# Rooting Evaluation of Kiwifruit (Actinidia chinensis) And Effects of Anaerobiosis on Bud Break

by

Bradley Jason Sims

A thesis submitted to the Graduate Faculty of Auburn University in partial fulfillment of the requirements for the Degree of Master of Science

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Keywords: dogwood, liriope, peach, chilling, flooding

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Approved by

James D. Spiers, Chair, Assistant Professor of Horticulture William A. Dozier, Jr. Professor of Horticulture Jeff L. Sibley, Professor of Horticulture

## Abstract

Two unrelated studies were conducted in completion of this degree. The first study evaluated different rates of growth hormone on softwood kiwifruit (*Actinidia chinensis*) cuttings to determine the proper rate for optimal root initiation. Potassium salt of indolebutyric acid (KIBA) in rates of 1000 ppm, 2500 ppm, 5000 ppm and 10,000 ppm were evaluated on two relatively new cultivars of a golden-fleshed kiwifruit (*Actinidia chinensis*) 'AU Golden Sunshine' and 'AU Golden Dragon'. Results indicated a higher rooting percentage in the 'AU Golden Sunshine' cuttings when using 5000 ppm and 10,000 ppm KIBA over the control (distilled water), 1000 ppm, and 2500 ppm KIBA. However, in the 'AU Golden Dragon' cuttings there was no significant difference among the treatments.

The second study evaluated the effects of anaerobiosis on bud break of peach (*Prunus persica* Batsch 'Contender') cuttings, white flowering dogwood (*Cornus florida* L.) seedlings, and whole plant liriope (*Liriope muscari* 'Variegata'). Anaerobiosis is a process in which plants are depleted of air by submersion under water. Results indicated that in the case of floral bud break, anaerobiosis could be substituted for some chilling hours because the two water submerged treatments had higher floral bud break at the 800 and 900 chilling hours than did the control. In the case of vegetative bud break, anaerobiosis did not substitute for any chilling and there was no vegetative bud break on cuttings that did not have at least 1000 hours of accumulated chilling. Once the cuttings

had accumulated 1000 hours of chilling, anaerobiosis did accelerate bud break and the warm water treatment resulted in higher vegetative bud break for all levels of chilling. In the liriope study anaerobiosis seemed to have a negative effect and the control had higher dry weights than the treatments that were submerged in water. The liriope dry weights actually decreased for the longer that the plants were submerged in water. The results from the white dogwood study were similar to the peach study in that the warm water treatment caused an increase in bud break and bud break to occur more rapidly. The control and cold water treatment had similar results in that bud break was not accelerated and the cumulative bud break numbers were similar.

# Acknowledgements

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I would also like to thank my lovely wife Jami for her unconditional love and support shown through this entire process and for the typing and computer skills that she has lent to the cause. Also, I have much appreciation for my wonderful children and their love and support through this process. Thanks to the entire Auburn University Horticulture Department for the positive impact on my life whether knowingly or unknowingly. This wonderful experience will always be a part of my life.

# Table of Contents

Abstract	ii
Acknowledgements	iv
List of Tables	v
List of Figures	vi
Chapter I. Literature Review	1
Chapter II. Kiwifruit Propagation	10
Chapter III. Effects of Anaerobiosis on Budbreak	23

# List of Tables

Table 1. Influence of KIBA on vegetative cuttings of Actinidia chinensis 'AU Golden   Sunshine'	
Table 2. Influence of KIBA on vegetative cuttings of Actinidia chinensis 'AU Golden   Dragon'	
Table 3. Influence of anaerobiosis on floral bud break on Prunus persica 'Contender'	
Table 4. Influence of anaerobiosis on vegetative bud break on <i>Prunus persica</i> 'Contender'	32
Table 5. Effects of anaerobiosis on rate of floral bud break in comparison to heat units   Prunus persica 'Contender'	
Table 6. Effects of anaerobiosis on rate of vegetative bud break in comparison to heat   units of <i>Prunus persica</i> 'Contender'	
Table 7. Dry weights of whole Liriope muscari 'Variegata' plants	35
Table 8. Cumulative bud break of white dogwood seedlings in response to anaerobic   conditions via submersion in water	36

# List of Figures

Figure 1. The percent rooting of Actinidia chinensis 'AU Golden Sunshine' in response to
KIBA
Figure 2. The percent rooting of Actinidia chinensis 'AU Golden Dragon' in response to
KIBA

# CHAPTER I LITERATURE REVIEW

# Section I. Kiwifruit

Kiwifruit (Actinidia sp.) is native to China and has been cultivated for centuries there. Plants were introduced into New Zealand in the early 1900's, which subsequently became the world's largest exporter of kiwifruit. The plants in New Zealand can be traced to a single seed source that arrived from China in 1904 (Ferguson, 1984). Two main species of kiwifruit are grown throughout the world: Actinidia deliciosa and Actinidia chinensis. Actinidia deliciosa is a green-fleshed fruit and the most prevelant species in production. Actinidia chinensis is a golden-fleshed fruit, and there are two new varieties that have been developed by Auburn University: 'AU Golden Sunshine' and 'AU Golden Dragon'. One advantage with the golden-fleshed varieties is that the fruit actually ripens on the vine as opposed to the green varieties that require cold storage to ripen, which may dramatically reduce the cost of production. The golden-fleshed varieties have a smooth skin as opposed to the hairy skin of the green-fleshed varieties. The golden-fleshed varieties also have a less tart and sweeter flavor than the green fleshed varieties. The two new cultivars that have been developed by Auburn University show promise for fruit growers in the Southeastern U.S. with a lower chilling requirement (800-900 hours) and the fact that these cultivars ripen at different times

allowing growers to extend the harvest time (Wall et al., 2008). Once these cultivars are released to commercial producers, the demand is expected to increase and therefore will require quick propagation techniques for true-to-type plants.

