

**Flowering Control and Production of *Strobilanthes dyerianus* Mast.
(Persian Shield)**

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(ABSTRACT)

Grown for its distinctive foliage, *Strobilanthes dyerianus* is a popular bedding and container plant. A problem in production is that over-wintered stock plants often flower. Once the plant becomes reproductive, stem elongation slows and floral buds arise from every node, rendering the plants useless for propagation. The objectives of this research were to examine the effectiveness of manipulating environmental factors and the application of ethephon on preventing floral bud initiation, as well as determine optimal nitrogen rate for stock plant culture.

The first experiment was performed in a glass greenhouse and ran 11 weeks, utilizing 8 h, 10 h, 12 h and a 4 h night interruption photoperiod treatments to determine critical photoperiod. None of the photoperiod treatments were significant for inhibiting flowering and there was a positive correlation between plant size and flowering. A second experiment was performed in growth chambers to create three photoperiods (8 h, 12 h and 16 h) with two temperature regimes (24°C day/ 21°C night and 17°C day/ 14°C night) for a total of six treatments. Neither photoperiod nor temperature inhibited bud initiation, and there was no correlation between plant size and flowering.

The third experiment examined potential use of ethephon to maintain vegetative plants. Six rates of ethephon (0, 150, 300, 450, 600 or 750 mg·L⁻¹) were applied at three frequencies (weekly, biweekly and monthly) over an eleven week period. Floral initiation was not totally inhibited, but flowering was highly correlated to plant size.

Additionally, plant growth response to nitrogen was examined to determine the optimal rate for stock plant production. Plants were treated with 0, 100, 200, 300 or 400 mg·L⁻¹ N from a 15 N – 2.2 P – 12.4 K fertilizer at each irrigation for eight weeks. There were no differences among plant quality ratings for plants receiving 100, 200, 300 or 400 mg·L⁻¹ N, and plants grown with 200 mg·L⁻¹ N had the largest leaf area and shoot dry weight.

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DEDICATION

This thesis is dedicated to my Mom, Elizabeth Gamrod, and to the memory of my Dad, Bruce Gamrod. Mom, I never could have done this without you. Thanks.

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CHAPTER 1: LITERATURE REVIEW

Introduction to *Strobilanthes dyerianus*

Strobilanthes dyerianus Mast., common name Persian Shield, has become a popular and successful ornamental foliage crop in the past few years. It is an old-fashioned plant, whose reentry into the industry has been fueled by inclusion in the Athens Select™ program, which consists of plants demonstrating exceptional performance under hot and humid conditions. A native of Burma in Southeast Asia, *S. dyerianus* is grown strictly for its foliage appeal, which is green overlaid with purple; older leaves exhibit a silver hue. The leaves are opposite, sessile and ovate to lanceolate, spanning 15 to 23 cm long and 7 to 10 cm wide, with toothed margins (Armitage, 2001). When grown in the landscape, the tropical perennial *S. dyerianus* can grow to a small shrub, hardy to USDA zone 9. This plant thrives in warm temperatures, growing quickly with the onset of hot summer days, and performs well in most of the country, either as a landscape or container plant (Armitage, 1997).

Strobilanthes dyerianus is classified in division Magnoliophyta, class Magnoliopsida, order Scrophulariales and family Acanthaceae (Watson and Dallwitz, 1992). The Acanthaceae family consists of approximately 250 genera and 2500 species, most of which are considered to be tropical shrubs or herbs (Watson and Dallwitz, 1992). Members of this family are native to Asia, Africa, South America, Central America, North America and Australia. Other cultivated genera in this family include *Acanthus*, *Fittonia*, *Pachystachys* and *Thunbergia*. While 250 species in the genus *Strobilanthes* have been identified by taxonomists, only recently has *S. dyerianus* found its way into the

retail spotlight, making it the only representative of this genus easily obtainable through garden centers (Armitage, 2001). The genus *Strobilanthes* thrives in areas with copious amounts of sun and is, on the whole, more appealing for its foliage, as flowers rarely appear in the landscape (Armitage, 2001). When flowers do appear, they do not contribute to the aesthetics of *S. dyerianus*. The pale blue flowers which form are tubular and five-lobed, with two, three or four stamens. The flowers are held at the top of the stems in petite, conical inflorescences (Armitage, 2001).

However, aesthetics are not the only reason flower production is considered undesirable by growers. Flower initiation is detrimental to cutting propagation by growers, for once *S. dyerianus* reaches the reproductive stage, all stem elongation ceases and buds arise from every node (Batson, 1982), rendering propagation extremely difficult. Post-reproductive stock plants decline, with leaves showing necrosis that begins on the margins and moves inward, which reduces the number of cuttings that may be obtained from the plant. Additionally, once the plant becomes reproductive, it does not easily revert to the vegetative stage (Batson, 1982). According to personal communication with growers and personal observation, bud initiation seems to be associated with the onset of winter. While the exact mechanism behind floral initiation is not known, the observed winter-time flowering indicates possible reproductive triggers could be photoperiod, temperature or a combination of the two.

Photoperiod Effects on Plant Growth and Reproduction

Flowering plants may be categorized as long day (LD), short day (SD) or day neutral (DN) according to their response to photoperiod. SD plants flower when the

period of darkness exceeds a critical length, whereas LD plants flower when the dark period does not exceed a critical length. DN plants flower in response to a stimulus other than photoperiod, such as temperature or total light energy, independent of the amount of day light (Dole and Wilkins, 1999). In addition, plants may be classified as either facultative or obligate SD or LD. A facultative SD or LD plant has the ability to flower under a wide range of daylengths, but flowers are produced more rapidly under either short or long days. A plant classified as obligate SD or LD flowers only under a specific photoperiod. Furuta (1954) noted that while *Dendranthema x grandiflorum* Kitam. (chrysanthemum) plants formed flower buds in a 14 hour photoperiod, shorter photoperiods (less than 13 hours) resulted in rapid flower bud development, thereby labeling chrysanthemum as a facultative SD plant. Other examples include *Salvia splendens* Ker-Gawl. (facultative SD) and *Euphorbia pulcherrima* Willd.ex Klotzsch (poinsettia – obligate SD) (Dole and Wilkins, 1999). *Pelargonium x hortorum* L.H. Bail. (geranium) is DN and are more quickly induced to flower by using higher light intensity (from supplemental lights) to achieve a higher total light energy. Many floriculture crops can only perceive photoperiods after they have reached maturity (Dole and Wilkins, 1999).

Pallez and Dole (2001) examined photoperiod manipulation to keep the LD plant *Gynura aurantiaca* Blume in a vegetative state to improve cutting quality and number. They found photoperiod of 8 hours kept the plant in a vegetative state, while photoperiods of 16 hours initiated flower buds on every shoot. They also found the 8 hour photoperiod produced plants with a more vibrant purple leaf color than the 12 or 16 hour photoperiods.

The floriculture industry utilizes photoperiodic flower initiation for several popular crops (Dole and Wilkins, 1999). Black cloth may be used to block light to shorten the natural photoperiod, called night extension, while supplemental lighting, often from high intensity discharge lamps, can be used to extend the natural photoperiod, called day continuation, resulting in better plant performance for some crops. However, incandescent lights are most often used when the goal is regulation of the natural photoperiod (Dole and Wilkins, 1999). Manipulation of photoperiod has been shown to influence floral bud initiation and development in plants such as chrysanthemum and poinsettia. Using photoperiod manipulation to control flowering on chrysanthemum, a short day plant, Furata (1954) found photoperiods over 14 hours were sufficient to inhibit flowering, while photoperiods less than 14 hours initiated flowering. Differences among cultivar's bud initiation response to photoperiod were examined by Seeley and Weise (1965) who performed a study comparing greenhouse and garden cultivars of chrysanthemum. They found all garden cultivars produced visible flower buds even at 24-hour photoperiods, while greenhouse cultivars formed crown buds or remained vegetative under long photoperiods. The total number of short or long photoperiods required for effective bud initiation varies as well. Post and Kamemoto (1950a) noted for some early chrysanthemum varieties, as few as four short photoperiods caused flower buds to initiate, while Kofranek and Halevy (1974) found one week of short day treatments fully initiated flower buds on chrysanthemums.

The number of photoperiods provided also impacts subsequent flower development. While buds initiate under fewer photoperiods, often additional photoperiod treatments are necessary for optimal flower development. Seeley and Weise (1965)

found photoperiod length influenced development of flower buds on greenhouse and garden cultivars of chrysanthemum, while Kofranek and Halevy (1974) found 3 or 4 weeks of short days were needed to produce high quality flowers on chrysanthemum. Armitage and Laushman (1989) found 14 photoperiod cycles were needed to induce flowering in the SD plant *Salvia leucantha* Cav., while 42 cycles were necessary for normal anthesis and elongation of the raceme. It has also been noted that buds initiated under short days which are then subjected to long days will cease development and fail to develop even if short days are reinstated, provided the long photoperiod treatment is utilized for a sufficient amount of time (Post and Kamemoto, 1950b).

Night interruption (NI), or night break lighting, is another method of manipulating photoperiod by providing the plants with supplemental lighting during the dark period. This process essentially divides the dark period into two “nights” and inhibits or delays floral initiation on short day plants or induces floral initiation on long day plants (Dole and Wilkins, 1999). For many species, NI lighting is more effective at inhibiting or inducing bud initiation than day continuation (Thomas and Vince-Prue, 1997). Stuart (1943) showed supplemental light applied in the middle of the night, as opposed to as an extension of the natural day, was as effective in inhibiting flowering. Cathey and Borthwick (1961) found cycles of lighting throughout the 4-hour night interruption were as effective as continuous illumination, provided the intensity and percentage of illumination per cycle were sufficient. Armitage and Laushman (1989) found night interruption inhibited flower bud formation in the short day plant *Salvia leucantha*.

Adams et al. (1998) found the critical photoperiod for petunia (*Petunia x hybrida* Hort. ex. Vilm.) to be 14.4 hours. Any days longer than this did not further hasten flower

bud initiation. Long (1939) found that the critical photoperiod in *Xanthium pennsylvanicum* Gandoger was affected by temperature as well as the age of the plant. As plants got older, they required a shorter photoperiod to induce flowering.

To date, no information has been published regarding the critical photoperiod of *S. dyerianus*.

Temperature Effects on Plant Growth and Reproduction

Temperature plays an important role in the growth and development of crops. Higher temperatures tend to encourage rapid growth, while cooler temperatures may be used to slow growth. Manipulation of temperature is often used in the floriculture industry to control growth and flowering response of many plant species. A specific temperature treatment, vernalization, is often used in floriculture production. Vernalization is the process of using cold temperature treatments to induce flowering in some species, such as *Tulipa gesneriana* L. and *Lilium* L. or to expedite flowering in plants such as *Echinacea purpurea* (L.) Moench. (Dole and Wilkins, 1999)

Post (1939) found low night temperatures inhibited bud formation in chrysanthemums. A chrysanthemum study by Cathey (1954c) found that a lower night temperature delayed bud formation and higher night temperatures produced the greatest number of flowers. Additionally, plants under all treatments did initiate flowers, though flowers on plants grown at the lowest and highest temperatures did not develop. Cathey (1954c) also noted that temperature provided to plants during the dark period had much more influence on flower initiation than did the temperature provided during the light period and the mean temperature was not correlated with flowering time. For instance,

plants grown at 15°C day and 15°C night temperature flowered in 65 days, while plants grown at 27°C day and 4°C night temperature flowered in 101 days, yet both treatments provided an average temperature of 15°C. Cockshull et al. (1982), however, showed that flower bud initiation in chrysanthemum was influenced by the average temperature over the 24-hour period, not specifically by the day or night temperature.

Cathey (1954b) found that certain varieties of chrysanthemum responded differently to various temperatures; some were induced to flower by low temperatures, while others were inhibited by low temperatures and flowered only under high temperatures. In an earlier study, Cathey (1954a) found that cuttings taken from stock plants of certain cultivars of chrysanthemum grown at various temperatures had different vegetative and reproductive growth. Stock plants of the cultivar Encore were grown at cooler temperatures (10°C and 12°C) produced plants that took longer to flower, by 16 days. Stock plants of the cultivar Shasta, grown under the same temperatures as 'Encore', showed no delay in flowering time. When stock plants of 'Encore' were grown at temperatures of 21°C and 27°C the plants did not respond to the pinch and flowered irregularly.

Photoperiod and Temperature Interactions

These two factors, which individually play roles in bud initiation on some species, can interact with each other to affect a plant's flowering ability. Critical photoperiod can also be affected by varying the temperature. *Xanthium pennsylvanicum* has a critical photoperiod of 8.5 hours at 21°C versus a critical photoperiod of 11 hours at 4°C (Long, 1939). In chrysanthemums, Cathey (1957) found that cooler temperatures extended the

critical photoperiod, while warmer temperatures shortened the photoperiod required for optimum flower development. *Primula malacoides* Hort. responded to temperature by delaying bloom data at higher temperatures (10°C-15°C vs. 15°C-21°C). Bloom time was also delayed at 10°C-15°C temperatures when the photoperiod was extended by 5 hours (Post, 1936). Later studies with *Primula obconica* Hance (Karlsson and Werner, 2002) also found interactions with time to flower and photoperiod and temperature. Flowering was delayed by 11 days when the long day photoperiod was kept consistent (16 hours) but the temperature was decreased from 20°C to 16°C. Similarly, the time to flower was delayed by 11 days when the photoperiod was reduced to short days, or 8 hours, and the temperature held consistent at 16°C.

Ethephon to Keep Plants Vegetative?

The phytohormone ethylene has many effects on most stages of the lifecycle of a plant. In floriculture research, it has been shown to keep plants in the vegetative state by aborting flower buds. This simple, two carbon molecule is involved in a variety of physiological responses, including germination of seeds; tissue differentiation; elongation of roots; flower initiation, opening, abscission and senescence; and fruit ripening and abscission, as well as promotion of flowering in *Bromus mango* Desv. (mango) and several species of *Bromeliaceae* (Salisbury and Ross, 1992). It is synthesized via the Yang cycle from the amino acid methionine, with 1-aminocyclopropane-1-carboxylic acid (ACC) as its immediate precursor, and is produced in plant meristems and senescing and wounded tissues (Beaudry and Kays, 1988).

Ethylene is also commonly referred to as the “stress hormone,” as synthesis of the hormone seems to increase as a result of various abiotic and biotic stressors. While plants produce small amounts of ethylene as part of their normal life cycle, “stress ethylene” production is induced in plants following encounters with stressors. These elevated levels of ethylene in plants have been linked to various physiological and growth responses, including reduced height, increases in the diameter of stems, abscission, rate at which fruit ripens and rapidity of senescence. These ethylene-related responses can be essential to a plant’s ability to survive the stress (Grichko and Glick, 2001). Depending on the species, stress ethylene production may be anywhere from two to 50 times the normal ethylene level, varying with each species’ sensitivity and the degree of stress encountered (Tingey, 1980).

A fairly common reaction of plants to stress is epinasty, in which a bending and contortion of the foliage occurs. Epinastic responses have been observed in plants exposed to a multitude of stressors, from mechanical to water. Epinastic responses of *Lycopersicon esculentum* Mill. (tomato) under waterlogged soil conditions were intensified with increasing ethylene concentrations in the petioles and other plant parts in comparison with the controls (Jackson and Campbell, 1975, 1976). Ethylene-induced epinasty can inhibit carbon dioxide assimilation by reducing the amount of leaf area available to absorb the light necessary for photosynthesis (Woodrow and Grodzinski, 1989). Thus severe epinastic responses in plants due to stress ethylene could lead to reduced growth and possibly failure to initiate flowers and set fruit.

Working with tomatoes exposed to chronic ethylene at either 0, 0.01, 0.05 or 0.1 $\mu\text{L}\cdot\text{L}^{-1}$, Blankenship and Kemble (1996) found increased epinasty in plants grown under

higher amounts of ethylene when compared to the controls, and also found reduced fruit set in plants exposed to $0.05 \mu\text{L}\cdot\text{L}^{-1}$ and virtual elimination of fruit set in plants grown under $0.1 \mu\text{L}\cdot\text{L}^{-1}$. Ethylene in plants therefore has been shown to be active in inducing responses at relatively low concentrations. Ethylene-induced epinasty has also been problematic with poinsettias during postharvest handling where the plants are sleeved, causing a mechanical stress (Saltveit et al., 1979; Staby et al., 1980).

Ethephon (2-chloroethylphosphonic acid), trade name Florel (Monterey Chemical Co., Fresno, Calif.), is a plant growth regulator that causes the release of ethylene gas once inside a plant's tissue. Ethephon works via a mechanism in which a central phosphorus atom found in the ethephon compound is attacked by a water ion or a hydroxyl ion, resulting in the simultaneous elimination of chlorine and release of ethylene (Beaudry and Kays, 1988). Increases in pH tend toward an increase in the ethylene release (Armitage, 1989; Beaudry and Kays, 1988). Translocation of ethephon is thought to occur from source to sink via the phloem pathway, and rates vary with leaf age and location (Beaudry and Kays, 1988).

Florel has an advantage over some other chemical growth regulators because its effects are threefold. In addition to maintaining compact growth through control of internode elongation, it also encourages lateral branching and holds the plant in a vegetative state, even under conditions conducive to promote flowering (Konjoian, 1998). Florel has been used in the production of several crops for many years, including chrysanthemum, geranium and *Fuchsia* L., and an expanded label was approved by the EPA in 1999 to include use on all floriculture crops (Konjoian, 1999). However, to date there was no literature available for the use of Florel on *S. dyerianus*.

Research on Florel's effects on geranium cultivars found treated stock plants yielded more cuttings that were faster rooting and of higher quality than untreated plants (Moorman and Campbell, 1980). Tsujita and Harney (1978) found Florel increased the yield of cuttings obtained from geranium plants, and that the increase was extremely noticeable in stock plants grown under high pressure sodium lighting. However, the authors also found a foliar application of 500 mg·L⁻¹ Florel resulted in a severely dwarfed stock plant, therefore yielding shorter cuttings. Tsujita and Harney (1978) and Samananda et al. (1972) found improvements in the rooting of geranium cuttings and chrysanthemum cuttings, respectively. Pallez and Dole (2001) found ethephon treatments at low rates induced flowering of purple velvet plants (*G. aurantiaca*). However, at rates high enough to maintain the plants in a vegetative state, severe stunting was observed, which made cutting harvest impossible (Pallez and Dole, 2001). Hayashi et al. (2001) found three treatments of ethephon at 1000 mg·L⁻¹ delayed flowering in the herbaceous perennials *Echinacea purpurea* Moench 'Bravado', *Monarda didyma* L. 'Blue Stocking', *Phlox paniculata* L. 'Mt. Fuji', and *Physostegian virginiana* Bentham 'Summer Snow'. Ho et al. (1985) found ethephon applications completely inhibited flowering of *Schlumbergera truncata* (Haw.) Moran.

Despite its status as a facultative short day plant, certain cultivars of chrysanthemum flower quickly under long days, which is problematic for propagators who supply vegetative cuttings (Stanley and Cockshull, 1989). Applications of ethephon to chrysanthemum plants, regardless of application site, delayed flower initiation, reduced apical dominance and stem elongation, and increased the number of leaves (Stanley and Cockshull, 1989). Carpenter and Carlson (1970, 1972a, 1972b) found

ethephon applications of $1000 \text{ mg}\cdot\text{L}^{-1}$ significantly delayed flowering of geraniums and chrysanthemums.

Proper Nutrition for Optimum Plant Growth

Adequate nutrition is essential to produce high quality, marketable plants.

