differentiation are concurrent with fruit development. Although induction can occur after harvest, few if any terminals are still growing at that time. Lateral flowers form from August through March, after those in the terminal buds of spurs, and seldom in a uniform manner (Zeller, 1960). Thus growth conditions during the season influence reproductive growth and development in both the developing fruit and the buds being formed for the next growing season.

Time of induction

Some time after terminal growth ceases, a physiological "switch" determines whether the bud will be vegetative or generative (flower induction). There is no visible evidence that induction has occurred, but cytochemical and histological differences between induced and non-induced buds have been observed prior to morphological changes. The level of DNA rises in both the apical zone and in the central meristem. This is followed by increased mitotic activity throughout the apex, and the central meristem can be seen immediately surrounded by the subdermatogen (Buban, 1981). Existing leaf primordia are also modified and develop into eight more scales. These form an enclosure as the meristem continues to initiate primordia of transition leaves, true leaves, and, if flower induction has occurred, bracts, a terminal (king) flower, and lateral flowers in the axils of the bracts and most apical true leaves. The first microscopic evidence that flower induction has occurred (flower initiation) is a flattening of the terminal dome of the apical meristem (Goff, 1899, as cited by Davis, 1957) 17-21 nodes (and several weeks) after the first bud scale (McLaughlin and Greene, 1991). A bourse shoot(s) is differentiated at a node proximal to those with flower primordia. This microscopic differentiation takes place over the growing season.

Understanding the biological switch that determines whether a given bud is vegetative or floral is an important key to understanding and controlling alternate bearing. Once the switch has been thrown, the fate of that bud for the following growing season is fixed. However, the vitality of these buds will largely depend on growing conditions during differentiation and maturation – after induction takes place.

Factors influencing flower induction

Genotype and juvenility. Juvenility is the first impediment to flower and fruit production in apple; however, in commercial apple production this problem is overcome by the use of mature clones. This imparts uniformity in growth habit of the trees and characteristics of the fruit, and all cuttings and bud sticks used for propagation are obtained from stock already in the adult phase; thus the new shoots are adult tissue and capable of flower bud formation. The scope of this study will be confined to the control of flowering of adult tissue. Genetic variation within the scion and rootstock, which are usually different genotypes, includes variation in precocity. A primary criterion for selecting a suitable rootstock is the influence it has on the scion in hastening and enhancing flower production. Although Hirst and Ferree (1995a, 1995b and 1996) reported that rootstocks affect vegetative growth and precocity, but not time or pattern of flower bud development, Nesterov et al. (1972, as cited by Buban, 1996) reported that dwarfing rootstocks caused induction to occur earlier in the season.

Environmental and cultural factors. Many environmental and cultural factors influence flower formation, including shading (Auchter et al., 1926; Gourley, 1920), temperature (Tromp, 1992, 1993), water supply (Sritharan and Lenz, 1988), nutrition

(Denker and Hansen, 1994; Hipps, 1992) and growth regulators, including naphthaleneacetic acid (Harley et al., 1958), benzyladenine (Wertheim and Estabrooks, 1994), carbaryl (McArtney et al., 1995), growth retardants (Ramirez and Hoad, 1981; Tromp, 1987), and gibberellins (Bangerth and Schroder, 1994; Greene, 1993; Looney et al., 1992).

Foliar application of GA inhibits flowering in apple, depending on time of application, concentration and the gibberellin used (Guttridge, 1962; Marcelle and Sironval, 1963). McArtney (1994) reduced the severity of the alternate bearing cycle in 'Braeburn' apple by applying GA_3 and GA_{4+7} at full bloom. Although there was a linear response to concentration, even the highest rate of GA_3 (330 ppm) did not entirely eliminate alternate bearing.

Environmental effects and chemical treatments often influence the current crop, making it difficult to determine whether effects on flowering are direct or indirect. Some investigators have proposed a direct effect of chemical thinning agents in stimulating flowering (Harley et al., 1958), but most consider the effect of thinning to be indirect via fruit removal. Branch orientation also affects flowering (Abbott,1960; Edwards, 1987). Emphasis in this thesis will be confined to the interactions within a tree between shoot, fruit and seed development in affecting flower induction.

Shoot length and leaf area. Flower induction is associated with the cessation of shoot growth and is dependent on adequate leaf area (Davis, 1957). In temperate regions, bloom is synchronized so that most flowers open within 2 - 5 days. These dates vary from year to year, and subsequent fruit development varies in a parallel manner. Thus full bloom is a convenient developmental event used to relate subsequent events in fruit development, usually expressed in days after full bloom (DAFB). How cessation of shoot growth or flower induction within buds is related temporally to flowering and fruit development is not known, but DAFB is the common reference used for lack of a better alternative.

Cessation of bourse shoot growth may be rather soon after full bloom in the "on" year of an alternate bearing tree or relatively later in an annual bearing one (Davis, 1957). Variations in shoot length affect other characteristics; longer shoots can have higher leaf numbers, more leaf area, and a later date of cessation of growth. Manual reduction of spur leaf area reduces flowering of "off" trees the following year depending on the time of defoliation (Harley et al., 1958; Li et al., 1995). Struckmeyer and Roberts (1942) found that removal of the two terminal leaves of 'Wealthy' spurs had an inhibitory effect on flower induction similar to that of removing all leaves except the two terminal ones, but less effect than if all leaves were removed. Caustic compounds used for fruit thinning reduced flowering when injury to foliage was pronounced. Clearly the presence of leaves on spurs is critical for flower induction, but what role they play is not well defined.

Roberts (1920), studying spurs (terminals with not more than 65 mm growth) in "off"-year trees, found that vegetative spurs were either long or short, averaging 2.6 mm vs. 18.1 mm of growth the previous year, respectively. Bourse shoots that flowered were intermediate in length; those that set fruit averaged 12.1 mm vs. 4.3 mm for those that did not. While length varied with cultivar and location, the relationship between length and flowering/fruiting was confirmed by several other investigators. Auchter and Schrader (1923) classified 1321 spurs (shoots 76 mm or less) of 'York Imperial' into five classes, and found that most of those that grew only 1 - 3 mm were continuously vegetative, whereas most of those that grew more than 5 mm were floral the following year, with percentage flowering increasing with shoot length. This was only true of trees with an annual bearing habit. When a heavy crop threw a tree into an "off" year, no spurs formed fruit buds, regardless of their growth the previous year. The next year nearly all spurs blossomed, including those that previously had been continuously vegetative. Growth of such spurs was stimulated in the "off" year, and the few that remained vegetative in the subsequent "on" year were the shortest ones, with few exceptions. "Off" year trees had average spur growth greater than "on" year trees (10.6 mm vs. 6.3 mm, respectively), but general vigor was good in these trees in all years. Thus flowering is correlated with shoot growth, but fruiting can affect flowering independently of shoot growth.

Leaf/fruit ratio. The dominant effect of fruiting on flowering is well established. The removal of the fruit from a tree or large branch allows flower buds to form in alternate-bearing cultivars, whereas on comparable fruiting units few or none will differentiate (Davis, 1957). Two alternative hypotheses have been proposed to explain this dominant effect: (a) overall crop acts collectively on all developing buds (crop effect) (Palmer, 1992) vs. (b) the fruits have a local effect only on bourse shoots arising from their own cluster bases (local effect) (Palmer et al., 1991). Auchter (1919), Roberts (1920) and Crow (1920) reported that spurs that bore fruit until 'June' drop rarely produced flower buds. Auchter and Schrader (1923) also reported that 50 percent of the non-blossoming spurs flowered the following year in a 'Stayman Winesap' tree bearing a light to medium crop. Response of flowering spurs declined as defloration/defruiting was delayed. Blossom/fruit removal at pink, full bloom, 'June' drop, and harvest resulted in 37, 32, 5, and 1 percent bloom the following year, respectively. Blossom removal increased secondary growth of spurs, and flowering was correlated with this growth. In reviewing studies on leaf:fruit ratio, Davis (1957) concluded that two to three times as many leaves were required to induce flower induction on non-ringed branches compared with ringed branches. Apparently some promoter(s) moved out of the non-ringed branch and more leaves were therefore required to permit induction.

Seeds. In his study characterizing flower initiation, Goff (1899, as quoted by Davis, 1957) stated "...We can now easily understand why a bountiful fruit crop so often causes reduced fruitage the following year. The nourishment that might otherwise contribute to the formation of flowers is absorbed by the numerous seeds that are maturing." Likewise, Heinicke (1917) noted, "It is conceivable that a tree bearing a heavy crop of many-seeded fruits is being devitalized to a far greater extent than another tree of the same variety bearing a crop of fruits equally heavy but having relatively few seeds." In listing reasons for early thinning (about the time of 'June' drop), Gourley (1922) stated, "The development of seeds and "pits" drains the energies of the tree." Gourley's inference was based on the data of Bigelow and Gore (1905, as cited by Gourley, 1922) for peach, which indicated that a large percentage of total solids occurred in the stone and kernel at maturity and that most pit development occurred early in fruit development, before pit hardening.

'Spencer Seedless' is a homeotic floral mutant apple cultivar that is homozygous recessive *ape ape* (Acquaah et al. 1992; Coen, 1991; Tobutt, 1994). This gene causes a second whorl of sepals to be substituted for the petals (Brase, 1937), and an additional 10 carpels are substituted in place of the stamens, making the flowers pistillate and capable of producing over 30 seeds per fruit. Although the apetalous flowers are not attractive to bees and thus not normally insect-pollinated, they set seedless fruits readily in most years, and form numerous flower clusters annually both on spurs and shoots. By hand pollinating the apetalous flowers, seeded and seedless fruits can be produced on the same tree. The fruits are of no commercial value.

Using a 'Spencer Seedless' apple tree at Geneva, New York, Chan and Cain (1967) discovered that when fruits were left until maturity, seeded fruits inhibited flower formation but seedless fruits did not. Further, by removing fruits at intervals during development, they were able to determine the time when developing seeds became active in preventing flower induction. When similar experiments were performed in a greenhouse, seeds failed to inhibit flowering to the same degree and this failure was associated with bourse shoots longer than 2 cm. Huet, (1972, 1973) confirmed the effects of seeds on flowering of 'Bartlett' pear, and showed that seeds were more inhibitory at low than at high leaf area. This supports the conclusion that a substance other than carbohydrate is responsible for the fruit's effect on flower induction. The data of Griggs et al. (1970) in California were more ambiguous; over 50 percent of the bourse shoots on spurs bearing seeded fruit still flowered in 'Bartlett' pear, and most of those were shorter than 25 mm.

Possible reasons for effects of seeds

Flower induction is controlled by genes that are activated under certain conditions. The unknown factors are the precise conditions required and the nature of the signal. From the above review, the time period during which a developing bud can be induced to become floral is fixed, and the signal must be sent at the appropriate time.

In apple, leaves promote and seeds inhibit flowering. Several hypotheses have been suggested to explain these facts. Both Goff (1899, as cited by Davis, 1957) and Heinicke (1917) suggested that seeds require "nutrients" over and above those needed by the fruit and therefore "drain the tree of nutrients". Heinicke (1917) demonstrated that seeds, "...supplement the forces that bring sap to the fruit." The compounds attracted by seeds could be carbohydrates and minerals, on the one hand, or hormones on the other. Their movement must be explainable in terms of the known transport mechanisms in plants (xylem and phloem).

Competition for carbohydrate. One alternative hypothesis is that fruits are superior competitive sinks for carbohydrate, thus buds are "starved" into being vegetative. Flowers and fruits accumulate dry matter slowly during the cell-division phase of development, but use large amounts of carbohydrate in respiration. This occurs when shoot growth is also rapid and leaf area is limited, creating strong competition for stored carbohydrates. Seeds enhance the sink effect of fruits for carbohydrate, thus reducing the flow of carbohydrates to the developing buds to a level that causes them to remain vegetative. This hypothesis assumes that floral-gene activation is mediated by carbohydrate supply or concentration. Little evidence exists for this mechanism.

Competition for water. Since developing fruits, leaves and buds all transpire, the competition could be driven by water relations rather than carbohydrate. Fruitlets can lose water and shrink during daily periods of high water demand, followed by recovery at night when transpiration demands are reduced. This suggests that it is unlikely that fruits have a lower water potential than the developing bud.

A major discrepancy that needs resolution with the "competing sinks" hypothesis is that both seeded and seedless apple fruits are sinks for carbohydrate and water, but the former inhibit flowering while the latter do not. The need for carbohydrate may be a quantitative one, since there is a correlation between seed number and individual fruit size. This correlation is mediated by spur size, flower size, flower position within the inflorescence, fruits borne on a spur and cropload (Heinicke, 1917).

Competition for "florigen". The positive effect of leaves on flowering of herbaceous plants led to coinage of the term "florigen" by Chailykyan (1937); however, this compound(s) has never been identified. Seedless fruits use carbohydrates and water, but conceivably do not attract "florigen" while seeded fruits do. Thus buds adjacent to seedless fruits can flower. This would require a mechanism in which developing seeds control loading or unloading of "florigen" from sap, thus altering the transport of "florigen" to the developing bud(s).

Seed-produced hormones. Seeds could have a direct effect on flowering by exporting an inhibitor of flowering to the buds. Such export would require movement against the general flow of metabolites. Transient or continuous flow of water out of fruitlets through the xylem could carry inhibitors to the bud.

Seeds are sources of several known hormones. Applications of auxin, ethylene and cytokinin can promote flowering, so there is little support for their roles as inhibitors of flowering; however, seed-produced gibberellins could inhibit flower induction. Gibberellins A_4 and A_7 were first extracted from apple seeds (Dennis and Nitsch, 1966), and many other gibberellins have been identified subsequently in seed extracts using gas chromatography-mass spectrometry (Hedden, et al., 1993; Hoad, 1980; Hoad and Ramirez, 1980; Lin et al. 1991, Luckwill et al., 1969).

Grochowska (1968) replaced seeds with cotton swabs soaked in growth regulators. Indoleacetic acid (IAA) at 20 $mg \bullet L^{-1}$ and gibberellic acid (GA_3) at 500 $mg \bullet L^{-1}$ had no effect, but GA_3 at 1000 $mg \bullet L^{-1}$ reduced flowering 33 percent and naphthaleneacetic acid (NAA) at 10 $mg \bullet L^{-1}$ promoted flowering. To my knowledge this experiment has never been repeated. Hoad and Ramirez (1980) and Hoad (1980) reported more gibberellins moving out of fruitlets of the strongly biennial ‘Laxton’s Superb’ than the more annual ‘Cox’s Orange Pippin’, as measured in diffusates collected from the pedicels *in vitro*. Stephan et al. (1997) reported export of GA_1, GA_3, GA_4, GA_7, GA_{20}, and GA_{34} by examining developing apple fruit exudates. One can question whether exudates from fruits represents a natural transport, since removing the fruits would disrupt natural transport mechanisms. They did not report the details of how these exudates were collected and how this process would compare to natural transport. Further, they collected over thirteen times as much total gibberellins from 'Spencer Seedless' fruits (without seeds) than from 'Elstar', a biennial cultivar. Most of the exudate gibberellin from 'Spencer Seedless' was GA_4, which is reported to even promote flowering (Looney, et al., 1985), but excluding GA_4, 'Spencer Seedless' still exuded 2.5 times as much inhibitory gibberellins from fruits. They did not report flowering response from spurs growing in similar conditions to those that were sampled for analysis.

Neither Green (1987), Stephan et al. (1997), nor Ban (1996) found significant transport of labeled GA from seeds to buds of apple. When Ban (1996) injected ^{14}C-GA_{12} into apple seeds *in vivo*, , no radioactivity occurred in the apex, with one exception. One unidentified polar metabolite was found in the cluster base and two metabolites were found in the fruit flesh. Less than 0.5 percent of the radioactivity was found outside the fruit. Green (1987) injected ^{3}H-GA_4 into apple seeds and found only 2-3 percent movement out of the seeds at most, and much less (0.0003-0.07 percent) was found in the bud when treated 49 - 77 days after full bloom. The cultivar that exhibited more movement in one year showed less movement the next, thus casting doubt on any correlation between alternate bearing and GA export from fruits. This contrasts with the reports of Hoad and Ramirez (1980) and Hoad (1980). They found movement of ^{3}H to buds following injection of ^{3}H-GA_4 into seeds, and this appeared to be greater in 'Laxton' than in 'Cox', but the values were small and the differences nonsignificant. Both Ban (1996) and Green (1987) found even less movement of GAs *in vitro* than *in vivo*. Movement of gibberellins may vary, not only with cultivar, but with growing conditions, or time and method of measurement. Also puzzling is the fact that while applied GAs inhibit flowering, they also reduce seed number of 'McIntosh' when applied at pink, full bloom and petal fall (Dennis and Edgerton, 1966).

Summary

Apple is an important fruit crop in an industry that depends on abundant annual flowering of each tree to produce economic yields of high value fruit. Alternate bearing continues to be a major problem in apple and the specific causes are still unknown. Shoots, seeds within developing fruits, crop load and applied gibberellins all have been shown to influence flower induction in some way. Further data is needed to better understand the interactions among these parameters that influence flower induction.

Literature Cited

Abbott, D.L. 1960. The bourse shoot as a factor in the growth of apple fruits. Ann. App. Biol. 48:434-438.

Abbott, D.L. 1970. The role of budscales in the morphogenesis and dormancy of the apple fruit bud, p. 65-82. In: L.C. Luckwill and C.V. Cutting (eds.). Physiology of Tree Crops. Academic Press, New York.

Acquaah, G., J.W. Saunders and L.C.Ewert. 1992. Homeotic floral mutations. Plant Breeding Rev. 9:63-99.

Auchter, E.C. 1919. Some influences of thinning, pollination, and fruit spur growth on the yearly performance record of fruit spurs and on the size of fruit produced. Proc. Amer. Soc. Hort. Sci. 16:118-131.

Auchter, E.C. and A.L. Schrader. 1923. Fruit spur growth and fruit bud production. Proc. Amer. Soc. Hort. Sci. 20:127-144.

Auchter, E.C., A.L. Schrader, F.S. Lagasse and W.W. Aldrich. 1926. The effect of shade on the growth, fruit bud formation and chemical composition of apple trees. Proc. Amer. Soc. Hort. Sci. 23:368-382.

Ban, J. 1996. Gibberelin metabolism and apple flowering. PhD Dissertation, Michigan State Univ.

Bangerth, F. and M. Schroder. 1994. Strong synergistic effects of gibberellins with the synthetic cytokinin N-(2-chloro-4-pyridyl)-N-phenylurea on parthenocarpic fruit-set and some other fruit characteristics of apple. Plant Growth Regul. 15:293-302.

Bigelow, W.D. and H.C. Gore. 1905. U.S. Dept. Agr. Bur. Chem. Bul. 97. (Cited by Gourley, 1922.)

Brase, K.D. 1937. The vascular anatomy of the flower of *Malus domestica* Borkh. f. *apetala* van Eseltine. M.S. Thesis, Cornell University.

Buban, T. 1981. Histological and cytological aspects of flower bud formation in apple. Acta Hort. 120:113-117.

Buban, T. 1996. Flower development and formation of sexual organs, p. 3-54. In: J. Nyéki and M. Soltész (eds.). Floral biology of temperate zone fruit trees and small fruits. Akadéniai Kiadó, Budapest.

Chailakhyan, M.Kh. 1937. Hormone theory of plant development (Russian) Moscow, Leningrad. Akad. Nauk. SSR.

Chan, B.G. and J.C. Cain. 1967. The effect of seed formation on subsequent flowering. Proc. Amer. Soc. Hort. Sci. 91:63-68.

Coen, E.S., 1991. The role of homeotic genes in flower development and evolution. Annu. Rev. Plant Physiol. Plant Mol. Biol. 42:241-279.

Crow, J.W. 1920. Biennial fruit bearing in the apple. Proc. Amer. Soc. Hort. Sci. 17:52-54.

Davis, L.D. 1957. Flowering and alternate bearing. Proc. Amer. Soc. Hort. Sci. 70:545-556.

Denker, I. and P. Hansen. 1994. Flower initiation and node development of axillary buds, growth partitioning, and bleeding sap production of young apple trees as affected by fertigation. J. Hort. Sci. 69:869-876.

Dennis, F.G., Jr. and L.J. Edgerton. 1966. Effects of gibberellins and ringing upon apple fruit development and flower bud formation. Proc. Amer. Soc. Hort. Sci. 88:14-24.

Dennis, F.G., Jr. and J.P. Nitsch. 1966. Identification of gibberellins A_4 and A_7 in immature apple seeds. Nature 211:781-782.

Edwards, G.R. 1987. Producing temperate-zone fruit at low latitudes: Avoiding rest and the chilling requirement. HortScience 22:1236-1240.

Goff, E.S. 1899. The origin and early development of the flower in the cherry, plum, apple, and pear. Wisconsin Agr. Expt. Sta. Annu. Rept. p. 289-303. (Cited by Davis, 1957.)

Gourley, J.H. 1920. The effect of shading some horticultural plants. Proc. Amer. Soc. Hort. Sci. 17:256-260.

Gourley, J.H. 1922. Text book of pomology. Macmillan, New York.

Green, J.R. 1987. The hormonal control of biennial bearing in cider apples. PhD Dissertation, Univ. of Bristol.

Greene, D.W. 1993. Effects of GA_4 and GA_7 on flower bud formation and russet development on apple. J. Hort. Sci. 68:171-176.

Griggs, W.H., G.C. Martin and B.T. Iwakiri. 1970. The effect of seedless versus seeded fruit development on flower bud formation in pear. J. Amer. Soc. Hort. Sci. 95:243-248.

Grochowska, M.J. 1968. The influence of growth regulators inserted into apple fruitlets on flower bud initiation. Bul. Acad. Polon. Sci., Ser. Biol. 16:581-584.

Guttridge, C.G.1962. Inhibition of fruit-bud formation in apple with gibberellic acid. Nature 196:1008.

Harley, C.P., H.H. Moon and L.O. Regeimbal. 1958. Evidence that post-bloom apple-thinning sprays of naphthaleneacetic acid increase blossom-bud formation. Proc. Amer. Soc. Hort. Sci. 72:52-56.

Hedden, P., G.V. Hoad, P. Gaskin, M.J. Lewis, J.R. Green, M. Furber and L.N. Mander. 1993. Kaurenoids and gibberellins, including the newly characterized gibberellin A_{88}, in developing apple seeds. Phytochemistry 32:231-237.

Heinicke, A.J. 1917. Factors influencing the abscission of flowers and partially developed fruits of the apple (*Pyrus malus* L.). Cornell Univ. Agr. Expt. Sta. Bul. 393.

Hipps, N.A. 1992. Fertigation of newly planted ‘Queen Cox’/M9 apple trees-establishment, early growth and precocity of cropping. J. Hort. Sci. 67:25-32.