There are four major types of propagation used to produce kiwifruit: budding, grafting, rooting of cuttings and micro-propagation. The most common forms of kiwifruit propagation are grafting to seedling rootstock and rooting of softwood cuttings (Tanimoto, 1994). Budding and grafting require the growing of seedling rootstock, which takes time, and the process of budding and grafting takes quite a bit of skill. The practice of growing rootstock seedlings is common and percent germination was reportedly as high as 94% in one study (Anderson and Lawes, 1980). As for grafting, most nurseries use the whip and tongue grafting method (Tanimoto, 1994). Micro-propagation, although being a speedy process, can be quite costly. One disadvantage to micro-propagation is that the thin hairless roots formed during micro-propagation tend to die shortly after potting (Kumar and Sharma, 2002).

Cutting propagation is the most convenient and feasible method of cloning plants (Hartman et. al., 2002). There have been many studies on the propagation of kiwifruit by cuttings with varying degrees of success being demonstrated by incorporating bottom heat, varying the timing of cuttings, and using different concentrations of rooting hormones (Testolin and Vitaglian, 1987). Research with the cultivar *Actinidia deliciosa* 'Monty' showed rooted cuttings at 61.5% for treated cuttings and 54% with the untreated cuttings (Abhi et.al., 1991). In a study conducted by Monrovia Nursery, cuttings of *Actinidia deliciosa* treated with IBA had poor rooting results (30%), but when IBA was combined with NAA the rooting percentages doubled to 70% (Connor, 1982). In another

study, herbaceous cuttings of Actinidia deliciosa had a higher rooting percentage than hardwood cuttings (cuttings taken prior to vegetative growth in spring) (Bartolini and Ianni, 1990). In Italy cuttings were taken at different times throughout the growing season, cuttings harvested July – August and treated with 4000 – 6000 ppm IBA gave the best rooting percentages (Biasi, et. al., 1990). In an Auburn University study, hardwood cuttings of Actinidia chinensis 'AU Golden Sunshine' had 40% rooting with no hormone treatment, while cuttings treated with KIBA rooted at 92.5% rooting (M. Harrison, personal communication). The same study at Auburn University used hardwood cuttings of another cultivar, Actinidia chinensis 'AU Golden Dragon', and achieved a 90% rooting with no hormone, while the KIBA treated cuttings showed a rooting percentage of 82.5%. A study that compared plant growth for 'Hayward' plants obtained by micro-propagation, hardwood cuttings, and by grafting showed that the micro-propagation and grafted plants showed greater vigor (Loreti and Piccotino, 1992). A study that compared fruit yields between kiwi plants from cuttings, grafting, and micro-propagation, showed the micropropagated plants had the highest cumulative yields over a 7-year period (Monastra and Testoni, 1991). In a study that compared root growth between plants obtained by cuttings, grafting, and micro-propagation over a two year period, the root systems of the grafted and micro-propagated plants were 50% larger than the plants from cuttings (Piccotino et.al., 1991). Another study showed that kiwi plants obtained by micro-propagation, had higher growth rates over plants from cuttings, but the fruit yields were the same over a six year period (Xiloyannis et.al., 1997).

# **Literature Cited**

Abhi, N., M.Z. Ahmadi, and H. Sadeghi. 1991. Effects of different factors on rooting percentage of hardwood and semi-hardwood kiwifruit 'Monty' cuttings in Mazandaran, Iran. N.Z. J. of Crop and Horticulture Sci. 19:365-367.

Anderson, D.R. and G.S. Lawes. 1980. Influence of temperature and gibberellic acid on kiwifruit (*Actinidia chinensis*) seed germination. N.Z. J. of Experimental Agric. 8:277-280.

Bartolini, G. and G. Ianni. 1990. Kiwi propagation test with herbaceous and hardwood cuttings. Acta Hort. 282:239-241.

Biasi, R., G. Marina, and G. Costa. 1990. Propagation of Hayward (*Actinidia deliciosa*) from soft and semi-hardwood cuttings. Acta Hort. 282:243-250.

Connor, D.M. 1982. Cutting propagation of *Actinidia chinensis* (kiwifruit). Combined Proc. Int. Plant Prop. Soc. 32: 329-333.

Ferguson, A.R. 1984 Kiwifruit: A Botanical review. Hort. Reviews 6:1-64.

Hartmann, H.T., D.E. Kester, F.T. Davies, Jr., R.L. Geneve Plant Propagation: Principles and Practices. New Jersey: Prentice Hall, 2002 (6<sup>th</sup> edition). 880 pages.

Kumar, S. and D.R. Sharma. 2002. In-vitro propagation of kiwifruit. J. of Horticultural Sci. & Biotechnology 77:503-508.

Loreti, F. and D. Piccotino. 1992. Effects of propagation techniques on vegetative growth and fruiting in kiwifruit. Acta Hort.

Monastra, F. and Testoni, A. 1991. Horticultural performance and quality characteristics of fruit from kiwifruit plants (cv. Hayward) obtained by in-vitro. Acta Hort. 1-297:197-203.

Piccotino, D., R. Massai, G. Baroni, and M. Boro. 1991. Root system conformation and growth of kiwifruit as affected by propagation technique. Acta Hort. 297: 391-399.

Sibley, J. L., W.A. Dozier, Jr., J.A. Pitts, A.W. Caylor, D.G. Himelrick and R.C. Ebel.2005. Kiwifruit cultivars differ in response to winter chilling. Small Fruits Review 4:19-29.

Tanimoto, G. Kiwifruit: Growing and Handling. California: ANR Publications. 1994.

Testolin, R. and C. Vitagliano. 1980. Influence of temperature and applied auxins during winter propagation. HortScience 22:573-574.

