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PRE-WILTING BURLEY TOBACCO TO ENHANCE MANUAL AND MECHANICAL HARVESTING AND HOUSING

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THESIS

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Biosystems and Agricultural Engineering in the College of Engineering at the University of Kentucky.

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ABSTRACT OF THESIS

PRE-WILTING BURLEY TOBACCO TO ENHANCE MANUAL AND MECHANICAL HARVESTING AND HOUSING

Traditionally, burley tobacco has been harvested by hand because the green plant weight, volume, and leaf fragility make mechanical harvesting very challenging. This study examined possible ways to wilt a plant still standing in the field (termed 'pre-wilting') to reduce weight, volume, and leaf fragility. Several methods of pre-wilting burley tobacco plants in the field were explored including: root pruning, stalk girdling, freezing with liquid nitrogen, and burning. Experiments were conducted in three locations over three consecutive years during the tobacco harvest season. Leaf breaking angle, leaf moisture content and time-lapse photography were investigated as methods to quantify treatment effects on wilting. The time-lapse photography helped reveal that wilting was most prevalent during the late afternoon, and that wilted plants sometimes began to recover after more than five days, apparently due to root re-growth. Root pruning was the only mechanical means that caused witling reliably during the first two years of testing, and even then the results were somewhat inconsistent. During the third year, a high-clearance tobacco sprayer was modified with a hydraulically actuated coulter disc in order to root-prune a large number of subjects.

Keywords: tobacco, wilting, pre-wilting, specialty crop, root prune, mechanization

Multimedia Elements Used: JPEG (.jpg); Video (.mp4)

Ben C Hebener
Signature

April 13th, 2018
Date

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For my family and friends.

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I would like to thank my parents and girlfriend for supporting me to finish this thesis despite the long and arduous journey. Unique circumstances popped up along the way, and without them quitting would have been all too easy. They did nothing but encourage and help me to achieve my goal of obtaining my Master's degree, and taught me quite a bit about perseverance along the way.

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CHAPTER 1 – INTRODUCTION

1.1 Introduction

Burley tobacco is a unique crop because it has such a high manual labor requirement for production compared to other crops which have become more mechanized. Typical burley tobacco crops require 150-200 hours per acre of labor (hr/ac) with approximately a third of that total required for harvest (Seebold and Pearce 2012). Throughout the last century, there has been a strong emphasis on developing mechanization solutions for tobacco harvesting. The problem with most of these harvesters is they were either damaging or losing leaves; burley tobacco's leaves are delicate especially while the plant is fully turgid. Examples of such leaf loss can be seen in evaluations of a low-cost harvester by Camenisch, Wells, Smith, & Duncan, 2002, as well as a trail-type harvester described by Bader, Walton, & Casada, 1990. Any damage to the leaves results in a lower market value and loss of product.

Traditional burley tobacco harvesting begins with a worker cutting down each plant individually. After a plant is cut the worker spears it onto a stick. When the stick reaches maximum capacity (typically six plants), the stick is tilted to the side and the plants are left in the field to wilt (Bailey et al., 2011). Wilting is important because it reduces the turgor pressure which makes the plants less delicate, lighter, and smaller. After approximately three days in the field the tobacco will be wilted enough to be sufficiently pliable for moving and hanging with minimal damage.

Merriam Webster defines turgor pressure as "the actual pressure developed by the fluid in a turgid plant cell". Plant cells naturally try to achieve maximum turgor pressure; however, during certain conditions such as a drought, the plant is forced to lose turgor pressure. High turgor pressure can be the cause of large losses from leaf breakage (Osamura et al., 2002). The higher the turgor pressure, the less flexible the plant becomes, so tasks like moving them onto a truck or throwing them can snap leaves off very easily (Walton et al., 1976). If the turgor pressure is lower, the leaves become more relaxed, and tasks like stacking the plants will cause less damage.

Wilting is essentially a plant not having enough water to remain fully turgid. This happens because the net transpiration rate of the tobacco plant exceeds the water uptake of the plant from the soil. Plants will wilt if they are subject to sufficient stress caused by one or more factors (Kramer and Boyer 1995) including physical damage (e.g. mechanical means, disease, or insects) and environmental factors (e.g. high temperature, drought, low humidity, or flooding).

Plants survive through a delicate balance between the soil and the atmosphere; this is called the soil-plant-atmosphere continuum. Water is constantly moved from the soil, through the plant, and out into the atmosphere as a necessity for the plant to grow and remain alive (Kramer and Boyer, 1995). This balance is largely a mechanical process, driven by a gradient of pressure potentials (Cowan, 1965). The plant will vary its rate of water usage - or transpiration - based on many factors including the stage of growth, stresses, and available water. When a plant's needed rate of of transpiration exceeds the rate at which the plant can uptake water, wilting will occur (Briggs and Shantz, 1912). While the maximum transpiration rate is still largely a mechanical process, dependent on the water potential gradient from soil to atmosphere, the plant can add resistance to the water's path, slowing transpiration (Begg and Turner, 1970). While stresses and conditions can significantly lower the transpiration rate, it can never reach zero – in others words while the plant is alive, some respiration has to occur. It should, then, be possible to force a plant to wilt if there was a way to inhibit water uptake in a tobacco plant such that the plant's water loss from transpiration is larger than the uptake.