Without proper nutrition, a plant cannot achieve optimal growth and may become more susceptible to stressors (Jones, 1998). Also, fertilizer runoff is a prominent environmental concern. As such, growers are trying to produce plants using the least amount of fertilizer that produces the quality plants the consumer demands. As there is currently no literature available on previous nutrition studies for *S. dyerianus*, determination of fertilization levels for optimum plant performance is essential.

In order to produce high quality, saleable plants, it is important to know the best concentration of fertilizer for the specific plant. Plants that are heavier feeders, such as poinsettias, require a higher concentration of nitrogen to produce quality plants (Whipker et al., 2001). Fertilizer recommendations for poinsettia productions vary in the range of $250 \text{ mg}\cdot\text{L}^{-1}$ to $300 \text{ mg}\cdot\text{L}^{-1}$ nitrogen (N) for constant liquid feed (Ecke et al., 1990). Kuehny et al. (2000) found quality poinsettias can be produced using reduced concentrations of fertilizer, provided a lower leaching fraction is used. Along with most bedding plants, *S. dyerianus* would be classified as a light feeder, with optimum electrical conductivity (soluble salts) levels of $1.0\text{-}2.6 \text{ mS}\cdot\text{cm}^{-1}$, as measured by the pour through method (Whipker et al., 2001). Frett et al. (1985) recommended a fertilizer rate of $200 \text{ mg}\cdot\text{L}^{-1}$ N for *Petunia x hybrida* 'Coral Sea,' which was found to produce the greatest dry weight, branch length and flowering. For *Impatiens wallerana* Hook (double impatiens),

the recommended irrigation rate is 150 to 200 mg·L⁻¹ N, varying by cultivar (Whipker et al., 1999). Campos and Reed (1993) presented a fertilizer range of 100-400 mg·L⁻¹ N for maximum growth in the salt-sensitive tropical plants *Spathiphyllum* Schott and *Dieffenbachia* Schott. Specifically recommended N rates were 100-200 mg·L⁻¹ for *Spathiphyllum* and 200-400 mg·L⁻¹ for *Dieffenbachia*, which gave the greatest leaf area. The authors tested concentrations of nitrogen as high as 3200 mg·L⁻¹ N and found higher concentrations of fertilizer to cause a reduction in growth and decline in the plant's appearance, while the highest concentration, 3200 mg·L⁻¹ killed all plants. As growing media can have an impact on the nutrient availability, a higher level of nitrogen is often recommended to avoid a deficiency in orchids when they are grown in 100% bark, which has little nutrient retention capabilities (Wang, 2000).

A good growing program should utilize information that can be obtained through monitoring the soluble salt content of the media (electrical conductivity or EC) and the concentration of hydrogen ions in the media (pH). Media EC is a measurement of the overall nutritional status of a crop (Wright et al., 1990), while pH expresses the hydrogen ion activity in a solution, an indication of the acidity or alkalinity of a solution (Foth and Ellis, 1997). Both of these measurements, obtained through the pour-through extraction method (Wright, 1986) can be used to determine ranges for optimum growth of plants.

The recommended pH level for soilless media is between 5.4 and 6.0; however this can vary depending on crop type (Dole and Wilkins, 1999). Media pH can be affected by the components of the media, the alkalinity of the tap water in the greenhouse and also the acidity/basicity of the fertilizer being used. Different media components decompose, releasing varying amounts of hydrogen ions. For example, peat moss and

pine bark have very acidic decompositions, while vermiculite is slightly alkaline. A pH that is too far outside of the optimal range can cause problems with nutrient availability, rendering certain nutrients unavailable for uptake by the plant. Melaheberhan et al. (2001) looked at the effect of sub-optimal pH with transplanting seedlings of *Prunus avium* L. At soil pH of 3.9, all seedlings were dead by 4 weeks after transplant, and increased levels of aluminum were found in the tissue with decreased levels of calcium, indicating increased availability and absorption of aluminum by the plants.

EC is monitored by passing electric current through the media leachate. The more salts in the solution the easier a current will pass through the solution. Plants require some concentration of salts in the medium; however excessive salts can lead to “physiological drought.” The condition of physiological drought occurs when high concentrations of soluble salts in the medium inhibit water uptake by the roots because of competitive osmosis (Dole and Wilkins, 1999). Physiological drought results in a wilted plant, despite moist medium. Aside from physiological drought, soluble salt concentrations that are too high or too low can be detrimental to plant growth and saleability. High ECs may contribute to necrotic leaf margins, poor or erratic rooting of cuttings and increased vulnerability to diseases effecting the root and crown (Dole and Wilkins, 1999). Morvant et al. (1997) found that lower concentrations of soluble salts in the medium encouraged root growth, while higher concentrations inhibited root growth. While some plants may show adverse effects from a high EC, some species can tolerate a higher EC level. One such plant is *Viola x wittrockiana* Gam., which exhibited optimal growth at a leachate EC ranging from 1.5 to 4.0 dS·m⁻¹ (van Iersel, 1999).

Foliar analysis is often used to determine the elemental status of the plant, which can then be used to recommend an appropriate fertilizer range, set sufficiency averages or ranges and to examine potential nutritional problems. Whipker and Hammer (1997) looked at hydroponically grown poinsettias for patterns in nutrient uptake. They found increasing the concentration of nitrogen and potassium in the fertilizer three-fold did not significantly affect uptake of phosphorus, potassium, calcium, magnesium, sodium, boron, copper, iron, manganese, molybdenum or zinc, but did influence the uptake of the ammonium ion form of nitrogen. From these data, they presented rates for production of poinsettias without leaching in the northern United States. Simmone et al. (1999) proposed sufficiency ranges for *Loropetalum* R. Br. using slow release fertilizers based on leaf analysis. Kovacic and Holcomb (1981) used five controlled-release fertilizers applied either as a top-dressing or incorporated into the media of subirrigated *Kalanchoe blossfeldiana* v. Poelln. 'Pixie.' Fertilizers and application method were evaluated for optimum plant performance. Leaf elemental concentrations were measured and presented in ranges for high quality kalanchoe plants. At present, no sufficiency averages or ranges have been published for *S. dyerianus*.

This research will examine environmental factors and their impacts on production of *S. dyerianus*. Predominantly, the research will focus on photoperiod and temperature to control unwanted flower production. Ethephon applications will be examined as a possible alternative to manipulation of environmental conditions as a method of inhibiting floral initiation. As there are no published data concerning the nutritional requirements of *S. dyerianus*, this experiment will also look at the species response to

levels of nitrogen in the fertilizer. All of the conclusions from this research will be combined to provide growers with guidelines for successful *S. dyerianus* production.

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CHAPTER 2: DETERMINATION OF THE APPROXIMATE CRITICAL PHOTOPERIOD AND EFFECTS OF TEMPERATURE ON FLORAL INITIATION OF *STROBILANTHES DYERIANUS* MAST.

Abstract. *Strobilanthes dyerianus* Mast. (Persian Shield) is a popular foliage annual in summer landscapes, where it rarely flowers. However, stock plants of *S. dyerianus* have been observed to flower in the winter in greenhouses, indicating potential photoperiod or temperature effects. Once it becomes reproductive, the plant quality declines and propagation by vegetative cuttings becomes difficult. The objective of this research was to determine the critical photoperiod under which *S. dyerianus* initiated buds and if manipulation of environmental factors could be used to keep stock plants in a vegetative state. The first experiment was performed in a glass greenhouse, with black cloth pulled over the benches to obtain the following photoperiods: 8 h, 10 h, 12 h and a 4 h night interruption. Plugs were potted in 2.8 L containers using soilless media, grown for 4 weeks under an extended photoperiod, and then placed under the black cloth treatments. None of the photoperiod treatments were significant for inhibiting flowering, and bud initiation was sporadic within the treatments. Shoot dry weight was positively correlated with bud initiation, and leaf area was positively correlated with bud initiation. The second experiment was held at the Southeastern Plant Environment Laboratory at N.C. State University. Plugs were transplanted into 15.24 cm pots using soilless media and grown for 10 days under 14 h photoperiods, before being placed in growth chambers. Reach in growth chambers were used to create three

photoperiods (8 h, 12 h and 16 h) with two day/night temperature regimes (24°C / 21°C and 17°C / 14°C) for a total of six treatments. Bud initiation was extremely sporadic, with only five out of 60 plants becoming reproductive by termination of the experiment at 15 weeks. Neither photoperiod nor temperature had a significant effect on bud initiation, and neither shoot dry weight nor leaf area were correlated with bud initiation.

Introduction

Strobilanthes dyerianus Mast., Persian Shield, has become a popular and successful ornamental foliage crop in recent years. It is part of the Athens Select™ program, a collection of plants demonstrating superior performance under hot and humid conditions. As a landscape plant, the tropical perennial *S. dyerianus* is particularly attractive with its purple-green foliage and can grow to a small shrub, hardy to USDA zone 9. It is currently the only representative of this genus easily obtainable through garden centers (Armitage, 2001).

Floral initiation on *S. dyerianus* in the landscape is generally not a concern, as flowers rarely appear and do not contribute to the aesthetics of the plant (Armitage, 2001). However, stock plants of *S. dyerianus* may become reproductive, rendering propagation extremely difficult, as the primary propagation method used for *S. dyerianus* is vegetative cuttings (personal communication with growers). Once *S. dyerianus* reaches the reproductive stage, all stem elongation ceases and buds arise from every node (Batson, 1982). Once the plant becomes reproductive, it does not easily revert to the vegetative stage (Batson, 1982). According to personal communication with growers and

personal observation, bud initiation seems to be associated with the onset of winter.

While the exact mechanism behind floral initiation is not known, the observed winter-time flowering indicates possible reproductive triggers could be photoperiod, temperature or a combination of the two.

Plants may be categorized as long day (LD), short day (SD) or day neutral (DN) based on their flowering response to photoperiod. Photoperiodic plants may be further classified as facultative (quantitative) or obligate (qualitative) SD or LD. A facultative SD or LD plant has the ability to flower under a wide range of daylengths, but flowers are produced more rapidly under either short or long days. A plant classified as obligate SD or LD flowers only under a specific photoperiod. Furata (1954) noted that while *Dendranthema x grandiflorum* Kitam. (chrysanthemum) plants formed flower buds in a 14 hour photoperiod, shorter photoperiods (less than 13 hours) resulted in rapid flower bud development, thereby labeling chrysanthemum as a facultative SD plant.

Night interruption (NI), or night break lighting, is a method of manipulating photoperiod by providing plants with supplemental lighting during the dark period. This process essentially divides the dark period into two “nights” and inhibits or delays floral initiation on short day plants or induces floral initiation on long day plants (Dole and Wilkins, 1999). For many species, NI lighting is more effective at inhibiting or inducing bud initiation than day continuation (Thomas and Vince-Prue, 1997). To date, no information has been published regarding the critical photoperiod of *S. dyerianus*.

Temperature plays an important role in the growth and development of crops. Higher temperatures tend to encourage rapid growth, while cooler temperatures may be

used to slow growth. Temperature is often a trigger for floral initiation. Certain species require vernalization, or a cool temperature treatment, in order to flower. Other species, which are capable of flowering without a cold treatment, flower more rapidly when given a vernalization treatment (Dole and Wilkins, 1999). Temperature manipulation is often used in the floriculture industry to control growth and flowering response of many plant species. Chrysanthemums have been shown to illustrate a range of responses varying with cultivar. Cathey (1954b) noted that certain varieties responded differently to various temperatures; some were induced to flower by low temperatures, while others were inhibited by low temperatures and flowered only under high temperatures. Post (1939) found inhibition of bud formation in chrysanthemums was possible with low nighttime temperatures.

These two environmental factors, which individually play roles in bud initiation on some species, can interact with each other to affect a plant's flowering ability. Critical photoperiod can be affected by varying the temperature. *Xanthium pennsylvanicum* Gandoger has a critical photoperiod of 8.5 hours at 21°C (70°F) versus a critical photoperiod of 11 hours at 4°C (40°F) (Long, 1939). In chrysanthemums, Cathey (1957) found that cooler temperatures extended the critical photoperiod, while warmer temperatures shortened the photoperiod required for optimum flower development.

The objectives of this research were to (1) determine the critical photoperiod for *S. dyerianus*, (2) examine inhibition of flowering through photoperiod manipulation, and (3) determine potential impacts of greenhouse temperature on the production of *S. dyerianus* as a floriculture crop.

Materials and Methods

Greenhouse Photoperiod Experiment

On 10 Oct. 2001, 126 *S. dyerianus* plugs provided by EuroAmerican Propagators (Bonsall, Calif.) were potted in 2.8 L (17.2 cm by 18.4 cm) containers using a potting mix consisting of peat moss, processed pine bark, perlite and vermiculite (Fafard 3B soilless potting mix, Fafard, Inc., Anderson, S.C.) and were irrigated as necessary with 15N-2.2P-12.4K (Miracle-Gro Excel 15-5-15 Cal-Mag, The Scotts Co., Marysville, Ohio) at 200 mg·L⁻¹ nitrogen as a constant liquid feed. The experiment took place in a glass greenhouse on the Virginia Tech campus in Blacksburg, Va. The average high temperature in the greenhouse during the experiment was 22°C ± 5°C (72°F ± 10°F) and the average low was 13°C ± 3°C (56°F ± 6°F). Substrate pH and EC (electrical conductivity) were monitored every 2 weeks using the pour through extraction method (Wright, 1986). The plants were grown on under supplemental lighting consisting of four 100 watt incandescent bulbs set on timers (from 1600 to 2200 HR) to extend the daylength to approximately 14 h for 4 weeks.

Four weeks after transplant, 36 plants were selected for uniformity of height and approximate leaf area. Plants were treated with four photoperiods, each on a separate bench: 8 h, 10 h, 12 h and 4 h night interruption, and there were nine replications on each bench. Benches were set up with black cloth that could be pulled over the plants to regulate photoperiod. Cloth was pulled off between 0700 and 0900 HR each morning and benches were recovered 8 h later. One-hundred watt incandescent bulbs were placed under the 10 h, 12 h and night interruption cloth; the night temperature under the cloths

was approximately 6°C higher than the air temperature in the greenhouse due to the light bulbs under the cloths. The 10 h and 12 h benches received additional supplemental lighting from the incandescent bulb for 2 h and 4 h, respectively, while the night interruption treatment received 4 h of supplemental lighting from 2200 to 0200 HR. Using a LiCor 250 light meter with a LI-190SA Quantum Sensor (Li-Cor, Inc., Lincoln, Nebr.) the photosynthetically active radiation (PAR) from the supplemental light under the cloths at pot level was measured at $3.35 \mu\text{mol}\cdot\text{s}^{-1}\text{m}^{-2}$, with greenhouse PAR light levels measured at $259.2 \mu\text{mol}\cdot\text{s}^{-1}\text{m}^{-2}$. The experiment was terminated 77 days after initiation of treatment (DAIT), at which time, plants were harvested and leaf area recorded using a Li-Cor LI-3000 Portable Area Meter with LI-3050A transparent belt conveyer accessory (Li-Cor, Inc., Lincoln, Nebr.) between 21 Jan. and 23 Jan. 2002. On 23 Jan. plants were placed in a drier at 65.5°C (150°F) for 5 days and on 28 Jan. shoot dry weight was recorded. Data were then analyzed by GLM, correlations and regressions using SAS Version 8 (SAS Institute, Inc., Cary, N.C.).

Growth Chamber Photoperiod Experiment

On 24 Oct. 2002, 117 *S. dyerianus* plugs (84 cells per tray, EuroAmerican Propagators, Bonsall, Calif.) were potted in 15.2 cm azalea pots (Dillen Products, Middlefield, Ohio) using a soilless potting mix consisting of peat moss, processed pine bark, perlite and vermiculite (Fafard 3B potting media, Fafard, Inc., Anderson, S.C.). Plants were grown on for 10 d using clear water irrigation. A string of six 100 watt incandescent light bulbs was used to extend the photoperiod to 14 h.

On 4 Nov., 10 days after transplant (DAT), 60 uniform plants were selected and transported to the phytotron at N.C. State University (Raleigh, N.C.), where they were fumigated with nicotine sulfate as part of facility regulation. Plants were placed in reach-in growth chambers 11 DAT. The chambers had 16 fluorescent tubes and 6 incandescent bulbs for a total photosynthetic photon flux density of $411 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$. As cool white fluorescent accentuates blue, green and yellow wavelengths, and incandescent accentuates red and far red, the quality of light for plants in the growth chamber should be comparable to that produced by sunlight. The growing space in the chambers was 0.91×1.22 m with a vertical height of 1.22 m. Temperatures may vary from the set point by $\pm 0.5^\circ\text{C}$. Treatments consisted of three photoperiods, 8, 12 and 16 h, with two temperature regimes, $17^\circ\text{C}/14^\circ\text{C}$ and $24^\circ\text{C}/21^\circ\text{C}$ day/night temperature, with 10 reps of each. Day and night temperature corresponded with the photoperiod: for example, under the 8 h warm treatment, plants received 8 h at 24°C and 16 h at 21°C . Plants were irrigated with a solution consisting of $40.44 \text{ g}\cdot\text{L}^{-1}$ potassium nitrate, $16 \text{ g}\cdot\text{L}^{-1}$ ammonium nitrate, $4.8 \text{ g}\cdot\text{L}^{-1}$ potassium phosphate (mono), $5.6 \text{ g}\cdot\text{L}^{-1}$ potassium phosphate (diabasic), $6.0 \text{ g}\cdot\text{L}^{-1}$ potassium sulfate, $6.8 \text{ g}\cdot\text{L}^{-1}$ sodium sulfate, $0.28 \text{ g}\cdot\text{L}^{-1}$ boric acid, $0.002 \text{ g}\cdot\text{L}^{-1}$ molybdic acid, $0.018 \text{ g}\cdot\text{L}^{-1}$ hampene zinc, $0.0816 \text{ g}\cdot\text{L}^{-1}$ manganous chloride, $0.012 \text{ g}\cdot\text{L}^{-1}$ hampol copper, $0.0004 \text{ g}\cdot\text{L}^{-1}$ sequestrene cobalt and $0.3 \text{ g}\cdot\text{L}^{-1}$ uranine (Phytotron Procedural Manual, 1991). Fertigation occurred Monday, Wednesday and Friday for 4 weeks, at which point fertigation was increased to 5 days a week. Data collected included days to visible bud, initial and final node count, with leaf area, shoot dry weight and overall plant quality being measured upon termination of the experiment, 15 weeks after initiation of treatment (WAIT). Plant quality was rated on a scale of 0 to 4 with 0

being dead, 1 having severe chlorosis and/or necrosis, 2 having moderate chlorosis and/or necrosis, 3 having mild chlorosis and/or necrosis, and 4 being an excellent looking plant with little or no chlorosis and/or necrosis. Data were analyzed using t-tests (LSD), GLM and correlations in SAS Version 8 (SAS Institute, Inc., Cary, N.C.).

Results and Discussion

Greenhouse Experiment

Bud initiation was not correlated with photoperiod treatment; however, there was a positive correlation between shoot dry weight and flowering and a positive correlation between leaf area and flowering (Table 2.1). Plants under each treatment flowered, with the earliest visible buds on the 8 h treatment at 51 DAIT. The last treatment to show visible buds was the NI, which showed buds 68 DAIT. Newly budded plants were continually observed on the NI treatment until 75 DAIT, while no new plants initiated buds for the duration of the experiment in the other three treatments. Thirty-three percent of plants in the study had visible buds at termination of the experiment. The NI treatment had the most plants in bud with 56%; the 8 h was 33%, 10 h was 22% and 12 h was 33% (Figure 2.1).