Wall, C., W. Dozier, R.C. Ebel, B.Wilkins, F. Woods, and W. Foshee. 2008. Vegetative and floral chilling requirements of four new kiwi cultivars of *Actinidia chinensis* and *Actinidia deliciosa*. HortScience 43:644-647.

Xiloyannis, C., V. Nuzzo, R. Massai, D. Piccotino, and G. Baroni. 1997. Vegetative growth, yield and root development in kiwifruit plants obtained by different propagation techniques. Acta Hort. 444: 144-148.

#### Section II. Effects of Anaerobiosis on Budbreak

Dormancy is a phase in a plant's development that allows it to survive winter conditions (Saure, 1985). It is also a state in which visible growth is temporarily suspended (Samish, 1954). Temperate plant species must be exposed to a certain period of chilling temperatures for dormancy release to occur. This period is known as the chilling requirement (Saure, 1985). With some crops, if the chilling requirement is not met, a chemical such as hydrogen cyanamide can be sprayed to overcome dormancy to a certain extent. A chilling hour is one hour at or below 45 degrees F (Powell et al., 1999). Some nursery grown plants for outplanting that do not get enough chilling in the field need more chilling hours in cold storage in order to break dormancy.

Generally speaking, bud break following completion of rest is temperature mediated. However, other stress factors can lead to initiation of bud break such as light levels, water stress, wounding, or flooding. Cuttings are often submerged briefly in water baths containing fungicides or insecticides to sanitize tissues prior to propagation. In a paper by Copes and Blythe (2011), cuttings were submerged for extended periods in water at various temperatures with no added chemicals to determine if pathogen control or rooting would be affected. However their study did not consider the possibility of a plant response such as an effect on flower or vegetative bud break due to anaerobic conditions.

The purpose of this study was to determine if anaerobiosis could accelerate bud break in dormant plants. Anaerobiosis is defined as a condition in which oxygen, carbon dioxide, and other gasses are completely absent or depleted, which is often the case with wetland species near water bodies. In the case of plants, oxygen depletion is not the

primary concern, but carbon dioxide. Since growers often modify lifting and transplanting schedules based on chilling accumulation, this study was conducted to determine if anaerobiosis could offset insufficient chilling to induce natural bud break. If so, seedlings of trees could be grown in areas of low chilling and still be marketed in other areas without the used of chemicals. It also would be possible for trees grown in greenhouses to be moved to the field without having to be in a cooler for an extended period.

Very little work has investigated the effects of anaerobiosis on bud break of dormant plants. In 1985, J.L. Regnard had an article "Breaking the dormancy of buds in woody plants by artificial means. Analysis of anoxia effects in poplar" in a French Journal. More recently, two articles in Korean carried out similar work with grapes (Kim, et al., 2000a and Kim et al., 2000b). In each of these articles, soaking plant parts or whole plants in water induced bud break. The optimal length of time appeared to be 24 hours across a range of temperatures.

# **Literature Cited**

Copes, W.E. and E.K. Blythe, 2011. Rooting response of azalea cultivars to hot water treatment used for pathogen control. HortScience (in press).

Kim, S.K., S.H. Kim, and Y.C. Lee. 2000a. Effects of anaerobiosis and subsequent treatment with Merit Blue and Allium Formulae on budbreak in 'Cambell Early' and 'Muscat Bailey A' grapevine cuttings. J. Kor. Soc. Hort. Sci. 41:269-272.

Kim, S.K., S.H. Kim, and Y.C. Lee. 2000b. Effects of anaerobiosis on budbreak in grapevine cuttings. J. Kor. Soc. Hort. Sci. 41:273-275.

Powell, A., D. Himlerick, W. Dozier, and D. Williams. 1999. Fruit culture in Alabamawinter chilling requirements. Alabama Coop. Ext. Sys. Bull. ANR-53-D.

Regnard, J.L. 1985. Breaking the dormancy of buds in woody plants by artificial means. Analysis of anoxia effects in poplar. Colloqre Surles Recherches Fruitieres. 1985: 187-200.

Samish, R.M. 1954. Dormancy in Woody plants. Ann. Rev. Plant Physiology 5:183-204.

Saure, M.C. 1985. Dormancy release in deciduous fruit trees. HortReviews 7:239-300.

# CHAPTER II KIWIFRUIT PROPAGATION

#### Introduction

Kiwifruit (Actinidia sp.) is native to China and has been cultivated for centuries there. Plants were introduced into New Zealand in the early 1900's, which subsequently became the world's largest exporter of kiwifruit. The plants in New Zealand can be traced to a single seed source that arrived from China in 1904 (Ferguson, 1984). Two main species of kiwifruit are grown throughout the world: Actinidia deliciosa and Actinidia chinensis. Actinidia deliciosa is a green-fleshed fruit and the most prevalent species in production. Actinidia chinensis is a golden-fleshed fruit. There are two new A. chinensis varieties that have been developed by Auburn University: 'AU Golden Sunshine' and 'AU Golden Dragon'. One advantage with the golden-fleshed varieties is that the fruit actually ripens on the vine as opposed to the green varieties that require cold storage to ripen, which may dramatically reduce the cost of production. The goldenfleshed varieties have a smooth skin as opposed to the hairy skin of the green-fleshed varieties. The golden-fleshed varieties also have a less tart and sweeter flavor than the green-fleshed varieties. The two new cultivars that have been developed by Auburn University show promise for fruit growers in the Southeastern U.S. with a lower chilling requirement (800-900 hours) and the fact that these cultivars ripen at different times allowing growers to extend the harvest time (Wall et al., 2008). Once these cultivars are