1.2 Purpose of Research

One of the reasons that it is so difficult to mechanize the harvesting of burley tobacco is that mechanically handling the plants causes so much leaf breakage due to the fragility of the plant in the field. If the plants were wilted, as happens in traditional harvesting practice, the plants would be less fragile, lighter, and smaller. High weight, while somewhat of a concern with traditional harvesting, can be an even greater factor in mechanical harvesting (Camenisch et al., 2002). Wilting is also important for plant dimensional considerations. When tobacco is cured, the plants are placed very close together in barns or other curing structures. Achieving the desirable plant density during curing would not be possible with an unwilted plant. Osamura et al., (2002) reported that, while five to six green (unwilted) tobacco plants could be placed on a traditional stick of 4.9 feet, up to eight plants could be safely placed on the same size stick after being considerably wilted.

1.3 Goal and Objectives

1.3.1 Goal

The overall goal of this study was to design a machine that can force tobacco plants to wilt while they are standing in the field.

To accomplish this goal, new and unique methods of causing tobacco plants to wilt had to be discovered and tested. This information was then used to design a machine that can wilt a large number of tobacco plants in the field in a cost-effective manner, without damaging them or knocking them over. The following specific objectives guided the investigations.

1.3.2 Objective 1

Identify and refine techniques to pre-wilt tobacco.

- Task 1: The first task was to identify a number of different methods that have potential to pre-wilt tobacco. To be adoptable, the method would have to be mechanizable.
- Task 2: The second task was to further refine several of these methods to determine which methods would be reasonably implementable.

1.3.3 Objective 2

Develop and test a mechanical system to pre-wilt tobacco.

After identification of the best potential wilting technique, the next objective was to mechanize that solution. This technique needed to not only pre-wilt tobacco, but also be cost-effective, require low minimal labor, incur low losses, and to be able to be used in conjunction with typical industry tobacco machinery and cultural practices.

1.4 Organization of Thesis

Because of the nature of tobacco harvesting, there are only a few weeks each year when data can be collected; as such, this study was conducted in three phases spanning three years of testing. Phase one, conducted in 2009-2010, addressed objective 1, task 1 by identifying potential wilting techniques. Phase 2, conducted in 2010, addressed objective 1, task 2 by further refining the wilting techniques with the highest mechanization potential. Phase 3, conducted in 2011, addressed objective 2 by developing a mechanized pre-wilting technique. The chapter organization of this thesis describes the three phases of this project as follows:

1. Introduction – explanation of project and why a solution was beneficial
2. Literature Review – identification of relevant research and studies regarding wilting, tobacco, and measuring plant water status
3. Phase One: Initial Testing – evaluation of various methods of pre-wilting and measurement of plant water status
4. Phase Two: Large Scale Testing and Refinement – narrowing all possible treatments down to the best candidate for mechanization and developing data collection and statistical analysis methodology
5. Phase Three: Design and Testing of Root Pruning Implement – development, testing, and evaluation of a mechanized version of a method for pre-wilting tobacco
6. Conclusion – summary and possible future work

CHAPTER 2– LITERATURE REVIEW

2.1 Explanation of Project and Background

Traditionally, tobacco is hand cut in the field. Five to six stalks are speared onto wooden sticks and left in the field for a period of time to wilt (Seebold and Pearce 2012). The tobacco is left very close to the spot it was cut and not transported immediately after cutting to minimize damage to the leaves. It is propped up in the field and left there until the plant is visibly wilted. Although a variety of factors affect wilting rate, usually sufficient wilting occurs in just a few days with an associated 5%-10% drop in leaf moisture content (Burton et al. 1989). Camenisch et al. (2002) stated that traditional field wilting usually takes two to three days. In this wilted state, the plant is more flexible and therefore less delicate; additionally, the plant weighs less.

In one test of a Burley tobacco harvester to evaluate its overall performance compared to traditional harvesting, one of the major concerns cited was excessive leaf loss (Bader et al. 1990). In this study, mechanical harvesting resulted in 1.3%-4% of the total amount of leaves being lost. The majority of leaves that were damaged or knocked off landed on the floor beneath the area where the tobacco was hung. The factors that appeared to contribute to the high incidence of leaf loss were overall plant size, maturity, and number of leaves per plant.

Labor availability is another challenge facing burley tobacco producers. Efforts have been made in virtually every step of tobacco production to reduce labor and production costs. One of the most sought after areas of development is that of harvest mechanization, which could reduce harvest labor requirements by as much an order of magnitude (Bader et al. 1990). Leaf loss and damage to plants has been a constant challenge with all mechanized harvesters because they cut and move the tobacco while it is fully turgid and brittle. Volume limitations on machines that load the tobacco onto racks for curing are another concern. The closer together that the tobacco is loaded, the greater the potential for physical damage to the leaves as well as mold damage during curing. The further the plants are spaced apart, the more stops the machine has to make for unloading, decreasing the efficiency and reducing the advantage of a mechanical harvesting. If the plants were wilted, they could be placed into a denser arrangement.