Photoperiod had no statistically significant effect on shoot dry weight, but longer photoperiods produced plants with a larger leaf area (Table 2.2). Means for shoot dry weight were not statistically different between the photoperiod treatments; however, there was a significant difference in the mean leaf area of the NI (long day) treatment and the 8 h treatment.

Growth Chamber Experiment

Photoperiod was not correlated with bud initiation, nor was temperature correlated with bud initiation (Table 2.3). Bud initiation was sporadic, with only 8% of the plants showing visible buds by termination of the experiment (five out of 60 plants). There was no correlation between the shoot dry weight and bud initiation or between leaf area and bud initiation (Table 2.4). There was also no correlation between node number and bud initiation (data not presented).

Photoperiod and temperature each had significant effects on shoot dry weight and leaf area (Table 2.5). The significant interactions were due to the fact that under the 12 h photoperiod there were one or two plants with lower shoot dry weight or leaf area than other plants grown under that treatment. This caused the data points to cross, resulting in an interaction (Figure 2.2 and Figure 2.3). Growing plants under warmer temperatures (24°C day / 21°C night) resulted in higher shoot dry weight and leaf area than the cooler treatment (Table 2.6 and 2.7). Plants grown under the 8 or 12 h photoperiod were significantly smaller than those produced under the 16 h photoperiod, regardless of temperature, but there was no significant difference between the 8 or 12 h photoperiod. Pearson et al. (1995) also found the half-hardy perennial *Osteospermum jucundum* (Phillips) Norlindh grew significantly larger under a 16 h photoperiod than under an 8 h photoperiod. The means of shoot dry weight were significantly different under all 3 photoperiods. The leaf area was statistically the same for the 8 and 12 h photoperiod, but the 8 and 12 h were statistically different than the 16 h treatment.

Photoperiod and temperature both had a significant effect on overall plant quality (Table 2.8). Plants produced under warmer temperatures showed a deeper purple leaf than those under cool temperatures, regardless of photoperiod. However, quality ratings were based on necrosis. The cooler temperature treatment had more necrosis than the warmer temperature, hence lower quality in the cooler treatment. While larger plants were produced under the 16 h treatment, the 8 h and 12 h photoperiod received higher quality ratings because they had less necrotic leaves (Table 2.9). The interaction between photoperiod and temperature can be explained by variations in the amount of necrosis on plants under the same treatments.

Strobilanthes dyerianus is a tropical plant, therefore the fact that the largest plants were produced under longer photoperiods and warmer temperatures is not a surprise. This further reinforces observations of better plant performance under warmer temperatures (Armitage, 1997), though the temperatures tested in this experiment were not comparable to those encountered by plants in a summer landscape.

The erratic bud initiation was consistent throughout all the treatments. This indicates that a factor other than photoperiod triggers the reproductive mechanism in *S. dyerianus*. Armitage (1988) found photoperiod induced flowering in *Trachelium caeruleum* Linn., a plant produced for cut flowers, but did not gain 100% bud initiation under the critical photoperiod. Armitage postulated another factor contributed to floral initiation, and noted that 100% flower initiation had been achieved with injections of $1000 \text{ mg}\cdot\text{L}^{-1} \text{ CO}_2$ (Geertsen and Bredmose, 1987).

The results of the greenhouse experiment indicated a potential link between plant size and flowering, based on positive correlations between shoot dry weight and flowering and between leaf area and flowering. Furata (1954) found flower bud initiation in chrysanthemum was correlated to growth in non-inductive photoperiods. Many ornamental crops exhibit flowering responses due to juvenility, or the period in a plant's life where it is insensitive to any conditions that would normally induce floral initiation (Bernier et al., 1981). Several floriculture crops can only perceive photoperiods after they have reached maturity (Dole and Wilkins, 1999). More often, plants exhibit an increasing sensitivity to reproductive triggers throughout their lifespan, and are rarely completely inhibited from flowering. Lyons and Booze-Daniels (1986) found *Eschscholzia californica* Cham. (California poppy) became increasingly sensitive to LD photoperiods as the plant aged, and at least 10 expanded leaves were necessary for the shortest time to flower; however, the plants were never entirely insensitive to photoperiod. They also found the amount of photosynthetic leaf area was not as critical to flowering as leaf number, indicating a potential influence of specific number of leaves on the flowering response. The highly positive correlation between plant size and flowering in the greenhouse experiment supports the theory that *S. dyerianus* responds after a period of juvenility. However, the data from the growth chamber experiment did not support the data from the greenhouse experiment, with growth chamber data showing no correlation between size of the plants and flowering.

Another flowering mechanism is light accumulation. *Pelargonium x hortorum* L.H. Bail. (geranium) exhibits flowering responses depending on the amount of total light energy received at the proper temperatures, but is otherwise classified as a DN plant

(Langton and Runger, 1985). Geraniums undergo a period of juvenility after which the amount of light is responsible for bud initiation and early growth (Bethke and Carlson, 1985). In the case of *S. dyerianus*, with plugs arriving from the same grower and being placed under the same conditions, it is doubtful that light accumulation was significantly higher for 33% of the plants in the greenhouse experiment or 8% of the plants in the growth chamber experiment.

Data from the greenhouse experiment showing days to visible bud indicate that *S. dyerianus* is a SD plant. Plants under the 8 h treatment did develop buds first, with a delay of 2 weeks before the next flower buds were noted. However, the fact that the NI, or long day, treatment continued to initiate buds until termination of the experiment indicates a facultative LD plant. The results from the growth chamber experiment were not as conclusive. A single plant in the 8 h, cool temperature (17°C / 14°C) treatment budded first, followed by one plant under the 12 h, cool temperature treatment 3 days later, and a week later one plant under the 16 h, cool temperature treatment. There was a 5 week delay before the next buds were noted, on two plants under the 8 h, warm treatment (24°C / 21°C). This does indicate a temperature effect on bud initiation: higher temperatures delayed bud initiation. Cathey (1954c) found that a lower night temperature delayed bud formation in chrysanthemums. Cathey (1954c) also noted that temperature provided to plants during the dark period had much more influence on flower initiation than did the temperature provided during the light period and the mean temperature was not correlated with flowering time. For instance, plants grown at 15°C day and 15°C night temperature flowered in 65 days, while plants grown at 27°C day and 4°C night temperature flowered in 101 days, yet both treatments provided an average temperature

of 15°C. Cockshull et al. (1982), however, showed that flower bud initiation in chrysanthemum was influenced by the average temperature over the 24-hour period, not specifically by the day or night temperature. *Primula obconica* Hance (Karlsson and Werner, 2002) found interactions with time to flower and photoperiod and temperature. Flowering was delayed by 11 days when the long day photoperiod was kept consistent (16 hours) but the temperature was decreased from 20°C to 16°C. Similarly, the time to flower was delayed by 11 days when the photoperiod was reduced to short days, or 8 hours, and the temperature held consistent at 16°C. Gartner and McIntyre (1957) found flowering of greenhouse-grown poinsettias could be delayed by using supplemental light. The amount of light was influenced by the temperature, with an additional month of lighting needed if the temperature was increased above 15°C in order to maintain the vegetative state of the plant and allow them to bloom for the Christmas holiday. The data from the two photoperiod experiments on *S. dyerianus* showed sporadic bud initiation under all treatments indicating an unclear flowering mechanism, but one that could potentially be linked to photoperiod and temperature.

At no time during this experiment was one-hundred percent bud initiation observed. It appears to be a facultative SD plant; however critical photoperiod could not be established due to sporadic bud initiation and conflicting results from the two experiments. It cannot be stated with certainty what affect photoperiod has on the flowering of *S. dyerianus* based on the data from this research. Likewise, a temperature effect on flowering cannot be ruled out either. Lower temperatures appeared to induce flowering in the growth chamber experiment, while warmer temperatures gave improved plant color and growth. Erratic bud initiation in these experiments indicates potential for

an additional factor at work in the bud initiation of *S. dyerianus*. Further work could examine potential residual effects on the cuttings from stock plants, as was noted by Cathey (1954a), who found that cuttings taken from stock plants of certain varieties of chrysanthemum grown at various temperatures had different vegetative and reproductive growth. Stock plants of the variety Encore were grown at cooler temperatures (10°C and 12°C) produced plants that took longer to flower, by 16 days. Stock plants of the variety Shasta, grown under the same temperatures as Encore, showed no delay in flowering time. When stock plants of Encore were grown at temperatures of 21°C and 27°C the plants did not respond to the pinch and flowered irregularly.

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Table 2.1. Correlation coefficients for bud initiation of *Strobilanthes dyerianus* and photoperiod, shoot dry weight and leaf area in the greenhouse photoperiod experiment. Shoot dry weight and leaf area were measured at harvest, 11 weeks after initiation of treatment.

	Bud initiation	P-Value
Photoperiod	0.16	0.36 ^{NS}
Shoot dry weight	0.78	<0.0001*
Leaf area	0.74	<0.0001*

NS, * Not significant or significant at P=0.05.

Table 2.2. Mean shoot dry weight and leaf area for *Strobilanthes dyerianus* treated with photoperiod, harvested at termination of the experiment, 11 weeks after initiation of treatment, in the greenhouse photoperiod experiment. (n=9)

Source	Photoperiod	Mean
Leaf area (cm ²)	NI ^z	1463 a
	12 h	1278 ab
	10 h	1178 ab
	8 h	1112 b
	LSD	311
P-Value		0.02*
Shoot dry weight (g)	NI	13.1 a
	12 h	12.7 a
	10 h	10.9 a
	8 h	10.9 a
	LSD	2.97
P-Value		0.07 ^{NS}

Means followed by the same letter are not different by LSD.

^z NI=night interruption treatment.

*,^{NS} Significant or not significant at P=0.05.

Table 2.3. Analysis of variance for bud initiation of *Strobilanthes dyerianus* treated with photoperiod and temperature for the growth chamber photoperiod experiment after termination of the experiment, 15 weeks after initiation of treatment.

Source	DF	Mean Square	P-Value
Photoperiod	2	0.07	0.44 ^{NS}
Temperature	1	0.02	0.65 ^{NS}
PPD x Temp	2	0.07	0.44 ^{NS}

^{NS} Not significant at P=0.05.

Table 2.4. Correlation coefficients for bud initiation and shoot dry weight and leaf area of *Strobilanthes dyerianus* harvested 15 weeks after initiation of treatment in the growth chamber photoperiod experiment.

	Flowering	P-Value
Shoot dry weight	-0.07	0.6 ^{NS}
Leaf area	-0.06	0.7 ^{NS}

^{NS} Not significant at P=0.05.

Table 2.5. Analysis of variance for the shoot dry weight and leaf area of *Strobilanthes dyerianus* treated with photoperiod (PPD) and temperature (Temp), harvested 15 weeks after initiation of treatment, in the growth chamber photoperiod experiment.

Source	DF	Mean Square	P-Value
Shoot dry weight			
PPD	2	2519	<0.0001*
Temp	1	8029	<0.0001*
PPD x Temp	2	617	<0.0001*
Leaf area			
PPD	2	9682717	<0.0001*
Temp	1	66315415	<0.0001*
PPD x Temp	2	3410112	0.0002*

* Significant at P=0.05.

Table 2.6. Mean shoot dry weight for *Strobilanthes dyerianus* treated with photoperiod and temperature, harvested at termination of the experiment, 15 weeks after initiation of treatment, in the growth chamber photoperiod experiment. (n=20 for photoperiod, n=30 for temperature)

Treatment	Shoot dry weight (g)
Photoperiod	
8 h	18.8 a
12 h	24.2 b
16 h	40.4 c
LSD	0.179
Temperature	
17°C	39.4 a
24°C	16.2 b
LSD	3.11

Means followed by the same letter are not different by LSD.

Table 2.7. Mean leaf area for *Strobilanthes dyerianus* treated with photoperiod and temperature, harvested at termination of the experiment, 15 weeks after initiation of treatment, in the growth chamber photoperiod experiment. (n=20 for photoperiod, n=30 for temperature)

Treatment	Leaf area (cm ²)
Photoperiod	Mean
8 h	1765 a
12 h	1937 a
16 h	3047 b
LSD	368
Temperature	
17°C	1198 a
24°C	3301 b
LSD	300

Means followed by the same letter are not different by LSD.

Table 2.8. Analysis of variance for overall plant quality rating of *Strobilanthes dyerianus* treated with photoperiod (PPD) and temperature (Temp) for the growth chamber experiment.

Source	DF	Mean Square	P-Value
PPD	2	5.40	<0.0001*
Temp	1	1.67	0.03*
PPD x Temp	2	4.07	<0.0001*

* Significant at P=0.05.

Table 2.9. Mean overall plant quality ratings for *Strobilanthes dyerianus* treated with photoperiod and temperature, harvested at termination of the experiment, 15 weeks after initiation of treatment, in the growth chamber photoperiod experiment. (n=20 for photoperiod, n=30 for temperature)

Treatment	Plant quality rating
Photoperiod	
	Mean
8 h	3.1a
12 h	3.1 a
16 h	2.2 b
LSD	0.38
Temperature	
17°C	2.6 a
24°C	3.0 b
LSD	0.31

Means followed by the same letter are not different by LSD.

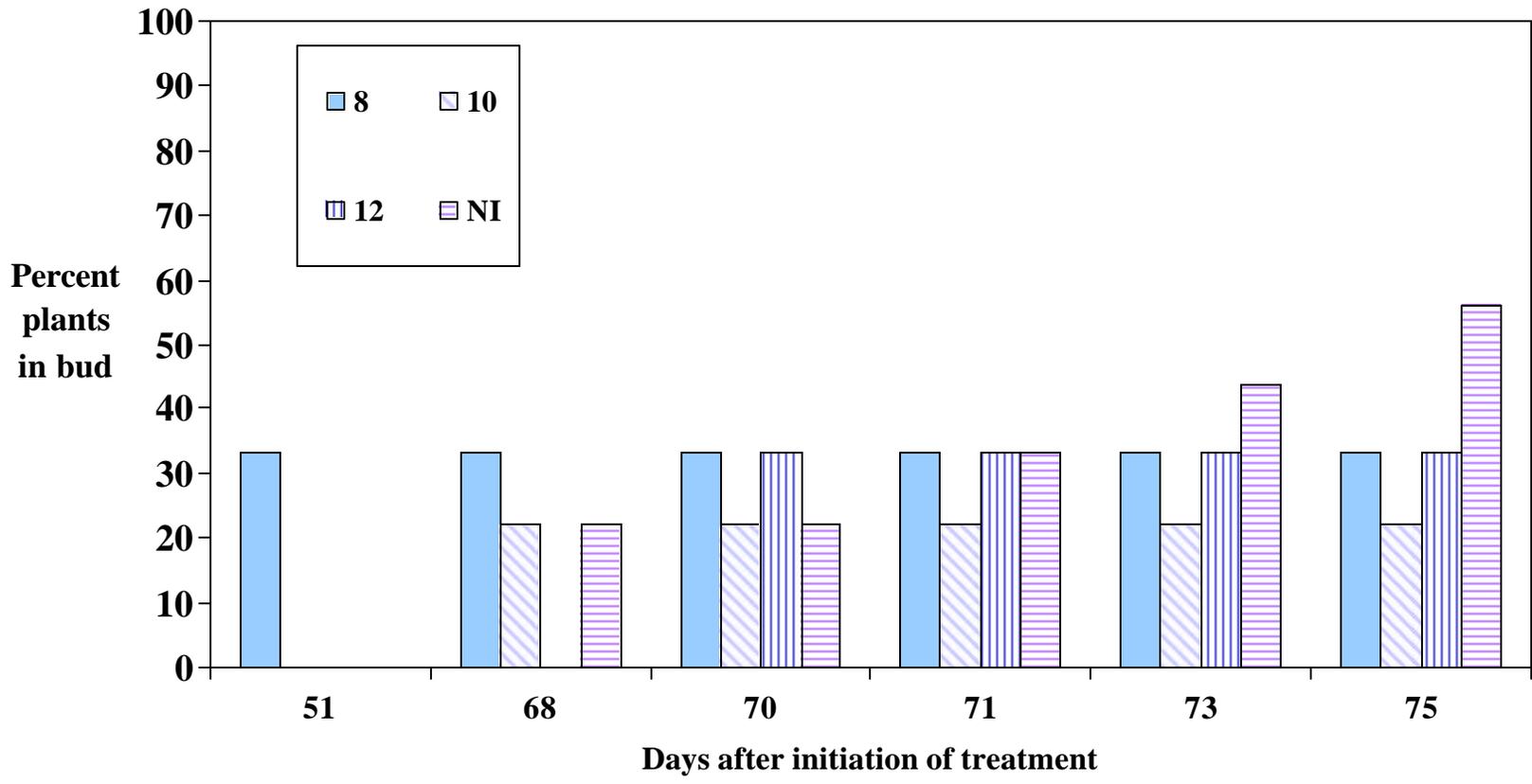


Figure 2.1. Percent of *Strobilanthes dyerianus* plants showing visible buds under each photoperiod (8 h, 10 h, 12 h or 4 h night interruption (NI)) in the greenhouse experiment. The first buds were noted 51 days after treatment initiation in the 8 h photoperiod and the experiment was terminated after 75 days.

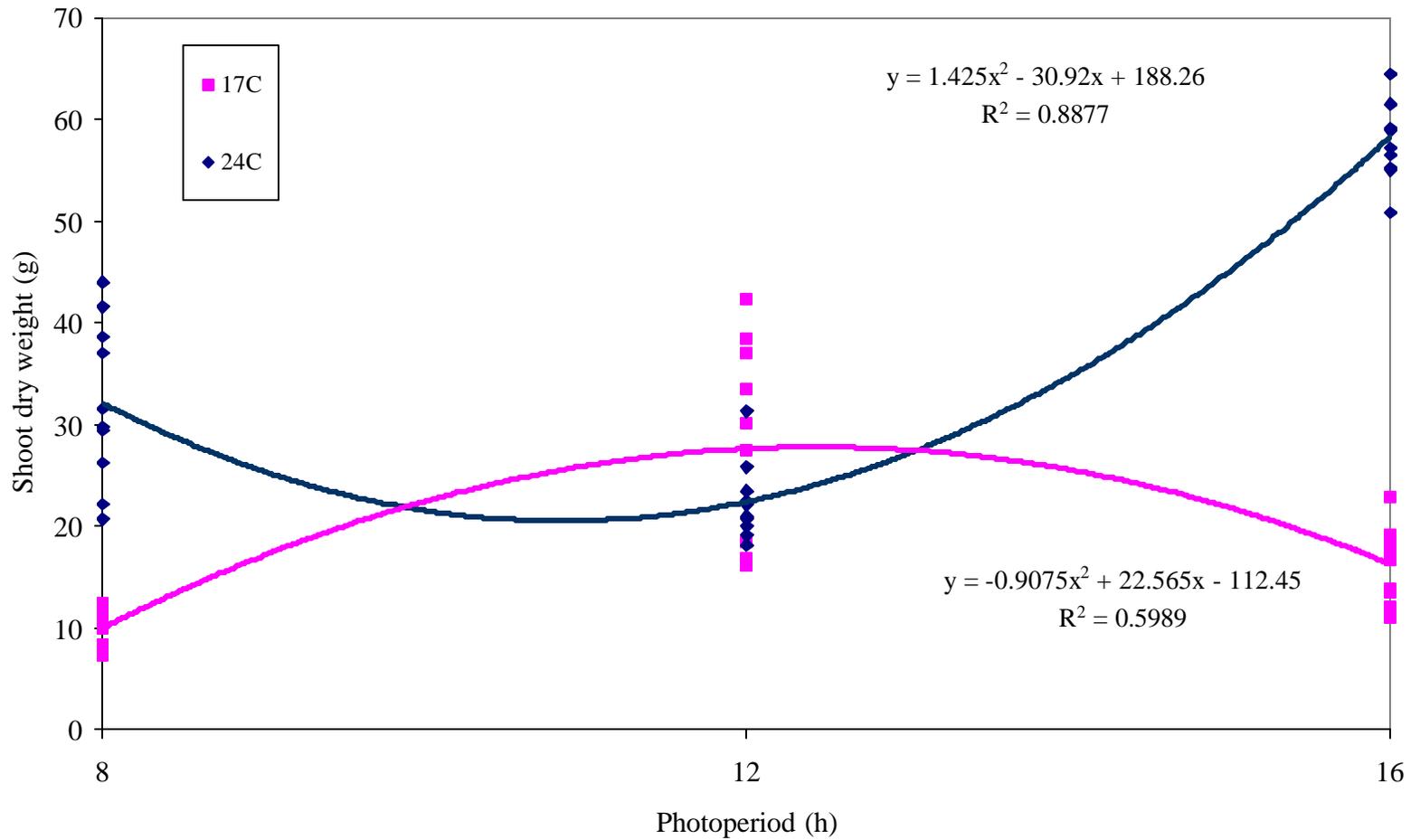


Figure 2.2. Shoot dry weight (g) of *Strobilanthes dyerianus* by photoperiod and temperature treatment, measured 15 weeks after initiation of treatment in the growth chamber experiment.