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# **Materials and Methods**

Semi-hardwood cuttings from (Actinidia chinensis) 'AU Golden Sunshine' and 'AU Golden Dragon' were collected from the Chilton Area Research and Extension Center on July 14, 2010. The cuttings were placed in coolers, packed in ice, and transported to Auburn University, where they were placed in a walk-in cooler overnight. The following day, July 15, 2010, the cuttings were transported to the greenhouse complex at Auburn University to initiate the experiment. The 'AU Golden Sunshine' cuttings were processed first and placed in 5 blocks of 4 different treatments, plus the control treatment. Each block contained 10 cuttings that were wounded at the base and dipped for 10 seconds in the treatment. After the cuttings were removed from the treatment they were placed into 4 inch plastic pots containing a standard 6:1 pinebark: sand mix amended using 7.1 kg Osmocote 18-6-12 N-P<sub>2</sub>O<sub>5</sub>  $-K_2O/m^3$  (12 lbs./yd<sup>3</sup>, approximately 1.0 kg N/m<sup>3</sup>), 0.6 kg/m<sup>3</sup> Micromax (1.0 lbs./yd<sup>3</sup>) (O. M. Scotts Co. Marysville, Ohio), and 5 lbs./yd<sup>3</sup> of dolomitic limestone and placed on expanded metal benches in the greenhouse. This process was repeated until all 5 treatments of 10 cuttings each were completed. The 5 treatments consisted of a control (distilled water), KIBA (potassium chloride dissolved in distilled water) at 1000 ppm, 2500 ppm, 5000 ppm, and at 10,000 ppm. This process was repeated until there were 5 blocks of 'AU Golden Sunshine' and the whole process was repeated again for the 'AU Golden Dragon' cuttings.

Once all the blocks were completed, they were covered with clear poly to hold in moisture. Data loggers were placed inside the poly coverings to monitor temperature. The

cuttings were watered with intermittent mist to keep from drying out. The cuttings were monitored periodically while remaining in the greenhouse for 12 weeks.

Once the cuttings had 12 weeks in the greenhouse, they were removed from the covered benches and the data collection was started. The data was evaluated per rep (10 cuttings per treatment). Cuttings were carefully removed from the plastic pots by turning them upside down and slowly pulling the pot away from the roots and soil. Soil was carefully washed away from the cutting and roots. The percentage of cuttings that rooted was recorded and the number of primary roots connected to the cutting were counted. The roots were also analyzed to determine the degree of root branching that was present, by assigning a number from 1-5 (1 being little branching and 5 being the highest degree of branching). Once these steps were completed, the roots were removed at the base of the cuttings and weighed to determine fresh weight of the roots. The roots were then placed in brown paper bags, placed in a dryer oven at 160°F for 72 hours, removed and weighed to determine the dry weight for the roots. The new shoots were removed and weighed to determine fresh weight. Finally, callus at the base of the cuttings was measured to determine the diameter of each callus. Data were subjected to analysis of variance in the General Linear Methods procedures and Duncan's Multiple Range Test in SAS (SAS, 1996).

#### **Results and Discussion**

The 'AU Golden Sunshine' cuttings had a higher rooting percentage for the 10,000 ppm KIBA and 5000 ppm KIBA treatments as compared to the control, 1000 ppm KIBA and 2500 ppm KIBA. The root weight (both fresh and dry), shoot weight, number of main roots and the degree of root branching was also higher for the 5000 ppm KIBA

and 10,000 ppm KIBA as compared to the control, 1000 ppm KIBA and 2500 ppm KIBA. There was no significant difference in the root lengths among the treatments (ranged from 50.44 cm to 81.29 cm). The callus size was significantly larger for the control, 1000 ppm and 2500 ppm versus the callus size of the 5000 ppm and 10,000 ppm treatments (Table 1; Fig. 1).

There were no differences in the 'AU Golden Dragon' cuttings in rooting percentages among the different treatments and the rooting percentages ranged from 30 to 54 percent rooting, 0.97g to 1.31g for root fresh weight, 3.21g to 3.68g for shoot fresh weight, 4 to 4.85 main root numbers, 56.89cm to 84.13cm for root length, 11.41 to 15.20 for callus size, 1.78 to 2.35 for the degree of root branching.

The rooting percentages coincide with previous research done at Auburn University where 'AU Golden Sunshine' cuttings had 40% rooting for the control and 92.5% rooting for KIBA treatment and 'AU Golden Dragon' cuttings had 90% rooting for the control and 82.5% rooting for KIBA treatment (M. Harrison, personal communication). This leads to a conclusion that KIBA increases the rooting ability of 'AU Golden Sunshine' but has no impact on the rooting ability of 'AU Golden Dragon'. In an earlier study, herbaceous cuttings of *Actinidia deliciosa* treated with 3000 ppm IBA had an 85% rooting, while hardwood cuttings treated with 3000 ppm IBA had less than 40% rooting (Bartolini and Ianni, 1990). This suggest that it would be best to obtained cuttings in late spring or early summer to ensure that they are still herbaceous and seem to root more easily than hardwood or semi-hardwood cuttings.

In conclusion, semi-hardwood cuttings taken during mid-summer showed that a higher ppm KIBA improved rooting percentages for 'AU Golden Sunshine.' Additional

research is needed to determine optional treatments to improve rooting percentages of 'AU Golden Dragon' and the optimal time to harvest cuttings for both cultivars.

# **Literature Cited**

Abhi, N., M.Z. Ahmadi, and H. Sadeghi. 1991. Effects of different factors on rooting percentage of hardwood and semi-hardwood kiwifruit 'Monty' cuttings in Mazandaran, Iran. N.Z. J. of Crop and Horticulture Sci. 19:365-367.

Anderson, D.R. and G.S. Lawes. 1980. Influence of temperature and gibberellic acid on kiwifruit (*Actinidia chinensis*) seed germination. N.Z. J. of Experimental Agric. 8:277-280.

Bartolini, G. and G. Ianni. 1990. Kiwi propagation test with herbaceous and hardwood cuttings. Acta Hort. 282:239-241.

Biasi, R., G. Marina, and G. Losta. 1990. Propagation of Hayward (*Actinidia deliciosa*) from soft and semi-hardwood cuttings. Acta Hort. 282:243-250.