Wilting plays a crucial role on not only in reducing damage incurred by the valuable leaves, but also in more efficiently utilizing space. A study accompanying a patent (Osamura et al. 2002) found that if wilted enough, eight tobacco stalks could fit on one standard stick and cure with no adverse reactions. If the plants just had mild wilting, however, six was the maximum number of plants recommended for one standard stick. An increase from six to eight plants facilitated by increased wilting would yield a 25% reduction in space used, either onboard a mechanized harvester or in a curing barn.

2.2 Physiology of Tobacco in Relation to Wilting

Plants are living organisms, and as such naturally vary and sometimes behave in ways that are not totally predictable. The evapotranspiration process through a plant is a good example of this and can vary due to things such as field location, soil type, etc. The one action that remains in direct correlation with plant transpiration is the opening and closing of the stomata on the plant's leaves (Jarvis and Mcnaughton 1986). There is no known safe way on a plant that will be used for human consumption to force it to open all of its stomatal guard cells. Krausche and Gilbert (1937) showed that copper sprays artificially increase leaf transpiration rates, even going against the well-being of the plant, but such sprays added to the leaf would not be tolerated in commercial tobacco. L. R. Walton et al. (1976) determined that a tobacco leaf's pliability is directly related to its moisture content. The location of the plant in the field, the height of the leaf on the plant and other interactions were not significant. This indicates that an accurate moisture content reading will show the wilting status of the plant; however, each individual plant and even each leaf have their own unique maximum moisture content.

The reason a plant wilts is simple: the transpiration needs exceed the absorption by the roots so the plant's moisture content decreases (Briggs and Shantz 1912). Transpiration capacity within a species can be related to the surface area of the leaves. Likewise, absorption can be directly related to the surface area of the roots. Knowing this, a leaf to root ratio can help predict how a plant will behave in certain situations. Parker (1949) showed that reducing the root surface area decreased transpiration nonlinearly. The root tips were shown to uptake far more soil moisture than any other part of the root ball.

Roots do not actively bring water into the plant; it is the pressure gradient between the soil and the atmosphere in the xylem of the plant that causes water to be taken up into the plant. This passive water movement concept is shown by Kramer (1933), who found that even after all the roots of some plants were killed, the plant would continue to uptake water and transpire (if the plant was left undisturbed). The function of the roots is to grow towards water and act like a conduit. Surprisingly, although roots do anchor the tobacco plant, more than half of its resistance to falling over from gravity or wind comes from the in-ground portion of its stalk rather than the roots (Casada, Walton, and Swetnam 1980).

Transpiration is directly affected by two major components: the plant itself, and the growing environment. Different plant species and even individual plants within each species can have different stomatal conductance, which is the average resistance produced by the stomata on the flow of water through it. Several environmental factors strongly impact the potential stomatal conductance: relative humidity, air temperature (both ambient and net radiation from the sun), and wind speed (Jarvis and Mcnaughton 1986). If a plant is held in a controlled environment with conditions being mild for that particular plant, then stomatal conductance becomes the only factor that affects transpiration. But because outside weather is constantly changing, so is each plant's transpiration rate. Plants measured at the edge of a row and at the inside of a plot will usually show different transpiration rates simply because of the difference in wind and radiation the plant receives. While the net radiation a plant receives does impact transpiration rates, the light intensity itself has been shown to have no correlation (Briggs and Shantz 1912). Raschke (1960) showed that on average 90% of the transpiration of a given plant is correlated to the net radiation. The decrease in wind on plants on the inside of a plot allows each leaf to accumulate a larger boundary layer; therefore, in most cases these plants will wilt less often than plants on the edge. The stomatal conductance of tobacco is much greater in the top leaves than it is in the bottom leaves (Figure 2-1). This is due to the increased radiation and wind that the top leaves encounter.

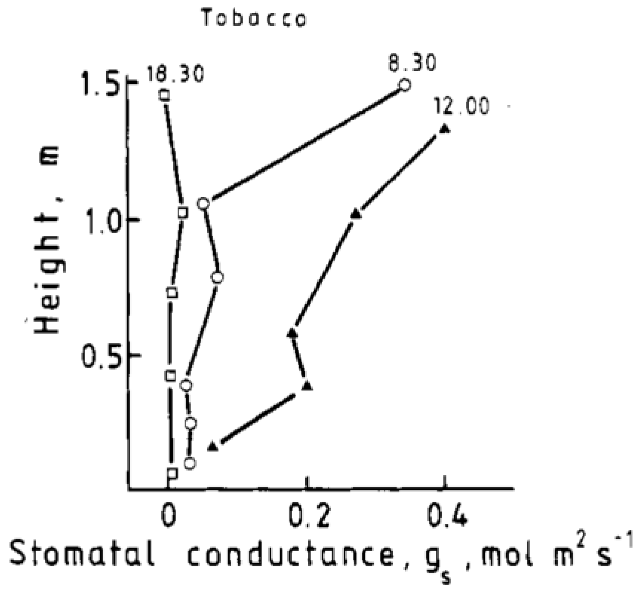


Figure 2-1 - The vertical distribution of abaxial stomatal conductance at three times of day in a canopy of tobacco. At 8:30 and 12:00 the top leaves transpire more than the bottom leaves; in the afternoon the transpiration rate reaches zero, this is the most likely time for a plant to wilt. After, Turner and Incoll (1971).