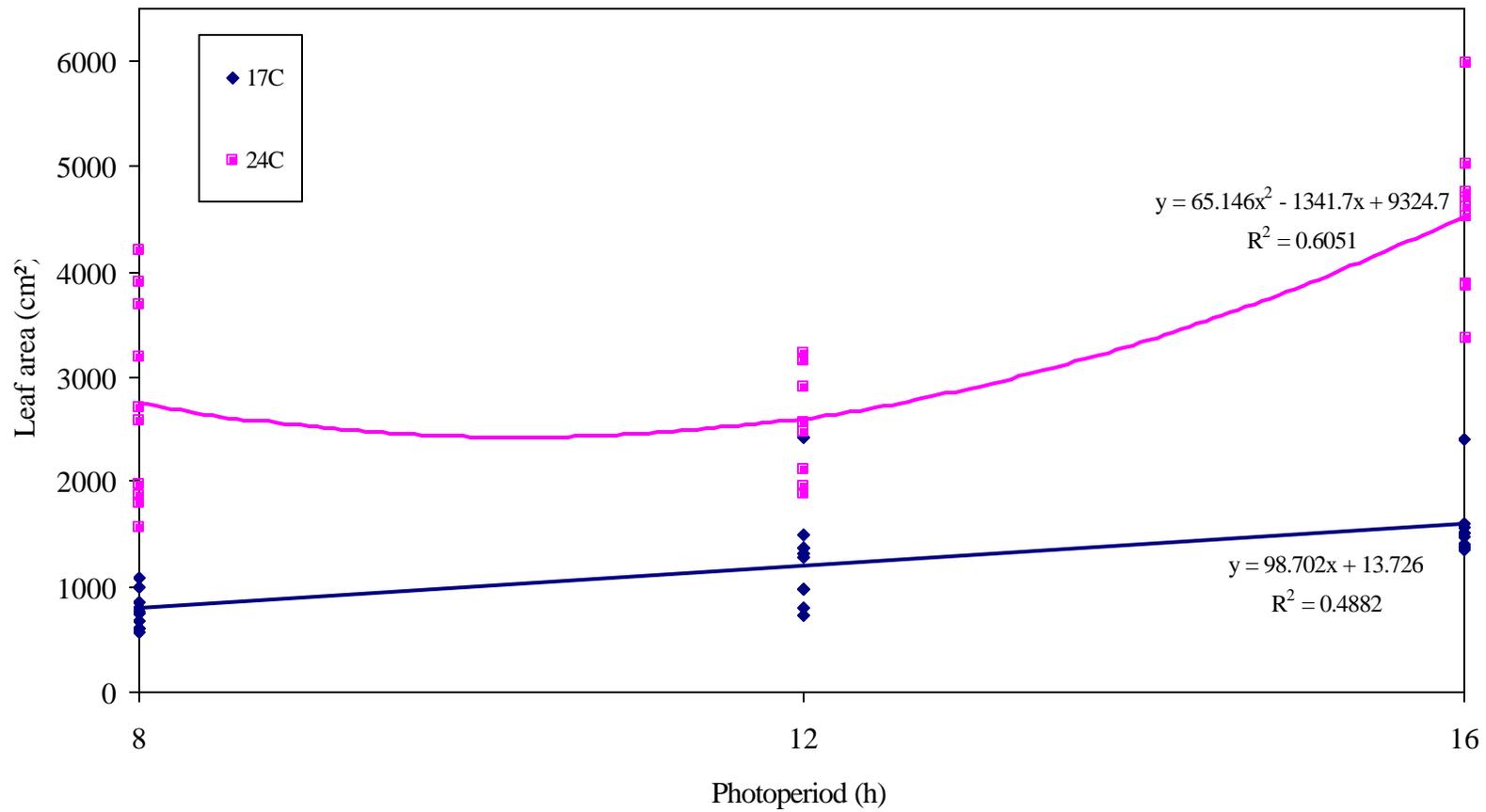


Figure 2.3. Leaf area (cm²) of *Strobilanthes dyerianus* by photoperiod and temperature treatment, measured 15 weeks after initiation of treatment in the growth chamber experiment.

CHAPTER 3: USING ETHEPHON SPRAYS TO MAINTAIN VEGETATIVE STOCK PLANTS OF *STROBILANTHES* *DYERIANUS* MAST.

Abstract. The purpose of this research was to determine the effectiveness of ethephon applications at various rates and frequencies on inhibiting flower bud formation in *Strobilanthes dyerianus* Mast., a popular annual in the landscape. Stock plants grown in the greenhouse have been observed to flower in conjunction with the onset of winter. When the plant becomes reproductive, stem elongation ceases and flower buds arise from every node, rendering propagation by vegetative cuttings extremely difficult. Ethephon, an ethylene releasing compound, has been found to keep some plants in a vegetative state. Plugs were transplanted into 2.8 L containers using a soilless potting mix. Plants were grown under an extended photoperiod for 4 weeks, at which point plants were treated with 0, 150, 300, 450, 600 or 750 mg·L⁻¹ ethephon at weekly, biweekly or monthly frequencies. The plants were then grown under an 8 hour photoperiod for 11 weeks. Ethephon applications were not successful in maintaining vegetative plants at the rates and frequencies used. There was a high positive correlation between leaf area and flowering and between shoot dry weight and flowering.

Introduction

Strobilanthes dyerianus Mast. (Persian Shield) has recently become a popular and successful ornamental foliage crop. It has been marketed as an Athens Select™ plant, a distinction earned due to its exceptional performance under hot and humid conditions (Armitage, 1997). *S. dyerianus* is a tropical perennial, native of Burma in Southeast Asia, and grown for its foliage appeal: large, purple, green and silver leaves. It is used in containers and landscapes in most of the United States as an annual, hardy to USDA Zone 9.

When flowers do appear, they do not contribute to the aesthetics of *S. dyerianus*. Propagation of *S. dyerianus* is mainly by vegetative cuttings, and floral initiation is detrimental to growers, for once it reaches the reproductive stage, all stem elongation ceases and buds arise from every node (Batson, 1982). Post-reproductive stock plants become necrotic and do not easily revert to the vegetative stage (Batson, 1982), reducing the number of cuttings that may be obtained from the plant. According to personal communication with growers and personal observation, bud initiation seems to be associated with the onset of winter, but the mechanism behind floral initiation is not known.

The phytohormone ethylene has many effects on most stages of the lifecycle of a plant. In floriculture research, it has been shown to keep plants in the vegetative state by aborting flower buds (Dole and Wilkins, 1999). Ethylene is a simple, two carbon molecule that is involved in many physiological responses, including germination, root

elongation, and fruit ripening and abscission and flower senescence and is produced in plant meristems and senescing or wounded tissues (Beaudry and Kays, 1988).

Ethephon [(2-chloroethyl) phosphonic acid] is an ethylene-releasing compound found in the plant growth regulator FlorelTM (Monterey Chemical Co., Fresno, CA). Ethephon initiates a release of ethylene gas once inside a plant's tissue (Beaudry and Kays, 1988). It aborts young flower buds, thereby encouraging more vegetative growth (Dole and Wilkins, 1999) and holds the plant in a vegetative state, even under conditions conducive to promote flowering (Konjoian, 1998). Hayashi et al. (2001) found three treatments of ethephon at 1000 mg·L⁻¹ delayed flowering in the herbaceous perennials *Echinacea purpurea* Moench 'Bravado', *Monarda didyma* L. 'Blue Stocking', *Phlox paniculata* L. 'Mt. Fuji', and *Physostegian virginiana* Bentham 'Summer Snow'. Flowering of *Schlumbergera truncata* (Haw.) Moran was also inhibited by ethephon sprays (Ho et al., 1985). Despite status as a facultative short day plant, certain cultivars of *Dendranthema x grandiflorum* Kitam. (chrysanthemum) flower quickly under long days, which is problematic for propagators who supply vegetative cuttings (Stanley and Cockshull, 1989). Applications of ethephon to chrysanthemum plants, regardless of application site, delayed flower initiation, reduced apical dominance and stem elongation and increased the number of leaves (Stanley and Cockshull, 1989). At present, there is no published research regarding the use of ethephon-containing plant growth regulators on *S. dyerianus*.

The objective of this research was to determine if applications of ethephon could be used to maintain vegetative stock plants of *S. dyerianus*, and to determine the optimum rate and frequency of application for *S. dyerianus*.

Materials and Methods

On 10 Oct. 2001, 126 *S. dyerianus* plugs provided by EuroAmerican Propagators (Bonsall, Calif.) were potted in 2.8 L (17.2 cm by 18.4 cm) containers using a potting mix consisting of peat moss, processed pine bark, perlite and vermiculite (Fafard 3B soilless potting mix, Fafard, Inc., Anderson, S.C.) and were irrigated as necessary with 15N-2.2P-12.4K (Miracle-Gro Excel 15-5-15 Cal-Mag, The Scotts Co., Marysville, Ohio) at 200 mg·L⁻¹ nitrogen as a constant liquid feed. The experiment took place in a glass greenhouse on the Virginia Tech campus in Blacksburg, Va. The average high temperature in the greenhouse during the experiment was 22°C ± 5°C and the average low was 13°C ± 3°C. Substrate pH and EC (electrical conductivity, or soluble salts) were monitored every 2 weeks using the pour through extraction method (Wright, 1986). The plants were grown on under supplemental lighting consisting of four 100 watt incandescent bulbs set on timers (from 1600 to 2200 HR) to extend the daylength to approximately 14 h for 4 weeks, at which point 90 plants were selected for uniformity of height and approximate leaf area.

The experimental design was a completely randomized design with a factorial treatment of six rates of plant growth regulator (PGR) and three frequencies of applications; there were five replications of each treatment. Data taken included initial and final height, plus overall plant quality rating, epinasty rating, leaf area and shoot dry weight, which were measured at conclusion of experiment. Plant quality ratings were taken on a scale of 0 to 4, with 0 being dead, 1 having severe chlorosis and/or necrosis, 2 having moderate chlorosis and/or necrosis, 3 having mild chlorosis and/or necrosis, and 4 being an excellent looking plant with little or no chlorosis and/or necrosis. Epinasty

ratings were on a scale of 0 – 3, with 0 being no epinasty present, 1 being mild epinasty, 2 being moderate epinasty and 3 being severe epinasty.

On 5 Nov. plants were sprayed with 0, 150, 300, 450, 600 or 750 mg·L⁻¹ ethephon (Florel, Monterey Chemical Co., Fresno, Calif.) using a hand-held CO₂ pressurized sprayer (R&D Sprayer, Opelousas, La.) with an 800VS nozzle. PGR was evenly applied at 210 mL·m⁻². Plants were sprayed weekly (10 total applications), biweekly (6 total applications) or monthly (3 total applications). Environmental conditions for each spray event are shown in Table 3.1. Black cloth was pulled over the benches from 1700 to 0900 HR to achieve an 8 h photoperiod from 6 Nov. 2001 to 21 Jan. 2002. On 21 Jan., 11 weeks after initiation of treatment, plants were harvested and leaf area was measured between 21 Jan. and 23 Jan. with a Li-Cor LI-3000 Portable Area Meter with LI-3050A transparent belt conveyer accessory (Li-Cor, Inc., Lincoln, Nebr.). Plants were then dried at 65.5°C for 5 days, and dry weight measured on 28 Jan. Height data was not analyzed due to highly variable growth habits. Data were analyzed by correlations and GLM ANOVA using SAS Version 8 (SAS Institute, Inc., Cary, N.C.).

Results and Discussion

Floral Initiation

Ethephon applications did not inhibit floral initiation in *S. dyerianus* at the rates tested, and there was no significant effect on bud initiation by rate or frequency (Table 3.2). By termination of the experiment, visible buds were noted on some plants in all treatments except the 600 mg·L⁻¹ weekly treatment (Table 3.3). There was no correlation

between ethephon rate and flowering (Table 3.4); however there was a positive correlation between shoot dry weight and flowering, and a positive correlation between leaf area and flowering. Rate of application, but not frequency, had a statistically significant effect on leaf area and shoot dry weight, with leaf area and shoot dry weight decreasing with increasing rates of ethephon (Table 3.5).

If rate was significant on leaf area and shoot dry weight, and leaf area and shoot dry weight were positively correlated with flowering, then a likely conclusion was that rate would have an effect on flowering. Plants sprayed at higher rates were smaller than those sprayed with lower rates (Figures 3.1 and 3.2), but there seemed to be no relationship between plants that flowered and rate of application as illustrated by Table 3.3. Bud initiation appeared to be “preset” such that no amount of ethephon used in this experiment was sufficient to keep the plants in a vegetative state. Plants that had initiated buds which were then treated again with ethephon did not appear to abort the buds, though the experiment was not carried on until flowers opened. The objective was to keep the plants from ever becoming reproductive, which is why the experiment was terminated before anthesis.

Ethephon is commonly used to induce flowering in *Bromus mango* Desv. (mango) and several species in the *Bromeliaceae* family (Salisbury and Ross, 1992). Hayashi et al. (2001) found ethephon treatments of 500 mg·L⁻¹ or 1000 mg·L⁻¹ sprayed one, two or three times hastened the time to flower in *Achillea millefolium* L. ‘Weser River Sandstone’, while time to flower was not affected in *Coreopsis verticillata* L. ‘Moonbeam’ or *Leucanthemum x superbum* Bergmans ex. J. Ingram ‘Thomas Killen’. These plants all had initiated inflorescences when sprayed with ethephon, and did not

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abort flower buds. *Gynura aurantiaca* Blume is a long day plant which is propagated by vegetative cuttings. Floral initiation on this species is detrimental to stock plant production and subsequent cuttings (Pallez and Dole, 2001). Ethephon applied at rates high enough to maintain the plants in a vegetative state resulted in severe stunting, which impeded cutting harvest. Ethephon treatments at low rates induced flowering of *G. aurantiaca* (Pallez and Dole, 2001).

Ethylene-Induced Epinasty

Adverse effects due to ethephon sprays were noted. One day after the initial treatment, epinasty was observed in the 300, 450, 600 and 750 mg·L⁻¹ rates. Plants sprayed at rates greater than 300 mg·L⁻¹ showed epinasty significant enough to severely reduce quality of the plant, and rate had a statistically significant effect on epinasty and overall plant quality (Table 3.5). Frequency of application was not statistically significant for epinasty or overall plant quality. Increasing the rate of application decreased overall plant quality due to increased epinasty (Figures 3.3, 3.4).

Ethylene is frequently referred to as the “stress hormone.” Plants often respond to stress with epinasty or malformation of the foliage. Ethylene-induced epinasty can be detrimental to plants by inhibiting carbon dioxide assimilation due to a reduced amount of leaf area available to absorb the light necessary for photosynthesis (Woodrow and Grodzinski, 1989). Severe epinastic responses in plants due to stress ethylene production may lead to reduced growth and failure to initiate flowers and set fruit.

Lycopersicon esculentum Mill. (tomato) exposed to chronic ethylene at either 0, 0.01, 0.05 or 0.1 µL·L⁻¹, exhibited increased epinasty in plants grown under higher

amounts of ethylene when compared to the controls, with reduced fruit set in plants exposed to $0.05 \mu\text{L}\cdot\text{L}^{-1}$ and virtual elimination of fruit set in plants grown under $0.1 \mu\text{L}\cdot\text{L}^{-1}$ (Blankenship and Kemble, 1996).

Moorman and Campbell (1980) noted a foliar application of $500 \text{ mg}\cdot\text{L}^{-1}$ ethephon resulted in severely dwarfed geranium stock plants, therefore yielding shorter cuttings. Hayashi et al. (2001) reported phytotoxicity on *Monarda* treated with $1000 \text{ mg}\cdot\text{L}^{-1}$ of ethephon at all spray frequencies (1, 2 or 3 times). For *S. dyerianus*, growers reported success with $750 \text{ mg}\cdot\text{L}^{-1}$ of ethephon applied early and then repeated applications approximately 2 and 4 weeks later (personal communication). This would correspond to the $750 \text{ mg}\cdot\text{L}^{-1}$ monthly treatment, which resulted in epinasty in our experiment. Ethephon is sensitive to environmental conditions when it is applied, which may have accounted for the epinasty.

Ethephon at the rates tested did not maintain vegetative stock plants of *S. dyerianus*. Plants in our experiment showed ethylene-induced epinasty at rates used successfully by growers. Both leaf area and shoot dry weight, were positively correlated with bud initiation.

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Table 3.1. Conditions for each application of ethephon on *Strobilanthes dyerianus* in 2001-2002.

Treatment/Date	Sprayer Pressure(psi)	Relative Humidity	Weather
1, 05 Nov	16	21%	Sunny, 25°C
2, 12 Nov	12	32%	Sunny, 23°C
3, 19 Nov	13	15%	Sunny, 18°C
4, 26 Nov	13	15%	Sunny, 18°C
5, 03 Dec	22	30%	Sunny, 25°C
6, 10 Dec	14	31%	Cloudy, 22°C
7, 17 Dec	24	53%	Cloudy, 22°C
8, 31 Dec*	26	29%	Sunny, 18°C
9, 07 Jan	28	42%	Cloudy, 18°C
10, 15 Jan	28	35%	Sunny, 23°C

* One week skipped for treatment due to Christmas holiday.

Table 3.2. Analysis of variance for the effect of ethephon rate and frequency of application on flowering of *Strobilanthes dyerianus* at termination of the experiment, 11 weeks after initiation of treatment.

Source	DF	Mean Square	P-Value
Rate	5	0.064	0.943 ^{NS}
Frequency	2	0.311	0.317 ^{NS}
Rate x Freq	10	0.231	0.568 ^{NS}

^{NS} Not significant at P=0.05.

Table 3.3. Number of *Strobilanthes dyerianus* plants with visible buds for each combination of rate x frequency of ethephon application observed at 11 weeks after initiation of treatment.

		Rate of ethephon (mg·L ⁻¹)						Total ^z
		0	150	300	450	600	750	
Application frequency	Monthly	2	1	4	3	2	3	15
	Biweekly	2	3	2	3	4	3	17
	Weekly	2	3	2	2	0	2	11
	Total ^y	6	7	8	8	6	8	43 ^x

^z Total out of 30 possible plants by frequency

^y Total out of 15 possible plants by rate

^x Total out of all plants in the experiment (n=90)

Table 3.4. Correlation coefficients for ethephon rate, shoot dry weight and leaf area with flowering for *Strobilanthes dyerianus* at termination of the experiment, 11 weeks after initiation of treatment.