Connor, D.M. 1982. Cutting propagation of *Actinidia chinensis* (kiwifruit). Combined Proc. Int. Plant Prop. Soc. 32: 329-333.

Ferguson, A.R. 1984 Kiwifruit: A Botanical review. Hort. Reviews 6:1-64.

Hartmann, H.T., D.E. Kester, F.T. Davies, Jr., R.L. Geneve Plant Propagation: Principles and Practices. New Jersey: Prentice Hall, 2002 (6<sup>th</sup> edition). 880 pages.

Kumar, S. and D.R. Sharma. 2002. In-vitro propagation of kiwifruit. J. of Horticultural Sci. & Biotechnology 77:503-508.

Loreti, F. and D. Piccotino. 1992. Effects of propagation techniques on vegetative growth and fruiting in kiwifruit. Acta Hort.

Monastra, F. and Testoni, A. 1991. Horticultural performance and quality characteristics of fruit from kiwifruit plants (cv. Hayward) obtained by in-vitro. Acta Hort. 1-297:197-203.

Piccotino, D., R. Massai, G. Baroni, and M. Boro. 1991. Root system conformation and growth of kiwifruit as affected by propagation technique. Acta Hort. 297: 391-399.

Sibley, J.L., W.A. Dozier, Jr., J.A. Pitts, A.W. Caylor, D.G. Himelrick and R.C. Ebel. 2005. Kiwifruit cultivars differ in response to winter chilling. Small Fruits Review 4:19-29.

Tanimoto, G. Kiwifruit: Growing and Handling. California: ANR Publications. 1994.

Testolini, R. and C. Vitagliano. 1980. Influence of temperature and applied auxins during winter propagation. HortScience 22:573-574.

Wall, C., W. Dozier, R.C. Ebel, B.Wilkins, F. Woods, and W. Foshee. 2008. Vegetative and floral chilling requirements of four new kiwi cultivars of *Actinidia chinensis* and *Actinidia deliciosa*. HortScience 43:644-647.

Xiloyannis, C., V. Nuzzo, R. Massai, D. Piccotino, and G. Baroni. 1997. Vegetative growth, yield and root development in kiwifruit plants obtained by different propagation techniques. Acta Hort. 444: 144-148.

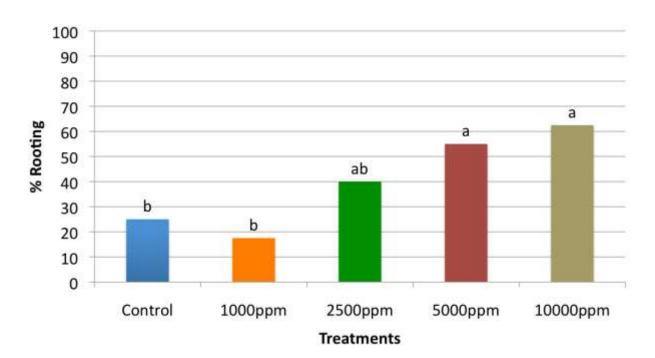


Figure 1. The percent rooting of *Actinidia chinensis* 'AU Golden Sunshine' in Response to KIBA.

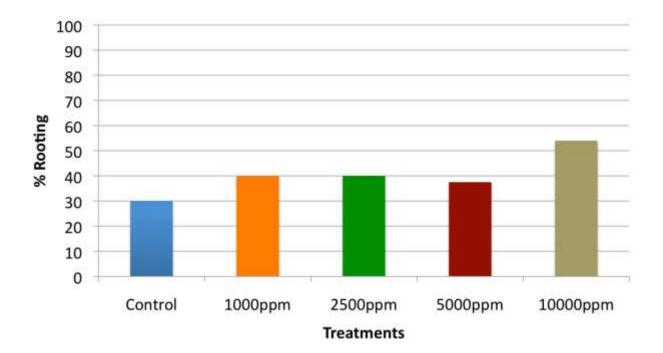


Figure 2. The percent rooting of *Actinidia chinensis* 'AU Golden Dragon' in Response to KIBA.

Treatment	R wt <sup>Y</sup>	S wt <sup>Y</sup>	R# <sup>Y</sup>	R lgth <sup>Y</sup>	Callus <sup>Y</sup>	Branch <sup>Y</sup>	Root <sup>Y</sup> DW
Control	1.30b <sup>x</sup>	1.51b	3.56b	81.29	20.57ab	1.7b	.15b
1000ppm	1.55b	2.15b	2.86b	60.90	23.40a	1.5b	.21b
2500ppm	.84b	1.85b	2.75b	57.44	21.07ab	1.5b	.14b
5000ppm	3.32a	5.03a	5.48a	70.17	16.06b	2.9a	.50a
10000ppm	3.78a	4.85a	16.32a	60.95	16.61b	2.9a	.37ab
Pr > F	.0001	.004	.027	NS	.054	.0001	.006

Table 1. Influence of KIBA on Vegetative Cuttings of *Actinidia chinensis* 'AU Golden Sunshine'.<sup>Z</sup>

<sup>z</sup>Cuttings prepared 15 July, 2010 and harvested 24 October, 2010. Cuttings kept under intermittent mist in climate controlled greenhouse.

<sup>Y</sup>RWT= Root weight fresh in grams; Swt= Shoot weight fresh in grams; R#= Root numbers; R lgth= length of longest root in cm; Callus measured with a caliper in millimeters; Branch= degree of root branching rated 0-5; Root DW= root dry weight in grams.

<sup>x</sup>Values in a column followed by the same letter are not significantly different. P $\leq$ 0.05, Duncan's Multiple Range Test.