Moinat (1932) points out that when a plant has reached permanent wilting it does not mean that a certain level of soil moisture has been reached. The soil moisture content and the state of wilting of the plant are related by the wilting coefficient. For a short period of time, plants remain in an “apparently permanent wilted state” while not actually being permanently wilted. In this state the plant is using water reserves from its own tissues.

The wilting coefficient is a way to describe the amount of moisture a certain soil will have to have to make a certain plant undergo a permanent reduction in moisture content. Even if the plant were placed in a chamber with 100% relative humidity, it would still need water to be added to the soil in order to reestablish itself (Briggs and Shantz 1912). As the definition states, there are two variables at play: 1) the water holding capacity of the particular type of soil, and 2) the plant phenotype. For example, clay soil that is denser and resists water movement will cause plants to have a harder time absorbing the water from the soil; therefore, these plants wilt more easily. Plant phenotype can alter the wilting coefficient by producing a plant with characteristics such as a more robust root system, or a bigger leaf surface area.

The root system of a plant not only serves as the conduit to absorb water and nutrients, but it is also the anchor that holds the plant in the ground (Kramer and Boyer 1995). The roots adapt as the plant is growing to both meet the needs of keeping it anchored and finding enough water to sustain the plant. Plants with root systems that develop to extend deeper often are more stable in soil moisture extremes and droughts. In the case of tobacco, the roots are essential since they are where nicotine synthesis occurs (Kramer and Boyer 1995). Primary roots and the taproot develop early on and usually exist for the life of the plant, whereas smaller fibrous roots, or root hairs, grow and die in a constant cycle. Most all of the absorption in the root system comes from the newly grown fibrous root tips. It can then be theorized that severing the root tips from a root ball would effectively diminish the potential water absorption by more than half. Root regrowth starts almost immediately, but roots will only grow in soil that is above the wilting coefficient. Jarvis & Mcnaughton (1986) showed that when water in a field reaches a level where the plant wants to uptake more water than is available, stomatal conductance changes and becomes very low. The plants will then respire as little as possible, trying to conserve water. The plant will not wilt until the osmotic potential is greater in the soil than it is in the roots.

Moinat (1932) measured plants that had lost 30% moisture content of the leaves when they reached permanent wilting. This number does not mean much because different species and strains of plant as well as age, soil type, etc., affect the moisture content where plants cannot recover. This study also determined that the moisture content of a plant of the same variety, age, and conditions will remain approximately constant when the plant reaches permanent wilting.

Another common practice in tobacco production is a technique called “topping”, where a worker cuts the upper flowering portion of the plant off (Steinberg and Tso 1958). Topping a tobacco plant has many benefits, such as higher alkaloid content and increased size and weight of leaves, but the osmotic pressure potential remains unchanged (Steinberg and Tso 1958) – meaning that this practice likely has no effect on the outcome of this study.

The transpiration process is an almost purely physical process that occurs when water moves from soil to plant to atmosphere (Begg and Turner 1970). This process is determined by the gradient between the water potential in the soil and in the atmosphere. The major resistances along this soil-plant-atmosphere continuum are at the entry and exit points of the plant: the roots and the stomata. The atmosphere is usually the controlling factor in evapotranspiration on a daily basis, with transpiration peaking usually around mid-day when the environmental conditions such as radiation have reached their peak. The whole soil-plant-atmosphere system can be thought of as an electrical circuit where the stomata and other restrictions are the resistances and the flow of water is the current, etc. Cowan (1965) diagrams what this soil-plant-atmosphere system would look like (Figure 2-2).