Hirst, P.M. and D.C. Ferree. 1995a. Rootstock effects on shoot morphology and spur quality of 'Delicious' apple and relationships with precocity and productivity. J. Amer. Soc. Hort. Sci. 120:622-634.

Hirst, P.M. and D.C. Ferree. 1995b. Rootstock effects on the flowering of 'Delicious' apple. I. Bud development. J. Amer. Soc. Hort. Sci. 120:1010-1017.

Hirst, P.M. and D.C. Ferree. 1996. Effects of rootstock on bud development and flower formation of 'Starkspur Supreme Delicious' apple. Fruit Var. J. 50:25-34.

Hoad, G.V. 1980. The role of seed derived hormones in the control of flowering in apple. Acta Hort. 80:93-103.

Hoad, G.V. and H. Ramirez. 1980. La función de las giberelinas sintetizadas en las semillas del fruto para el control de la floración en manzanos. Turrialba 30:284-288.

Huet, J. 1972. Étude des effêts des feuilles et des fruits sur l'induction florale des brachyblastes du Poirier. Physiol. Vég. 10:529-545.

Huet, J. 1973. Floral initiation in pear trees. Acta Hort. 34:193-198.

Li, S.H., S.Q. Meng, T.H. Li, H.Z. Liu and Y.C. Tu. 1995. Critical period of flower bud induction in 'Red Fuji' and 'Ralls Janet' apple trees. Gartenbauwissenschaft 60:240-245.

Lin, J.-T., A.E. Stafford and G.L. Steffens. 1991. Identification of endogenous gibberellins in immature apple seeds. Agr. Biol. Chem. 55:2183-2185.

Looney, N.E., R.L. Granger, C.L. Chu, S.J. McArtney, L.N. Mander and R.P. Pharis. 1992. Influences of gibberellins A_4, A_{4+7}, and A_4+iso-A_7 on apple fruit-quality and tree productivity. I. Effects on fruit russet and tree yield components. J. Hort. Sci. 67:613-618.

Looney, N.E., R.P. Pharis, and M. Noma. 1985. Promotion of flowering in apple trees with gibberellin A_4 and C-3 epi-gibberellin A_4. Planta 165:292-294.

Luckwill, L.C., P. Weaver and J. MacMillan. 1969. Gibberellins and other growth hormones in apple seeds. J. Hort. Sci. 44:413-424.

Marcelle, R. and C. Sironval. 1963. Effect of gibberellic acid on flowering of apple trees. Nature 197:405.

McArtney, S.J. 1994. Exogenous gibberellin affects biennial bearing and the fruit shape of 'Braeburn' apple. New Zealand J. Crop and Hort. Sci. 22:343-346.

McArtney, S.J., D.S. Tustin, S. Seymour, W. Cashmore and N.E. Looney. 1995. Benzyladenine and carbaryl effects on fruit thinning and the enhancement of return flowering of 3 apple cultivars. J. Hort. Sci. 70:287-296.

McLaughlin, J.M. and D.W. Greene. 1991. Fruit and hormones influence flowering of apple. I. Effect of cultivar. J. Amer. Soc. Hort. Sci. 116:446-449.

Nesterov, Y.S., G.G. Anisimova and N.I. Savelev. 1972. Vestnik Sel'skokhov. Nauki 10:114-118. (Cited by Buban, 1996.)

Palmer, J.W. 1992. Effects of varying crop load on photosynthesis, dry-matter production and partitioning of 'Crispin'/M.27 apple trees. Tree Physiol. 11:19-33.

Palmer, J.W., Y.L. Cai and Y. Edjamo. 1991. Effect of part-tree flower thinning on fruiting, vegetative growth and leaf photosynthesis in 'Cox Orange Pippin' apple. J. Hort. Sci. 66:319-325.

Ramirez, H. and G.V. Hoad. 1981. Effects of growth substances on fruit-bud initiation in apple. Acta Hort. 120:131-136.

Roberts, R.H. 1920. Off year bearing and apple spur growth. Wisconsin Agr. Expt. Sta. Bul. 317.

Singh, L.B. 1948. Studies in biennial bearing. II. A review of the literature. J. Hort. Sci. 24:45-65.

Sritharan, R. and F. Lenz. 1988. Effects of water-stress in apple. I. Effect of water-supply on flower formation and fruit-set of the apple cultivar 'Golden Delicious'. Gartenbauwissenschaft 53:223-226.

Stephan, M., F. Bangerth and G. Schneider. 1997. Transport and metabolism of the gibberellins A_1, A_3, and A_4 after application to developing apple fruits of *Malus domestica* cv. Jonagold. Acta Hort. 463:113-119.

Struckmeyer, B.E. and R.H. Roberts. 1942. Investigations on the time of blossom induction in Wealthy apple trees. Proc. Amer. Soc. Hort. Sci. 40:113-119.

Tobutt, K.R. 1994. Combining apetalous parthenocarpy with columnar growth habit in apple. Euphytica 77:51-54.

Tromp, J. 1987. Growth and flower-bud formation in apple as affected by paclobutrazol, daminozide, and tree orientation in combination with various gibberellins. J. Hort. Sci. 62:433-440.

Tromp, J. 1992. The effect of soil-temperature on lateral shoot formation and flower-bud formation in apple in the 1st year after budding. J. Hort. Sci. 67:787-793.

Tromp, J. 1993. Lateral shoot formation and flower-bud formation in apple in the 1st year after budding as affected by air-temperature and exposure to red-light. J. Hort. Sci. 68:255-260.

Weigel, D. and O. Nilsson. 1995. A developmental switch sufficient for flower initiation in diverse plants. Nature 377:495-500.

Wertheim, S.J. and E.N. Estabrooks. 1994. Effect of repeated sprays of 6-benzyladenine on the formation of sylleptic shoots in apple in the fruit tree nursery. Scientia Hort. 60:31-39.

Zeller, O. 1960. Entwicklungsgeschichte der Blütenknospen und Fruchtanlangen an einjährigen Langtrieben von Apfelbüschen. I. Entwicklungsverlauf und Entwicklungsmorphologie der Blüten am einjährigen Langtrieb. Z. für Pflanzenz. 44:175-214.

SECTION 1. EFFECTS OF SEED NUMBER, FRUIT REMOVAL, BOURSE SHOOT LENGTH AND CROP DENSITY ON FLOWERING IN 'SPENCER SEEDLESS' APPLE.

Abstract

The objective of this study was to determine the effects of seed number, bourse shoot length, and crop density on flowering in 'Spencer Seedless' apple. 'Spencer Seedless' is an apetalous cultivar that is facultatively parthenocarpic; pollination results in seeded fruits. Individual spurs bearing either seedless or seeded fruits were tagged and the fruits were removed at various times between bloom and harvest in 1989-1992. The fruits were weighed, seeds counted and bourse shoots measured for each spur. Seventy to 100 percent of the spurs bearing seedless fruits flowered the following year. The percentage flowering in spurs bearing seeded fruits decreased as defruiting was delayed; this reduction was variable among years, with 10 to 70 percent of the spurs flowering when fruits were left until harvest. In 1994 and 1996 spurs were tagged at harvest, and grouped into classes based on fruit weight, seed number, shoot length, and crop density. Flowering of each bourse shoot was recorded the following year. Percentage flowering decreased from 94 to 38 (1995) or 15 (1997) as seed number increased from 0 to >15. Flowering increased with bourse shoot length; no bourse shoots less than 2 mm long flowered, whereas almost all bourse shoots longer than 16 mm flowered regardless of seed number. Seeds had the greatest inhibitory effect in shoots 2-4 mm long or when crop density was greater than four fruits per cm^2 branch cross-sectional area. There was little inhibitory effect of fruit mass independent of seeds.

Introduction

Critical to studying flower induction is having a model plant system that can be manipulated to produce floral or vegetative buds with a high degree of predictability. Most research on the control of flowering has been conducted with plants in which flowering is controlled by photoperiod (Salisbury, 1982), but this work has not been of use in solving the alternate bearing problem found in tree fruits, particularly apple, in which photoperiod has little effect on flower formation, as evidenced by the production of apple year-round at low latitudes (Edwards, 1987; Gorter, 1955).

Excessive cropping reduces or eliminates flowering in apple. When fruits are removed within 30 days of bloom, flower formation is enhanced for the following year (Harley, et al. 1942). 'Spencer Seedless' is a flower mutant apple cultivar that is homozygous recessive *ape ape* (Acquaah*et al.* 1992; Coen, 1991; Tobutt, 1994). This gene causes a second whorl of sepals to be substituted for the petals (Brase, 1937), and an additional 10 carpels are substituted for the stamens, making the flowers pistillate and capable of producing over 30 seeds per fruit. Although the apetalous flowers are not attractive to bees and thus not normally insect-pollinated, they set seedless fruits readily in most years, and form numerous flower clusters annually both on spurs and shoots. By hand pollinating the apetalous flowers, seeded and seedless fruits can be produced on the same tree. The fruits are of no commercial value.

Using a 'Spencer Seedless' apple tree at Geneva, New York, Chan and Cain (1967) discovered that when fruits were left until maturity, seeded fruits inhibited flower formation but seedless fruits did not. Further, by removing fruits at intervals during development, they were able to determine the time when developing seeds became active in preventing flower induction. When similar experiments were performed in a greenhouse, seeds failed to inhibit flowering to the same degree and this failure was associated with bourse shoots longer than 2 cm. Huet (1972, 1973) confirmed the effects of seeds on flowering of 'Bartlett' pear, and showed that seeds were more inhibitory at low than at high leaf area.

We repeated Chan and Cain's work as a means to determine the time of flower induction for sampling of spurs for a related study of the role of endogenous gibberellins in flowering. In this paper we report on the effects of several factors that modify the inhibitory effects of seeds on flowering.

Materials and Methods

Plant material. Two 'Spencer Seedless' apple trees on M.7 rootstock, planted in 1969 at the Horticultural Research and Teaching Center, East Lansing, Michigan, were used.

Pollination and fruit removal. By hand pollinating flowers of selected branches of each tree with mixtures of 'Red Delicious', 'Golden Delicious', 'Rome Beauty' or 'McIntosh' pollen (collected locally or purchased from Antles Pollen Supplies Inc., Wenatchee, Wash., or Firman Pollen Co., Inc., Yakima, Wash.), both seeded and seedless fruits were produced on the same trees in 1989, 1990, 1991, 1992, 1994 and 1996. One or two flowers per spur were hand pollinated and the remaining flowers removed.

Effects of fruit removal, seed number, bourse shoot length and fruit mass on flower induction. To establish when flowering was inhibited by seeds, fruits from pollinated and non-pollinated flowers were removed 28, 35, 56 and 147 days after full bloom (DAFB) in 1988; 10, 27, 84 and 149 DAFB in 1989; 32, 68 and 138 DAFB in 1990 and 15, 21, 28, 35, 49, 70, 110, 138, 168 DAFB in 1991, and the fruits and spurs labeled. The fruits were weighed and their seeds counted. Bourse shoot length and flowering status were recorded the following spring. When more than one fruit was borne on a spur, the fruit mass and seed number were totaled for each spur. Because 'Spencer Seedless' frequently produces two bourse shoots per spur, one spur can produce two flowering or non-flowering terminal buds or one of each. These shoots were counted independently when tabulating data.

Effects of seed number, crop density and bourse shoot length. In late October, 1994, a detailed observation of spur characteristics relative to return flowering was undertaken to identify sources of variation in flowering response. Portions of the same trees were hand pollinated in May 1994 as described previously. Twenty-five branches with varying crop loads were selected and their fruit number and base diameters recorded. Individual bearing spurs were then tagged and labeled. The fruit-bearing spurs on the remainder of the trees were also labeled. The fruits were removed, labeled as to source, weighed, and their seeds counted. In the spring of 1995 the length of the bourse shoots was measured and the type of terminal bud (flowering or vegetative) recorded. Spurs were then grouped into classes based on various parameters [seed number, shoot length, total fruit mass borne, crop density (no. of fruits per cm^2 branch cross-sectional area = BCSA)] and the percentages of the terminal buds within these classes that were floral were determined. Seed number classes were selected by observation of numbers of bourse shoots with a given number of seeds, and apparent break points in percentage flowering as seed number increased from 0 to over 15. Regression analysis was performed on seed number and percentage flowering where the relationship appeared linear. In 1996-97 this experiment was repeated except that the crop density was not determined.

Results

Effects of fruit removal, seeds, bourse shoot length and fruit mass on flower induction. In all years seedless fruits of 'Spencer Seedless' did not inhibit flowering even when left attached to the spurs until harvest (Figure 1). In contrast, seeded fruits consistently inhibited flowering. The degree of inhibition varied with year, seeds being most inhibitory in 1990. Little inhibition occurred if fruits were removed 0-20 DAFB, but inhibition increased as defruiting was delayed. Late fruit removal in 1992 resulted in less inhibition than mid-season removal. Overall among the labeled spurs, seeded fruits always reduced percentage flowering but frequently 40 to 50 % still flowered, demonstrating great heterogeneity in the spur population.

Following treatment in 1989 and 1990, bourse shoots on spurs bearing fruits containing more than six seeds were mostly vegetative, those on spurs bearing fruits with less than six seeds mostly floral, regardless of shoot length (Figure 2). In 1991 and 1992 the exceptions were more numerous, and flowering was not strongly correlated with shoot length alone. Spurs with few seeds flowered regardless of shoot length,

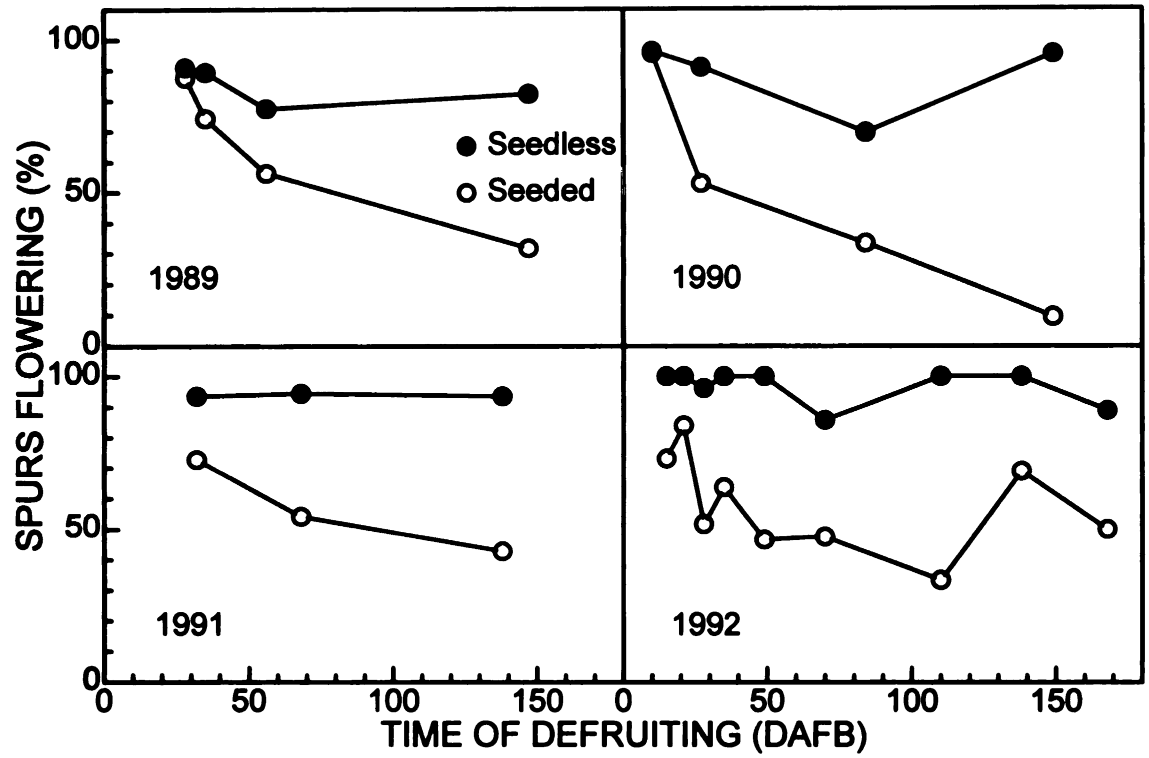

Figure 1. Effects of seeds and time of fruit removal the previous year on flowering of terminal buds of 'Spencer Seedless' apple bourse shoots. Each point represents from 6 to 45 bourse shoots. Year indicates time of fruit removal.

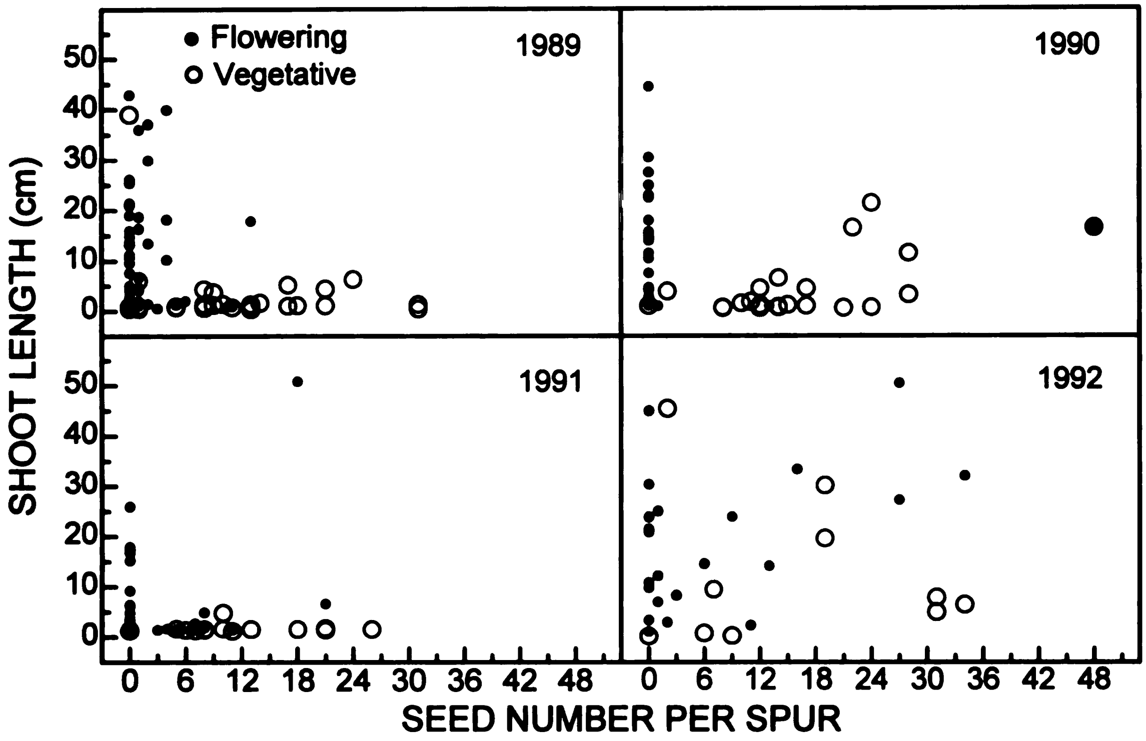

Figure 2. Effects of shoot length and seed number the previous year on flowering of terminal buds of bourse shoots of 'Spencer Seedless' apple. All fruits were harvested at maturity. Each point represents one bourse shoot.

while those with many seeds flowered less frequently. In 1989 - 1991, few of the spurs bearing seeded fruits had bourse shoots longer than 10 cm and very few flowered. In contrast, the 1992 sample of spurs bearing seeded fruits had few shoots shorter than 10 cm and flowering was more frequent (Figure 2).

At harvest fruit mass per spur varied from 0.05 to 0.5 kg for both flowering and vegetative shoots and generally increased with seed number. Seeded fruits were generally larger than seedless fruits, but with multiple fruits per spur the total fruit mass per spur was comparable for both floral and vegetative shoots (Figure 3). Spurs bearing fruits with less than five seeds were floral and those bearing fruits with five or more seeds were vegetative with few exceptions, regardless of fruit mass.

Effects of seed number, crop density and bourse shoot length. When seed number alone was plotted against flowering, percent flowering decreased from near 90 to 40 (1994) or 10 (1996) percent as seed number increased from 0 to more than 15 per spur (Figure 4). Nearly 40 % of spurs that bore fruits with more than 15 seeds still flowered in 1994, similar to observations from previous years. The fact that near 10 percent of the spurs that bore no seeds did not flower is also noteworthy. When the spurs in seed-number classes were further separated by shoot length, large differences were observed (Figure 5). Shoots less than 2 mm long showed little evidence of leaf scars and were essentially buds that formed directly on the bourse. None of these flowered. Shoots longer than 10 mm almost always flowered (11 of 333 were vegetative in 1994) regardless of seed number. For shoots from 2 to 4 mm long percent flowering decreased sharply as seed number increased. In shoots 5 to 9 mm long flowering

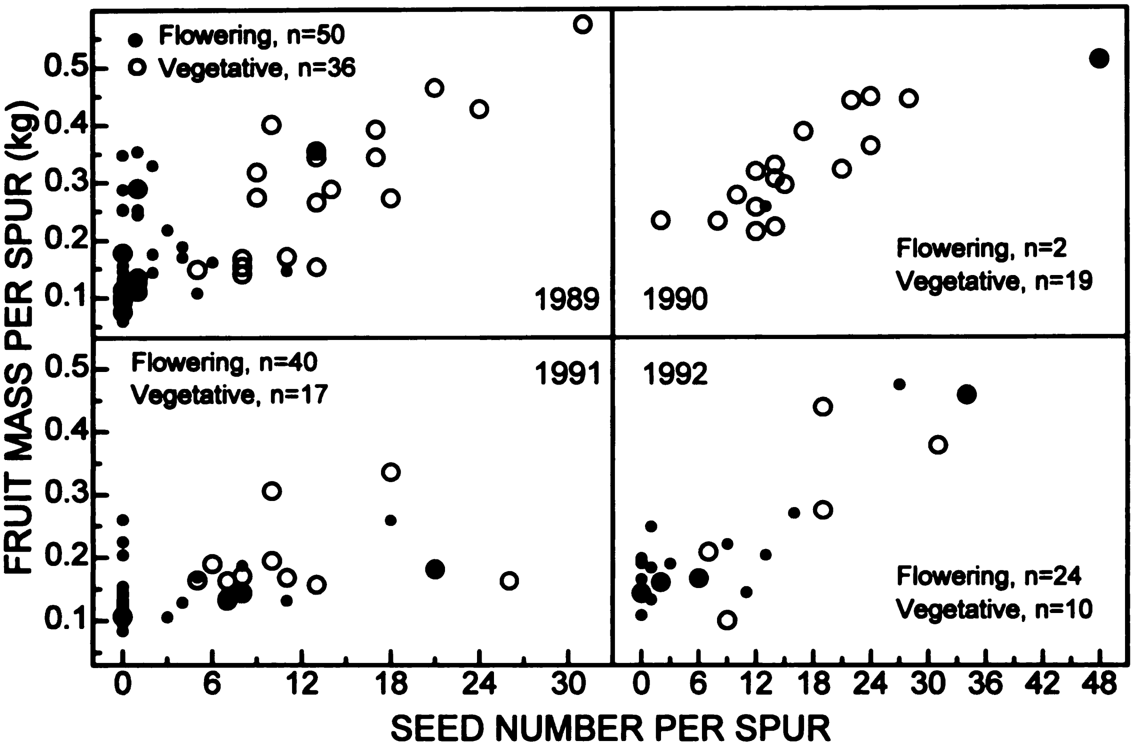

Figure 3. Relationship between total seed number and total weight of fruit per spur at harvest in 1989, 1990, 1991, and 1992 for 'Spencer Seedless' apple spurs that were flowering or vegetative the following season. All fruits were harvested at maturity. Only fruits from hand-pollinated branches were weighed in 1990. Each point represents one bourse shoot.