Treatment	<b>R</b> wt <sup>Y</sup>	S wt <sup>Y</sup>	<b>R</b> # <sup>Y</sup>	R lgth <sup>Y</sup>	Callus <sup>Y</sup>	Branching <sup>Y</sup>	R DW <sup>Y</sup>
Control	1.19 <sup>x</sup>	3.65	4.27	82.99	13.49	2.27	.104
1000ppm	1.25	3.68	4.00	84.13	11.41	2.35	.138
2500ppm	1.20	3.21	4.85	56.89	13.35	2.15	.159
5000ppm	1.31	3.7	4.24	69.92	12.42	2.12	.155
10000ppm	.97	3.36	4.37	82.61	15.20	1.78	.117
Pr > F	NS	NS	NS	NS	NS	NS	NS

Table 2. Influence of KIBA on Vegetative Cuttings of Actinidia chinensis 'AU Golden Dragon'.<sup>Z</sup>

<sup>z</sup>Cuttings prepared 15 July, 2010 and harvested 24 October, 2010. Cuttings kept under intermittent mist in climate controlled greenhouse.

<sup>Y</sup>RWT= Root weight fresh in grams; Swt= Shoot weight fresh in grams; R#= Root numbers; R lgth= length of longest root; Callus measured with a caliper in millimeters; Branch= degree of root branching rated 0-5; Root DW= root dry weight in grams.

<sup>x</sup>Values in a column followed by the same letter are not significantly different.  $P \le 0.05$ , Duncan's Multiple Range Test.

# CHAPTER III EFFECTS OF ANAEROBIOSIS ON BUDBREAK

#### Introduction

Dormancy is a phase in a plant's development that allows it to survive winter conditions (Saure, 1985). It is also a state in which visible growth is temporarily suspended (Samish, 1954). Temperate plant species must be exposed to a certain period of chilling temperatures for dormancy release to occur. This period is known as the chilling requirement (Saure, 1985). With some crops, if the chilling requirement is not met, a chemical such as hydrogen cyanamide can be sprayed to overcome dormancy to a certain extent. A chilling hour is one hour at or below 45 degrees F (Powell et al., 1999). Some nursery grown plants for out planting that do not get enough chilling in the field need more chilling hours in cold storage in order to break dormancy.

Generally speaking, bud break following completion of rest is temperature mediated. However, other stress factors can lead to initiation of bud break such as light levels, water stress, wounding, or flooding. Cuttings are often submerged briefly in water baths containing fungicides or insecticides to sanitize tissues prior to propagation. In a paper by Copes and Blythe (2011), cuttings were submerged for extended periods in water at various temperatures with no added chemicals to determine if pathogen control or rooting would be affected. However their study did not consider the possibility of a plant response such as an effect on flower or vegetative bud break due to anaerobic conditions.

The purpose of this study was to determine if anaerobiosis could accelerate bud break in dormant plants. Anaerobiosis is defined as a condition in which oxygen, carbon dioxide, and other gasses are completely absent or depleted, which is often the case with wetland species near water bodies. In the case of plants, oxygen depletion is not the primary concern, but carbon dioxide. Since growers often modify lifting and transplanting schedules based on chilling accumulation, this study was conducted to determine if anaerobiosis could offset insufficient chilling to induce natural bud break. If so, seedlings of trees could be grown in areas of low chilling and still be marketed in other areas without the use of chemicals. It also would be possible for trees grown in greenhouses to be moved to the field without having to be in a cooler for an extended period.

Very little work has investigated the effects of anaerobiosis on bud break of dormant plants. In 1985, J.L. Regnard had an article "Breaking the dormancy of buds in woody plants by artificial means. Analysis of anoxia effects in poplar" in a French Journal. More recently, two articles in Korean carried out similar work with grapes (Kim et al., 2000a and Kim et al., 2000b). In each of these articles, soaking plant parts or whole plants in water induced bud break. The optimal length of time appeared to be 24 hours across a range of temperatures.

#### **Materials and Methods**

1. Effects of anaerobiosis on bud break of Prunus persica 'Contender'

Chilling hours were determined by using the Modified 45-model (Powell et al., 1999) where all hours below 45 °F and above 32 °F were counted as 1 chill hour. Bud

sticks (12-inches long) of *Prunus persica* 'Contender', a 1000 chill-hour peach, were collected on November 15, 2009 from the Chilton Area Research and Extension Center, Clanton, AL with 400 hours of ambient chilling. The cuttings were transported to Auburn University greenhouses and placed in a 38 °F cooler. Seven levels of forced chilling were applied (total of 800-1400 chilling hours, including ambient chilling).

Upon accumulation of 800 chilling hours, 54 stems were removed from the cooler. Half of the stems (27) were submerged in room temperature (68 °F) water for 24 hours and the other 27 stems were submerged in 75 °F water for 24 hours. After 24 hours, the stems were removed from the water and 27 more stems were removed from the cooler. All the stems were then divided into treatments that consisted of 9 replications of 3 stems each. The stems were placed in pint jars filled with water and wrapped in aluminum foil to exclude light from the water, then placed on greenhouse benches and placed in a standard double-poly greenhouse maintained at a base temperature of 68 °F. Remaining shoot segments remained in the cooler at 38°F with bases in water, with 27 cuttings removed from the cooler in 100 hour increments and placed into the greenhouse as described above.

The study was conducted using a randomized complete block design (RCBD) with 7 blocks. Each block was arranged in a completely randomized design (CRD). This process was repeated at 100 hour chilling increments until 1400 hours of chilling. After placement in the greenhouse, stems were monitored twice weekly for floral and foliar bud break, which was record as soon as a floral or vegetative bud began to open. This was done until the termination of the study on February 25 2010.