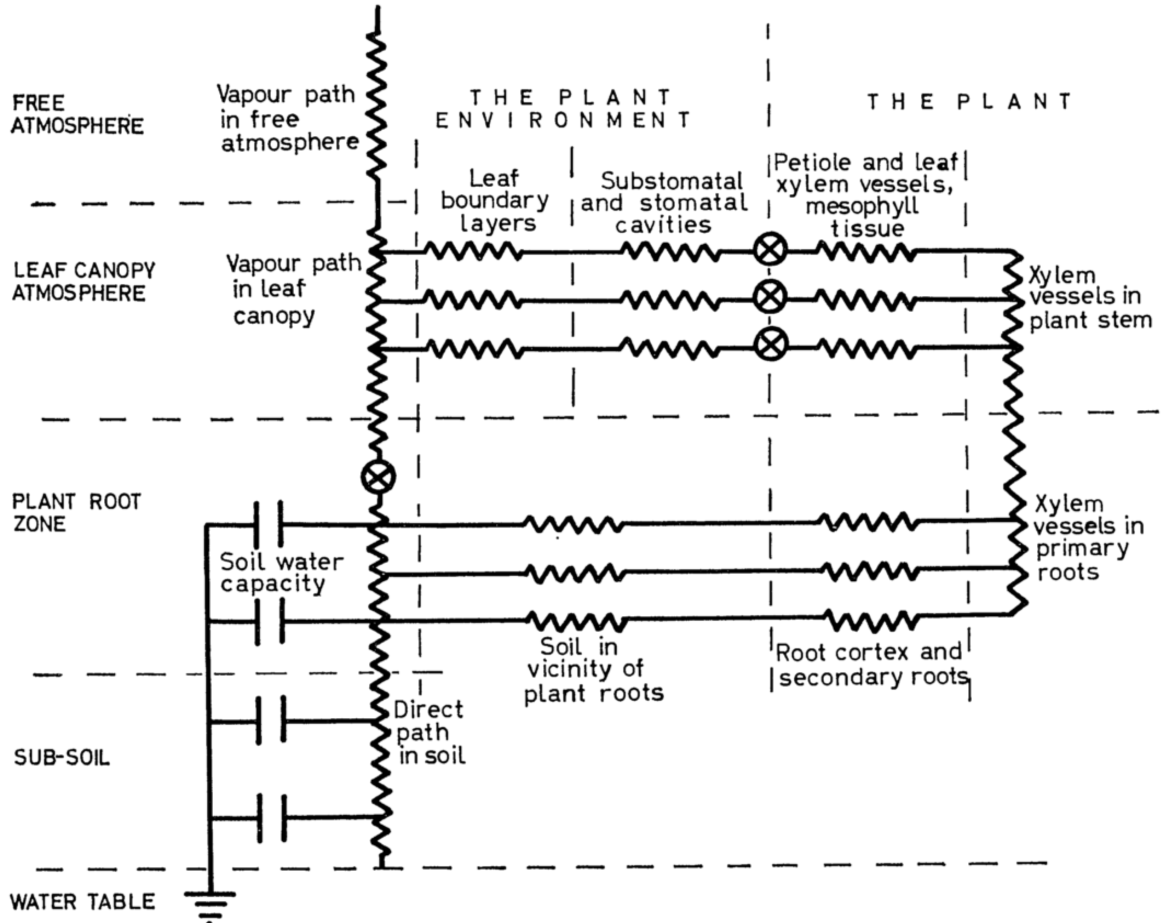


Figure 2-2 - Representation of the pathways that water follows in the soil-plant-atmosphere system. Diagram uses electrical engineering notation to represent things such as water pathway resistance and water holding capacitance (Cowan 1965)

The majority of water is taken up by root-tips and newly formed sections of root which are usually white (Bottomley, Rogers, and Foster 1986). The depth that the root ball grows within a certain strain of tobacco depends on the soil conditions while the plant was maturing. A plant experiencing a dry growth phase compared to a plant in soil with optimum soil moisture will have a deeper and more robust root system (Comas, Eissenstat, and Lakso 2000).

Environmental stresses such as frost, drought and excess salt, as well as non-environmental stressors like mechanical damage, can temporarily lower cell water potential (Hincha et al. 1987). These stresses usually lead to membrane dehydration and slowed transpiration. This condition can be seen as wilting, but it is not the same process as wilting due to an imbalance in the transpiration-absorption system (Schneider and Childers 1941). Another possible environmental plant stress is low soil temperature. Low soil temperatures stress the plant and affect the root system in several ways that inhibit the absorption of water including: slowed root cell activity, increased viscosity of water, slowed root growth, and decreased permeability of root cells (Kramer 1940).

Another stress that can contribute to tobacco plants wilting is an anaerobic root zone, which happens when constantly heavy rain or a flood is encountered (Figure 2-3 and Figure 2-4). Not only is this a stress, but inhibiting respiration of the root zone completely stops plant respiration (Kramer and Jackson 1954). When respiration stops, transpiration also stops and plants will stop taking up water since they cannot utilize the water without oxygen.

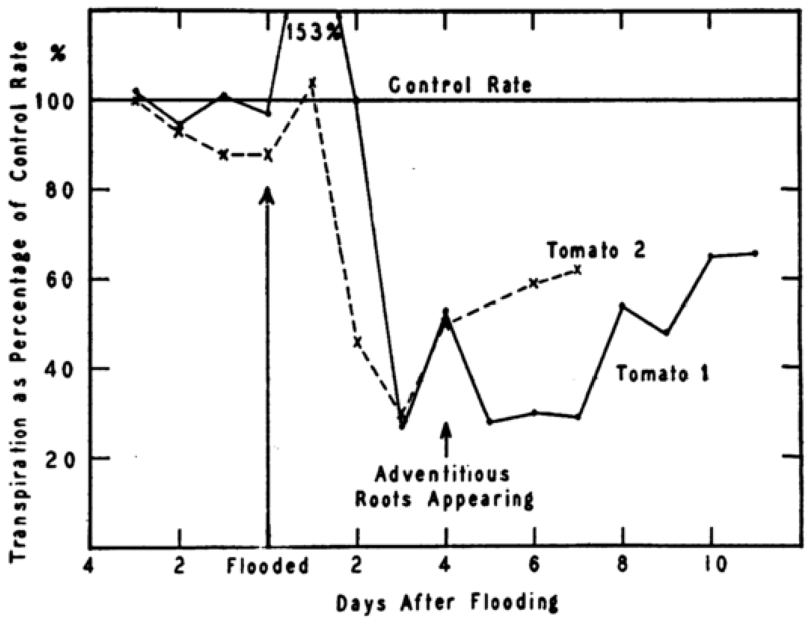


Figure 2-3 - Transpiration rate is negatively impacted by flooding the root zone. (Kramer 1951)