	Flowering	P-Value
Ethephon rate	0.05	0.67 ^{NS}
Shoot dry weight	0.47	<0.0001*
Leaf area	0.40	<0.0001*

^{NS}, *Not significant or significant at P=0.05.

Table 3.5. Analysis of variance for rate and frequency of ethephon application on leaf area, shoot dry weight, overall plant quality and epinasty ratings of *Strobilanthes dyerianus* at 11 weeks after initiation of treatments.

Source	DF	Mean Square	P-Value
Leaf area			
Rate	5	1008580	<0.0001*
Frequency	2	213987	0.086 ^{NS}
Shoot dry weight			
Rate	5	103	<0.0001*
Frequency	2	19.0	0.136 ^{NS}
Overall quality ratings			
Rate	5	23.56	<0.0001*
Frequency	2	1.24	0.36 ^{NS}
Epinasty ratings			
Rate	5	23.25	<0.00001*
Frequency	2	0.23	0.04*

^{NS}, * Not significant or significant at P=0.05.

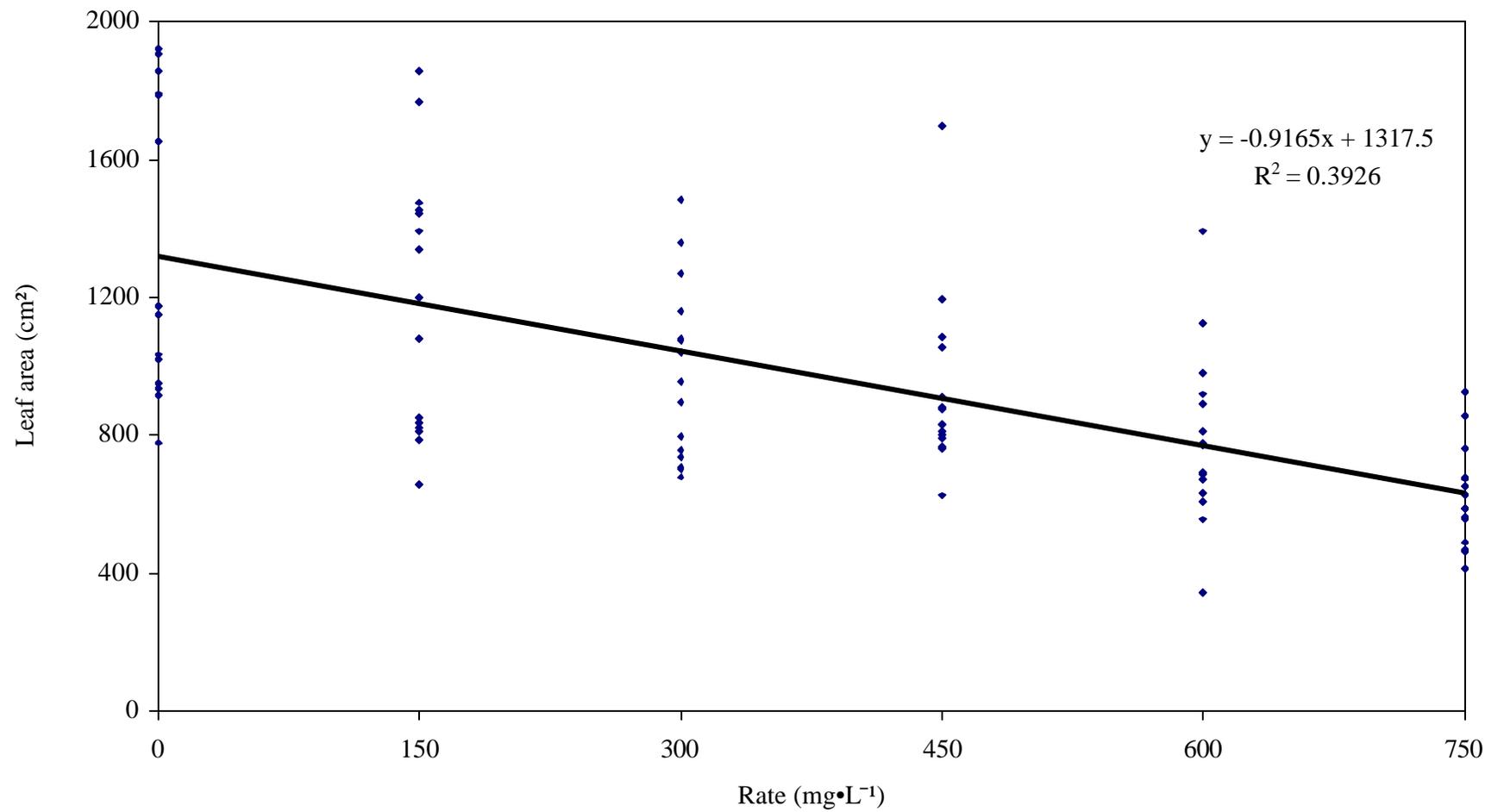


Figure 3.1. Total leaf area (cm²) of *Strobilanthes dyerianus* 11 weeks after initiation of treatments regressed over rate of ethephon.

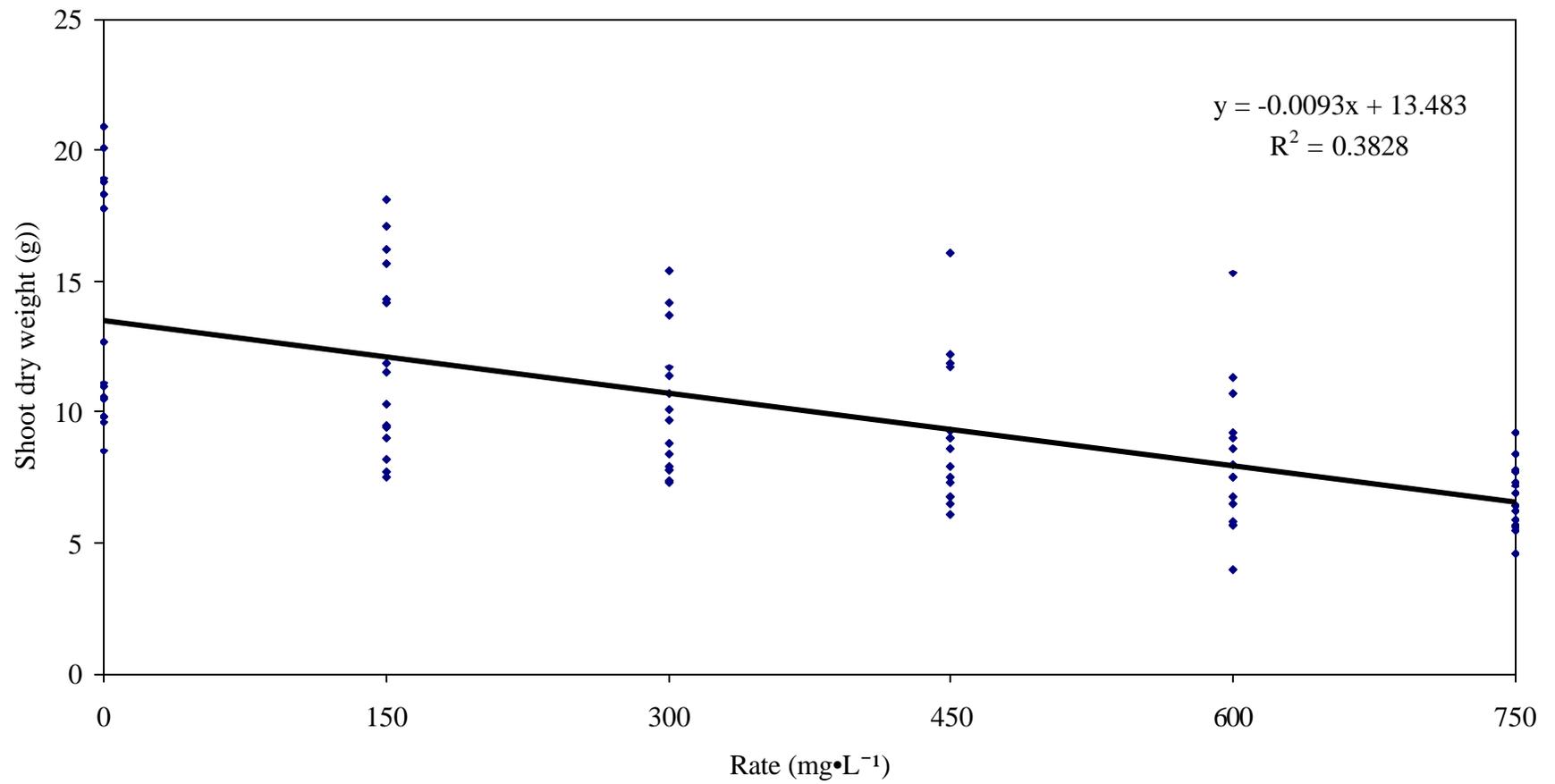


Figure 3.2. Shoot dry weight (g) of *Strobilanthes dyerianus* 11 weeks after initiation of treatments regressed over rate of ethephon.

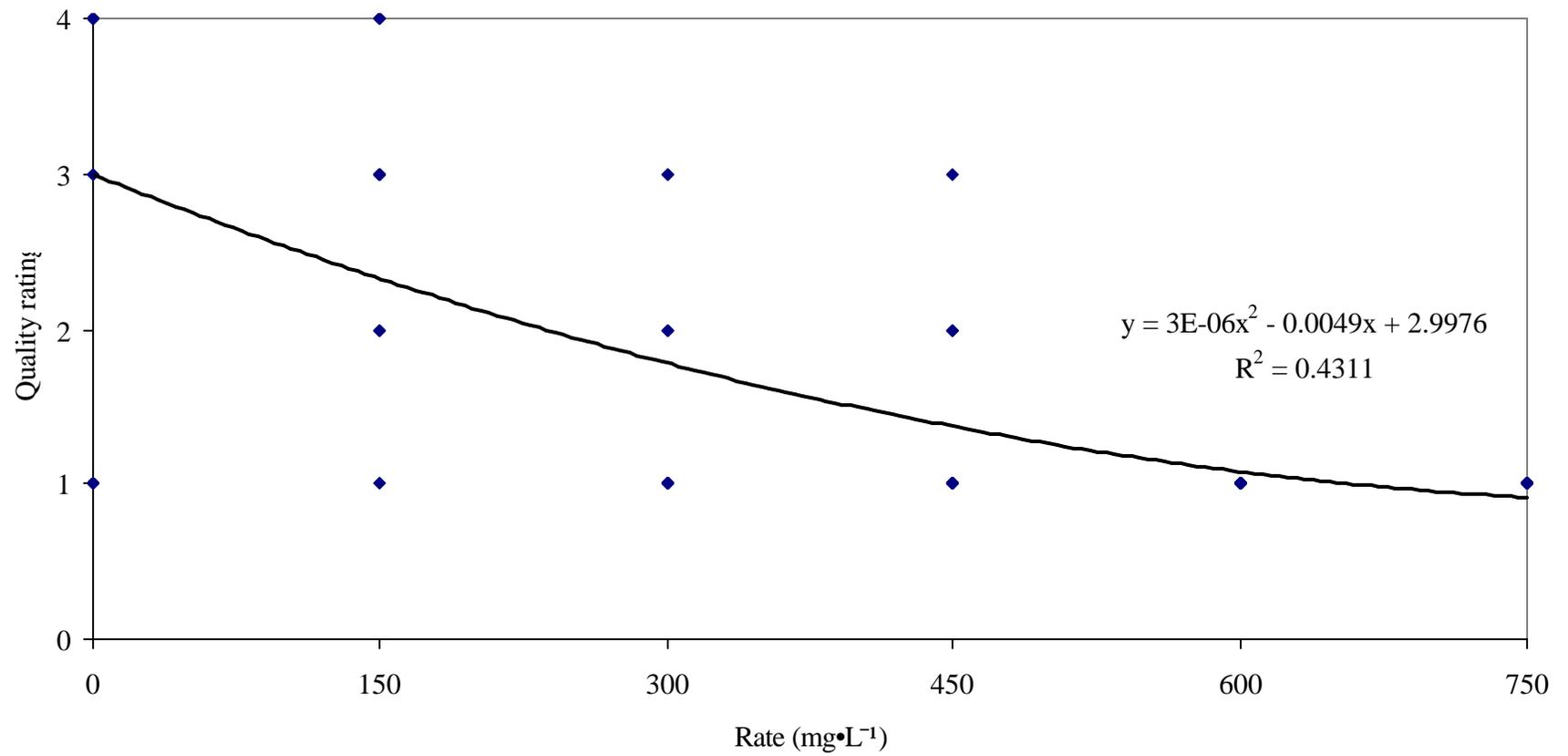


Figure 3.3. Overall plant quality rating of *Strobilanthes dyerianus* 11 weeks after initiation of treatment regressed over rate of ethephon.

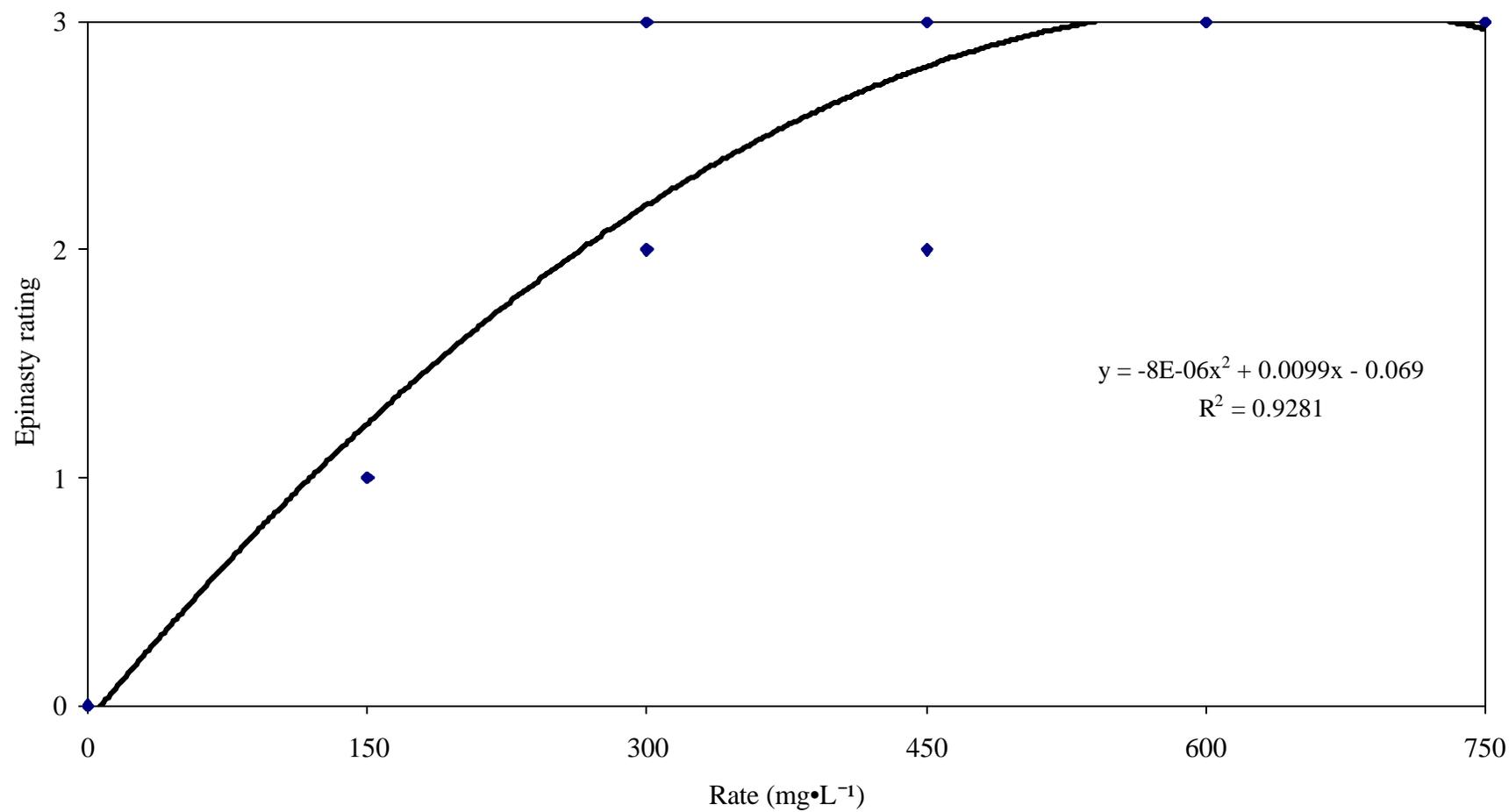


Figure 3.4. Epinasty ratings of *Strobilanthes dyerianus* 11 weeks after initiation of treatment regressed over rate of ethephon.

CHAPTER 4: DETERMINATION OF THE OPTIMAL FERTILIZER RATE AND TISSUE ELEMENTAL CONTENT OF *STROBILANTHES DYERIANUS* MAST.

***Abstract.* *Strobilanthes dyerianus* Mast. was examined for response to different levels of nitrogen. Grown as an annual in most of the United States, *S. dyerianus* has become increasingly popular in summer landscapes partially due to its superior performance in hot and humid conditions. At present, there is no published research on the nutritional requirements of *S. dyerianus*. Rooted cuttings were transplanted into 1.5 L pots using soilless media. Plants were grown for one week with clear water irrigation. A 15 N – 2.2 P – 12.4 K Cal-Mag fertilizer was applied as a constant liquid feed at 0, 100, 200, 300 and 400 mg·L⁻¹. Plants were irrigated with 450 mL whenever the volumetric water content of the media was <20% as determined by measuring with a Theta Probe moisture meter. Weekly pH and electrical conductivity (EC) were monitored using the pour through method. Eight weeks after initiation of treatment, plants were harvested and leaf area was measured. The plants were then dried at 65.5°C for 7 days and shoot dry weight was recorded. Recently mature leaf tissue was ground and analyzed for total N, P, K, Ca, Mg, S, Fe, Mn, B, Cu, Zn and Mo. There were no significant differences in plant quality under the 100, 200, 300 or 400 mg·L⁻¹ N treatments. The largest plants, based on leaf area and shoot dry weight, were produced with 200 mg·L⁻¹ N.**

Introduction

Proper nutrition is essential to grow a quality crop. Once a proper nitrogen level has been established, foliar analysis may be performed. The results of the foliar analysis, the elemental content of the tissue, are used to establish sufficiency ranges or averages for the essential elements. Sufficiency ranges and averages have been established for many species of commercially produced plants and provide growers with an approximate number by which they can diagnose potential nutrient deficiencies or toxicities and adjust their nutrition program accordingly.

Strobilanthes dyerianus Mast., (Persian Shield), has recently become a popular and successful annual foliage crop. It is a member of the Athens Select™ program, which is made up of plants demonstrating exceptional performance under hot and humid conditions. A native of Burma in Southeast Asia, *S. dyerianus* is grown for its foliage: large, sessile leaves that are purple, green and silver.

In order to produce high quality, marketable plants, the nutritional requirements of a plant must be established. Without proper nutrition, a plant cannot achieve optimal growth and may become more susceptible to stressors (Jones, 1998). Also, fertilizer runoff is a prominent environmental concern. As such, growers are trying to produce plants using the least amount of fertilizer that produces the quality plants the consumer demands. Currently, there is no published literature regarding the nutritional requirements for *S. dyerianus*.