# 2. Effects of anaerobiosis on growth of Liriope muscari 'Variegata'

Bare root plants of Liriope muscari 'Variegata' were purchased and kept in a walk in cooler until the experiment began. Three hundred and twenty plants (80 plants per 4 treatments) were removed from the cooler and taken to a greenhouse and the experiment was started. The study consisted of the control (bare root plants potted up directly from the cooler), treatment 1(bare root plants submerged in water for 24 hours), treatment 2 (bare root plants submerged in water for 48 hours) and treatment 3 (bare root plants submerged in water for 72 hours). The treatments consisted of eight replications of ten plants for each treatment. The bare root whole plants of Liriope muscari 'Variegata' were potted into four-inch pots in a standard 6:1 pine bark: sand mix amended using 7.1 kg Osmocote 18-6-12 N-P<sub>2</sub>O<sub>5</sub>  $-K_2O/m^3$  (12 lbs./yd<sup>3</sup>, approximately 1.0 kg N/m<sup>3</sup>), 0.6 kg/m<sup>3</sup> Micromax (1.0 lbs./yd<sup>3</sup>) (O. M. Scotts Co. Marysville, Ohio), and 5 lbs./yd<sup>3</sup> of dolomitic limestone. The potted plants were placed on expanded metal benches inside a 72 °F greenhouse where they were watered daily and remained there for 6 weeks. The study was conducted using a randomized complete block design (RCBD) with 4 blocks of 8 reps. Once the 6 weeks were over, the plants were harvested for data collection. Each ten plant replication was harvested by cutting the entire plant above the soil line with scissors and placing the plant into a paper bag. This process was completed for each replication of the four treatments. Once all the treatments were harvested, they were placed in a dryer oven at 160 °F for 72 hours. The samples were removed from the drier oven and weighed. Data were subjected to analysis of variance in the General Linear Methods procedures and Duncan's Multiple Range Test in SAS (SAS, 1996).

#### 3. Effects of anaerobiosis on bud break of Cornus florida

Locally available, field-grown, dormant, bare-root, 18 to 24 inch tall white flowering dogwood seedlings (Cornus florida) were purchased in January 2011 following about 850 hours of ambient chilling. Trees were stored in a cooler at 38°F for an additional 120 hours. On January 25, 25 trees were completely submerged in a water bath inside a cooler where the water had acclimated for two days to 38 °F. An additional 25 trees were submerged in a water bath inside a greenhouse where the water had acclimated to 68 °F for two days. On January 26, the same procedure was repeated in separate water baths for an additional 25 trees each, followed by the same procedures on January 27. On January 28, all trees were removed from the water baths, along with 25 additional trees direct from cooler storage in matted wet newsprint and Celluwet. All 175 trees were potted on January 28 following this incremental treatments of 0 (25 trees), 24 hours at 38 or 68 °F (25 trees each), 48 hours at 38 or 68 °F (25 trees each), and 72 hours at 38 or 68 °F (25 trees each), into trade gallon containers in a standard 6:1 pine bark: sand mix amended using 7.1 kg Osmocote 18-6-12 N-P<sub>2</sub>O<sub>5</sub>  $-K_2O/m^3$  (12 lbs./yd<sup>3</sup>, approximately 1.0 kg N/m<sup>3</sup>), 0.6 kg/m<sup>3</sup> Micromax (1.0 lbs./yd<sup>3</sup>) (O. M. Scotts Co. Marysville, Ohio), and 5 lbs./yd<sup>3</sup> of dolomitic limestone. Potted trees were placed in a double layer poly covered greenhouse at 68°F for subsequent observation of bud break, which was recorded twice weekly from January 28 through March 28, 2011. The study was conducted using a randomized complete block design (RCBD).

#### Results

1. Effects of anaerobiosis on bud break of Prunus persica 'Contender'

The rate of floral bud break was accelerated by increasing the level of chilling until the 1200 hours of accumulated chilling, then the rate of floral bud break started decreasing. The study also shows that anaerobiosis leads to decreased heat required to stimulate bud break as the two treatments both had significant bud break prior to the control, and there was actually floral bud break for the two treatments at only 800 hours of chilling, while the control had no floral bud break. At 900 hours of chilling, both treatments had significant bud break and the control had very little bud break. In the case of floral bud break, anaerobiosis did seem to take the place of chilling. The 1000 and 1100 hours of chilling cuttings still showed a significantly higher rate of bud break for the two treatments over the control. At the 1200, 1300, and 1400 hours of chilling the control actually showed a higher rate of bud break over the warm water treatment, while being about the same as the cold water treatment (Table 5 & 7).

There was no vegetative bud break until the cuttings had received their required 1000 hours of accumulated chilling that the cultivar 'Contender' needs before it can overcome its dormancy phase. The water treatment did not take the place of chilling, but did increase bud break numbers for this peach. The study did seem to show that once the cuttings had reached their required chilling hours, anaerobiosis did accelerate the rate of vegetative bud break, less heat units were required for the two water treatments to break bud. The number of open buds was zero for the 800, 900 and 1000 hours of accumulated chilling for the control and the two treatments. However, there were more open buds in response to the water bath treatments at 1100, 1200, 1300 and 1400 hours of accumulated

chilling when compared to the control treatment. The warm water bath had a significant higher bud total than the cold water bath (Table 4 & 6).

# 2. Effects of anaerobiosis on growth of Liriope muscari 'Variegata'.

The dry weights of the liriope were the highest among the control plants and the 24 hour treatment, which statistically the same. The water submersion actually decreased the growth of the liriope plants, with the weights decreasing the longer the plants were submerged under water. The plants that were submerged for 48 hours and 72 hours had the lowest dry weights and were statistically the same weight (Table 7).

#### 3. Effects of anaerobiosis on bud break of Cornus florida

The bud break for the dogwood seedlings was accelerated by the warm water treatment, which began to break bud sooner than the control and cold water treatment. However, the overall cumulative bud break totals were the same among all treatments at the conclusion of the study (Table 8).