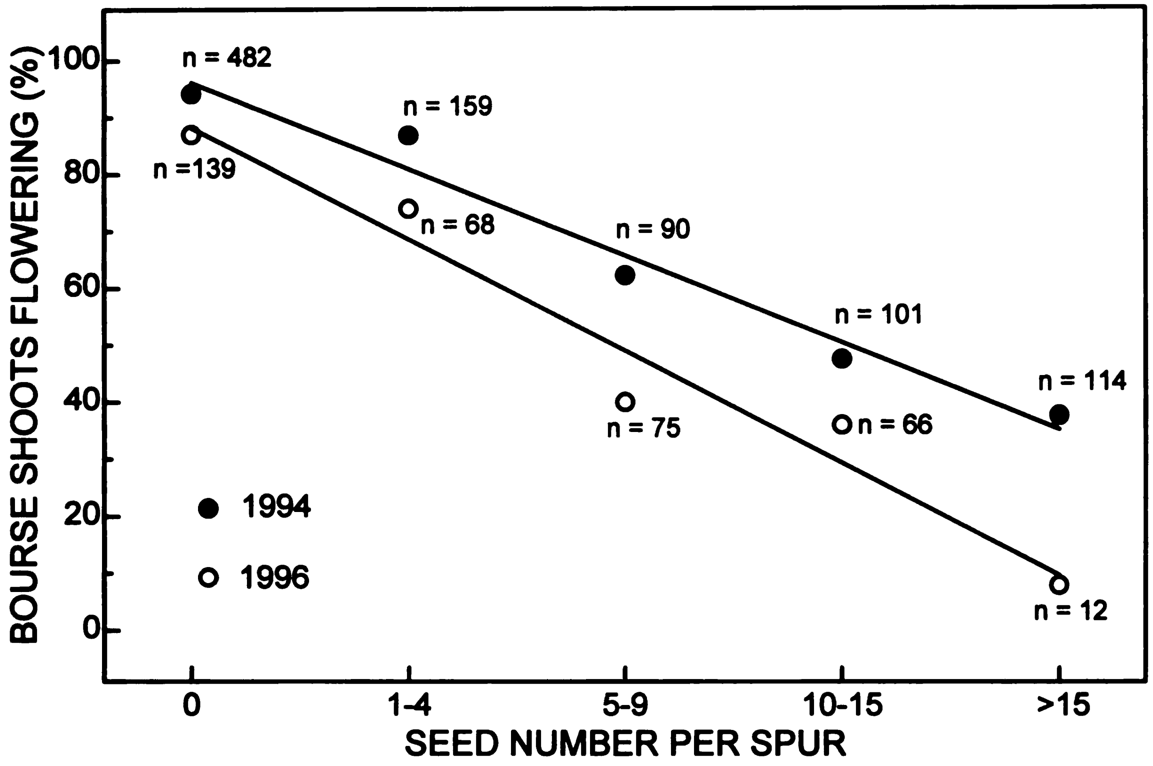

Figure 4. The relationship between seed number per spur and percentage of 'Spencer Seedless' apple bourse buds that flowered the following growing season in 1994 ($y = 96.1 - 15.2x$, $R^2 = 0.97$) and 1996 ($y = 88.2 - 19.6x$, $R^2 = 0.96$). Seed number classes are based on observation of apparent break points and sample size.

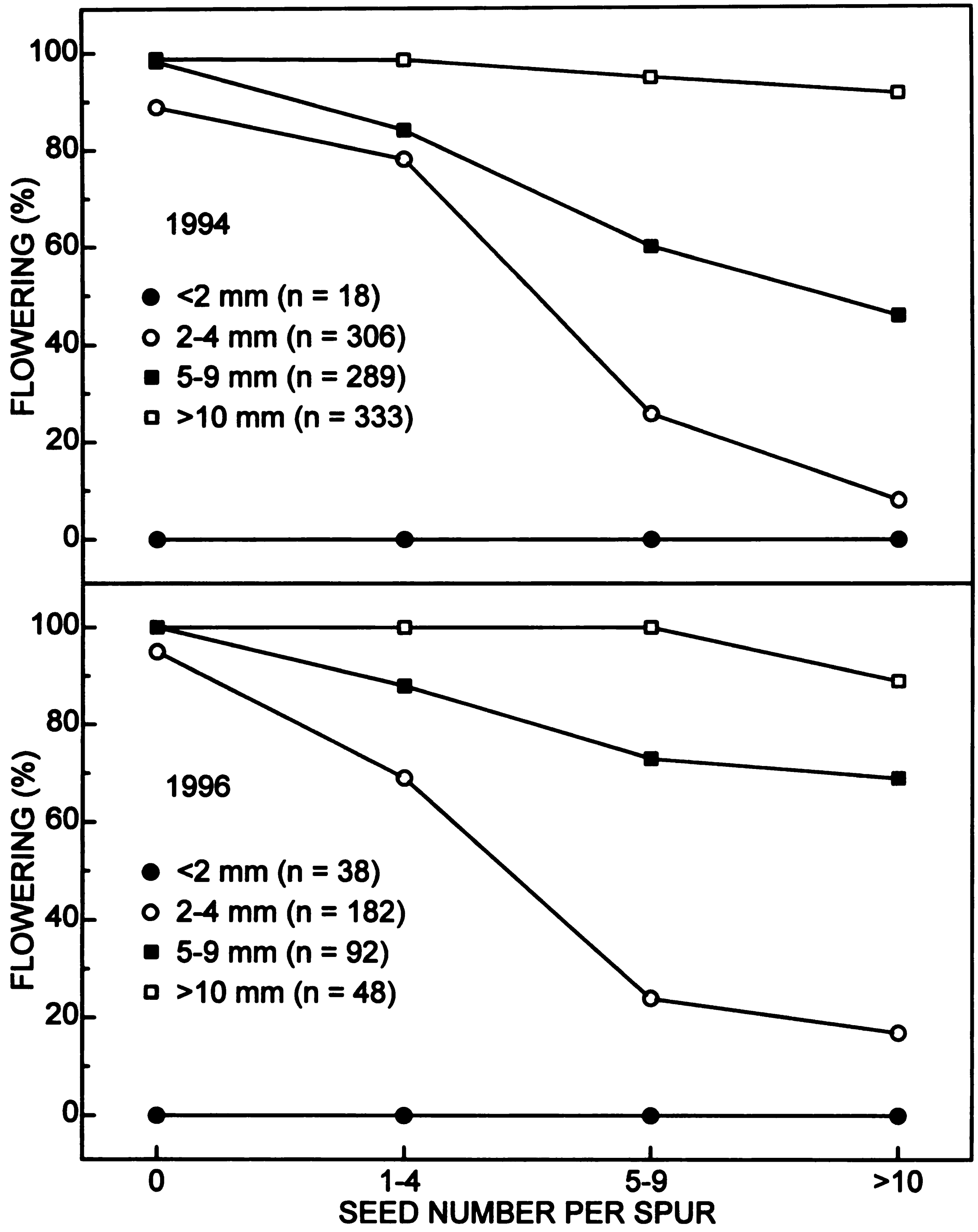

Figure 5. The relationship between seed number and shoot length of bourse shoots of 'Spencer Seedless' apple in 1994 and 1996 vs. percentage of bourse buds that flowered the following growing season. Seed number and shoot length classes were based on observation of apparent break points and sample size.

decreased as seed number increased, but response was less than in shorter shoots.

When bearing spurs were sorted by total fruit mass per spur, the number of spurs in classes less than 80 g or more than 240 g were insufficient to determine percent flowering with confidence (Figure 6). Large sample sizes are required for binomial data, such as flowering status of buds, to calculate confidence limits at $P = 0.95$. For calculations to be valid, based on an approximation of a normal distribution, a minimum of 30 spurs per class is required for 50 % flowering, whereas the sample size required increases to n = 1400 at 95% flowering. If tables based directly on the binomial distribution are used, n = 300 for each class is required for a percentage of 96% (Steel and Torrie, 1980). Still larger values for n will narrow the confidence limit. Data were available for over 1200 spurs in 1994-95, providing a much larger population than the 360 spurs studied in 1996-97. The similar distributions provide confidence in the results. When spurs were classified based on the total fruit mass borne per spur, fruit mass had little effect on percentage flowering, whereas seeds clearly inhibited flowering (Figure 7). When classified by total seed number borne per spur, less than five seeds were not inhibitory regardless of whether the spurs bore fruit mass above the median or below (Figure 8). When five or more seeds were present, higher fruit mass was associated with increased flowering in 1994 but the reverse was true in 1996.

As crop density increased, flowering decreased (Figure 9), with maximum effect when crop density exceeded four fruits/cm^2 BCSA. Crop density greater than four (maximum was 6.0) alone was not sufficient to prevent flowering by seedless fruits, but the inhibitory effect of seeded fruits was greatest when the crop density was high.

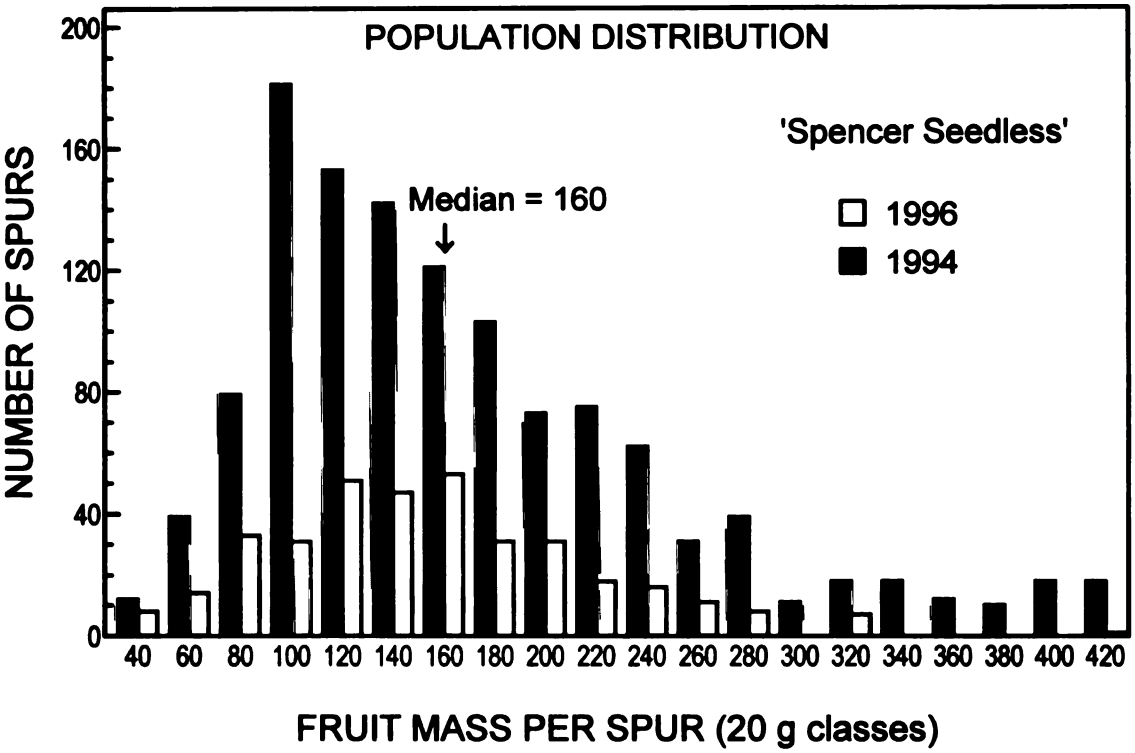

Figure 6. Distribution of bearing spurs of 'Spencer Seedless' apple by total fruit mass borne per spur in 1994 and 1996. No spurs bore less than 40 g. of fruit. 40 g = 40 to 59 g; 60 g = 60 to 79 g, etc.

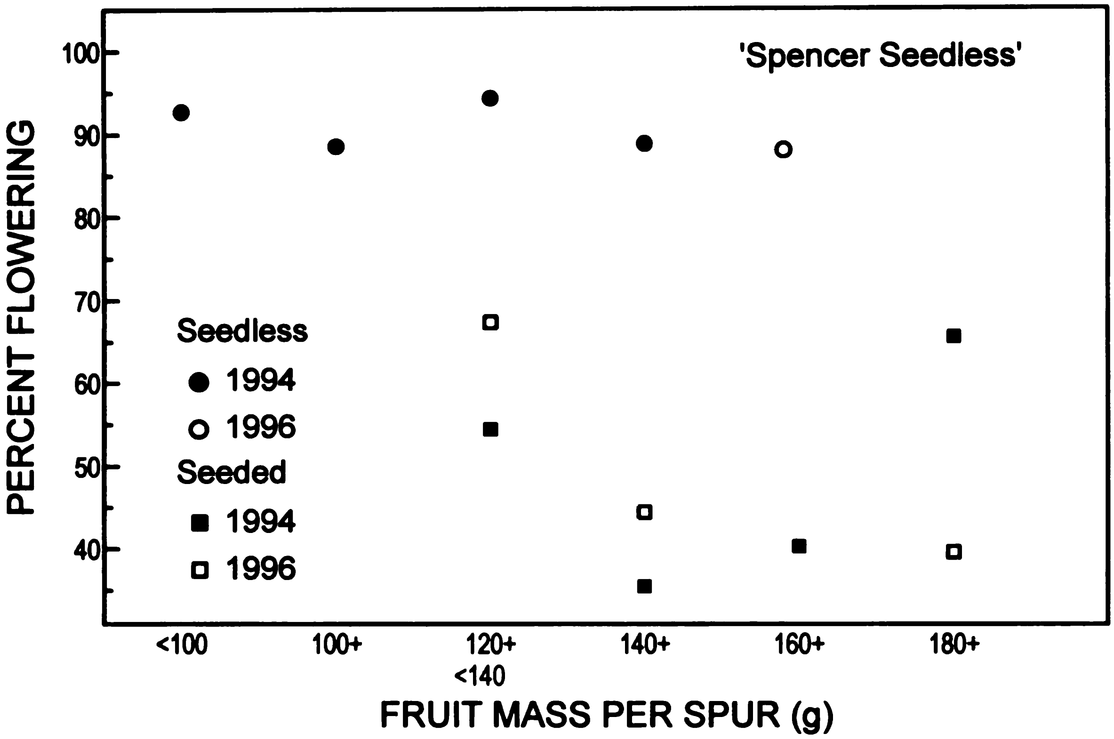

Figure 7. Effect of total 'Spencer Seedless' apple fruit mass (g) borne per spur on percentage of spurs that flowered the following year, in 1994 and 1996. Spurs bearing seedless and seeded fruits were ranked by total fruit mass (20 g increments). Classes were combined when necessary to give sufficient numbers to determine *P* = 0.95 confidence limits. Number of spurs per class varied from 49 to 337, as required to calculate confidence limits for binomial data. Classes represent spurs with fruit mass from the mass given to the next class above for a given treatment, except for the lowest class, which represents all fruits less than the value given. Insufficient spurs bore seedless fruit in 1996 to classify, so mean fruit mass of all seedless spurs is reported.

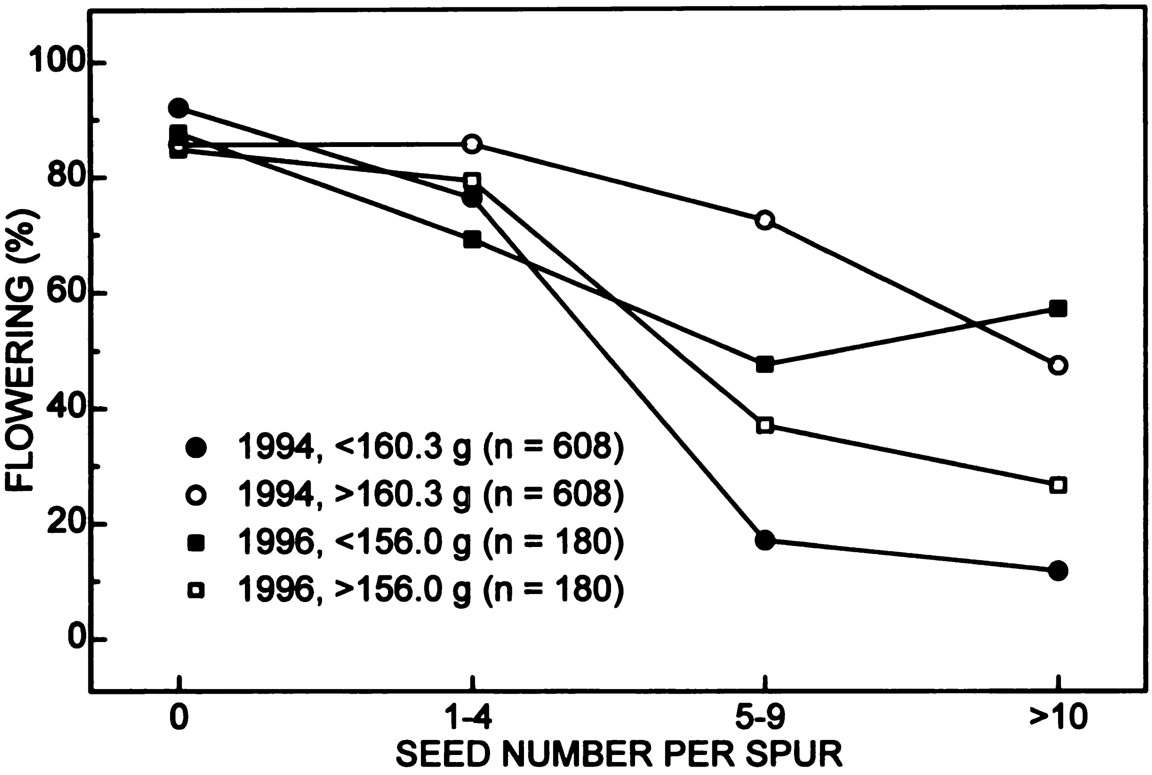

Figure 8. The relationship between seed number per spur in 'Spencer Seedless' apple and percentage of bourse buds that flowered the following growing season on spurs bearing less than or greater than the median fruit mass. Seed number classes are based on observation of apparent break points and sample size.

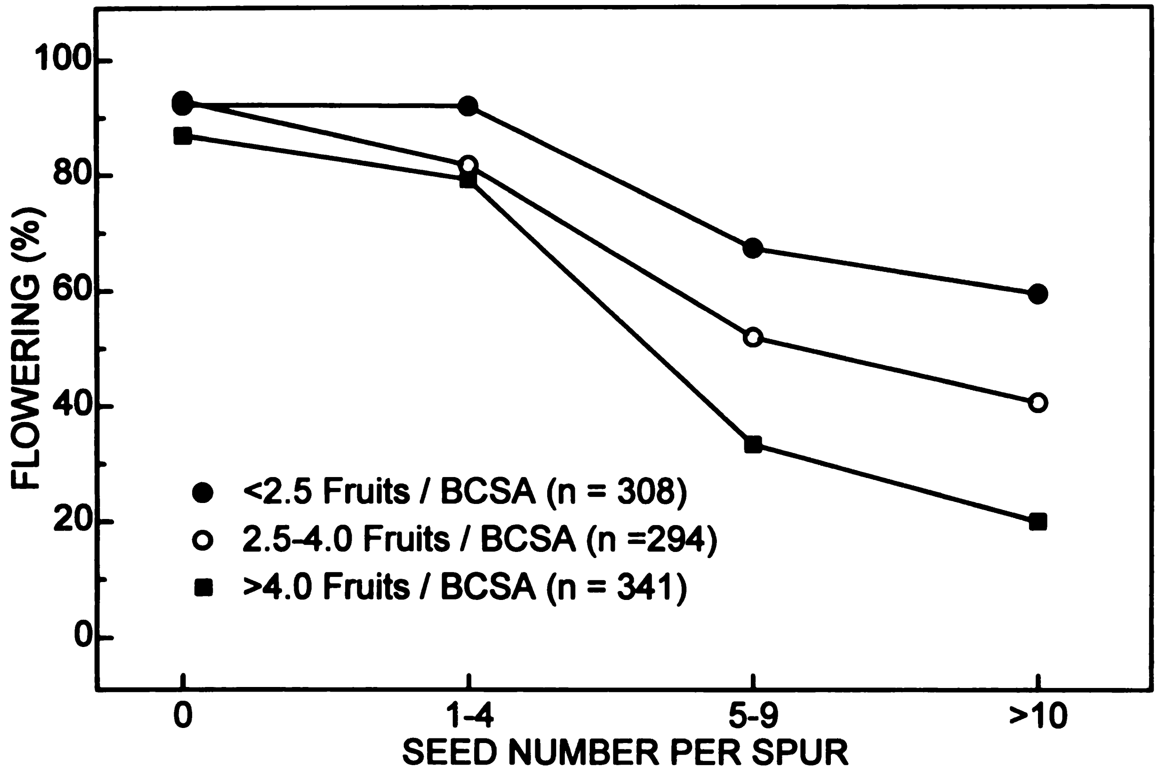

Figure 9. The relationship between seed number and crop density per cm^2 branch cross-sectional area in 'Spencer Seedless' apple in 1994 vs. percentage of bourse buds that flowered the following growing season. Seed number and crop density classes are based on observation of apparent break points and sample size.

Discussion

In this study, seedless fruits of 'Spencer Seedless' did not inhibit flowering, regardless of total fruit mass per spur or shoot length. Among spurs bearing seeded fruit, as the aggregate number of seeds per spur increased, flowering decreased in the terminal buds of the subtending bourse shoots less than 10 mm long. However, unexpectedly, seeds had little inhibitory effect in bourse shoots longer than 10 mm. These results only partially agree with those of Chan and Cain (1967), who found that one to five seeds inhibited flowering 65 to 98% under orchard conditions. Their work was done in New York State under climatic conditions similar to those in E. Lansing, but they did not provide data on shoot length, and the tree that they used may have been less vigorous than those in E. Lansing. However, Chan and Cain (1967) did report that the effects of seeds were less pronounced in greenhouse experiments, where, among spurs bearing seeded fruits, all bourse shoots longer than 50 mm, and 33 to 35% of those shorter than 50 mm, flowered. Only 72-85% of spurs bearing seedless fruits flowered under these conditions. The authors mentioned that "the axillary buds of the young spurs were often forced out and grew into axillary shoots in the same season, contrary to the field experiments in which the axillary bud remained in bud form. The longer the axillary shoots grew the more of them flowered." Whether they are referring to axillary or terminal buds of bourse shoots, which are axillary to the flower cluster, is not clear.