EFFECTS OF VARIOUS FLOODING AND GAS TREATMENTS ON TOBACCO PLANTS *

TREATMENT	REACTION ON FIRST DAY OF FLOODING	REACTION ON SECOND DAY OF FLOODING	REACTION AFTER FOUR DAYS OF FLOODING
1. Soil flooded without aeration	Severely wilted at midday. Plants flooded 1 day and drained recovered.	Slightly wilted at midday. Plants flooded 2 days and drained did not recover.	Severely wilted early in morning. Severe chlorosis of leaves.
2. Soil flooded with aeration	No wilting.	No wilting.	No wilting at any time. Very slight chlorosis.
3. Soil at field capacity, saturated with CO_2	Severely wilted within an hour. Recovery in late afternoon.	Wilted earlier and more severely than plants with other treatments.	Unwilted early in morning and only slight wilting at midday. Moderate chlorosis.
4. Soil at field capacity, saturated with N_2	Moderately wilted at mid-day, recovered in late afternoon.	Moderately wilted by noon.	Lower leaves slightly wilted early in morning, only slight wilting at noon. Moderate chlorosis.
5. Roots washed free of soil and flooded without aeration	Less severe wilting than in plants with roots in soil (Tr. 1).	Upper leaves severely wilted by noon and all leaves wilted in afternoon.	Severe wilting early in morning. Some death of leaf tissue. Severe chlorosis of leaves.
6. Roots washed free of soil and flooded with aeration	No wilting.	Slight wilting at midday.	Slight wilting at midday. Slight chlorosis.

* Six plants per treatment.

Figure 2-4 - Effects of flooding and gas treatments on wilting of tobacco plants. (Kramer and Jackson 1954)

Stresses and periods of drought or flooding can impact the plants differently depending on how mature they are. Maw, Stansell, & Mullinix (1997) found that with certain varieties of tobacco, a drought during weeks eight and nine of plant growth was the most detrimental to the crop, while a drought during the weeks preceding and succeeding those weeks was slightly less harsh on the crop. They also verified the belief that slight undersupplied soil water availability leads to enhanced tobacco root development. In cases of drought, flooding, or severe stress, tobacco often wilts immediately (Kramer 1951). When root death occurs, roots are fairly slow to start regrowing.

2.3 History of Attempts to Pre-wilt Tobacco

Few attempts have been made with tobacco to purposely hurt the crop or make it wilt. Work that has been done was mostly looking for things to avoid doing to tobacco plants so that they did not wilt. P J Kramer (1940) found that when the soil temperature was dropped, root activity and absorption decreased, which considerably slowed transpiration and plant growth (including root growth). But a side effect of a massive cut off in water absorption is wilting. Tobacco plants wilted when soil temperatures were lowered to between 3^o and 5^o Celsius. When soil temperatures were raised, the wilting effect dissipated.

Kramer & Jackson (1954) showed that flooding can cause similar temporary wilting of tobacco plants. They believed that this wilting occurred because the roots were starved for oxygen so that respiration could not occur. Testing was done to flood the soil with CO_2 and N_2 gasses; not surprisingly, severe wilting occurred, with permanent plant damage occurring after only three days. Two different types of wilting were shown. The first type is a short and temporary wilting induced by flooding within one hour of treatment; the second is permanent wilting effects that stay with the plant as the soil is kept flooded for more than 24 hours straight.

Andrews & Newman (1968) point out some interesting phenomenon describing the effects of root pruning on various plants. They showed that while most of the water uptake is in fact reduced by cutting off the new root tips, plants did not have a linear correlation between amount of roots lost and transpiration rate. Cutting half of the roots lowered the transpiration rate by 30% in some cases, while cutting 75% of the roots would cut 80% of the transpiration rate in other cases. As expected, they found that the plant transpiration rate was further reduced in soils with lower water content – crops with soils at field capacity did not transpire as fast.

Peroxidases are ubiquitous in all contemporary higher-order plants. This chemical is a type of glycoprotein that helps the plant oxidize a wide variety of compounds. Peroxidase-induced wilting has been researched on tobacco plants that have been genetically altered and works to some extent (Lagrimini, Bradford, and Rothstein 1990). This method works by increasing the rate of moisture that stomata transpire as well as decreasing water uptake by the roots. It is not likely that genetically altered plants will be sought after for their increased tendencies to wilt so long as the wilting cannot be easily controlled. If a crop were subject to drought midway between planting and harvest, for example, wilting would be undesirable -- and possibly even disastrous.