A good growing program should utilize the information that can be obtained through monitoring the soluble salt content (electrical conductivity or EC) and pH of the medium. EC is a measurement of soluble salts, or total dissolved salts, in the medium

(Whipker et al. 2001) which provides a representation of the overall nutritional status of a crop (Wright et al., 1990). pH expresses the hydrogen ion activity in a solution, an indication of the acidity or alkalinity of a solution (Foth and Ellis, 1997). Both of these measurements, obtained through the pour-through extraction method (Wright, 1986) can be used to determine ranges for optimal growth of plants. The best concentration of fertilizer varies depending on the plant. Plants that are heavier feeders, such as *Euphorbia pulcherrima* Willd. ex. Klotzsch (poinsettia), require a higher concentration of nitrogen than light feeders, and consequently may be grown at a higher EC range (Whipker et al., 2001).

Foliar analysis is often used to determine the elemental status of the plant, which can then be used to recommend an appropriate fertilizer range, set sufficiency averages or ranges and to examine potential nutritional problems. The objectives of this experiment were to (1) determine a nitrogen level for optimal plant growth and (2) present tissue elemental content data which could be used as a reference by growers, as no sufficiency averages or ranges are currently available.

Materials and Methods

Cuttings were taken from stock plants of *S. dyerianus* on 24 May 2002 and rooted in a mist propagation bed utilizing rooting hormone with thiram (Rootone, Dragon Corp., Roanoke, Va.). Cuttings were rooted in four weeks, at which time 45 cuttings were transplanted into 1480 cm³ pots (10.8 cm x 10.8 cm x 12.7 cm) using a potting mix consisting of 45% peat moss, 25% processed pine bark, 15% perlite and 15% vermiculite (Fafard 3B soilless potting mix, Fafard, Inc., Anderson, S.C.). Plants were grown for one

week using clear water to irrigate, at which point 30 plants were selected for uniformity of height and approximate leaf area. The experiment took place in a glass greenhouse on the Virginia Tech campus in Blacksburg, Va. The average day temperature in the greenhouse was $30^{\circ}\text{C} \pm 1.5^{\circ}$ ($86^{\circ}\text{F} \pm 4^{\circ}$) and the average night temperature was $21^{\circ}\text{C} \pm 2.19^{\circ}$ ($70^{\circ}\text{F} \pm 2.6^{\circ}$).

The experimental was arranged in a randomized complete block design with six replications. Within a block, plants were treated with five rates of fertilizer ($\text{mg}\cdot\text{L}^{-1}$ based on nitrogen): $0 \text{ mg}\cdot\text{L}^{-1}$ (tap water), $100 \text{ mg}\cdot\text{L}^{-1}$, $200 \text{ mg}\cdot\text{L}^{-1}$, $300 \text{ mg}\cdot\text{L}^{-1}$ or $400 \text{ mg}\cdot\text{L}^{-1}$ of 15N-2.2P-12.4K (Miracle-Gro Excel 15-5-15 Cal-Mag, The Scotts Co., Marysville, Ohio) as a constant liquid feed. Plants were irrigated with 450 mL to achieve a 0.20 - 0.30 leaching fraction when the volumetric substrate water content was 20% or less, measured with ThetaProbe (ThetaProbe Soil Moisture Sensor, Delta-T Devices, Ltd, Cambridge, United Kingdom; TH20 meter, Dynamax, Inc., Houston, Texas). The ThetaProbe uses a 100 MHz sinusoidal signal, which helps to insure the measurement based on the dielectric constant of water is minimally influenced by ionic conductivity. This works because the dielectric constants of media components are negligible ($\sim 1-5$) in comparison with water (~ 80). The measurement is then converted into a ratio between the volume of water present and the volume of the entire sample, or the volumetric water content.

Data taken included weekly measurement of substrate pH and EC (electrical conductivity) using the pour-through extraction method (Wright, 1986), plant quality, dry weight and leaf area at termination of the experiment. Plant quality was rated on a scale of 0 to 4 with 0 being dead, 1 having severe chlorosis and/or necrosis, 2 having moderate

chlorosis and/or necrosis, 3 having mild chlorosis and/or necrosis and 4 being an excellent looking plant with little or no chlorosis and/or necrosis.

Eight weeks after initiation of treatments, plants were harvested and leaf area measured using a Li-Cor LI-3000 Portable Area Meter with LI-3050A transparent belt conveyer accessory (Li-Cor, Inc., Lincoln, Nebr.). Plants were then dried at 65.5°C (150°F) for 7 days, and dry weight was recorded. Leaf tissue from recently mature leaves was then ground, combining two random plants from each treatment into one sample, for a total of 15 samples. Total nitrogen was determined using the Dumas method (Mills and Jones, 1996) using a CNS 2000 analyzer (LECO Corp., St. Joseph, Mich.), while phosphorous, potassium, calcium, magnesium, sulfur, boron, copper, iron, manganese, molybdenum and zinc were determined by dry ashing and inductively coupled plasma spectrophotometry (Jarrell-Ash ICAP 9000; Thermo Jarrell Ash Corp., Franklin, Mass.) (Jones and Case, 1990) at Micro-Macro Laboratories (Athens, Ga.). Data were analyzed by GLM and regressions using SAS Version 8 (SAS Institute, Inc., Cary, N.C.).

Results and Discussion

Plant Quality and Growth

The rate of nitrogen applied affected overall plant quality, shoot dry weight and leaf area of *S. dyerianus* (Table 4.1). There was little visible difference between plants receiving fertilizer, and all plants treated with 100, 200, 300 or 400 mg·L⁻¹ N were given overall quality ratings of 4 (excellent) (data not presented). Fertilizer concentrations between 200 and 400 mg·L⁻¹ gave plants with the largest shoot dry weight and leaf area

(Figures 4.1 and 4.2). Despite similar quality, plants fertilized with $100 \text{ mg}\cdot\text{L}^{-1}$ N did not grow as much as those receiving higher levels of nitrogen. Looking at the regression lines, the peak appears to be around $300 \text{ mg}\cdot\text{L}^{-1}$ N, however the data points are very close to each other in the 200, 300 and $400 \text{ mg}\cdot\text{L}^{-1}$ N treatments. Kuehny et al. (2000) noted no significant differences in shoot dry weight or quality of five poinsettia cultivars grown under different rates of fertilizer. Kovacic and Holcomb (1981) found *Kalanchoe blossfeldiana* v. Poelln 'Pixie' (kalanchoe) grown with MagAmp (Grace Chem. Co., Inc., Cambridge, Mass.) 7-17.5-5 fertilizer had no difference in quality when grown at $11.8 \text{ kg}\cdot\text{m}^{-3}$ or two or four times that rate.

Electrical Conductivity (EC)

The rate of nitrogen applied had a significant effect on the electrical conductivity, (EC) over the course of the experiment (Table 4.2). Increasing rates of N caused an increase in the EC. Plants receiving no fertilizer had the lowest EC levels (Figure 4.3) and exhibited chlorotic foliage and stunted growth, compared to the other treatments. A low EC is associated with a malnourished plant, which may be visually noted by stunted growth or leaf discoloration (varying depending on the deficient nutrient) (Whipker et al. 2001). On the contrary, a high EC may contribute to necrotic leaf margins, poor or erratic rooting of cuttings and increased vulnerability to diseases effecting the root and crown (Dole and Wilkins, 1999). Morvant et al. (1997) found that lower concentrations of soluble salts in the medium encouraged root growth, while higher concentrations of media soluble salts inhibited root growth.

Strobilanthes dyerianus is grown as an annual and would be classified as a bedding plant. Utilizing information on recommended EC levels for bedding plants, the treatments receiving 300 and 400 mg·L⁻¹ N had excessively high soluble salts. Similar salt accumulations at higher rates of N were seen by Campos and Reed (1993) for *Spathiphyllum* Schott. and *Dieffenbachia* Schott. which ultimately led to plant death. For *S. dyerianus*, the 200 mg·L⁻¹ N treatment maintained the EC within the recommended range for bedding plants of 1.0 to 2.6 mS·cm⁻¹ (Whipker et al., 2001) throughout the duration of the experiment. The fact that there was no difference in overall plant quality suggests that *S. dyerianus* is tolerant of excess media soluble salts. Similar performance was noted in *Viola x wittrockiana* Gam., which produced optimal leaf area and shoot dry weight at a leachate EC ranging from 1.5 to 4.0 dS·m⁻¹ (van Iersel, 1999).

pH

The substrate pH was also affected by N treatment. The recommended pH range for soilless media is 5.4 and 6.0; however this can vary depending on crop type (Dole and Wilkins, 1999). Only plants receiving 0 mg·L⁻¹ N had a substrate pH that fell into this range (Figure 4.4). By termination of the experiment, pH for the 100, 200, 300 and 400 mg·L⁻¹ N treatments had fallen as low as 3.4, decreasing with increasing N concentrations. James and van Iersel (2001) also reported decreases in media pH associated with increasing fertilizer concentrations. Media pH can be affected by the breakdown of the components in the media, the alkalinity of the tap water in the greenhouse and also the acidity/basicity of the fertilizer being used. A pH that is too far outside of the optimal range can cause problems with nutrient availability, rendering certain nutrients, particularly micronutrients, unavailable for uptake by the plant

(Whipker et al., 2001). However, the plants did not show any visible nutrient deficiency or toxicity symptoms, and the tissue elemental content was in agreement with two similar species from the *Acanthaceae* family (Table 4.3).

Optimal Fertilizer Concentration

Since there was no difference in plant quality and very little increases in shoot dry weight and leaf area with higher concentrations, 200 mg·L⁻¹ N is recommended as an appropriate fertilizer concentration based on the concentrations examined in this experiment. An N rate of 200 mg·L⁻¹ also kept the EC in the recommended range for bedding plant production. Another bedding plant, *Impatiens wallerana* Hook. (double impatiens) has a recommended N rate of 150 or 200 mg·L⁻¹ depending on cultivar (Whipker et al., 1999). Frett et al. (1985) recommended a fertilizer rate of 200 mg·L⁻¹N for *Petunia x hybrida* Hort. ex. Vilm. 'Coral Sea,' which was found to produce the greatest dry weight, branch length and flowering. Campos and Reed (1993) recommended rates of 100-200 mg·L⁻¹ for the tropical plant *Spathiphyllum* Schott. and 200-400 mg·L⁻¹ for the tropical plant *Dieffenbachia* Schott., as these rates produced plants with the greatest leaf area.

Tissue Elemental Content

Most elements increased linearly with increasing N rates and were significant linear models (Table 4.3, [Appendix Table 1, and Appendix Figures 1 – 12]). Exceptions were sulfur, iron, and molybdenum, which exhibited quadratic increases, and manganese, calcium and magnesium which exhibited a quadratic decrease. The decrease in Ca and Mg can most likely be attributed to increased K from higher concentrations of fertilizer

(Mengel and Kirkby, 2001). K has a competitive effect with Ca and Mg, all three of which are cations. While Ca and Mg are often present in higher concentrations than K in soils, K is taken up much more quickly. Ca can only be taken up by young root tips, while the uptake mechanism of Mg is not yet known (Mengel and Kirkby, 2001). Decreases in Ca due to increased K have been noted in double impatiens (Whipker et al., 1999) and in poinsettia (Whipker and Hammer, 1997).

The elemental content averages of three samples per treatment are reported in Table 4.3. The tissue elemental content for the 200 mg·L⁻¹ N treatment was compared to two other species in the *Acanthaceae* family, *Aphelandra squarrosa* (Zebra plant) and *Fittonia verschaffeltii argyroneura* (Silver Nerve plant) as presented in Mills and Jones (1996). The tissue elemental content of *S. dyerianus* was comparable to the sufficiency data from these two species (Table 4.4).

There was no visible difference in the quality of plants produced with 100, 200, 300 or 400 mg·L⁻¹ N, and the largest plants (quantified by shoot dry weight and leaf area) were produced by 200 and 300 mg·L⁻¹ N. EC values indicate high soluble salts in the medium at 300 mg·L⁻¹ N, while the 200 mg·L⁻¹ N kept the EC within the range recommended for growing bedding plants. With mounting environmental concerns, a nutrition regime consisting of 200 mg·L⁻¹ is appropriate for producing high quality *S. dyerianus* plants. Elemental tissue content from foliar analysis may be used by growers as a guideline for diagnosing possible nutritional problems.

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Table 4.1. Analysis of variance for nitrogen rate on dry weight, leaf area and overall plant quality for *Strobilanthes dyerianus*. Plant quality ratings were determined on a scale of 0-4, with 0 being dead and 4 being excellent.

	Source	DF	Mean Square	P-Value
Shoot dry weight (g)	Model	9	432	<0.0001*
	Error	20	14.3	
Leaf area (cm ²)	Model	9	8828693	<0.0001*
	Error	20	139777	
Quality	Model	9	2.13	<0.0001*
	Error	20	0	

* Significant at P=0.05.

Table 4.2. The effect of rate of nitrogen in the fertilizer on pH and electrical conductivity (EC) of *Strobilanthes dyerianus* media leachate.

Source	DF	Mean Square	P-Value
EC			
N Rate	4	154	<0.0001*
Error	25	0.12	
EC			
N Rate	4	7.2	<0.0001*
Error	25	0.11	

*Significant at P=0.05.

Table 4.3. The effect of nitrogen rate on foliar element content of *Strobilanthes dyerianus* harvested 8 weeks after initiation of treatment and the significant effects from regression analysis. The values presented in the table are the means of 3 samples per N treatment.

	N	P	K	Ca	Mg	S	Fe	Mn	B	Cu	Zn	Mo
mg·L ⁻¹ N	g·kg ⁻¹						μg·g ⁻¹					
0	11.8	2.66	17.9	24.0	19.1	0.93	30.9	242	34.1	2.10	40.5	0.12
100	31.0	4.73	29.3	15.1	10.8	1.70	61.4	127	42.4	3.79	36.0	5.19
200	39.0	5.87	39.4	14.1	9.90	1.60	70.6	141	67.7	5.02	52.5	3.86
300	43.0	5.77	48.3	14.0	9.57	1.53	68.4	144	89.4	5.54	54.1	3.91
400	46.5	6.12	52.2	12.7	8.87	1.67	66.8	133	103	6.46	53.9	3.42
Significance*	L	L	L	L	L	Q	Q	Q	L	L	L	Q

* Linear (L) or Quadratic (Q) effects at P ≤ 0.05.

Table 4.4. Tissue elemental content for three species of *Acanthaceae* plants.

	N	P	K	Ca	Mg	S	Fe	Mn	B	Cu	Zn	Mo
Species	----- g·kg ⁻¹ -----						----- μg·g ⁻¹ -----					
<i>Strobilanthes dyerianus</i> ^z	39.0	5.87	39.4	14.1	9.90	1.60	70.6	141	67.7	5.02	52.5	3.86
<i>Aphelandra squarrosa</i> *	20-30	2-4	10-20	4-20	5-10	2-3	50-300	50-300	35-50	10-50	20-200	0.12-0.5
<i>Fittonia verschaffeltii</i>												
<i>argyroneura</i> *	41.6	7.60	56.9	22.4	13.4	6.30	114	80.0	41.0	9.00	61.0	3.00

^z Mean tissue elemental content from 3 samples of *S. dyerianus* treated with 200 mg·L⁻¹ N.

* From Plant Analysis Handbook II (Mills and Jones, 1996).

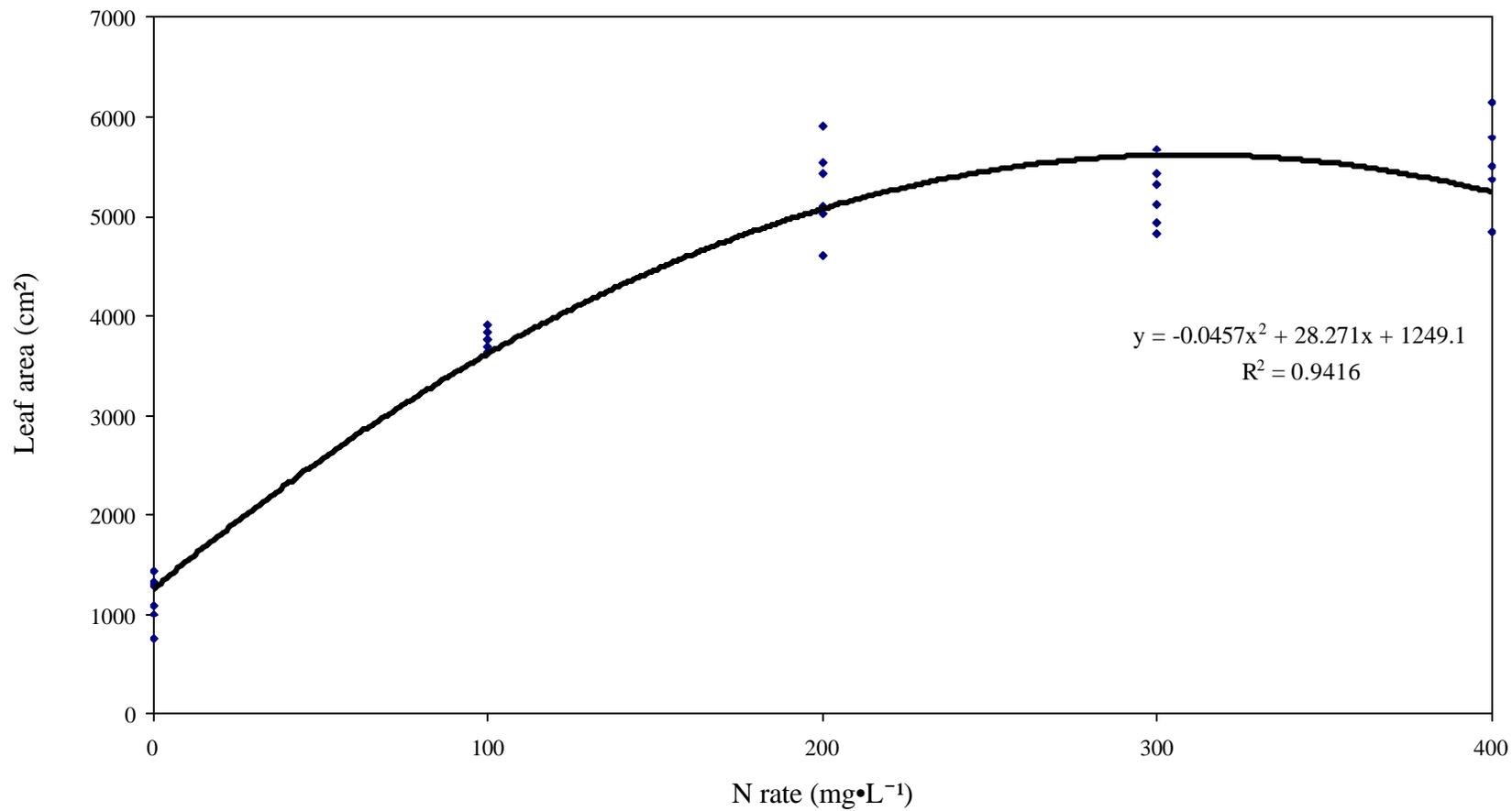


Figure 4.1. Leaf area of *Strobilanthes dyerianus* measured at harvest, 8 weeks after initiation of treatments, regressed over concentration of nitrogen in the fertilizer.