Similar results to these have been found with seeded and seedless 'Bartlett' pear. Huet (1972), working in France, reported a positive relationship between leaf area and flowering. Shoots on spurs bearing seeded fruits required a larger leaf surface to flower than did those on spurs bearing seedless fruits. The data of Griggs, et al. (1970) in California are more difficult to interpret. Flower buds were produced on 69 % of spurs bearing seeded fruits, compared with 94 % of spurs bearing seedless fruits, if fruits were left attached 31 DAFB. However, seeded and seedless fruits were equally inhibitory if they remained attached longer, and the inhibition was similar to that in spurs bearing seeded fruits at 31 DAFB. They found no relationship between flowering and axillary (bourse) shoot length, but presented data for only 32 spurs per category (seeded/seedless x fruit no. per spur). These were divided among shoot length classes, and most shoots were less than 25 mm long. How they calculated percentages or analyzed the data is not apparent.

Presumably extension of the shoot axis must cease before a terminal flower bud can form. The first indication of growth cessation is the abscission of the lamina and petiole of the eighth leaf primordium from the apical meristem (Abbott, 1970). One way in which long shoots differ from shorter ones may be the time of cessation of growth. The time of growth cessation relative to the timing of defoliation and defruiting treatments and flower induction has seldom been reported. This is understandable, given that apple flower induction is not sensitive to photoperiod (Gorter, 1955). In temperate climates with adequate chilling, all terminals begin growth and development simultaneously. In many cultivars, including ‘Spencer Seedless’, bourse shoots on spurs typically form two to four leaves with very short internodes and cease growth relatively synchronously within a few days after anthesis. Crow (1920) observed that most 'Oldenberg' and 'Wealthy' apple spurs grew for only four to ten days before setting terminal buds, which is about the time of fruit set. Terminal shoots with elongated internodes can bear 3 to 20 or more leaves, and therefore cessation of growth is later and flower induction may occur over an extended period of time. Hipps (1992) reported that fertigation and irrigation increased shoot length by accelerating, rather than prolonging, growth in newly planted 'Queen Cox' trees on M.9 rootstock, so that shoots of various lengths set terminal buds at about the same time. He did not report how many days after bloom this occurred. A better understanding of the relationships between shoot length and the period of shoot elongation might help in predicting when flower induction occurs and which factors are causal vs. which are parallel responses to environmental or internal signals.

Luckwill (1970) proposed that gibberellins in developing seeds inhibited flowering. This hypothesis was based on the inhibitory effects of applied gibberellins on flowering of apple, and the high concentrations of endogenous gibberellins in the seeds. However, he reported peak production of seed gibberellins in 'Emneth Early' at 9 weeks after full bloom , which coincides with rapid embryo development rather than with the period of flower induction 3 weeks earlier. Similar discrepancies with regard to timing of these processes have been reported by others (e.g., Dennis, 1976). However, timing and rate of transport would be more important than relative concentrations in determining flowering response (Bangerth, 1997). Grochowska (1968), for example, showed that the rate of diffusion of auxin from immature apple seeds following excision from the fruit differed markedly from the quantities that could be extracted from similar seeds. Longer shoots may provide greater spatial separation of the terminal bud from the developing seeds, with greater opportunity for dilution or metabolism of the gibberellins along the way. To date, however, studies of transport of gibberellins from seed to bourse shoot have provided little support for their role in inhibiting flowering (Ban, 1996; Green, 1987; Stephan, et al., 1997).

Several hypotheses, not involving gibberellins, could explain the relationship between seeds and shoot length in affecting flowering. Two of these relate to a hypothetical flowering-promoting hormone ("florigen"), and to the promotive effect of carbohydrates. The first assumes that seeds have priority over buds for "florigen". Another explanation involving "florigen"is that the greater leaf area on long shoots may produce an abundance of "florigen" that overrides the inhibitory effect of seeds. Molecular biology using model species may be helpful in identifying the reason for the effects of leaves on flower induction. This research should be performed on adult tissue of a polycarpic plant to separate flower induction responses from other maturation and senescence processes and to increase the likelihood that the results will be applicable to fruit trees. If "florigen" could be identified, preferential transport to seeded fruits, or increasing abundance with increasing leaf number, could be evaluated to test these hypotheses. Ethylene stimulates flowering in bromeliads. If it plays a role in flowering of apple, differences between seeded and seedless fruit in ACC content or transport to bourse buds could be determined. Gibberellins may inhibit this process.

Long bourse shoots have a higher leaf number and more leaf area than short ones, and therefore may furnish more carbohydrate to enhance flower induction.

However, assuming that fruit mass is an indicator of carbohydrate supply to a spur, carbohydrate cannot be the limiting factor for flower induction in 'Spencer Seedless', since spurs with seedless fruits flower over a wide range of fruit mass per spur whereas spurs bearing seeded fruit of similar mass fail to flower. Stutte and Martin (1986b) used light intensity and CO_2 enrichment to create varying carbohydrate levels in bearing and nonbearing olive trees, and found that fruits were inhibitory to flowering regardless of the carbohydrate concentration. They also reported that killing the seed prior to endocarp sclerification overcame alternate bearing without altering dry matter accumulation by the fruit (Stutte and Martin, 1986a). A similar experiment using apple would confirm the independence of flower induction from carbohydrate production.

While seeds have an inhibitory effect on flower induction of 'Spencer Seedless' apple, there appears to be an interacting effect of bourse shoot length and possibly cropload. Because of lack of sufficient numbers of trees, no replication was possible in these experiments and therefore statistical analysis was limited to regression based on spur data classified by various criteria. The main source of confidence in these results is the repeatability over several years. Large samples (> 100) increase one's confidence in discrete data where proportions (flowering vs. nonflowering) are the data of interest. The relationship of seeds, shoots, and crop to flower induction should be explored further in commercial cultivars, and with seeded and seedless fruits where adequate numbers of small uniform trees are available to allow better replication and analysis of variance. Cultivars should be selected to represent the diversity of morphological and physiological traits that influence flowering in apple. Regardless of the mechanism whereby shoot length influences flowering, it plays a more dominant role than seeds for shoots less than 2 mm or longer than 10 mm in 'Spencer Seedless'. For spurs of intermediate length seeds play a dominant role. Study of the mechanism whereby seeds inhibit flowering in 'Spencer Seedless' may need to be confined to spurs producing shoots 2 to 10 mm long.

Literature Cited

Abbott, D.L. 1970. The role of budscales in the morphogenesis and dormancy of the apple fruit bud. p. 65-82 In: L.C. Luckwill and C.V.Cutting (eds.). Physiology of Tree Crops. Academic Press, New York.

Acquaah, G., J.W. Saunders and L.C.Ewert. 1992. Homeotic floral mutations. Plant Breeding Rev. 9:63-99.

Ban, J. 1996. Gibberelin metabolism and apple flowering. PhD Dissertation, Michigan State Univ.

Bangerth, F.K. 1997. Can regulatory mechanism in fruit growth and development be elucidated through the study of endogenous hormone concentrations? Acta Hort. 463:77-87.

Brase, K.D. 1937. The vascular anatomy of the flower of Malus domestica Borkh. f. apetala van Eseltine. M.S. Thesis, Cornell University.

Chan, B.G. and J.C. Cain. 1967. The effect of seed formation on subsequent flowering. Proc. Amer. Soc. Hort. Sci. 91:63-68.

Coen, E.S., 1991. The role of homeotic genes in flower development and evolution. Annu. Rev. Plant Physiol. Plant Mol. Biol. 42:241-279.

Crow, J.W. 1920. Biennial fruit bearing in the apple. Proc. Amer. Soc. Hort. Sci. 17:52-54.

Dennis, F.G. Jr. 1976. Gibberellin-like substances in apple seeds and fruit flesh. J. Aner. Soc. Hort. Sci. 101:629-633.

Edwards, G.R. 1987. Producing temperate-zone fruit at low latitudes: Avoiding rest and the chilling requirement. HortScience 22:1236-1240.

Gorter, C.J., 1955. Photoperiodism of flowering in apple trees. Rep. XIVth Int. Hort. Cong., p 351-354.

Green, J.R. 1987. The hormonal control of biennial bearing in cider apples. PhD Dissertation, Univ. of Bristol.

Griggs, W.H., G.C. Martin, and B.T. Iwakiri. 1970. The effect of seedless versus seeded fruit development on flower bud formation in pear. J. Amer. Soc. Hort. Sci. 95:243-248.

Grochowska, M.J. 1968. Translocation of indole-3-acetic acid-2-^{14}C injected into seeds of five-week-old apple fruits. Bul. Acad. Polon. Sci., Ser. Biol. 16:577-580.

Harley, C.P., J.R. Magness, M.P. Masure, L.A. Fletcher and E.S. Degman. 1942. Investigations on the cause and control of biennial bearing in apple trees. U.S.D.A. Tech. Bul. 792.

Hipps, N.A. 1992. Fertigation of newly planted 'Queen Cox'/M9 apple trees: Establishment, early growth and precocity of cropping. J. Hort. Sci. 67:25-32.

Huet, J. 1972. Étude des effêts des feuilles et des fruits sur l'induction florale des brachyblastes du Poirier. Physiol. Vég. 10:529-545.

Huet, J. 1973. Floral initiation in pear trees. Acta Hort. 34:193-198.

Luckwill, L.C. 1970. The control of growth and fruitfulness of apple trees. p. 237-254 In: L.C. Luckwill and C.V.Cutting (eds.). Physiology of Tree Crops. Academic Press, New York.

Salisbury, F.B. 1982. Photoperiodism. Hort. Rev. 4:66-105.

Steel, R.G.D. and J.H. Torrie. 1980. Principles and Procedures of Statistics. 2nd Edition. McGraw-Hill Book Company, Inc., New York.

Stephan, M., F. Bangerth and G. Schneider. 1997. Transport and metabolism of the gibberellins A_1, A_3, and A_4 after application to developing apple fruits of *Malus domestica* cv. Jonagold. Acta Hort. 463:113-119.

Stutte, G.W. and G.C. Martin. 1986a. Effect of killing the seed on return bloom of olive. Scientia Hort. 29:107-113.

Stutte, G.W. and G.C. Martin. 1986b. Effect of light intensity and carbohydrate reserves on flowering of olive. J. Amer. Soc. Hort. Sci. 111:27-31.

Tobutt, K.R. 1994. Combining apetalous parthenocarpy with columnar growth habit in apple. Euphytica 77:51-54.

SECTION 2. EFFECTS OF FRUIT REMOVAL, CROP DENSITY, AND TRUNK GIRDLING ON FLOWER FORMATION IN 'PAULARED' APPLE: GENERAL VS. LOCAL EFFECTS OF FRUITS.

Abstract

The purpose of this study was to determine the general vs. local effect of developing apple fruits on flower induction. Whole 'Paulared' were defruited at several times from 20 to 104 days after full bloom (DAFB) in 1991 and 1992 to determine the critical time at which fruit removal could no longer stimulate flowering. Twenty fruiting terminals were tagged on each tree to determine flowering response the following season. Nearly 100 percent of tagged terminals flowered if fruits were removed before 60 DAFB, but flowering declined sharply thereafter. To determine the effect of crop density, fruit number per spur and girdling, the crop on whole trees was estimated as light, medium or heavy, and then adjusted by hand on 14 June 1996 (24 DAFB) to target crop densities of three, six or nine fruits per cm^2 trunk cross-sectional area (TCSA). On each tree, half of the scaffold limbs were thinned to two fruits per spur and half to one fruit per spur. Half of the trees with each of the target crop densities were girdled by removing a strip of bark 5 mm wide 30 cm above the soil level and applying grafting compound to the wound. Thirty spurs bearing one fruit and 30 spurs bearing two fruits around the periphery of each tree were marked and the fruit mass, seed number, bourse shoot length and flowering response recorded. Thirty new vegetative shoots per tree were also randomly selected, measured and flowering response recorded. Flowering was moderate to heavy in all cases, and girdling did not increase flowering significantly. Nearly all spurs flowered on trees with light crops, and

approximately 70 percent of the spurs on trees with medium or heavy crops. Flowering was significantly greater in non-bearing shoots than in terminals bearing one or two fruits. More lateral flowers formed on non-fruiting shoots on trees with light or medium crops than on trees with heavy crops, or on spurs bearing fruit at any cropping level. As crop density increased, flowering declined in non-girdled, but not in girdled, trees. The limited effect of fruit thinning on flowering in this biennial cultivar was unexpected, and suggests that weather conditions or other uncontrolled factors favored flowering over treatment effects.

Introduction

Developing fruits can inhibit the formation of floral buds in apple, leading to alternate bearing, but this effect varies greatly with cultivar. Some cultivars, e.g., ‘McIntosh’, are annual bearers, while others, e.g., ‘Paulared’, are biennial. Auchter (1919) and Crow (1920) suggested that alternate bearing results from too few "resting" (non-bearing) spurs in a given year, implying that the inhibitory effect of fruits is local and does not affect buds beyond the closest bourse shoot. Auchter and Schrader (1923) observed that a high percentage of spurs were continuously vegetative on ‘York Imperial’ trees. McKee and Forshey (1966) found that 22.8 percent of fruiting spurs on 'McIntosh' trees flowered the following season (repeat bloom) when cropping was moderate, but only 7.3 percent did so following a heavy crop. This suggests that alternate bearing is more complex than simply biennial bearing by individual spurs. Chemical thinning is standard practice to promote annual bearing, but results vary from year to year. Ideal crop density for annual bearing is not well defined and varies with cultivar and growing conditions.

Why developing fruits inhibit flowering is unknown. However, Chan and Cain (1967) demonstrated that the seeds were responsible for much of the inhibitory effect of the fruits on flower induction within individual spurs of 'Spencer Seedless', a facultatively parthenocarpic apple cultivar. Huet (1972, 1973) confirmed these results with seedless fruits of ‘Bartlett’ pear, and observed that flowering of spurs bearing seeded fruit increased as leaf area increased. Neilsen and Dennis (1997) found that bearing spurs of 'Spencer Seedless' may flower repeatedly if the number of seeds in the developing fruits is less than 5 or bourse shoot length is greater than 10 mm. Furthermore, the inhibitory effect of seeds declines as bourse shoot length increases. A given seed number was more inhibitory at high crop density, suggesting that there is a general inhibitory effect of a crop in addition to the local effect of developing seeds.

'Paulared' is an apple cultivar that produces long bourse shoots with terminal flower buds and forms few spurs, thus it is characterized as a "terminal bearer" with a "weeping growth habit". It also has a strong tendency toward alternate bearing. The fruits mature in late August to early September in Michigan, which makes it one of the earliest of the major commercial cultivars. These experiments were conducted to determine the effects of bourse shoot length and crop load on flower induction in a cultivar that bears seeded fruit. Because flowering was expected to be very limited when crop load was heavy, half of the trees were girdled to encourage flowering.

Materials and Methods

Expt. 1. Effects of time of fruit removal on flower initiation. To determine the influence of the time of fruit removal on flower induction, three replicate 'Paulared' trees planted at the Clarksville Horticultural Experiment Station, Clarksville, Mich., in 1980 were entirely defruited on each of four or five dates from 20 to 104 days after full bloom (DAFB) in 1991 and 1992. On each tree twenty terminal bourse shoots were tagged and evaluated for shoot length and return bloom.

Expt. 2. Effects of crop density, fruit number per spur, and girdling on flower initiation. To determine the influence of crop density, fruit number per spur and girdling on flower induction, bloom density on 21 May 1996 was visually graded on a scale from 0 (no flower clusters) to 10 (no visible vegetative terminals). Trunk circumferences were measured , and their cross-sectional areas (TCSA) calculated. Trees with light bloom were left untreated. Six replicate trees (18 total) with moderate to heavy bloom were assigned to one of three target crop density treatments (3, 6 or 9 fruits per cm^2 TCSA) and the cropload adjusted by selecting scaffold limbs representing approximately half of the bearing surface on each tree and on 14 June removing fruits from the fruiting terminals to leave two fruit per fruiting terminal (fruiting branch with bourse shoots up to 570 mm); the remaining scaffolds were thinned to one fruit per fruiting terminal. Fruits that were removed were collected, counted, and the total mass and average seed number determined. Three of the six replicate trees in each crop density group were girdled on 14 June by removing a 5 mm wide band of bark 30 cm above the soil level, after which the wound was covered with grafting compound.

Total fruit remaining per tree was estimated by counting selected branches and if too many fruits remained to reach the target crop density, additional fruiting terminals were defruited on 26 June (36 DAFB) to result in a uniformly distributed crop throughout the tree; half of which was borne two fruits per spur, half was one fruit per spur. Final mean crop densities were 3.4, 6.1 and 8.2 for the respective target crop density classes. At harvest, fruits and spurs (30 per tree bearing 2 or 1 fruits, respectively) were labeled, and the fruits were weighed and their seeds counted. Nonflowering shoots were also labeled (30 per tree) to represent zero fruits. The entire crop per tree was also counted and weighed and the crop density and yield efficiency calculated. Data from the treated trees were analyzed using a split-plot design with girdling and crop load as main plots and fruit number per spur as the split plot. Fruiting spurs on additional trees with light crops were also labeled, their fruits weighed and seeds counted to allow regression analysis of responses to crop density from very low to high levels.

Results

Expt. 1. Effects of time of fruit removal on flower initiation. When whole trees were defruited within 60 DAFB in 1991, no inhibition of flowering occurred, but delaying fruit removal to 100 DAFB reduced flowering by 60 percent (Figure 1). Similar data were obtained in 1992, except that flowering was also severely inhibited when defruiting was delayed to 73 or 103 DAFB (Figure 1). In 1992 several of the terminal buds had died and a greater percentage of those that survived were vegetative at the earliest defruiting date. In addition to terminal flowers, flowers developed from lateral buds on the bourse shoots when fruits were removed early. This rarely occurs in 'Paulared' trees bearing moderate crops. Only terminals were selected for recording flowering, and flowering did not appear to be related to shoot length, which ranged from 60 to 300 mm per tree at the time of fruit removal (Table 1).

Expt. 2. Effects of crop density, fruit number per spur, and girdling on flower initiation. Flowering in 1997 was unexpectedly heavy regardless of treatment, with 70 to 98 percent of the terminals flowering (Table 2). Girdling appeared to increase flowering, but the difference was statistically significant only at $P = 0.17$. Although values for trees with lowest crop density were greater than those for trees with heavier

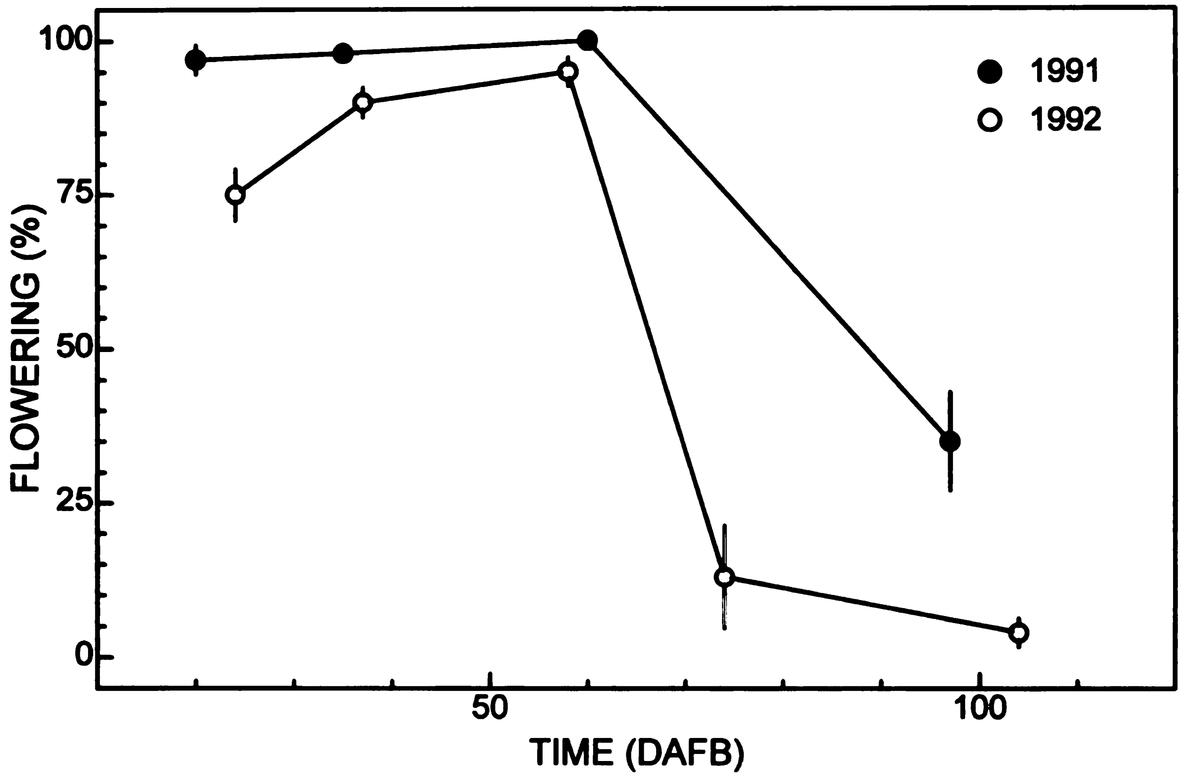

Figure 1. Effect of time of defruiting entire 'Paulared' apple trees in 1991 and 1992 on percentage of terminal buds of fruiting spurs flowering the following year. Shoots with dead terminal buds were excluded. Bars represent the standard errors of the means for three replicate trees.

Table 1. Effect of time of defruiting entire 'Paulared' apple trees in 1991 and 1992 on mean shoot length (± standard error) in the same growing season and number of lateral flowers per shoot the following year.

Time of defruiting (DAFB)		Shoot length (mm)		No. of lateral flowers[z]	
1991	1992	1991	1992	1992	1993
20	24	298 ± 38	109 ± 24	5.28 a	1.80
35	37	190 ± 37	138 ± 50	4.57 a	2.21
58	60	212 ± 24	62 ± 04	4.03 a	0.20
-	74	-	164 ± 33	-	0.67
97	104	238 ± 16	173 ± 13	0.42 b	0.00
P=		0.24	0.20	0.019	0.063

[z] Count data transformed by $\sqrt{y + 0.5}$ before analysis.

crops, the differences were again non-significant. Among trees with moderate and heavy crop densities (6.1 and 8.2 average fruits per TCSA, respectively) flowering of bourse shoots on spurs bearing no fruits was significantly greater than that on those bearing one or two fruits (Table 2), but the difference was small. Further, there were no significant interactions between crop density, girdling or fruit number per spur and flowering of terminal buds. However, the interaction between crop density and number of fruits borne per spur on the number of lateral flower buds per bourse shoot was significant (Figure 2); lateral flowering was greater on non-bearing shoots, but only at low crop densities. As would be expected, seed number and fruit mass per spur increased with crop density and with number of fruits per spur, but were not affected by girdling (Table 2).