# Discussion

Based on the results of these studies, it appears reasonable that smaller sized bareroot trees and other dormant liners could be induced to break bud and begin growth sooner in the spring. For greenhouse grown trees or shrubs, earlier bud break could decrease the time needed to reach size suitable for point of sale. Submerging entire plants in water for a period of time prior to out planting is a practical production method that could have direct application for plants not obligate to a fixed chilling requirement.

# **Literature Cited**

Copes, W.E. and E.K. Blythe, 2011. Rooting response of azalea cultivars to hot water treatment used for pathogen control. HortScience (in press).

Kim, S.K., S.H. Kim, and Y.C. Lee. 2000a. Effects of anaerobiosis and subsequent treatment with Merit Blue and Allium Formulae on budbreak in 'Cambell Early' and 'Muscat Bailey A' grapevine cuttings. J. Kor. Soc. Hort. Sci. 41:269-272.

Kim, S.K., S.H. Kim, and Y.C. Lee. 2000b. Effects of anaerobiosis on budbreak in grapevine cuttings. J. Kor. Soc. Hort. Sci. 41:273-275.

Powell, A., D. Himlerick, W. Dozier, and D. Williams. 1999. Fruit culture in Alabamawinter chilling requirements. Alabama Coop. Ext. Sys. Bull. ANR-53-D.

Regnard, J.L. 1985. Breaking the dormancy of buds in woody plants by artificial means. Analysis of anoxia effects in poplar. Colloqre Surles Recherches Fruitieres. 1985: 187-200.

Samish, R.M. 1954. Dormancy in Woody plants. Ann. Rev. Plant Physiology 5:183-204.

Saure, M.C. 1985. Dormancy release in deciduous fruit trees. HortReviews 7:239-300.

Accumulated Chilling Hours	800	900	1000	1100	1200	1300	1400
Treatment 1 Control	0.001 <sup>ZY</sup>	0.157kl	0.764kji	1.83gh	6.007b	5.00c	5.635bc
Treatment 2 Cold water	0.39kjl	2.078gf	2.418gef	7.399a	7.36a	5.922b	5.588bc
Treatment 2 Warm water	1.46ji	2.641ef	2.235gf	3.837d	3.863d	3.052e	1.259hi

Table 3. Influence of anaerobiosis on floral bud break on Prunus persica 'Contender'.

<sup>Z</sup>Average buds per treatment.

<sup>Y</sup>Values in a column followed by the same letter are not significantly different.

Accumulated Chilling Hours	800	900	1000	1100	1200	1300	1400
Treatment 1 Control	0.00e <sup>ZY</sup>	0.00e	0.00e	0.00e	0.131e	0.778d	1.012d
Treatment 2 Cold water	0.00e	0.00e	0.00e	0.837d	1.425c	1.327c	0.953d
Treatment 3 Warm water	0.00e	0.00e	0.098e	1.758b	3.105a	3.085a	2.882a

Table 4. Influence of anaerobiosis on vegetative bud break on *Prunus persica* 'Contender'.

<sup>z</sup>Average buds per treatment.

<sup>Y</sup>Values in a column followed by the same letter are not significantly different.

Accumulated Chilling Hours	800	900	1000	1100	1200	1300	1400
Treatment 1 Control	NBB <sup>Z</sup>	34608a <sup>YX</sup>	31680a	29568a	25536a	22698a	20160a
Treatment 2 Cold water	31416a	25237b	25872b	23968b	23445b	21354b	20966a
Treatment 3 Warm water	27384a	24192b	24117b	23968b	22814b	20757b	20160a

Table 5. Effects of anaerobiosis on number of heat units required for floral bud break on *Prunus persica* 'Contender'.

<sup>Z</sup>NBB= no buds broke on any cuttings in the treatment.

<sup>Y</sup>The average number of heat units accumulated before first bud break; 1 heat unit = 1 degree above 40 °F/hour (example: 68 °F= 28 heat units/hour).

<sup>X</sup>Values in a column followed by the same letter are not significantly different.

Table 6. Effects of anaerobiosis on number of heat units required for vegetative bud break on *Prunus persica* 'Contender'.

Accumulated Chilling Hours	800	900	1000	1100	1200	1300	1400
Treatment 1 Control	NBB <sup>Z</sup>	NBB	NBB	NBB	35448a <sup>YX</sup>	25632a	22982a
Treatment 2 Cold water	NBB	NBB	NBB	26400a	24288b	28747a	19264a
Treatment 3 Warm water	NBB	NBB	35280	27404a	21504b	18592b	18547a

 $^{Z}$ NBB= no buds broke on any cuttings in the treatment.

<sup>Y</sup>The average number of heat units accumulated before first bud break; 1 heat unit = 1 degree above 40 °F/hour (example: 68 °F= 28 heat units/hour).

<sup>X</sup>Values in a column followed by the same letter are not significantly different.

Table 7. Dry weights of whole Liriope plants following six weeks of growth in response to total submersion in water for zero to 72 hours.

		Liriope Data		
	Treatment 1 (control)	Treatment 2 (24hr submersion)	Treatment 3 (48hr submersion)	Treatment 4 (72hr submersion)
Dry weight in grams	7.370a <sup>z</sup>	6.610a	3.276b	3.286b

<sup>z</sup>Values in a row followed by the same letter are not significantly different.

Treatment	21 February	25 February	1 March	5 March	10 March	15 March	21 March
1 control (no soaking)	0	1	2	6	7	9	12
2 (24 hr. warm water)	2	4	6	10	10	10	11
3 (48 hr. warm water)	1	2	2	3	3	3	3
4 (72 hr. warm water)	2	3	4	7	9	12	14
5 (24 hr. cold water)	0	1	1	2	4	4	7
6 (48 hr. cold water)	0	1	3	5	6	7	7
7 (72 hr. cold water)	0	4	6	8	8	9	11

Table 8. Cumulative bud break of white dogwood seedlings in response to anaerobic conditions via submersion in water.