2.4 Measurement of Water Status in Plants

2.4.1 *Introduction*

The best way to directly measure wilting in a plant is to measure the turgor pressure of the plant. An alternative is to indirectly measure wilting by measuring its effects on the plant, i.e.: drooping leaves, change in color, etc. (Turner 1981).

2.4.2 *Direct Measurement*

The most common method for measuring the turgor pressure of a leaf is the use of a pressure bomb chamber. Using this chamber, a leaf's excised stem is open to the atmosphere while the other side is closed in a sealed chamber and pressurized (Turner 1981). When water is pressed from the leaf's tissue out of the stem, a balance in pressure between the xylem and the atmosphere has been achieved. This pressure is equal to that of the turgor pressure (Wei, Tyree, and Bennink 2000). While this can be done in the field, it is not a very rapid procedure and not suited for a large sample size as the gas that is used to fill the chamber only lasts for a limited number of readings. The instruments are better suited for small leaves; big leaves, such as tobacco leaves, require a big chamber. These instruments are more expensive and use far more gas than more common models. At the time of writing, an entry level field version of a pressure bomb for a tobacco leaf was around $2000.

If cost was no issue, a product made exclusively by ZIM Plant Technology, described in Zimmermann et al. (2008) and Hüsken, Steudle, & Zimmermann (1978) would be one of the best options. It is a real-time, constantly monitoring probe that can be placed on any plant leaf and is capable of accurately measuring turgor pressure by relating the thickness of the leaf to the pressure inside of the leaf. Additionally, this method is non-destructive and field suitable while being highly precise. However, at the time of writing, this product was very expensive – costing more than $10,000 to continuously measure nine subjects at once. A similar intracellular pressure probe exists, but it is also very expensive and measurements are not suited for rapid, field type measurements.

The moisture content of a leaf can be determined using a simple and common process (Turner 1981). Plant leaves can be collected and weighed immediately to determine a wet weight. Then they must be oven dried at 60 to 105 Celsius for 24 hours. A dry weight is then measured and moisture content on a wet basis can then be calculated. This directly relates to turgor pressure but is not turgor pressure itself. To obtain relative water content, a fully turgid state must be acquired. To do this a leaf can be submerged in water or placed in a humidity chamber until the leaf is fully turgid. If samples cannot be weighed immediately, they must be sealed from the atmosphere so no moisture can enter or leave.

2.4.3 *Indirect Measurement*

Hunt & Rock (1989) describe a method of measuring the relative water content of a leaf by near and middle-infrared reflectance. Relative water content is defined as the current volume of water contained in a leaf divided by the maximum volume of water a leaf can achieve (fully turgid). Leaf reflectance was related to leaf weight by simultaneous measurement. Calibration of the reflectance method and the subsequent measurements of actual data must be done in a controlled setting (not field settings).

Ball tonometry, described by Philip M. Lintilhac, Chunfang Wei, Jason J. Tanguar 2000, is a method in which a glass sphere is pressed into thin-walled plant leaf tissue to obtain the contact area of the sphere on the leaf. The advantages of this method are that it can be done rapidly, accurately, and nondestructively; the disadvantages are that it is an indirect measurement.

Calibration of the relationship between how the glass sphere affects the leaf and the leaf's relative water content has to be determined for the variety of burley tobacco being tested. Also, while this can be done rapidly, it is likely very hard to perform in the field requiring both a high technical skill as well as expensive equipment. In the study mentioned above, the subjects were analyzed in a laboratory while submerged in water.

Several methods exist to determine the water potential gradient in a tobacco plant, which would give the evaporative demand created by the soil-plant-atmosphere system (Begg and Turner 1970). Though this would allow for measurement of transpiration, it does not necessarily tell the current water status of the plant (Turner 1981). It is possible for a plant that is fully turgid to have a similarly low transpiration rate as that of a plant that is wilted, thereby making the rate at which the plant uses water not a main factor. It should be noted that if both the osmotic potential and the water potential are known, turgor pressure can be calculated. To utilize this method, a thermocouple psychrometer chamber must be used, and the relatively slow process must be conducted in a laboratory (Nonami, Boyer, and Steudle 1987).

Work was done by Seginer, Elster, Goodrum, & Rieger (1992) to use computer-vision tracking to monitor and determine the wilting of a given plant. This method is nondestructive, continuous, and can be related to vertical movement of leaves, plant water potential, angle of leaf droop, and other features. Unfortunately, this was done in a closed lab setting with a special backdrop and no wind. Because the tobacco in this study will be in a field setting, a computer is not likely to be able to track leaf tips with the background and foreground potentially containing similar objects. While automated tracking may not be feasible in this situation, picture analysis or time-lapse photography may be useful, although it is not as easily or quickly quantifiable (Turner 1981).

Mechanical stress on the stalk of tobacco can be measured, similar to how breaking stress can be measured on the midrib of a tobacco leaf. These tests can be done quite accurately, but because tobacco has a woody stalk, neither the moisture content of the plant nor the fragility of the leaves are related to the strength of the stalk (Fiedeldey, Walton, and Walker 1991).