Table 2. Main effects of girdling, crop density and number of fruits per spur in 1996 on flowering of 'Paulared' apple in 1997. Percentages calculated from 21 to 46 bourse shoots per tree for each category. Three replicate trees were used for each treatment in a split-plot design.

Main effect	Percent terminals flowering	Mean seed number per spur	Mean fruit mass per spur (g)
Girdling [z]			
Girdled	89	6.9	128
Control	70	6.6	129
P =	0.17		
Crop density class [y] (fruits/cm^2 TCSA)			
3.45±0.28	98	6.1	153
6.14±0.58	70	7.0	117
8.19±0.27	70	7.0	116
P =	0.12		
Fruits per spur [x]			
0	85 a[w]	-	-
1	79 b	6.4 a	133 a
2	74 b	13.7 b	252 b

[z] Means for 9 trees x 3 treatments (fruits per spur) per tree.

[y] Means for 6 trees x 3 treatments (fruits per spur) per tree, mean ± SE.

[x] Means for 18 trees.

[w] Mean separation within columns and factors by DMRT, $P = 0.05$.

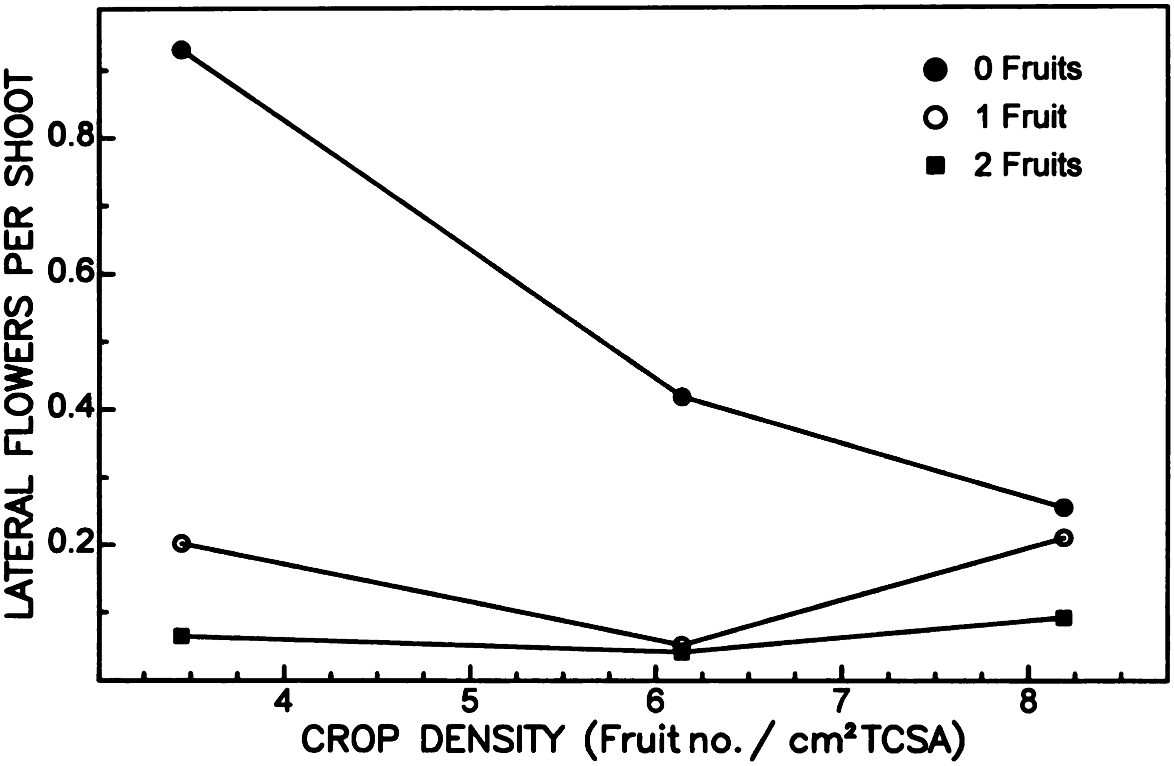

Figure 2. Effects of crop density and number of fruits borne on a spur on the average number of lateral flowers per bourse shoot.

Using regression analysis, all tagged spurs from all trees, including those with very light crop density, were ranked based on crop density, fruit number per spur, and shoot length. Only crop density significantly affected the percentage of terminals that flowered ($R^2 = 0.32$). The correlation was improved by excluding the trees that were girdled (Figure 3). The three trees with exceptionally low flowering (one girdled and two non-girdled) were the smaller trees, suggesting less vigor than others. Unexpectedly, flowering was not correlated with number of fruits (or seeds) per spur, or with bourse shoot length. The unusually high amount of return bloom in non-girdled trees with medium to high crop densities warrants specific examination of individual trees. Flowering response varied greatly among these six trees (Table 3), suggesting that factors other than crop density or fruits (seeds) per spur had a strong effect on flowering. In two trees with nearly identical crop density (4.3-4.9 fruits/cm^2 TCSA), almost all of the buds flowered in one tree regardless of the number of fruits borne on the spurs, while in the other flowering decreased as fruit number per spur increased. An even more dramatic difference was observed with three trees with crop density near 7.6 fruits/cm^2 TCSA. Two were unaffected by fruit number per spur; one with nearly all spurs flowering and the other with none. The remaining tree exhibited reduced flowering as fruit number increased. This variation suggests unknown influences on flowering within this experiment.

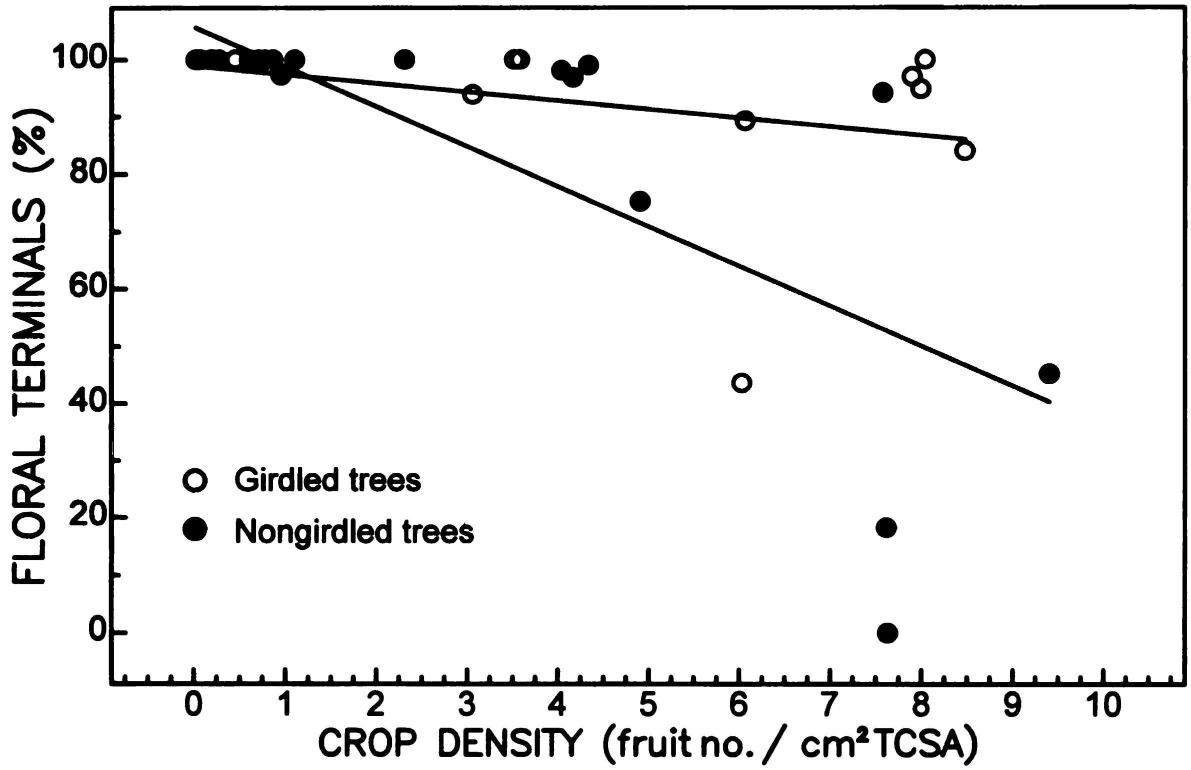

Figure 3. Effect of crop density on percentage of terminals flowering (90 shoots per tree) on girdled ($y = 98.9 - 1.5x$, $R^2 = 0.093$, $P = 0.34$) and nongirdled ($y = 105.7 - 7.0x$, $R^2 = 0.56$, $P = 0.000056$)'Paulared' trees.

Table 3. Effect of medium (M) and high (H) crop density on flower induction of non-bearing shoots and spurs bearing 1 or 2 fruits on individual, non-girdled 'Paulared' apple trees.

Crop density class and replicate (tree)	Crop density (fruit no. / cm^2 TCSA)	Mean shoot length (mm)	Flowering (%)^z of shoots on spurs bearing:		
			0 fruits	1 fruit	2 fruits
M-1	4.34	75	100.0	100.0	97.1
M-2	4.91	79	93.1	70.0	59.1
M-3	7.58	99	96.7	90.5	96.9
H-1	7.62	74	42.9	0.0	17.2
H-2	7.63	25	0.0	0.0	0.0
H-3	9.41	109	60.0	48.3	24.0

^{z} n = 30

Discussion

Based upon published data, 60 DAFB should be too late for flower induction to be affected appreciably by fruit removal, especially for a biennial cultivar such as 'Paulared'. Hand thinning is considered to be effective in enhancing return bloom only if performed before "June" drop (around 40 DAFB). Flowering response declines much more rapidly in biennial cultivars, such as 'Yellow Newtown', than in annual ones, such as 'Jonathan' (e.g., Harley, et al., 1942), and some early work, (e.g., Auchter, et al., 1926; McCormick, 1933) suggested that in biennial trees even excessive flowers were capable of inhibiting flowering in "on" years. However, I am aware of no data similar to that obtained for 'Paulared' in 1992, when percentage flowering in response to fruit removal dropped from nearly 100 % to nearly 0 % within 2 weeks.

The response of 'Spencer Seedless' (Section 1) resembled that of 'Jonathan', rather than that of 'Paulared'. The time of flower induction in 'Paulared' was well synchronized within trees and across years, suggesting that this was not a chance occurrence.

The earliest Chan and Cain (1967) removed seeded fruit of 'Spencer Seedless' was at 20 DAFB, when flowering was already reduced to 50 percent. Further delay in fruit removal decreased flowering to 20-30 percent by 60 DAFB, with minor further declines when seeded fruits remained until harvest. Harley et al., (1942) found steep declines in percentage flowering on 'Yellow Newtown' as defruiting was delayed, but not with 'Jonathan', which exhibited a gradual decline with increasing delay. In the results from Section 1 on 'Spencer Seedless' both patterns are apparent, depending on the year.

This variation among cultivars and experiments may be explained by variation in the time the bourse shoots stopped growing. Spurs have been the primary focus of other experiments to determine the time of flower induction. All labeled branch terminals of 'Paulared' in 1991 and 1992 had one or two long bourse shoots, as is typical for this cultivar. Most of these had stopped growing by 60 DAFB, but the precise date of cessation of growth was not recorded. Since flower induction cannot occur until shoot growth ceases, induction in these shoots may have been delayed relative to that in spurs, which cease growth earlier. This assumes that seeds are uniformly inhibitory over the entire time, which may not be true. The dramatic decrease in percent flowering that occurred when entire 'Paulared' trees were defruited later than 60 DAFB could be because overall crop density was heavier than on the 'Spencer Seedless' trees, particularly if only seeded fruits are considered to have an inhibitory effect. Alternatively, the difference in bearing habit (terminal bearing in 'Paulared' vs. spur-bearing in 'Spencer Seedless') could be important.

Overall, flowering was much greater than expected, given the crops produced; even in nongirdled trees with high crop density nearly 50 percent of the spurs flowered. One variable that may have influenced flowering was that the trees were not irrigated until mid-August, and the soil surface was noticeably dry. How much drought stress existed and what influence this had on flower formation is unknown. Drought conditions are considered by many to favor flowering, but actual data are limited.

In contrast with the results with 'Spencer Seedless', seeded fruits of 'Paulared' did not exhibit a consistent local effect on flowering, nor did shoot length appear to be related to flower formation; rather, crop density had the greatest influence, at least in non-girdled trees. The few bearing spurs on trees with very light crops still flowered, including those with 3-5 fruits. Conversely, flowering was frequently inhibited in non-fruiting shoots on heavily cropping, non-girdled trees, indicating an overall inhibitory effect of crop. Lakso (personal communication) found an interaction between cropload and fruit number per spur on 'Empire' apple in New York. One fruit per spur inhibited flowering only when cropload was light, whereas two fruits per spur were inhibitory regardless of cropload.

Auchter (1919) recorded data for large numbers of individual spurs for several cultivars to determine the relationships between growth, flowering and fruiting. In 'Baldwin', 'Northern Spy', 'Wagner', 'Ben Davis' and 'Tompkin's King' few or no spurs flowered two years in succession, whereas in 'Rome Beauty', 'Grimes Golden', 'Smokehouse' and 'Delicious' 20 to 50 % of the spurs flowered in two successive years; however, the spurs that did so did not set fruit. When spurs of these "annual cultivars" bore fruit, less than one percent flowered the following year. The difference between biennial and annual cultivars may be the degree to which overall crop inhibits flower induction, even on nonbearing spurs or shoots. Auchter (1919) further observed that 'Rome Beauty' and 'Smokehouse' bear terminally, and the side growth (long bourse shoot) often produces a flower bud and fruits the following year; thus these cultivars are considered annual bearers. This condition also occurs with 'Yellow Transparent' and 'Ben Davis' when the trees are young. This is not the case with 'Paulared', which exhibits similar terminal bearing habit, but is still biennial. Auchter (1919) also found that fruit removal at June drop reduced flowering the following year, except for 10 unusually long spurs of one 'Delicious' tree that produced terminal flower buds. This is similar to the pattern observed with 'Spencer Seedless' reported earlier.

Girdling generally overcame the inhibitory effect of heavy cropping, with 90 percent of the terminals flowering. The one exceptional tree was notably low in vigor. A local inhibitory effect of developing fruits on flowering was apparent in some of the 'Paulared' trees used, but this was not related to crop density or other known factors.

'Paulared' appears to differ from many other biennial cultivars in that fruits appear to become inhibitory to flowering very late in their development. This, plus its terminal bearing habit, make it an interesting subject for further investigation. Further work with 'Paulared' should focus on determining how the flower induction period varies within a given tree or cultivar and how overall cropping interacts with local distribution of fruits to influence bourse shoots. Methods of altering seed number mechanically or chemically may help determine the specific role of the fruit. The time when shoots cease growth relative to treatments also should be noted. Comparison of the biennial 'Paulared' with another terminal bearing cultivar with an annual bearing habit, such as 'Rome Beauty' may help explain the difference in flowering response. Care should be taken to use uniform trees with ample replication and a minimum of environmental variables.

Literature Cited

Auchter, E.C. 1919. Some influrences of thinning, pollination and fruit spur growth on the yearly performance record of fruit spurs and on the size of fruit produced. Proc. Amer. Soc. Hort. Sci. 16:118-131.

Auchter, E.C. and A.L. Schrader. 1923. Fruit spur growth and fruit bud production. Proc. Amer. Soc. Hort. Sci. 20:127-144.

Auchter, E.C., A.L. Schrader, F.S. Lagasse and W.W. Aldrich. 1926. The effect of shade on the growth, fruit bud formation and chemical composition of apple trees. Proc. Amer. Soc. Hort. Sci. 23:368-382.

Chan, B.G. and J.C. Cain. 1967. The effect of seed formation on subsequent flowering. Proc. Amer. Soc. Hort. Sci. 91:63-68.

Crow, J.W. 1920. Biennial fruit bearing in the apple tree. Proc. Amer. Soc. Hort. Sci. 17:52-54.

Harley, C.P., J.R. Magness, M.P. Masure, L.A. Fletcher and E.S. Degman. 1942.

Investigations on the cause and control of biennial bearing of apple trees. U.S. Department of Agriculture Tech. Bul. 792, 57 p.

Huet, J. 1972. Etude des effets des feuilles et des fruits sur l'induction florale des brachyblastes du Poirier. Physiol. Vég. 10:529-545.

Huet, J. 1973. Floral initiation in pear trees. Acta Hort. 34:193-198.

McCormick, A.C. 1933. Control of biennial bearing in apples. Proc. Amer. Soc. Hort. Sci. 30:326-329.

McKee, M.W. and C.G. Forshey. 1966. Effects of chemical thinning on repeat bloom of 'McIntosh' apple trees. Proc. Amer. Soc. Hort. Sci. 88:25-32.

Neilsen, J.C. and F.G. Dennis, Jr. 1997. Effects of seed number, fruit removal, bourse shoot length and crop density on flowering in 'Spencer Seedless' apple. Acta Hort. (In press)

SECTION 3. EFFECTS OF DEFRUITING, SEED REMOVAL AND REPLACEMENT OF SEEDS WITH GIBBERELLINS ON FLOWER FORMATION IN APPLE.

Abstract

Experiments were conducted to determine the effects of defruiting, adjusting seed number and replacement of seeds with gibberellin on flowering of 'Golden Delicious', 'Jonagold', 'Red Delicious', 'Spencer Seedless' and 'Ohio No. 3' (another seedless cultivar) apple. Seed number was altered by cutting fruits to remove or injure seeds in late June, approximately 30 days after full bloom when fruits were about 30 mm in diameter. Gibberellin A_3 or A_{4+7} was mixed with lanolin (1000 mg•L^{-1}) and 0.02 mL applied to each empty locule (seeds removed or naturally seedless). Flowering the following spring was greater than expected in both controls and gibberellin-treated spurs. Neither cutting fruits nor seed removal increased flowering significantly. The treatments were applied late in the period of flower induction when defruiting had little effect upon response. Nevertheless the results do not support the hypothesis that seed-produced gibberellins are responsible for inhibiting flowering in apple.

Introduction

The inhibition of flowering in apple by heavy crops is a serious limiting factor to profitable fruit production. Chan and Cain (1967) provided evidence that seedless fruits do not inhibit flowering. Developing apple seeds produce hormones, including gibberellins (Dennis and Nitsch, 1966; Hedden, et al., 1993; Lin et al., 1991), and spraying whole trees or foliage with gibberellins will inhibit flowering, suggesting that seed-produced gibberellins may be responsible for inhibition.

One can test the hypothesis that seed-produced gibberellins inhibit flower induction in apple by substituting gibberellins for seeds. In a study on fruit set, Abbott (1959) described techniques for reducing seed numbers in fruit by injecting ethanol into locules or cutting fruits transversely and replacing seeds with lanolin paste containing growth regulators. Grochowska (1968) replaced seeds with cotton swabs soaked in growth regulators. Seed removal alone was performed too late to affect flowering. Indoleacetic acid (IAA) at 20 $mg•L^{-1}$ and gibberellic acid (GA_3) at 500 $mg•L^{-1}$ had no effect, but GA_3 at 1000 $mg•L^{-1}$ reduced flowering 33 percent and naphthaleneacetic acid (NAA) at 10 $mg•L^{-1}$ increased flowering. To my knowledge this experiment has never been repeated. Hoad (1980) and Hoad and Ramirez (1980) reported more gibberellins moving out fruitlets of the strongly biennial 'Laxton's Superb' than the more annual 'Cox's Orange Pippin', as measured in diffusates collected from the pedicels *in vitro*. The purpose of my work was to determine if replacing seeds with gibberellin can inhibit flowering in seeded and/or seedless apple fruits.

Materials and Methods

Expt. 1. Seeded cultivars. On 25 June 1996 (32 DAFB) five 'Golden Delicious'/M 26 trees planted in 1991 at the Clarksville Horticulture Experiment Station, Clarksville, Mich., were selected. On each tree spurs with bourse shoots 2-20 mm long bearing two or more fruits were treated in the following manner:

A. Fruiting control.

B. Fruits removed.

C. Fruits cut transversely through locules and seeds, but apex of fruit not severed.

D. Fruits cut transversely, all but three seeds removed, and top of fruit replaced and fixed in place with Parafilm®.

E. Fruits cut as in D, all but one seed removed.

F. Fruits cut as in D, all seeds removed and a total of approximately 0.1 mL of lanolin paste placed in the empty locules (0.02 mL in each) with a one-mL syringe.

G. Same as F except that the lanolin paste contained 1000 $mg \bullet L^{-1}$ GA_3.

H. Same as F except that the lanolin paste contained 1000 $mg \bullet L^{-1}$ GA_{4+7}.

Two fruits were treated on each spur, and any additional fruits were removed. Each tree bore about five spurs per treatment (25 total) except for treatment A with a total of 100 spurs being tagged throughout the five trees. At harvest, fruits were weighed and all seeds counted. Trunk diameter was also measured and total crop was counted and weighed (Table 1). The following spring (1997) tagged spurs were evaluated for flower buds, and entire trees were rated visually from 0 to 10 based on estimated bloom density.

A nearly identical experiment was conducted on 'Jonagold'/M 9 trees planted in 1990 at the Horticulture Teaching and Research Center, E. Lansing, Mich., except that the 25 spurs per treatment were distributed among six trees and not all seed number and cropping data were collected. Treatments A - C were also applied to spurs on seven Redchief 'Delicious' trees planted in 1981 at the Clarksville Horticulture Experiment Station, Clarksville, Mich. No data were available on seed number per fruit because the crop was harvested before the treated fruits could be collected.

Expt. 2. Seedless cultivars. Similar experiments were also conducted in 1996 on 'Spencer Seedless' and 'Ohio No. 3' (both facultatively parthenocarpic) trees planted at the Horticulture Teaching and Research Center, E. Lansing, Mich., in 1969. Hand pollination during bloom provided some spurs bearing seeded fruits for treatment A. The 25 spurs used per treatment were all on one 'Spencer Seedless' tree, but were distributed between two 'Ohio No. 3' trees. Treatments C - E were omitted and treatments F - H applied to seedless fruits.