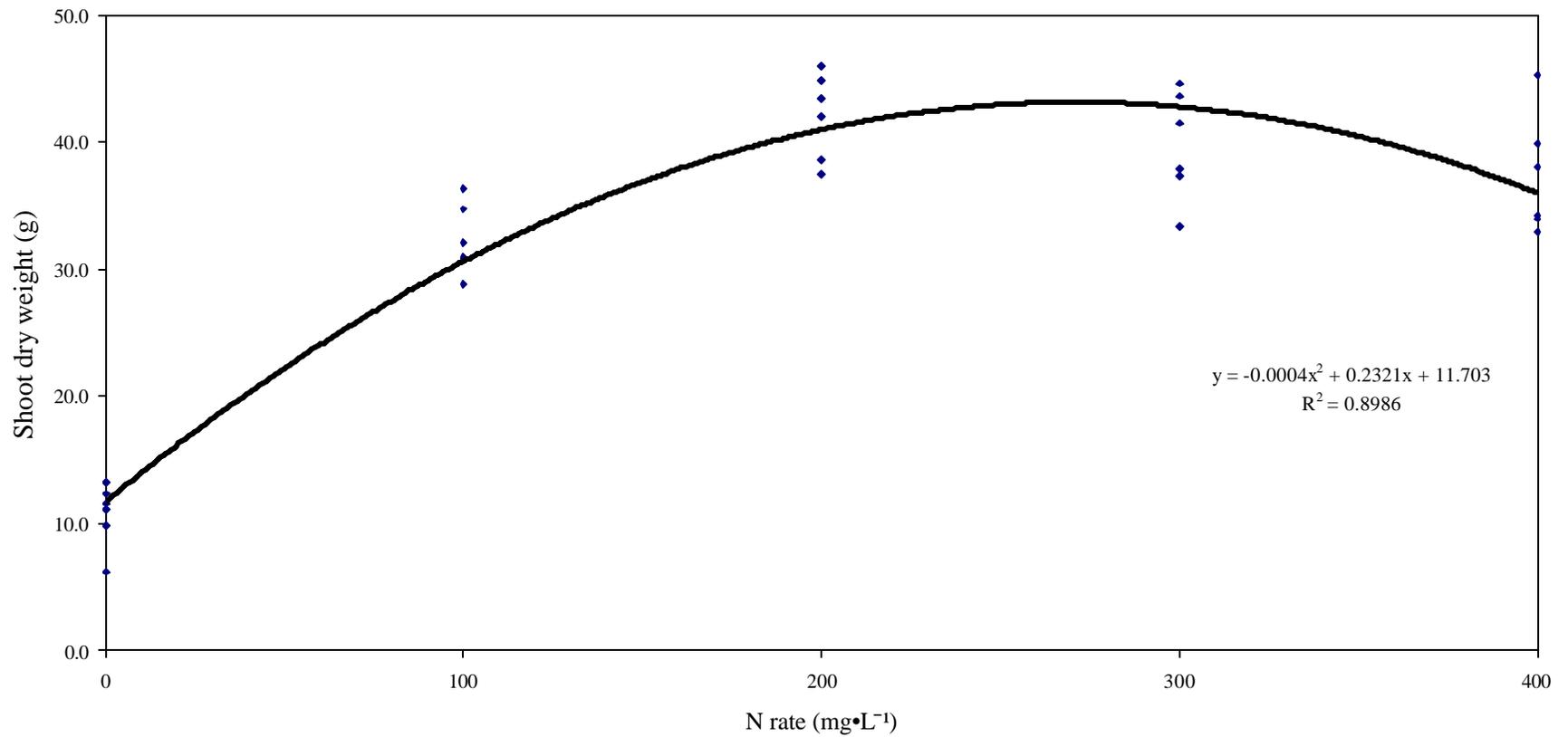


Figure 4.2. Shoot dry weight of *Strobilanthes dyerianus*, measured at harvest, 8 weeks after initiation of treatments, and regressed over concentration of nitrogen in the fertilizer.

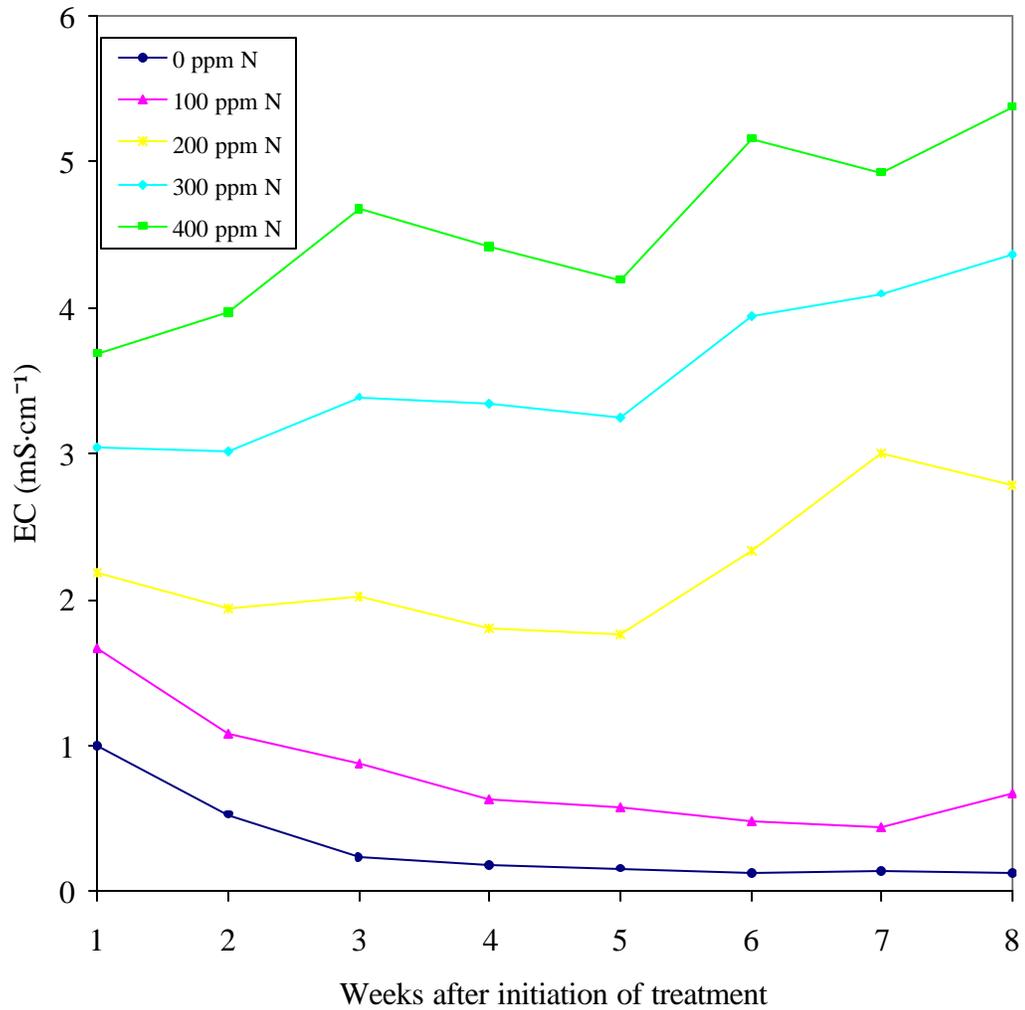


Figure 4.3. Electrical conductivity (EC) of the substrate leachate of *Strobilanthes dyerianus* measured each week during the experiment using the pour through extraction method.

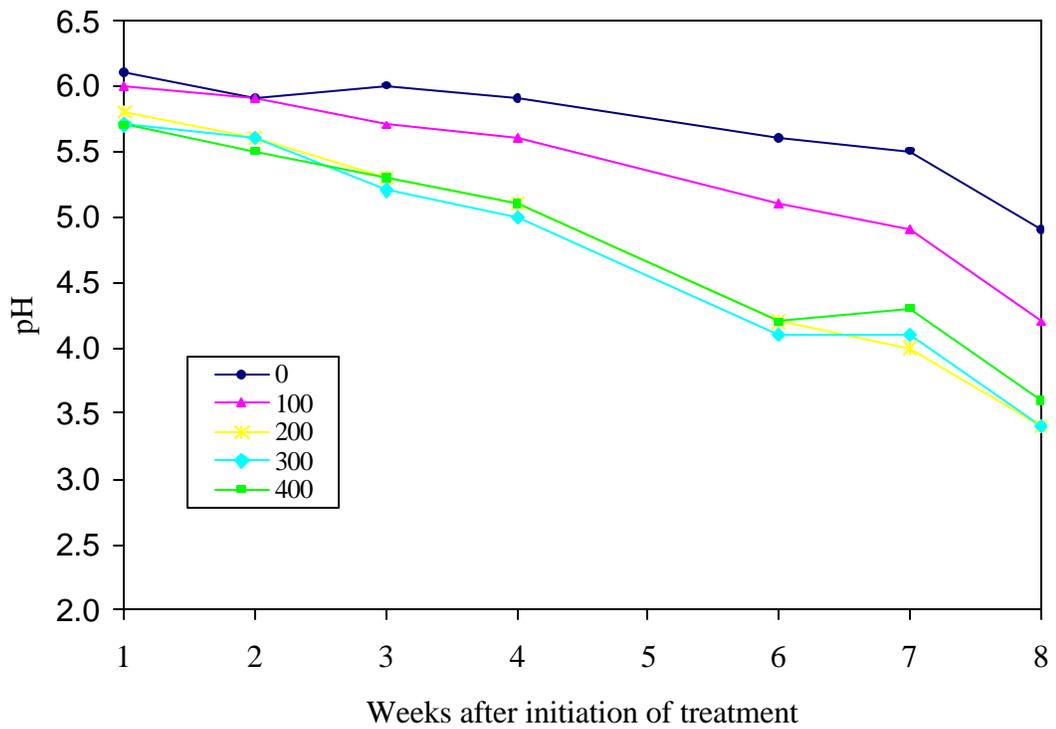


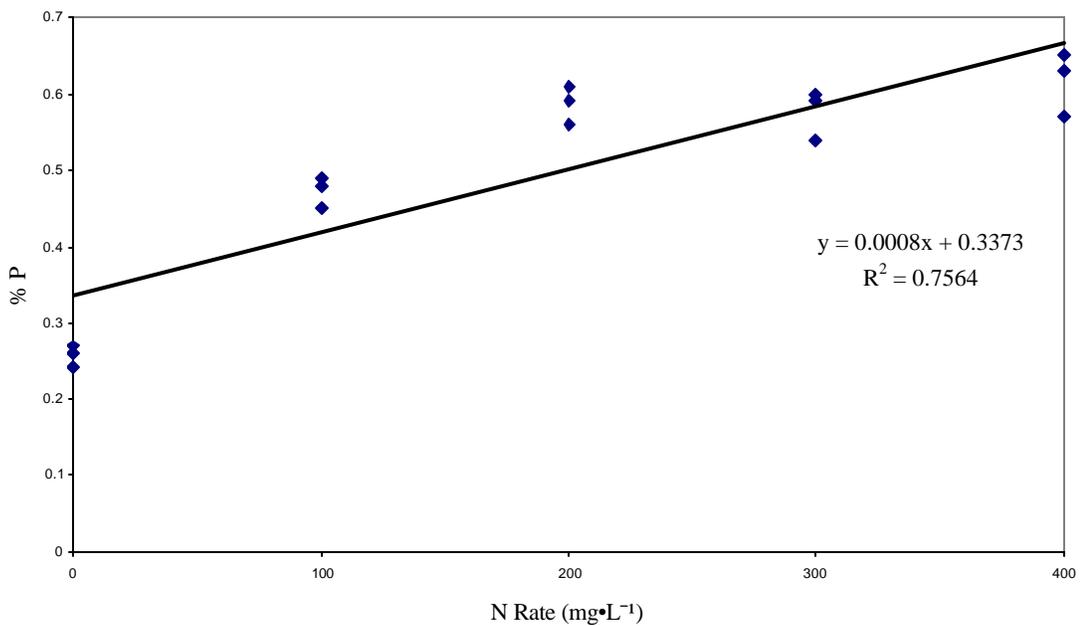
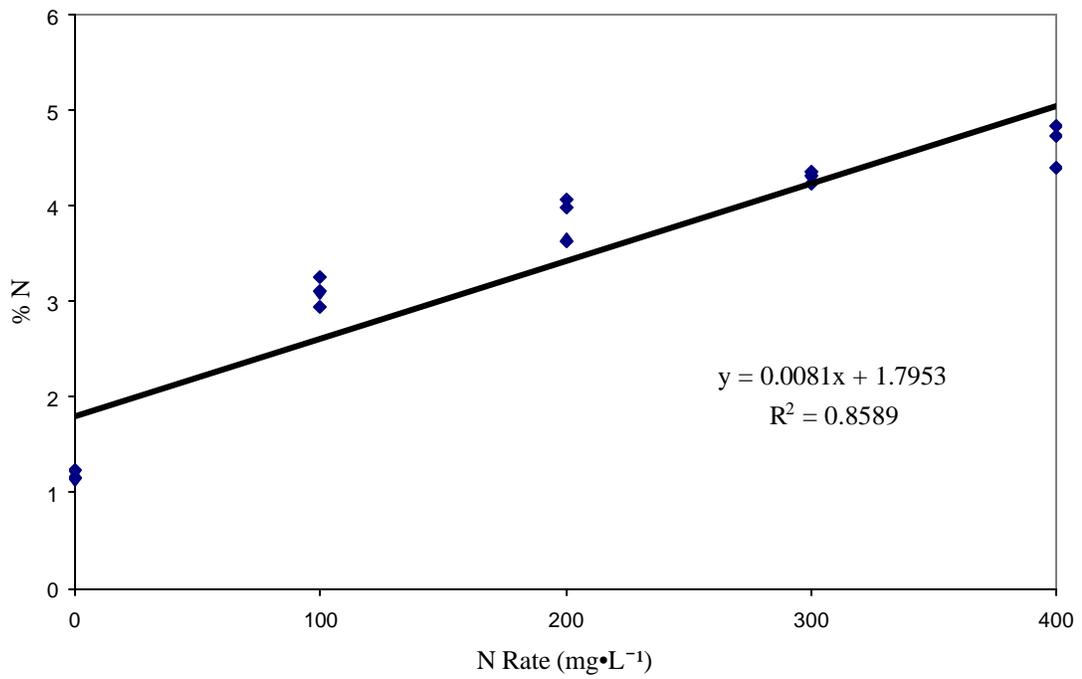
Figure 4.4. *Strobilanthes dyerianus* substrate pH measured weekly by the pour through extraction method for the duration on the experiment. (Week 5 data excluded due to equipment malfunction).

APPENDIX

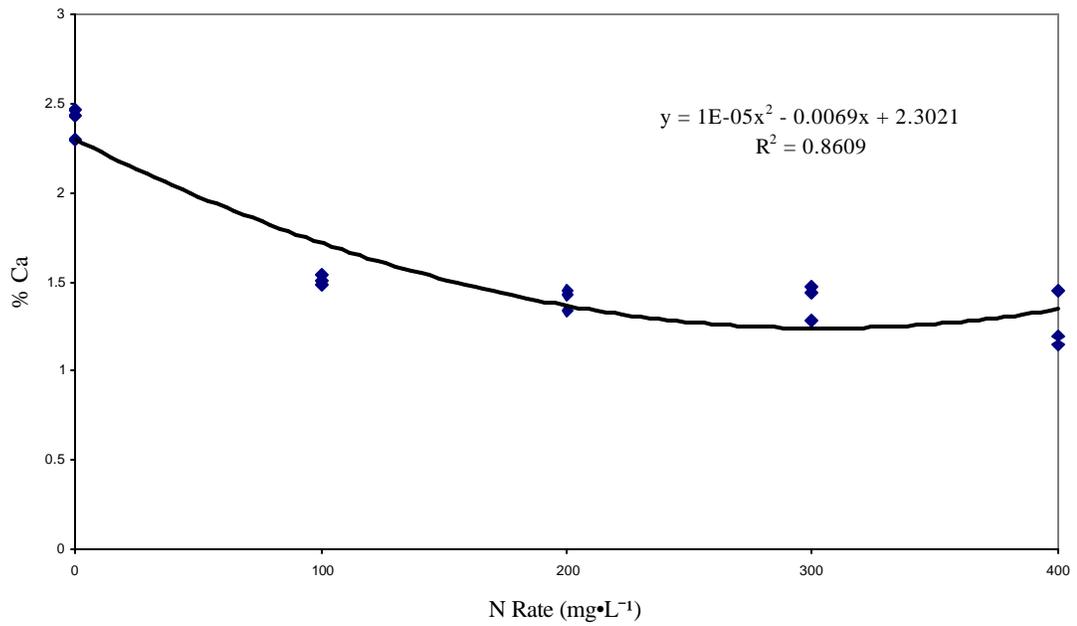
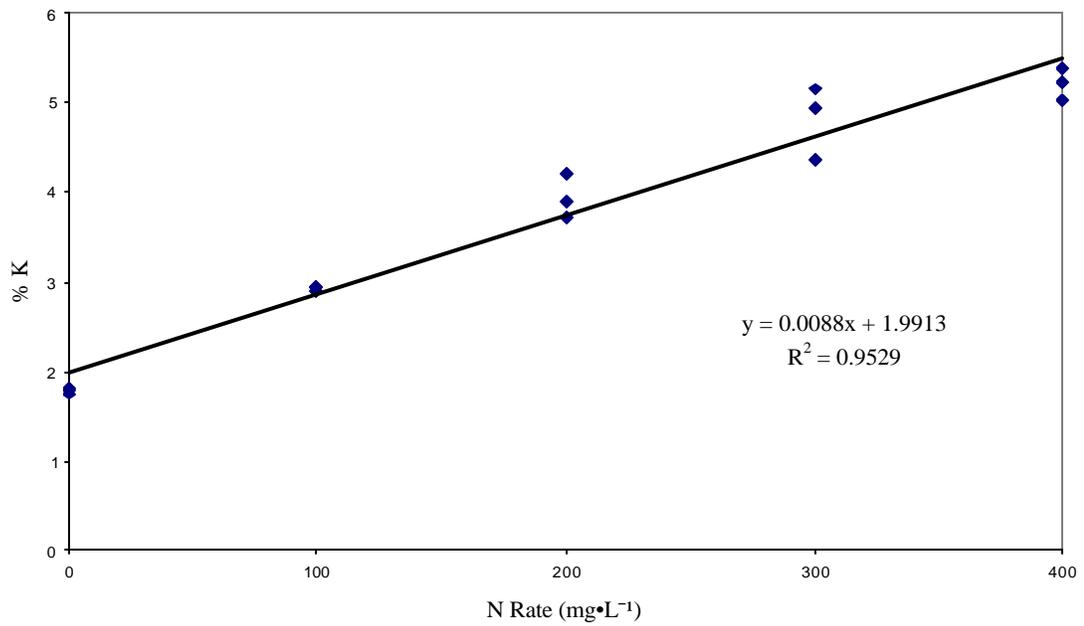
Appendix Table 1. Regression models for the foliar elemental content of *Strobilanthes dyerianus* harvested 8 weeks after initiation of treatment (single degree of freedom contrasts, n=15).