Results

Expt. 1. Seeded cultivars. In 'Golden Delicious' return flowering in bearing spurs was greater than expected, given the biennial bearing habit of this cultivar, with 67.5% of the buds on control spurs flowering (Table 1,2). Because of lack of replication, only large differences are of interest. Defruiting enhanced flowering only

Table 1. Harvest data for five 'Golden Delicious' apple trees used in Expt. 1. Clarksville Horticulture Experiment Station, 1996 - 1997.

Tree no.	Fruit (no./tree)	Fruit mass (kg/tree)	Trunk dia. (cm)	Crop density (fruit no./ cm^2 TCSA)	Yield efficiency ($g \cdot cm^{-2}$ TCSA)	Relative return bloom (0-10)
1	71	10.32	6.2	2.35	342	10.0
2	88	12.48	5.8	3.33	473	9.8
3	68	11.28	4.9	6.61	598	6.5
4	71	11.71	6.3	2.28	376	8.9
5	283	35.55	6.2	9.37	1177	5.2

Table 2. Effects of defruiting, fruit cutting, seed removal, and replacement of seeds with lanolin pastes on flowering of 'Golden Delicious' apple. Clarksville Horticulture Experiment Station, 1996 - 1997. Treatments were applied to 5 trees on 26 June 1996.

Treatment	Total buds (no.)	Flower buds (no.)	Flowering (%)	Number of mature seeds		
				Mean	S.E.	C.V.
Defruited control	22	17	77	7.2^{z}	0.2	3.4
Fruiting control	114	77	68	7.7	0.4	46.1
Locules cut	25	21	84	1.5	0.4	123.4
3 seeds left	19	17	90	0.9	0.3	151.8
1 seed left	21	20	95	0.0		
0 seeds left + lanolin paste						
Control	24	23	96			
GA_3	25	21	84			
GA_{4+7}	25	23	92			

z Immature seeds

slightly. Cutting locules reduced average seed number per spur, but up to five seeds still developed to maturity. Both seed removal treatments also reduced seed number, although the number of seeds remaining varied widely, and appeared to increase flowering relative to the fruiting control. Replacement of seeds with gibberellins had no appreciable effect on flowering. Results with 'Jonagold' were similar to those with 'Golden Delicious' (Table 3), except that almost all buds on fruiting spurs flowered. Although no seed number data were collected for 'Delicious', both defruiting and cutting the locules of developing fruits appeared to increase flowering (Table 4).

Table 3. Effects of defruiting, fruit cutting, seed removal, and replacement of seeds with lanolin pastes on flowering of 'Jonagold' apple. Horticulture Teaching and Research Center, 1996 - 1997. Treatments were applied to 6 trees on 29 June 1996.

Treatment	Total buds (no.)	Flower buds (no.)	Flowering (%)	Number of mature seeds		
				Mean	S.E.	C.V.
Defruited control	22	19	86	4.3[z]	0.4	37.0
Fruiting control	17	16	94	4.4	0.3	29.9
Locules cut	25	20	80			
3 seeds left	19	18	95			
1 seed left	20	20	100			
0 seeds left + lanolin paste						
Control	19	19	100			
GA_3	19	18	95			
GA_{4+7}	22	20	91			

[z] Immature seeds

Table 4. Effects of defruiting and fruit cutting on flowering of 'Delicious' apple. Clarksville Horticulture Experiment Station, 1996 - 1997. Treatments were applied to 7 trees on 30 June 1996.

Treatment	Total buds (no.)	Flower buds (no.)	Flowering (%)
Fruiting control	37	23	62
Defruited	36	31	86
Locules cut	31	30	97

Expt. 2. Seedless cultivars. In 'Spencer Seedless', as the seed number borne per spur increased, percentage flowering decreased (Table 5). Defruiting, or cutting and treating seedless fruits with the control lanolin paste did not affect response. Treating locules with gibberellin appeared to decrease the percentage flowering slightly in 'Spencer Seedless', but not in 'Ohio No. 3' (Table 6) where flowering was 90 to 100 percent in all treatments.

Discussion

The data for 'Spencer Seedless' demonstrated the quantitative inhibitory effect of seeds, but substituting gibberellin for the seeds was not as inhibitory as expected, and failed to inhibit flowering in 'Ohio No. 3'. Flowering of fruiting control spurs of seeded cultivars was greater than expected (62 - 94 %), which can best be attributed to the moderate crop most of these trees carried. In 'Golden Delicious' return bloom rating was proportional to crop density. Taken together, this suggests an effect of cropload that is stronger than the local effect of seeds within fruits borne on the spur; similar gibberellin treatments might have been more inhibitory had cropload been heavier.

Table 5. Effects of seed number and of applications of lanolin pastes to locules of cut fruits of 'Spencer Seedless' at the Horticulture Teaching and Research Center, 1996 - 1997. Treatments were applied to fruits on one tree on 29 June 1996.

Treatment	Mean seed number	Total buds (no.)	Flower buds (no.)	Flowering (%)
Defruited control		24	23	96
Fruiting control	0	126	119	94
	1-4[z]	66	49	74
	5-9	73	30	41
	10-15	63	23	36
	>15	13	20	15
Locules cut + lanolin paste				
Control		19	19	100
GA_3		22	18	82
GA_{4+7}		20	16	80

[z] Data in seed number classes taken from Section I for comparison.

Table 6. Effects of defruiting and applications of lanolin pastes to locules of cut fruits of 'Ohio No. 3' apple at the Horticulture Teaching and Research Center, 1996 - 1997. Treatments were applied to fruits on 2 trees on 25 June 1996.

Treatment	Total buds (no.)	Flower buds (no.)	Flowering (%)
Defruited control	47	46	98
Fruiting control	65	58	89
Locules cut + lanolin paste			
Control	41	39	95
GA_3	39	39	100
GA_{4+7}	46	46	100

This effect of crop was also observed in 'Paulared' as discussed in Section 2. Other possible explanations for the small and inconsistent differences between defruited and fruiting controls may be the small numbers of spurs used to calculate percentages (about 20 per cultivar in most cases), and the need to use several trees of each cultivar without being able to factor out crop density and other tree differences. Had more spurs been available per tree, trees could have been used as blocks.

In 'Spencer Seedless' 5 to 9 seeds were sufficient to limit flowering to 41 %, whereas in seeded cultivars similar numbers of seeds has less effect (68 % flowering with 7.7 seeds per spur in 'Golden Delicious'; 94 % with 4.4 seeds per spur in 'Jonagold'). Note that return bloom was greater in 'Jonagold', with fewer seeds per spur, than in the biennial cultivar 'Golden Delicious'.

The results of this study fail to support the hypothesis that seed-produced gibberellins inhibit flowering. Gibberellins applied to foliage are inhibitory, even when applied to trees bearing few or no fruits (Dennis and Edgerton, 1966; Li et al., 1995; McArtney, 1994), so the inhibitory effect of gibberellins is well established. Whether or not seed gibberellins are involved in the control of flowering is less clear. Several experiments in which GAs have been substituted for seeds have had little or no effect on flowering in apple.

Grochowska (1968) applied gibberellins to locules in cotton swabs, which may make limited contact with the surface of the locule, dry out, or result in rapid absorption by the fruit tissue, providing a nearly instantaneous dose for metabolism or transport, and therefore be ineffective. I assumed that lanolin paste would provide better contact with the locular surface and would not dry out, and that diffusion of GA into the fruit tissue would be slower, but more prolonged, better simulating the effects of seeds as sources of GAs. Applying GAs directly to the bourse bud in lanolin might verify that they can diffuse out of lanolin paste into the tissue and inhibit flowering, but would not test whether they can diffuse from seeds in sufficient quantities to do so.

Although application of GAs in lanolin may be preferable to application in cotton swabs, neither method is ideal, for seeds are attached to the fruits by specialized placental tissue, and transport of substances through this tissue is metabolically regulated. Furthermore, this tissue provides a very small "target" relative to the entire suface of the locule.

Application may also have been after flower induction had occured. The time of flower induction varies considerably with cultivar and bearing condition, as noted in

Sections 1 and 2 above. Selected spurs had bourse shoots 5 to 20 mm. long that should have had high potential to flower (Auchter and Schrader, 1923; Bobb and Blake, 1938) except that they were bearing fruit. At 30 DAFB they had ceased growth, but the precise time of flower induction was not determined.

Another possibility is that the amount of GA applied was insufficient. This is unlikely, however, for approximately 0.1 mg of GA was applied per fruit, which is more by several orders of magnitude than the levels found in seeds. Green (1987) injected ^3H-GA_4 into apple seeds and found that only 2-3 percent moved out of the seeds at most; much less (0.0003-0.07 percent) was found in the bud. More was transported later than earlier in the growing season and the cultivar that exhibited more movement in one year showed less movement the next, eliminating any correlation between alternate bearing and GA export from fruits. Stephan et al. (1997) injected ^3H-GA_1, ^3H-GA_3 and ^3H-GA_4 into the core of apple fruits at 49 DAFB and found that much less than five percent was transported out of the fruit and much less than one percent occurred in the bourse shoot. Of these limited amounts, GA_1 and GA_3 was transported preferentially to GA_4. In contrast, Hoad (1980) and Hoad and Ramirez (1980) reported movement of ^3H to bourse shoots following injection of ^3H-GA_4 into seeds, with more transport in 'Laxton' (biennial) than in 'Cox' (annual), though the values were small and the differences nonsignificant.

Movement of gibberellins may vary, not only with cultivar, but with growing conditions, or time and method of measurement. However, in general, this mechanism is not well supported by the data at hand and must be viewed as only one of several alternative hypotheses (e.g., "florigen", auxin transport, cytokinin/gibberellin balance, ethylene production or sensitivity) to explain the role of seeds in alternate bearing.

Literature Cited

Abbott, D.L. 1959. The effects of seed removal on the growth of apple fruitlets. Annu. Rep. Long Ashton Agr. Hort. Res. Sta. 1958. p 52-56.

Auchter, E.C. and A.L. Schrader. 1923. Fruit spur growth and fruit bud production. Proc. Amer. Soc. Hort. Sci. 20:127-144.

Bobb, A.C. and M.A. Blake. 1938. Annual bearing in the 'Wealthy' apple was induced by blossom thinning. Proc. Amer. Soc. Hort. Sci. 36:321-327.

Chan, B.G. and J.C. Cain. 1967. The effect of seed formation on subsequent flowering. Proc. Amer. Soc. Hort. Sci. 91:63-68.

Dennis, F.G. Jr., and L.J. Edgerton. 1966. Effect of gibberellins and ringing upon apple fruit development and flower bud formation. Proc. Amer. Soc. Hort. Sci. 88:14-24.

Dennis, F.G., Jr. and J.P. Nitsch. 1966. Identification of gibberellins A_4 and A_7 in immature apple seeds. Nature 211:781-782.

Green, J.R. 1987. The hormonal control of biennial bearing in cider apples. PhD dissertation, Univ. of Bristol.

Grochowska, M.J. 1968. The influence of growth regulators inserted into apple fruitlets on flower bud initiation. Bul. Acad. Polan. Sci. Ser. 16:581-584.

Hedden, P., G.V. Hoad, P. Gaskin, M.J. Lewis, J.R. Green, M. Furber and L.N. Mander. 1993. Kaurenoids and gibberellins, including the newly characterized gibberellin A_{88}, in developing apple seeds. Phytochemistry 32:231-237.

Hoad, G.V. 1980. The role of seed derived hormones in the control of flowering in apple. Acta Hort. 80:93-103.

Hoad, G.V. and H. Ramirez. 1980. La función de las giberelinas sintetizadas en las semillas del fruto para el control de la floración en manzanos. Turrialba 30:284-288.

Li, S.H., Z.Q. Meng, T.H. Li, H.Z. Liu and Y.C. Tu. 1995. Critical period of flower bud induction in 'Red Fuji' and 'Ralls Janet' apple trees. Gartenbauwissenschaft 60:240-245.

Lin, J.-T., A.E. Stafford and G.L. Steffens. 1991. Identification of endogenous gibberellins in immature apple seeds. Agr. Biol. Chem. 55:2183-2185.

McArtney, S.J. 1994. Exogenous gibberellin affects biennial bearing and the fruit shape of 'Braeburn' apple. New Zealand J. Crop and Hort. Sci. 22:343-346.

Stephan, M., F. Bangerth and G. Schneider. 1997. Transport and metabolism of the gibberellins A_1, A_3, and A_4 after application to developing apple fruits of *Malus domestica cv.* Jonagold. Acta Hort. 463:113-119.

SECTION 4. EFFECT OF GIBBERELLIN SPRAYS ON FLOWER FORMATION IN LATERAL BUDS OF 'GALA' APPLE.

Abstract

The objective of this experiment was to determine if treatment of bearing 'Gala' apple trees with gibberellins could prevent flowering in lateral buds without affecting flowering in terminal buds. A mixture of gibberellins A_4 and A_7 ($GA_{4/7}$, ratio not stated) in the commercial formulation ProVide® was sprayed on trees at 150, 300 and 600 mg•L^{-1} on each of three dates (3 July, 19 July and 5 August 1996) in a factorial arrangement. On 3 July two unformulated mixtures of $GA_{4/7}$ with differing ratios (64/27 and 48/42 percent) were also applied at 150 mg•L^{-1}. Non-treated controls were not sprayed. Response was measured by subjectively rating bloom on the entire tree, and by counting flower buds on 10 shoots on each tree, distinguishing between terminal and lateral flower buds. There was no significant difference ($P = 0.05$) in response to treatments applied on 3 July 1997, when compared alone, nor was interaction between time of application and concentration significant. However, the effects of time of application on subjective bloom rating, and on percentages of terminal and lateral buds flowering, were significant. The early application (3 July 1996) reduced bloom compared to later applications. Values for the latter two dates were similar to those of the non-treated controls. Interpretation of the data was complicated by variability in crop density among the trees selected for treatment. Using crop density as a covariant did not affect the analysis of variance for flowering parameters, so unadjusted means are reported.

Introduction

Bobb and Blake (1938) pointed out that apple can produce many more flowers on spurs alone than are required to produce a full crop, and thatlateral flowers just add to the excess. They also reported the inhibition of spur leaf area caused by this overabundance of flowers. 'Wealty', 'Wagner', 'Oldenburg' and 'Jonathan' often produce enough fruit from lateral flowers to be considered annual bearing (Auchter, 1919), but fruits developing from these flowers are smaller than those from terminal flower buds (Volz et al., 1994). 'Gala' is a new commercial cultivar in many regions of the United States, and size is often smaller than optimum. It produces many lateral flower buds on current season's growth, thus reducing the average size. Fruits from lateral flowers also mature later, requiring multiple harvests or resulting in large variations in maturity of fruit going into storage. Zeller (1960) reported that lateral flowers on terminal shoots are initiated later than are spur buds. Gibberellin sprays inhibit flower formation in apple (Dennis and Edgerton, 1966; Guttridge, 1962). Greenhalgh and Edgerton (1967) found that potassium gibberellate (KGA_3) treatments of 100, 200 and 400 $mg•L^{-1}$ applied 2 days after full bloom (DAFB) inhibited flower induction of 'McIntosh' apple. By 25 DAFB the same concentrations were much less inhibitory. Marino and Greene (1981) found that GA_3 at 30 $mg•L^{-1}$ selectively reduced flowering of lateral buds when applied 10 DAFB to mature 'Early McIntosh' trees, while GA_{4+7} at the same rate was ineffective. At 300 $mg•L^{-1}$ both treatments reduced flowering of both spurs and laterals and a second identical treatment 20 DAFB did not significantly increase the effect. McArtney (1994) reduced the severity of the alternate bearing cycle of 'Braeburn' apple by applying GA_3 and GA_{4+7} at full bloom. Although there was a linear response to concentration, even the highest rate of GA_3 (330 ppm) did not eliminate alternate bearing. None of the treatments influenced flower bud formation on one-year wood, which accounted for a high proportion of the total flower clusters. When applied 6, 9 or 12 weeks after full bloom, GA_3 or GA_7 inhibited flowering on one-year wood but not on spurs of vigorously growing 'Braeburn' shoots that had been top-worked on 'Royal Gala'/MM. 106 apple trees (McArtney and Li, 1998). GA_7 was more effective than GA_3 and response increased as treatment was delayed. My purpose was to determine the optimum concentration and timing for application of gibberellin sprays for inhibiting flowering in lateral buds of 'Gala' apple.

Materials and Methods

'Gala' apple trees propagated on 'Mark' rootstock and planted in 1991 at the Clarksville Horticulture Experiment Station, Clarksville, Mich., were used in 1996. Prior to treatment trees were visually rated for crop load and blocked accordingly. On 3 July, 19 July and 5 August 1996 (40, 56 and 73 DAFB, respectively) whole trees were sprayed with 150, 300 or 600 $mg \cdot L^{-1}$ GA_{4+7}, formulated as 'ProVide®' (courtesy of Abbott Laboratories, North Chicago, Ill.). In addition, two mixtures of GA_{4+7} (64/27 and 48/42 percent GA_4 and GA_7, also from Abbott Laboratories) in powder form were applied on the earliest date for comparison with the commercial product. All sprays were applied with a hand gun to drip. Nontreated trees were included as controls. At harvest, fruit number, yield and trunk diameter were recorded for each tree to better control variation due to crop density during data analysis. In spring 1997, 10 current-season shoots with at least 10 nodes (approximately 20 to 30 cm long) were selected on each tree and the number of lateral flowers were counted. In addition, four persons visually rated each entire tree for bloom density on a scale from 0 to 10, and the four scores were averaged. All treatments applied on 3 July 1996 and the nontreated control were compared statistically to determine the effects of GA concentration, formulation and ratio. A second factorial analysis was performed on the data for three concentrations of $GA_{4/7}$ repeated over three dates to determine the effects of both timing and concentration. Further, bloom rating and number of lateral flowers were correlated with crop density to determine how closely these parameters were related across treatments. All proportional values were transformed ($\sqrt{y + 0.5}$) to ensure near normal distribution of values for analysis of variance.

Results

Though most of the trees in this experiment carried moderate to heavy crops, return bloom was still adequate for a full crop the following year (Table 1). When considered by themselves, none of the treatments applied 3 July inhibited flowering significantly at $P \leq 0.05$, as evaluated by subjective rating of the whole trees or by sampling shoots (Table 1). Although mean values for all treatments except one were less than the control value, variability was also great. Considering the time by concentration factorial treatments, bloom rating and lateral flowers per shoot increased as time of application was delayed (Tables 2 and 3), but no interaction between

Table 1. Effect of $GA_{4/7}$ sprays applied 3 July 1996 to trees with varying crop density on bloom rating and percentage of lateral buds flowering in 1997 in 'Gala' apple at Clarksville Horticulture Experiment Station, Clarksville, Mich. All values are means for 5 trees.

Treatment	Concn. ($mg \bullet L^{-1}$ a.i.)	Crop density (Fruit no./ cm^2 TCSA)	Bloom rating[z]	Percentage lateral buds flowering[y]
Nontreated		8.61	5.20	8.3
ProVide	150	10.40	4.00	4.2
	300	8.99	3.50	3.2
	600	9.35	3.73	3.1
$GA_{4/7}$ (64/27 %)	150	8.04	4.65	0.9
$GA_{4/7}$ (48/42 %)	150	8.20	5.30	2.4
P[x]		0.156	0.166	0.488

[z] Mean of four evaluators' visual bloom rating (0-10) at full bloom. Transformation ($\sqrt{y + 0.5}$) performed before analysis for bloom rating.

[y] Based on node counts of 10 shoots per tree. Percentages transformed (arcsin square-root) before analysis.

[x] Probability (*P*) of differences being due to random variability based on analysis of variance.

Table 2. Effect of timing and concentration of ProVide® treatments in 1996 on crop density and bloom rating in 1997 in 'Gala' apple at Clarksville Horticulture Experiment Station, Clarksville, Michigan. Five trees per treatment.

	Crop density (Fruit no./cm^2 TCSA)				Bloom rating[z]			
	ProVide concn.(mg•L^{-1})				ProVide concn.(mg•L^{-1})			
Timing	150	300	600	Mean	150	300	600	Mean
3 Jul 96	10.4	9.0	9.4	9.6	4.0	3.5	3.7	3.7
19 Jul 96	6.6	11.0	9.2	8.9	5.5	5.0	5.6	5.4
5 Aug 96	7.0	9.9	8.5	8.5	6.3	4.8	6.0	5.7
P[y]								0.014
Mean	8.0	10.0	9.0	0.0005[x]	5.3	4.4	5.1	

[z] Mean of four evaluators' visual bloom rating (0-10) at full bloom. Transformation ($\sqrt{y + 0.5}$) performed before analysis.

[y] Probability (*P*) of differences being due to random variability based on analysis of variance. *P* values near 1 not shown.

[x] Significance of interaction of timing x concentration.

Table 3. Effect of timing and concentration of ProVide® treatments in 1996 on flowering of terminal and of lateral buds in 1997 in 'Gala' apple at Clarksville Horticulture Experiment Station, Clarksville, Michigan. Analysis of variance based on transformed (arcsin square-root) data.

	Terminal buds flowering (%)[z]				Lateral buds flowering (%)[y]			
	Provide concn.(mg•L^{-1})				Provide concn.(mg•L^{-1})			
Timing	150	300	600	Mean	150	300	600	Mean
3 Jul 96	28	20	18	22.0	4.3	3.2	3.1	3.5
19 Jul 96	60	54	50	54.7	13.1	5.0	13.1	10.4
5 Aug 96	66	56	52	58.0	14.2	12.6	14.1	13.6
P^x				0.003				0.042
Mean	51.3	43.3	40.0		10.5	6.9	10.1	

[z] Based on 10 selected shoots per tree.

[y] Based on total node no. of 10 selected shoots per tree.

[x] Probability (*P*) of differences being due to random variability based on analysis of variance. *P* values near 1 not shown.

concentration and time of treatment was detected (Figures 1 and 2). As crop density increased, bloom rating declined significantly ($P < 0.0001$) (Figure 3). Crop density also appeared to have a stronger effect on lateral flowering than did gibberellin treatments. Many trees with more than eight fruits per cm^2 TCSA produced few or no lateral flowers (Figure 4). Within the range of 5 - 13 fruits per cm2 TCSA, percentage of lateral buds that were floral varied from 0 to 45.

Discussion

A specific objective of this experiment was to determine the effect of GA sprays on lateral flower induction in bearing 'Gala' apple trees. When young dwarf trees bear widely varying crops, controlling alternate bearing with chemical thinning can be very difficult. If the undesirable lateral flowers could be prevented from developing without affecting flowering of spurs, more uniform flowering and cropping could be achieved.

Lateral flowering is especially abundant in New Zealand, where photosynthetic activity and development can continue for some time after harvest (R. Giuliani, personal communication). McArtney and Li (1998) applied GA to 'Braeburn' apple topworked on 'Royal Gala'/MM 106. The trees were extremely biennial and all were totally vegetative in the year of treatment, resulting in control trees having 83 percent of the lateral buds flowering the following year. Treatment with surfactant alone (Regulaid) reduced lateral flowering to 70 percent, but treatment with GA_3 or GA_7 (100 $mg \cdot L^{-1}$) reduced this to 59 percent. Flowering decreased to 38 percent as the concentration was raised to 400 $mg \cdot L^{-1}$. In contrast, I found that flowering of lateral buds of 'Gala' varied

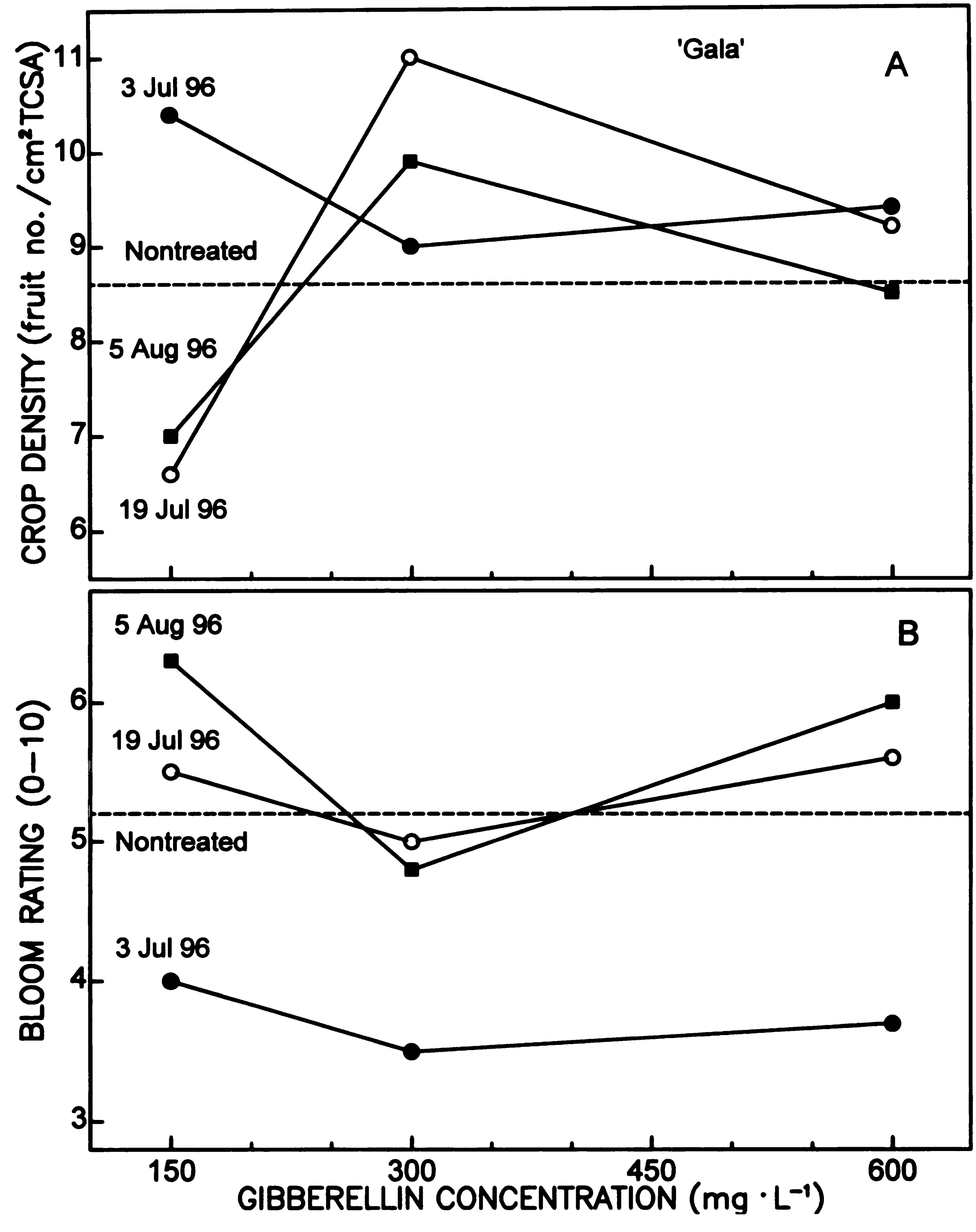

Figure 1. Crop density on 'Gala' apple trees used (A) and effects of concentration and time of application of gibberellin in the form of ProVide® in 1996 on bloom rating in 1997 (B). Each point is the mean for five replicate trees.

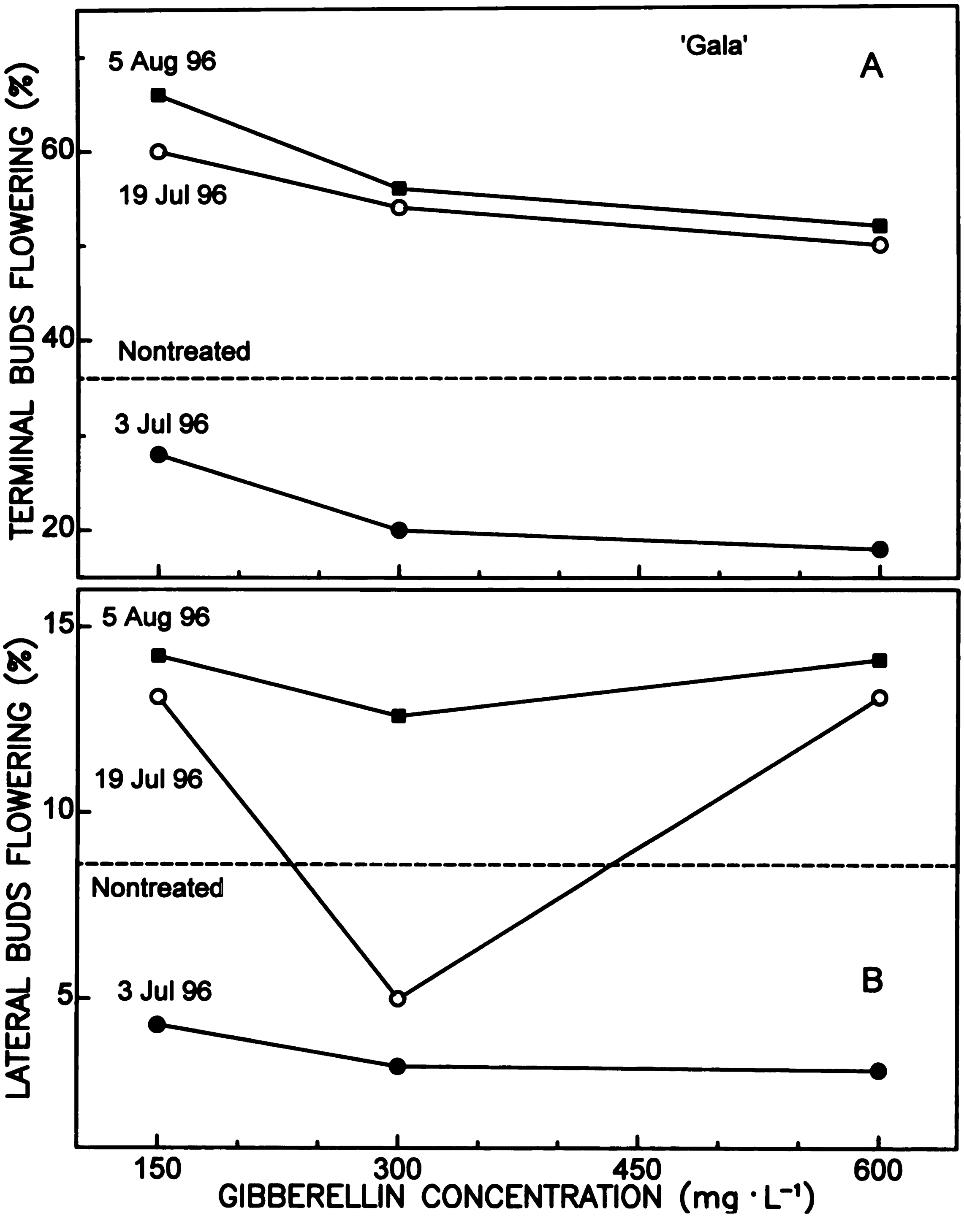

Figure 2. Effects of concentration and time of application of gibberellin in the form of ProVide® on percentage terminal (A) and lateral (B) buds flowering the following year on 'Gala' apple. Each point is the mean for five replicate trees.

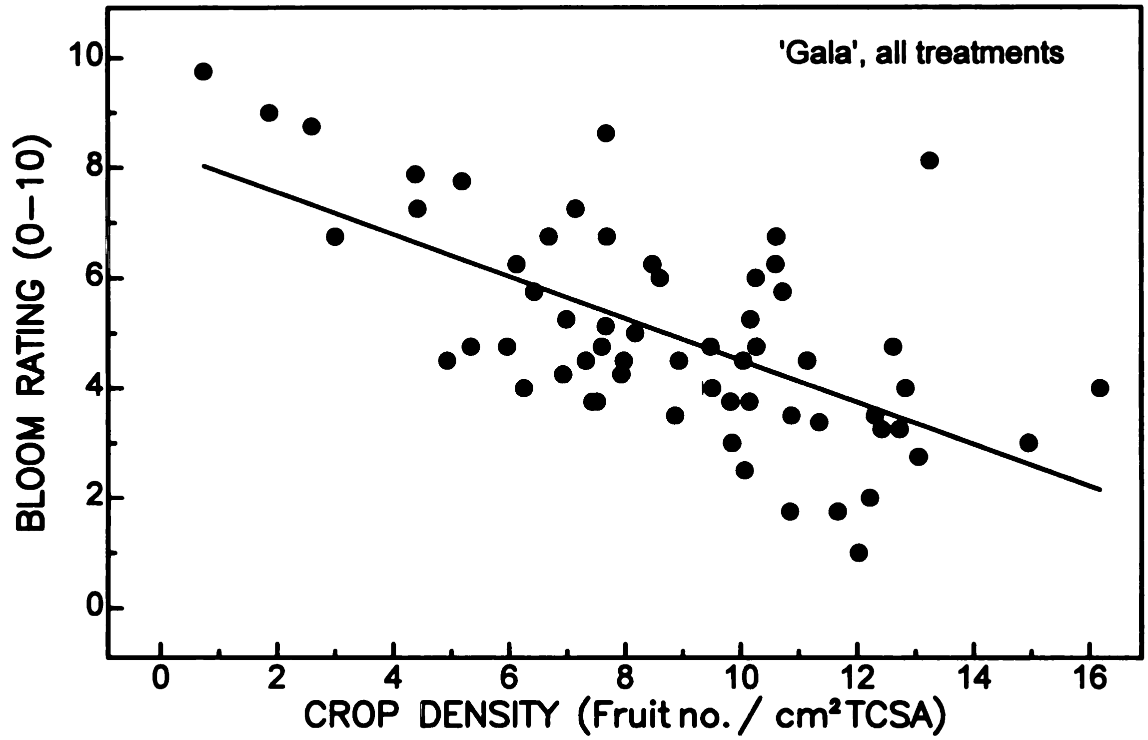

Figure 3. Relationship ($y = 8.32 - 0.38x$, $R^2 = 0.38$, $P < 0.0001$) between return bloom rating in 1997 and crop density in 1996 in 'Gala' apple over three treatment dates and three concentrations of ProVide® at Clarksville Horticulture Experiment Station.

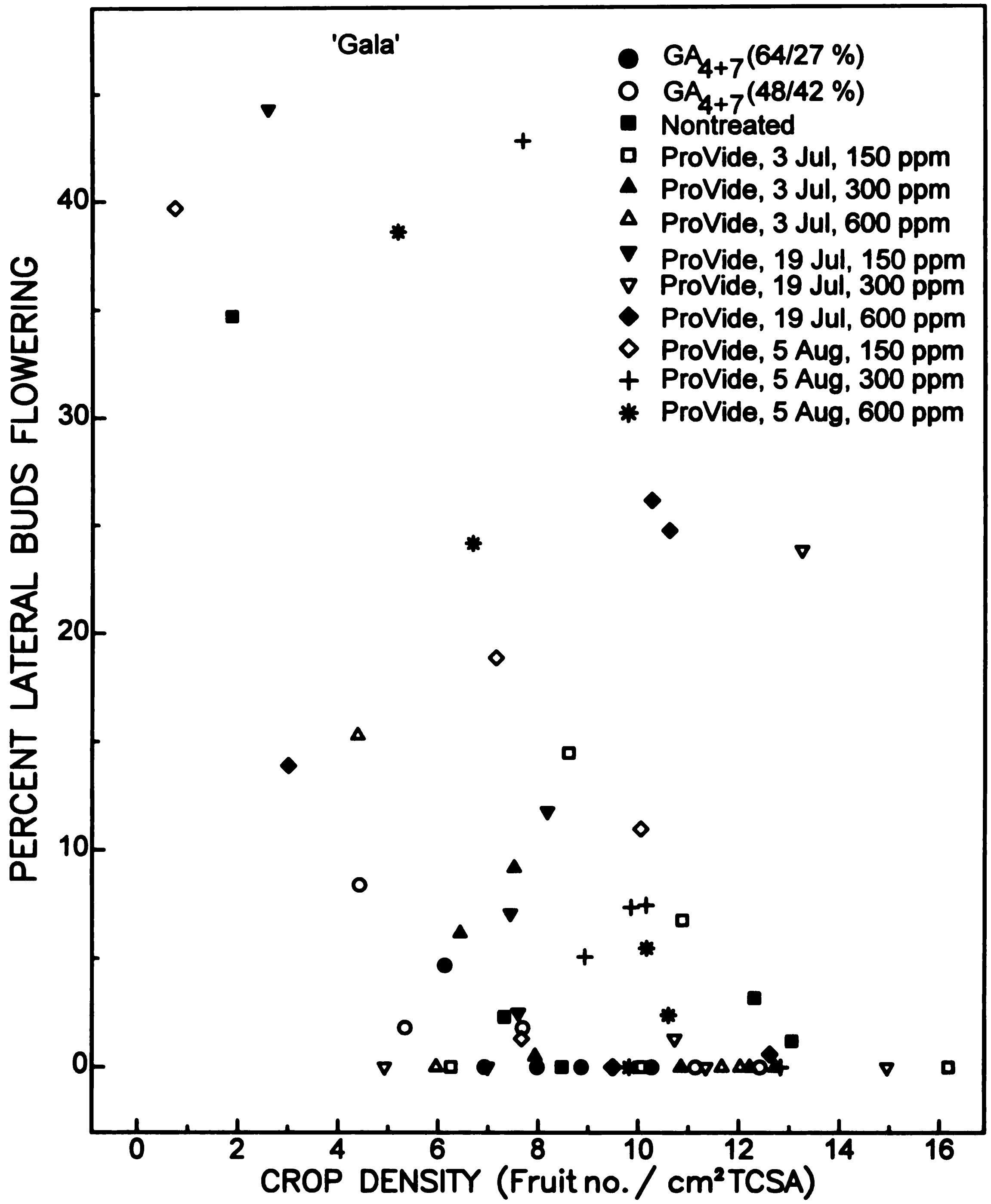

Figure 4. Relationship between crop density in 1996 in 'Gala' apple and percentage of lateral nodes flowering in 1997 over three treatment dates for ProVide® concentrations from 150 to 600 $mg \bullet L^{-1}$ at Clarksville Horticulture Experiment Station.

from 0 to 44.3 percent; of the 60 trees used, 24 formed no lateral flower buds. Although the range in percentage flowering was approximately the same in both experiments, the absolute values in ‘Gala’ were much lower than in ‘Braeburn’.

GA did not inhibit flowering of spurs in ‘Braeburn’ (McArtney and Li, 1998), presumably because the earliest treatment (42 DAFB) was applied after flower induction had occurred. A linear reduction in flowering of lateral buds occurred as time of GA application was delayed; maximum sensitivity could have occurred even later than 84 DAFB – their latest treatment. In contrast, although application times were similar, my earliest GA treatments were applied too late to establish the time of maximum sensitivity. ‘Gala’ is an early-maturing cultivar, so these trees had considerable time for bud development between harvest and leaf fall, but terminal growth had ceased prior to harvest. If early crop removal alone favored flower induction, summer apple varieties as a group would be expected to flower annually, yet they are notoriously alternate-bearing in the United States. Other important differences may be the effects of climate and cultivar on cessation of growth. The moderate temperatures in New Zealand allow shoot growth to occur later than in continental climates, such as that of Michigan (McArtney and Li, 1988). McArtney and Li (1998) used top-worked trees with a few vigorous branches developing per trunk, and growth may have been more uniform. This is supported by the fact that differences of 10 percent were highly significant in their study, but nonsignificant in mine.

Marino and Greene (1981) also obtained selective inhibition of flowering in lateral buds by applying GA_3 ($30\ mg \cdot L^{-1}$) at 10 DAFB to mature ‘Early McIntosh’ trees in Massachusetts; GA4+7 was ineffective at this concentration. At 300 mg·L^{-1} both GAs reduced flowering of both spurs and laterals; a second identical treatment 20 DAFB had no additional effect. This is much earlier than my treatments (40-70 DAFB), but differences in response could also reflect differences in growth habit among cultivars. Although the effects on 'Early McIntosh' were statistically significant, they were small, in terms of the potential for lateral flower buds to develop, based on node number. Marino and Greene (1981) expressed the results as number of blossom clusters per meter of shoot length, rather than as percentage of total buds, and 300 mg·L^{-1} reduced lateral flowering from 17.0 (control) to 6.7 buds per meter. If expressed similarly, the data for the 60 'Gala' trees I used varied from 0 to 33.3 lateral clusters per meter of one-year-old wood, but this difference was related more to crop density than to GA treatment. The shoots selected (average length 245 mm) had an average of 61 nodes per meter.

In contrast with the trees used by McArtney and Li (1998) and Marino and Greene (1981), which bore no crop the year of treatment, these 'Gala' trees all were cropping. Even though the cropload varied widely, flowering generally, including lateral flowering, was unexpectedly abundant, and cropload influenced flowering more than did the applied gibberellins. This was particularly true for some trees with heavy crops, where the combination of cropload and GA treatment was expected to be very inhibitory to flowering.

My selection of GA_{4+7} was primarily because ProVide® is already registered for use on apple in early summer as a means of reducing fruit russeting and cracking. No differences were found in response to the two samples of pure chemical with differing ratios of GA_4 and GA_7 vs. the ProVide® formulation of GA_{4+7} (unknown ratio). GA_4 is reported to be less inhibitory to flowering than is GA_7 (Looney et al., 1985; Tromp, 1982; Wertheim, 1982). Greene (1993) applied GA_4 or GA_{4+7}, each at 10-40 $mg \cdot L^{-1}$, to 'Golden Delicious' at weekly intervals for 3-4 weeks beginning at petal fall. GA_4 promoted flowering in 2 years and inhbited it in one, whereas GA_{4+7} inhibited flowering in 3 out of 4 years, although response varied with year and concentration.

Li et al. (1995) found that GA_3 treatment could be used to determine the time of flower induction. They also reported that flower induction in 'Red Fuji' occurred earlier in spurs than in terminal buds of new shoots. The different types of buds in 'Ralls Janet' were more uniform within a given year, but the period of high sensitivity to GA_3 was shorter (10 days) in an off-year than in an on-year (40 days), indicating an interaction between sensitivity to GA and cropload. This was not the case in my work with 'Gala', probably because the treatments were applied too late. Marino and Greene (1981) confirmed earlier reports that flower induction in spurs begins much later in the "off-year" (42 DAFB) than in the "on-year" (0 DAFB). Assuming that induction in trees with moderate crops occurs between these two extremes, the time of induction in both spurs and lateral buds of the 'Gala' trees used in this study could vary depending on crop load and type of bud (spur, shoot terminal and shoot lateral).

A better understanding of the timing of induction is needed to be able to apply treatments to prevent flowering in lateral buds. Denker and Hansen (1994) found that initiation in lateral buds does not occur until the shoots stop elongating. Terminal flower induction also appears to be related to time of cessation of growth, but greater specificity would allow better timing of treatments. Two critical questions remain to be answered: how far in advance of growth cessation does flower induction occur, and does induction occur simultaneously in terminal and lateral buds or does it progress basipetally, starting with the apex? A related question is how long is GA effective in inhibiting induction?

In this study, only those shoots that formed terminal flowers initiated lateral flower buds, with very few exceptions, and abortion of terminal flowers might account for these. Zeller (1960), working in Germany, reported that lateral flowers can be initiated very late in the growing season and even the following spring. Repeated application of gibberellins at low concentrations may be necessary to prevent such late induction. This would be similar to the protocol used for russet control in 'Golden Delicious' (Greene, 1993), except that treatment would begin after spur buds have been initiated, or about 40 DAFB for light-cropping trees.

Most of the shoots sampled in my work were longer than 20 cm. To better determine the timing of initiation in terminal buds of spurs and in terminal and lateral buds of shoots, samples of each should be collected for dissection. The fruiting status of the spurs and the cropload for the tree should be recorded, as a clearer understanding of flower induction can be obtained only if all interacting factors are known.

Literature Cited

Auchter, E.C. 1919. Some influences of thinning, pollination, and fruit spur growth on the yearly performance record of fruit spurs and on the size of fruit produced.

Proc. Amer. Soc. Hort. Sci. 16:118-131.

Bobb, A.C. and M.A. Blake. 1938. Annual bearing in the 'Wealthy' apple was induced by blossom thinning. Proc. Amer. Soc. Hort. Sci. 36:321-327.

Denker, I. and P. Hansen. 1994. Flower initiation and node development of axillary buds, growth partitioning, and bleeding sap production of young apple trees as affected by fertigation. J. Hort. Sci. 69:869-876.

Dennis, Jr., F.G. and L.J. Edgerton. 1966. Effect of gibberellins and ringing upon apple fruit development and flower bud formation. Proc. Amer. Soc. Hort. Sci. 88:14-24.

Greene, D.W. 1993. Effects of GA_4 and GA_7 on flower bud formation and russet development on apple. J. Hort. Sci. 68:171-176.

Greenhalgh, W.J. and L.J. Edgerton. 1967. Interaction of Alar and gibberellin on growth and flowering of the apple. Proc. Amer. Soc. Hort. Sci. 91:9-17.

Guttridge, C.G.1962. Inhibition of fruit-bud formation in apple with gibberellic acid. Nature 196:1008.

Li, S.H., Z.Q. Meng, T.H. Li, H.Z. Liu and Y.C. Tu. 1995. Critical period of flower bud induction in 'Red Fuji' and 'Ralls Janet' apple trees. Gartenbauwissenschaft 60:240-245.

Looney, N.E., R.L. Granger, C.L. Chu, S.J. McArtney, L.N. Mander and R.P. Pharis. 1992. Influences of gibberellins A_4, A_{4+7}, and A_4+iso-A_7 on apple fruit-quality and tree productivity. I. Effects on fruit russet and tree yield components. J. Hort. Sci. 67:613-618.