Element	Source	MS	P-Value
N	Linear	19.91	<0.0001
	Quadratic	2.66	<0.0001
	Cubic	0.35	0.0087
P	Linear	0.2	<0.0001
	Quadratic	0.05	<0.0001
	Cubic	0.01	0.0146
K	Linear	22.97	<0.0001
	Quadratic	0.57	0.0076
	Cubic	0.04	0.3739
Ca	Linear	1.69	<0.0001
	Quadratic	0.55	<0.0001
	Cubic	0.25	0.0007
Mg	Linear	1.4	<0.0001
	Quadratic	0.53	<0.0001
	Cubic	0.18	<0.0001
S	Linear	0.0051	0.0005

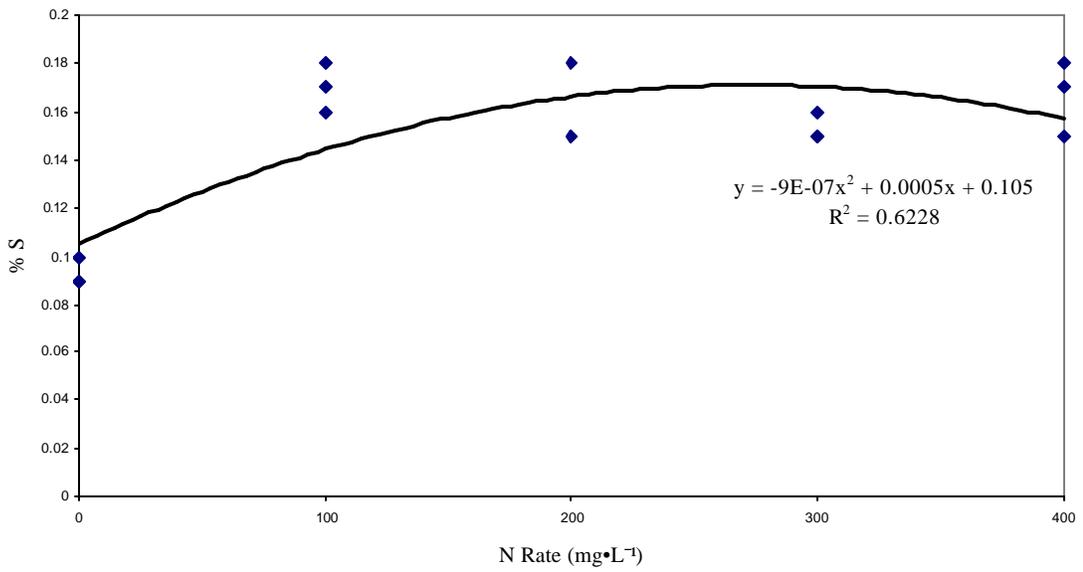
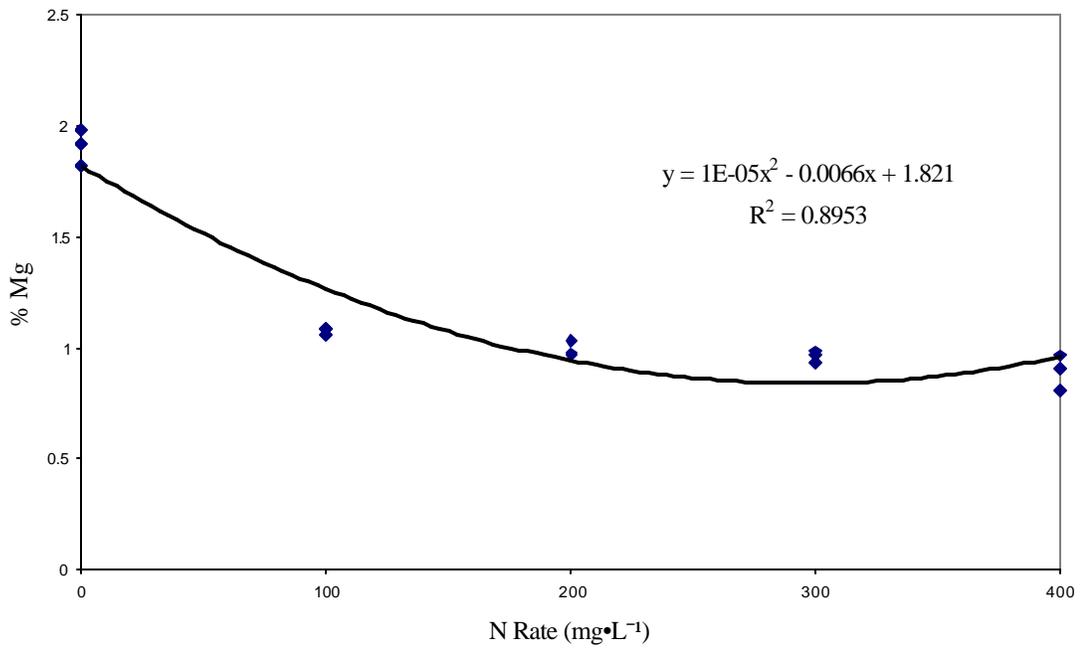
	Quadratic	0.0033	0.0021
	Cubic	0.0034	0.0018
Fe	Linear	1868.19	<0.0001
	Quadratic	1228.61	<0.0001
	Cubic	142.44	0.0351
Mn	Linear	12070.5	<0.0001
	Quadratic	8382.2	<0.0001
	Cubic	6115.55	0.0002
B	Linear	10313.06	<0.0001
	Quadratic	12.17	0.5265
	Cubic	186.05	0.0322
Cu	Linear	32.82	<0.0001
	Quadratic	1.08	0.1546
	Cubic	0.22	0.4951
Zn	Linear	604.62	0.0005
	Quadratic	8.7	0.5236
	Cubic	153.27	0.0232
Mo	Linear	8.52	0.0039
	Quadratic	20.31	0.0003
	Cubic	10.29	0.0023



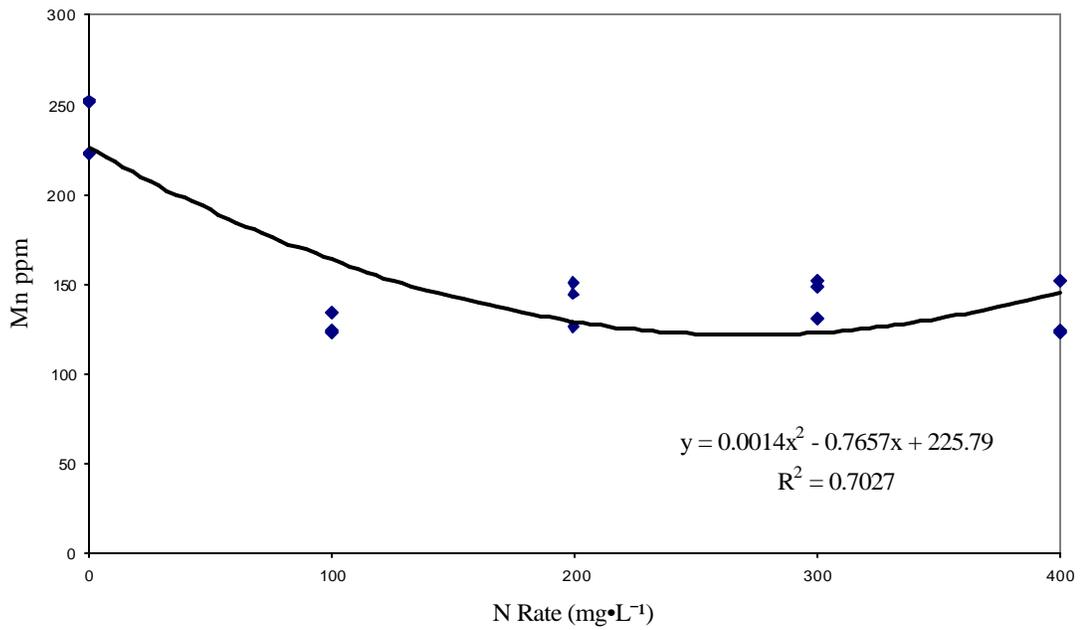
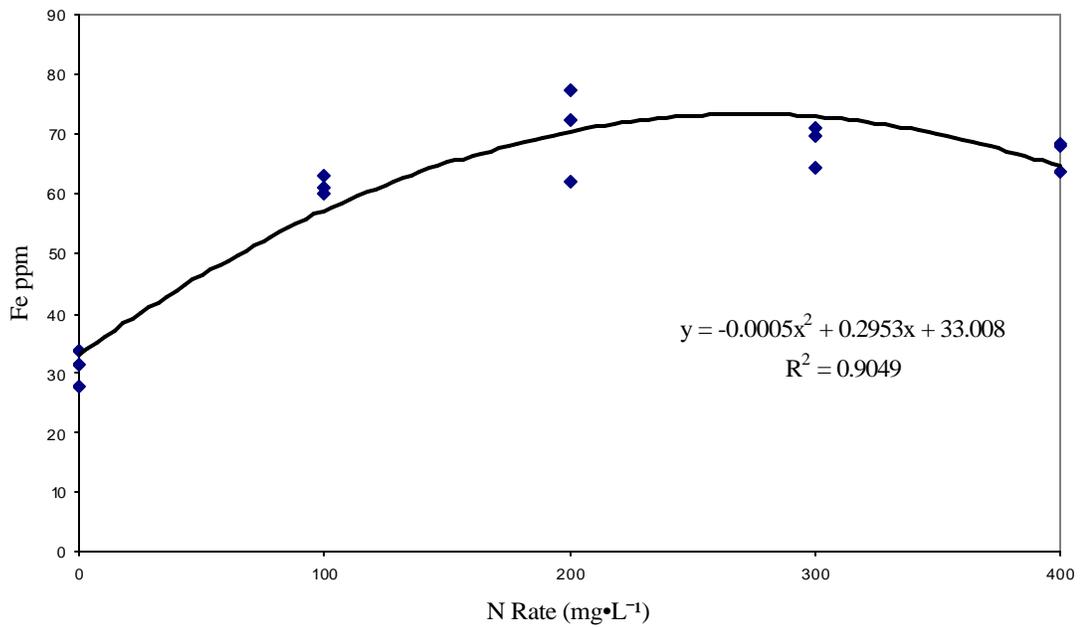
Appendix Figure 1 and 2. Foliar content of nitrogen and phosphorous (%) of *Strobanthes dyerianus* treated with different N rates harvested 8 weeks after initiation of treatment regressed over N rate.



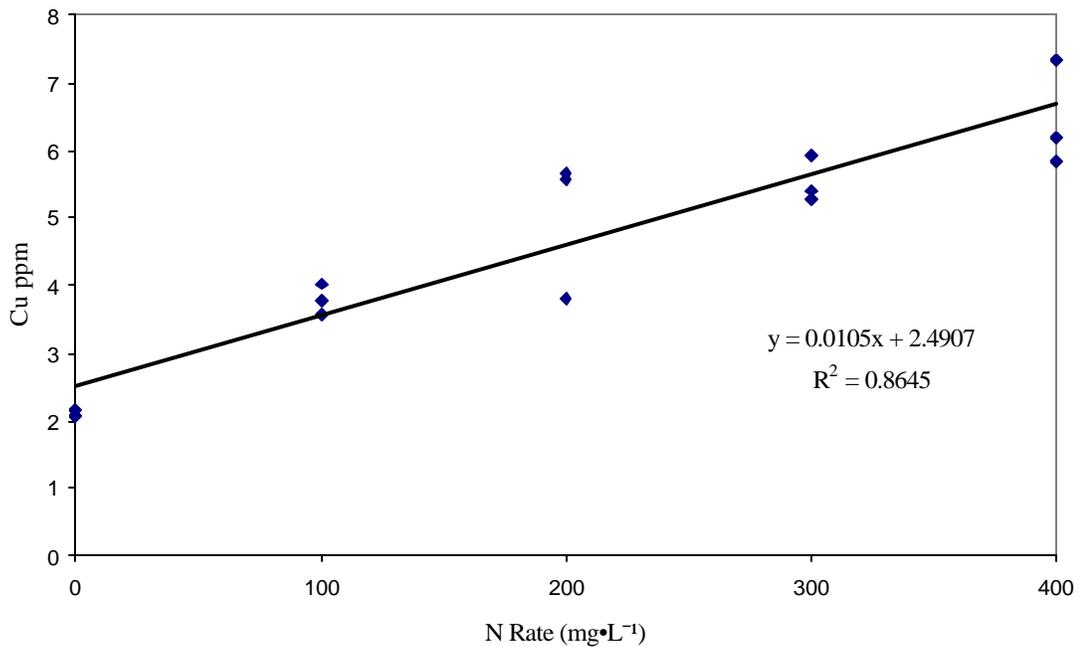
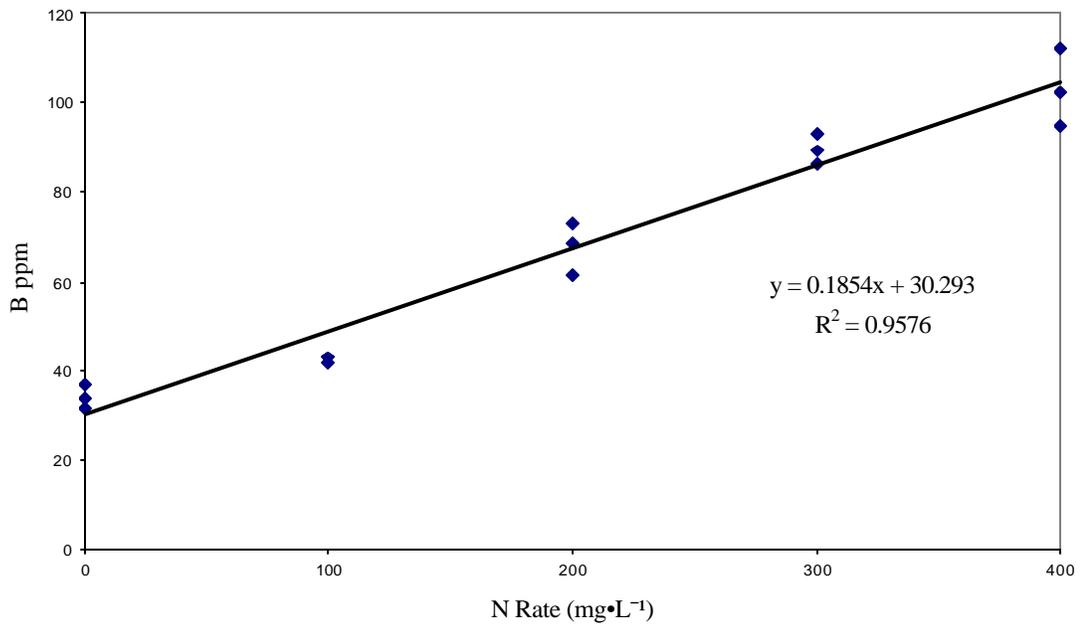
Appendix Figure 3 and 4. Foliar content of potassium and calcium (%) of *Strobilanthes dyerianus* treated with different N rates harvested 8 weeks after initiation of treatment regressed over N rate.



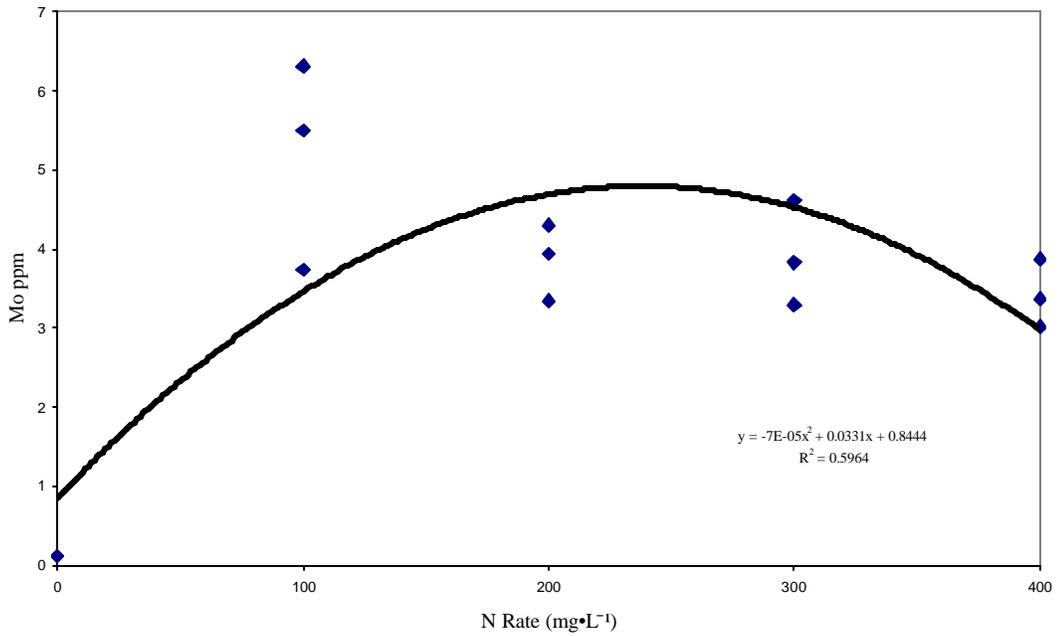
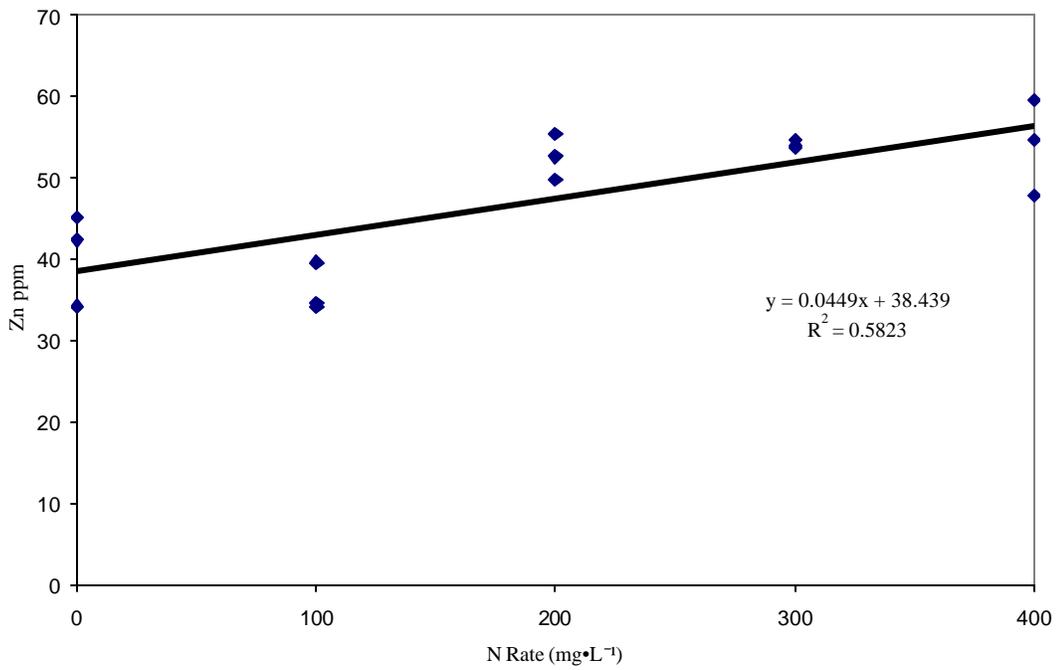
Appendix Figure 5 and 6. Foliar content of magnesium and sulfur (%) of *Strobilanthes dyerianus* treated with different N rates harvested 8 weeks after initiation of treatment regressed over N rate.



Appendix Figure 7 and 8. Foliar content of iron and manganese (ppm) of *Strobilanthes dyerianus* treated with different N rates harvested 8 weeks after initiation of treatment regressed over N rate.



Appendix Figure 9 and 10. Foliar content of boron and copper (ppm) of *Strobilanthes dyerianus* treated with different N rates harvested 8 weeks after initiation of treatment regressed over N rate.



Appendix Figure 11 and 12. Foliar content zinc and molybdenum (ppm) of *Strobilanthes dyerianus* treated with different N rates harvested 8 weeks after initiation of treatment regressed over N rate.

VITA

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