Marino, F. and D.W. Greene. 1981. Involvement of gibberellins in the biennial bearing of 'Early McIntosh' apples.

McArtney, S.J. 1994. Exogenous gibberellin affects biennial bearing and the fruit shape of 'Braeburn' apple. New Zealand J. Crop and Hort. Sci. 22:343-346.

McArtney, S.J. and S.H. Li, 1998. Selective inhibition of flowering on 'Braeburn' apple trees with gibberellins. Hortscience (in press).

Tromp, J. 1982. Flower-bud formation in apple as affected by various gibberellins. J. Hort. Sci. 57:277-282.

Volz, R.K., I.B. Ferguson, E.W. Hewett and D.J.Wooley. 1994. Wood age and leaf area influence fruit size and mineral composition of apple fruit. J. Hort. Sci. 69:385-395.

Wertheim, S.J. 1982. Fruit russeting in apple as affected by various gibberellins. J. Hort. Sci. 57:283-288.

Zeller, O. 1960. Entwicklungsgeschichte der Blütenknospen und Fruchtanlangen an einjährigen Langtrieben von Apfelbüschen I. Entwicklungsverlauf und entwicklungsmorphologie der Blüten am einjährigen Langtrieb. Z. für Pflanzenzüchtung 44:175-214.

SUMMARY

Seedless fruits of 'Spencer Seedless' did not inhibit flowering regardless of total fruit mass per spur or shoot length. Flowering was not inhibited unless the number of seeds exceeded five. This contrasts with the data of Chan and Cain (1967), who found that one to five seeds were sufficient to inhibit flowering 65 to 85 %. When bourse shoot length was less than 2 or greater than 10 mm, seeds had no effect; none of the very short shoots formed flowers, whereas almost all of the long ones did. This latter observation agrees with the data of Huet (1972, 1973) for 'Bartlett' pear, which indicated that the inhibitory effects of seeds could be overcome provided leaf surface per spur were sufficient. My attempts to reduce seed number per fruit in commercial cultivars were successful in reducing seed number and appeared to increase flowering, but the effects were too small to be significant. Further experiments using this technique may help elucidate the interaction between seeds and cropload.

For 'Spencer Seedless' spurs of intermediate length, seeds played a dominant role in controlling flowering. This contrasts with the results I obtained with 'Paulared', for most shoots were much longer than 10 mm, yet a heavy crop severely inhibited flowering if left until harvest. Crop density appeared to affect flowering both in 'Spencer Seedless' in the one year (1994) that it was recorded, and in 'Paulared', in which bourse shoots of spurs bearing up to five seeded fruits flowered when crops were light. When cropload was moderate to heavy in 'Paulared', the number of fruit (and seeds) per fruiting terminal had little effect on flower induction in the subtending bourse bud. Responses of individual trees, however, varied widely, some being sensitive to fruit number per spur and others either flowering or failing to flower regardless of fruit number borne on the terminal. This suggests that in 'Paulared', at least, the aggregate number of seeds per branch or tree is more important than the presence or absence of seeds *per se*.

The high return bloom in 1997 on 'Paulared', 'Golden Delicious', 'Jonagold', and 'Gala' trees bearing good crops in 1996 suggests that the effect of cropload can be modified by environmental factors. This may explain why some cultivars are annual in some locations and biennial in others. The profound effects of extreme alternate bearing on shoot growth, spur leaf area, and time of flower induction indicate that heavy bloom and cropping have a number of important effects. The effect on flowering may be direct, via GAs emanating from the seeds (Luckwill, 1970; Marino and Greene, 1981), or indirect, either by reducing spur leaf area and therefore the supply of "florigen", or by excessive use of carbohydrate reserves by fruits. However, Stutte and Martin (1986b) demonstrated that reduced carbohydrate content was not responsible for the inhibitory effect of fruiting on flowering of olive. This should be confirmed in apple. Hoad (1980) and Hoad and Ramirez (1980) reported the movement of ^3H to buds following injection of ^3H-GA_4 into seeds. Somewhat more radioactivity was recovered from buds of 'Laxton' (biennial) than from those of 'Cox' (annual); however, the values were small and the difference nonsignificant. These studies suggest that if endogenous GAs are responsible for alternate bearing, the mechanism is likely to be considerably more complex than has been assumed heretofore. My attempt to substitute GAs for seeds resulted in no detectable inhibition of flowering. In further trials, application directly to the buds would be an important control to verify that GAs are indeed inhibitory at the time of treatment, and thus separate the question of timing from that of transport. Defruited and fruiting controls are also needed for comparison of gibberellin treatment vs. fruit effect. The time required for seed removal from individual fruits limits the number of spurs that can be treated. Seedless fruits can simply be injected, but precise placement of the injected GAs is difficult, and their uptake, metabolism and transport is not likely to mimic that of seed-produced GAs.

When GAs were sprayed on whole 'Gala' trees, the earliest treatment (3 July) appeared to inhibit lateral flowering of both entire trees and lateral buds, but the effect was not statistically significant when compared with nontreated controls. Later sprays were without effect, regardless of concentration. Flowering was more closely associated with crop density than with GA treatment. In contrast with the trees used by McArtney and Li (1998) and Marino and Greene (1981), cropload varied considerably in the 'Gala' trees. Li et al. (1996) reported that bearing trees of 'Ralls Janet' were more sensitive to GA_3 application, and at an earlier date than were non-bearing trees. Marino and Greene (1981) clearly demonstrated that flower induction occurred earlier (0 DAFB) in "on-year" than in "off-year" trees (42 DAFB) of 'Early McIntosh'. In my study flowering was unexpectedly abundant on several heavily cropping 'Gala' trees that were sprayed with GA. The best explanation for this discrepancy is that the treatments were applied too late to affect flowering. Flowering of spurs was not recorded. In subsequent studies, flowering of spur buds, in addition to that of terminal and lateral buds on shoots, should be recorded. Also, repeated treatment with GA may be necessary to prevent lateral flower induction, which occurs over a long time period (Zeller, 1960). Though the role of endogenous GAs remains to be determined, GA application is a valuable tool to both investigate the mechanisms of flower induction and to control flower induction commerically.

Greenhalgh and Edgerton (1967) measured or derived 123 variables that could affect flowering of apple, including treatment with daminozide (Alar) and GA. Nine of the variables were significantly associated ($P = 0.05$) with return bloom, and accounted for 78 % of the total variance. The variable with the largest positive coefficient was log mean fruit mass, for which the authors had no physiological explanation; this was probably an indicator of reduced fruit set following daminozide application. The variable with the largest negative coefficient was log GA concentration, confirming other reports (Dennis and Edgerton, 1966; Guttridge, 1962; Marcelle and Sironval, 1963). Less important but still significant positive associations were found with both insoluble nitrogen content of spur leaves in June and time from full bloom until cessation of shoot growth. Although flowering was not associated with shoot length *per se*, it increased with duration of shoot growth. This parallels, in some respects, at least, my observations with regard to flowering in spurs bearing seeded fruits of 'Spencer Seedless'. They also reported that although GA treatment increased both the duration of shoot growth and shoot length, yet it inhibited flowering, indicating that GAs may have several effects, some positive and some negative, with respect to flower induction.

A comprehensive model to describe control of flowering in apple is still needed, including a more detailed study of the interaction of cropload, shoot and bud development, and fruit (seed) number per spur in several commercial cultivars. Care should be taken to allow for ample replication and to consider previous cropping history in order to eliminate as much variability as possible. Gibberellin and other hormones, both applied or endogenous, should be researched to determine whether they influence flower induction directly or indirectly.

LIST OF REFERENCES

LIST OF REFERENCES

Abbott, D.L. 1959. The effects of seed removal on the growth of apple fruitlets. Annu. Rept. Long Ashton Agr. Hort. Res. Sta. 1958. p 52-56.

Abbott, D.L. 1960. The bourse shoot as a factor in the growth of apple fruits. Ann. App. Biol. 48:434-438.

Abbott, D.L. 1970. The role of budscales in the morphogenesis and dormancy of the apple fruit bud, p. 65-82. In: L.C. Luckwill and C.V. Cutting (eds.). Physiology of Tree Crops. Academic Press, New York.

Acquaah, G., J.W. Saunders and L.C.Ewert. 1992. Homeotic floral mutations. Plant Breeding Rev. 9:63-99.

Auchter, E.C. 1919. Some influences of thinning, pollination, and fruit spur growth on the yearly performance record of fruit spurs and on the size of fruit produced. Proc. Amer. Soc. Hort. Sci. 16:118-131.

Auchter, E.C. and A.L. Schrader. 1923. Fruit spur growth and fruit bud production. Proc. Amer. Soc. Hort. Sci. 20:127-144.

Auchter, E.C., A.L. Schrader, F.S. Lagasse and W.W. Aldrich. 1926. The effect of shade on the growth, fruit bud formation and chemical composition of apple trees. Proc. Amer. Soc. Hort. Sci. 23:368-382.

Ban, J. 1996. Gibberelin metabolism and apple flowering. PhD Dissertation, Michigan State Univ.

Bangerth, F.K. 1997. Can regulatory mechanism in fruit growth and development be elucidated through the study of endogenous hormone concentrations? Acta Hort. 463:77-87.

Bangerth, F. and M. Schroder. 1994. Strong synergistic effects of gibberellins with the synthetic cytokinin N-(2-chloro-4-pyridyl)-N-phenylurea on parthenocarpic fruit-set and some other fruit characteristics of apple. Plant Growth Regul. 15:293-302.

Bigelow, W.D. and H.C. Gore. 1905. U.S. Dept. Agr. Bur. Chem. Bul. 97. (Cited by Gourley, 1922.)

Bobb, A.C. and M.A. Blake. 1938. Annual bearing in the 'Wealthy' apple was induced by blossom thinning. Proc. Amer. Soc. Hort. Sci. 36:321-327.

Brase, K.D. 1937. The vascular anatomy of the flower of Malus domestica Borkh. f. apetala van Eseltine. M.S. Thesis, Cornell University.

Buban, T. 1981. Histological and cytological aspects of flower bud formation in apple. Acta Hort. 120:113-117.

Buban, T. 1996. Flower development and formation of sexual organs. p. 3-54. In: J. Nyéki and M. Soltész (eds.). Floral biology of temperate zone fruit trees and small fruits. Akadéniai Kiadó, Budapest.

Chailakhyan, M.Kh. 1937. Hormone theory of plant development (Russian.) Moscow, Leningrad. Akad. Nauk. SSR.

Chan, B.G. and J.C. Cain. 1967. The effect of seed formation on subsequent flowering. Proc. Amer. Soc. Hort. Sci. 91:63-68.

Coen, E.S., 1991. The role of homeotic genes in flower development and evolution. Annu. Rev. Plant Physiol. Plant Mol. Biol. 42:241-279.

Crow, J.W. 1920. Biennial fruit bearing in the apple. Proc. Amer. Soc. Hort. Sci. 17:52-54.

Davis, L.D. 1957. Flowering and alternate bearing. Proc. Amer. Soc. Hort. Sci. 70:545-556.

Denker, I. and P. Hansen. 1994. Flower initiation and node development of axillary buds, growth partitioning, and bleeding sap production of young apple trees as affected by fertigation. J. Hort. Sci. 69:869-876.

Dennis, F.G. Jr. 1976. Gibberellin-like substances in apple seeds and fruit flesh. J. Amer. Soc. Hort. Sci. 101:629-633.

Dennis, F.G., Jr. and L.J. Edgerton. 1966. Effects of gibberellins and ringing upon apple fruit development and flower bud formation. Proc. Amer. Soc. Hort. Sci. 88:14-24.

Dennis, F.G., Jr. and J.P. Nitsch. 1966. Identification of gibberellins A_4 and A_7 in immature apple seeds. Nature 211:781-782.

Edwards, G.R. 1987. Producing temperate-zone fruit at low latitudes: Avoiding rest and the chilling requirement. Hortsci. 22:1236-1240.

Goff, E.S. 1899. The origin and early development of the flower in the cherry, plum, apple, and pear. Wisconsin Agr. Expt. Sta 16th Annu. Rept. p. 289-303. (Cited by Davis, 1957.)

Gorter, C.J., 1955. Photoperiodism of flowering in apple trees. Rep. XIVth Intern. Hort. Cong., p 351-354.

Gourley, J.H. 1920. The effect of shading some horticultural plants. Proc. Amer. Soc. Hort. Sci. 17:256-260.

Gourley, J.H. 1922. Text book of pomology. p. 115. Macmillan Co., New York.

Green, J.R. 1987. The hormonal control of biennial bearing in cider apples. PhD Dissertation, Univ. of Bristol.

Greene, D.W. 1993. Effects of GA_4 and GA_7 on flower bud formation and russet development on apple. J. Hort. Sci. 68:171-176.

Greenhalgh, W.J. and L.J. Edgerton. 1967. Interaction of Alar and gibberellin on growth and flowering of the apple. Proc. Amer. Soc. Hort. Sci. 91:9-17.

Griggs, W.H., G.C. Martin, and B.T. Iwakiri. 1970. The effect of seedless versus seeded fruit development on flower bud formation in pear. J. Amer. Soc. Hort. Sci. 95:243-248.

Grochowska, M.J. 1968. The influence of growth regulators inserted into apple fruitlets on flower bud initiation. Bul. Acad. Polon. Sci., Ser. Biol. 16:581-584.

Grochowska, M.J. 1968. Translocation of indole-3-acetic acid-2-^{14}C injected into seeds of five-week-old apple fruits. Bul. Acad. Polon. Sci., Ser. Biol. 16:577-580.

Guttridge, C.G.1962. Inhibition of fruit-bud formation in apple with gibberellic acid. Nature 196:1008.

Harley, C.P., J.R. Magness, M.P. Masure, L.A. Fletcher and E.S. Degman. 1942. Investigations on the cause and control of biennial bearing in apple trees. U.S.D.A. Tech. Bul. 792.

Harley, C.P., H.H. Moon and L.O. Regeimbal. 1958. Evidence that post-bloom apple-thinning sprays of naphthaleneacetic acid increase blossom-bud formation. Proc. Amer. Soc. Hort. Sci. 72:52-56.

Hedden, P., G.V. Hoad, P. Gaskin, M.J. Lewis, J.R. Green, M. Furber and L.N. Mander. 1993. Kaurenoids and gibberellins, including the newly characterized gibberellin A_{88}, in developing apple seeds. Phytochemistry 32:231-237.

Heinicke, A.J. 1917. Factors influencing the abscission of flowers and partially developed fruits of the apple (*Pyrus malus* L.). Cornell Univ. Agr. Expt. Sta. Bul. 393.

Hipps, N.A. 1992. Fertigation of newly planted 'Queen Cox'/M9 apple trees-establishment, early growth and precocity of cropping. J. Hort. Sci. 67:25-32.

Hirst, P.M. and D.C. Ferree. 1995a. Rootstock effects on shoot morphology and spur quality of 'Delicious' apple and relationships with precocity and productivity. J. Amer. Soc. Hort. Sci. 120:622-634.

Hirst, P.M. and D.C. Ferree. 1995b. Rootstock effects on the flowering of 'Delicious' apple. I. Bud development. J. Amer. Soc. Hort. Sci. 120:1010-1017.

Hirst, P.M. and D.C. Ferree. 1996. Effects of rootstock on bud development and flower formation of 'Starkspur Supreme Delicious' apple. Fruit Var. J. 50:25-34.

Hoad, G.V. 1980. The role of seed derived hormones in the control of flowering in apple. Acta Hort. 80:93-103.

Hoad, G.V. and H. Ramirez. 1980. La función de las giberelinas sintetizadas en las semillas del fruto para el control de la floración en manzanos. Turrialba 30:284-288.

Huet, J. 1972. Étude des effêts des feuilles et des fruits sur l'induction florale des brachyblastes du Poirier. Physiol. Vég. 10:529-545.

Huet, J. 1973. Floral initiation in pear trees. Acta Hort. 34:193-198.

Li, S.H., S.Q. Meng, T.H. Li, H.Z. Liu and Y.C. Tu. 1995. Critical period of flower bud induction in 'Red Fuji' and 'Ralls Janet' apple trees. Gartenbauwissenschaft 60:240-245.

Lin, J.-T., A.E. Stafford and G.L. Steffens. 1991. Identification of endogenous gibberellins in immature apple seeds. Agr. Biol. Chem. 55:2183-2185.

Looney, N.E., R.L. Granger, C.L. Chu, S.J. McArtney, L.N. Mander and R.P. Pharis.

1992. Influences of gibberellins A_4, A_{4+7}, and A_4+iso-A_7 on apple fruit-quality and tree productivity. I. Effects on fruit russet and tree yield components. J. Hort. Sci. 67:613-618.

Looney, N.E., R.P. Pharis, and M. Noma. 1985. Promotion of flowering in apple trees with gibberellin A_4 and C-3 epi-gibberellin A_4. Planta 165:292-294.

Luckwill, L.C. 1970. The control of growth and fruitfulness of apple trees. p. 237-254 In: L.C. Luckwill and C.V.Cutting (eds.). Physiology of Tree Crops. Academic Press, New York.

Luckwill, L.C., P. Weaver and J. MacMillan. 1969. Gibberellins and other growth hormones in apple seeds. J. Hort. Sci. 44:413-424.

Marcelle, R. and C. Sironval. 1963. Effect of gibberellic acid on flowering of apple trees. Nature 197:405.

Marino, F. and D.W. Greene. 1981. Involvement of gibberellins in the biennial bearing of 'Early McIntosh' apples. J. Amer. Soc. Hort. Sci. 106:593-596.

McArtney, S.J. 1994. Exogenous gibberellin affects biennial bearing and the fruit shape of 'Braeburn' apple. New Zealand J. Crop and Hort. Sci. 22:343-346.

McArtney, S.J. and S.H. Li, 1998. Selective inhibition of flowering on 'Braeburn' apple trees with gibberellins. Hortscience (in press).

McArtney, S.J., D.S. Tustin, S. Seymour, W. Cashmore and N.E. Looney. 1995. Benzyladenine and carbaryl effects on fruit thinning and the enhancement of return flowering of 3 apple cultivars. J. Hort. Sci. 70:287-296.

McCormick, A.C. 1933. Control of biennial bearing in apples. Proc. Amer. Soc. Hort. Sci. 30:326-329.

McKee, M.W. and C.G. Forshey. 1966. Effects of chemical thinning on repeat bloom of 'McIntosh' apple trees. Proc. Amer. Soc. Hort. Sci. 88:25-32.

McLaughlin, J.M. and D.W. Greene. 1991. Fruit and hormones influence flowering of apple. I. Effect of cultivar. J. Amer. Soc. Hort. Sci. 116:446-449.

Neilsen, J.C. and F.G. Dennis Jr. 1997. Effects of seed number, fruit removal, bourse shoot length and crop density on flowering in 'Spencer Seedless' apple. Acta Hort. (In press).

Nesterov, Y.S., G.G. Anisimova and N.I. Savelev. 1972. Vestnik Sel'skokhov. Nauki

10:114-118. (Cited by Buban, 1996.)

Palmer, J.W. 1992. Effects of varying crop load on photosynthesis, dry-matter production and partitioning of 'Crispin'/M.27 apple trees. Tree Physiol. 11:19-33.

Palmer, J.W., Y.L. Cai and Y. Edjamo. 1991. Effect of part-tree flower thinning on fruiting, vegetative growth and leaf photosynthesis in 'Cox Orange Pippin' apple. J. Hort. Sci. 66:319-325.

Roberts, R.H. 1920. Off year bearing and apple spur growth. Wisconsin Agr. Expt. Sta. Bul. 317.

Ramirez, H. and G.V. Hoad. 1981. Effects of growth substances on fruit-bud initiation in apple. Acta Hort. 120:131-136.

Salisbury, F.B. 1982. Photoperiodism. Hort. Rev. 4:66-105.

Singh, L.B. 1948. Studies in biennial bearing II. A review of the literature. J. Hort. Sci. 24:45-65.

Sritharan, R. and F. Lenz. 1988. Effects of water-stress in apple. I. Effect of water-supply on flower formation and fruit-set of the apple cultivar 'Golden Delicious'. Gartenbauwissenschaft 53:223-226.

Steel, R.G.D. and J.H. Torrie. 1980. Principles and Procedures of Statistics, 2nd Edition. McGraw-Hill Book Company, Inc., New York.

Stephan, M., F. Bangerth and G. Schneider. 1997. Transport and metabolism of the gibberellins A_1, A_3, and A_4 after application to developing apple fruits of *Malus domestica* cv. Jonagold. Acta Hort. 463:113-119.

Stutte, G.W. and G.C. Martin. 1986a. Effect of killing the seed on return bloom of olive. Scientia Hort. 29:107-113.

Stutte, G.W. and G.C. Martin. 1986b. Effect of light intensity and carbohydrate reserves on flowering of olive. J. Amer. Soc. Hort. Sci. 111:27-31.

Struckmeyer, B.E. and R.H. Roberts. 1942. Investigations on the time of blossom induction in Wealthy apple trees. Proc. Amer. Soc. Hort. Sci. 40:113-119.

Tobutt, K.R. 1994. Combining apetalous parthenocarpy with columnar growth habit in apple. Euphytica 77:51-54.

Tromp, J. 1982. Flower-bud formation in apple as affected by various gibberellins. J. Hort. Sci. 57:277-282.

Tromp, J. 1987. Growth and flower-bud formation in apple as affected by paclobutrazol, daminozide, and tree orientation in combination with various gibberellins. J. Hort. Sci. 62:433-440.

Tromp, J. 1992. The effect of soil temperature on lateral shoot formation and flower-bud formation in apple in the 1st year after budding. J. Hort. Sci. 67:787-793.

Tromp, J. 1993. Lateral shoot formation and flower-bud formation in apple in the 1st year after budding as affected by air-temperature and exposure to red-light. J. Hort. Sci. 68:255-260.

Volz, R.K., I.B. Ferguson, E.W. Hewett and D.J.Wooley. 1994. Wood age and leaf area influence fruit size and mineral composition of apple fruit. J. Hort. Sci. 69:385-395.

Weigel, D. and O. Nilsson. 1995. A developmental switch sufficient for flower initiation in diverse plants. Nature 377:495-500.

Wertheim, S.J. 1982. Fruit russeting in apple as affected by various gibberellins. J. Hort. Sci. 57:283-288.

Wertheim, S.J. and E.N. Estabrooks. 1994. Effect of repeated sprays of 6-benzyladenine on the formation of sylleptic shoots in apple in the fruit tree nursery. Scientia Hort. 60:31-39.

Zeller, O. 1960. Entwicklungsgeschichte der Blütenknospen und Fruchtanlangen an einjährigen Langtrieben von Apfelbüschen I. Entwicklungsverlauf und entwicklungsmorphologie der Blüten am einjährigen Langtrieb. Z. für Pflanzenzüchtung 44:175-214.