Roots can be excised from the ground to be examined both for root weight and state of the roots. To do this a known volume of root ball is excised; the roots are thoroughly washed, examined and dried. A mass can then be measured, giving a root density (Comas et al. 2000).

2.5 Soil Cutting Resistance

To design any sort of implement capable of root-pruning in a field setting, an estimation of expected forces is necessary. Unfortunately, there is very little literature concerning the forces exerted when cutting soil using a smooth coulter disc at varying depth, soil moisture content, soil type, angle of incision, and compaction. Recently, Afify and Kushwaha (2011) published results from a series of tests that have been completed to approximate the implement force requirements. The authors described the use of smooth, tractor drawn 0.46-meter diameter coulter discs and measured the forces exerted on them. The factors that affect these forces are summarized in Figure 2-5. Moisture, soil compaction, angle of cut, and depth were all found to be highly significant at a 1% confidence level. Pitla, Wells, & Shearer (2009) performed a similar test on a 0.81-meter diameter disc. This size disc was likely closer in size to that needed for tobacco plant root pruning, and additionally the test was conducted in Kentucky soil. The disc was fixed vertically, and at a depth of 0.3 meters had a required horizontal force of about 9.5KN and a required vertical force of about 13KN. These values are a good starting point, but unless exact conditions and soil type are copied, exact force requirements can only be obtained experimentally.

Results of analysis of variance for draft, vertical force, side force, and furrow wall strength (460 mm diameter disc).

Factor	Type	Levels
Moisture (M)	fixed	2
Compaction (C)	fixed	3
Angle (A)	fixed	7
Depth (D)	fixed	3

Source	DF	Draft	Vertical force	Side force	Furrow wall strength
M	1	0.000**	0.000**	0.000**	0.820-
C	2	0.000**	0.000**	0.000**	0.000**
A	6	0.000**	0.000**	0.000**	0.000**
D	2	0.000**	0.000**	0.000**	0.000**
M*C	2	0.365-	0.000**	0.061-	0.153-
M*A	6	0.000**	0.000**	0.000**	0.000**
M*D	2	0.000**	0.000**	0.000**	0.702-
C*A	12	0.004**	0.000**	0.000**	0.022*
C*D	4	0.000**	0.257-	0.003**	0.140-
A*D	12	0.000**	0.000**	0.000**	0.027*
M*C*A	12	0.445-	0.001**	0.010**	0.014*
M*C*D	4	0.378-	0.095-	0.192-	0.479-
M*A*D	12	0.001**	0.000**	0.000**	0.506-
C*A*D	24	0.487-	0.437-	0.222-	0.708-

** Highly significant at 1% level of confidence.

* Significant at 5% level of confidence.

\- None significant.

Figure 2-5 - An investigation into what factors affect soil penetration resistance are examined. (Afify and Kushwaha 2011)

CHAPTER 3 – Initial Testing

3.1 Introduction

Regardless of the phenotype, tobacco grown in the area in and around Kentucky is harvested once per year, typically between September and October. Because there was limited time for experimentation, testing was started almost immediately after the beginning of the project in September 2009. This first phase of the project addressed Objective 1, Task 1 of the project goal, which was to identify and refine techniques to pre-wilt tobacco. Using very early research and brainstorming, several prewilting treatments and measurements were devised. These methods were tested, iterated, and refined using the following specific sub-objectives.

3.1.1 Sub-Objectives

3.1.1.1 Sub-Objective 1: Find an optimal candidate for mechanically pre-wilting tobacco plants.

The ideal wilting method resulting from this research would have to encompass several key components. First, the proposed method must wilt tobacco in a robust manner. Second, the method must be able to be mechanized efficiently. Last, the treatment cost per plant must be low enough per plant to make this process economically viable.

3.1.1.2 Sub-Objective 2: Find and evaluate appropriate methods to test and measure the wilted state of a plant.

Wilting is a difficult process to adequately quantify while plants remain standing in the field. Because of this difficulty, various methods and schedules were attempted to better translate the wilting effect into objective data.

3.1.1.3 Sub-Objective 3: Determine the optimal times to both apply the experimental treatments and measure the results.

Wilting is the result of a confluence of factors, some of which relate to the time of day. An optimal window of time to apply the treatments so that maximum wilting can occur was important to determine. The time of maximum wilting was then to be determined so that data collection was able to be performed at time of maximum effect.

3.2 Experimental Methods & Data Collection

3.2.1 Introduction

In the first phase of testing, two locations were used to carry out preliminary trials on burley tobacco. Some of the treatments, methods, and plant water status measurements that were proposed were later changed due to difficulty in the field. During this phase of testing the primary focus was trying to wilt standing tobacco by any means necessary, and then performing tests on it to accurately quantify that wilted state.

3.2.1.1 Locations and Subjects

Two locations were used for phase one of testing. One plot was located in Lexington, Kentucky at the UK Spindletop Research Farm (38° 6'32.20"N 84°29'50.91"W), which will be subsequently referred to as Plot A. Another plot used that same year was located in Princeton, Kentucky at the UK Princeton Research Farm (37° 5'41.62"N 87°51'49.10"W), which will be referred to as Plot B.

All tobacco used in this phase of testing was commercial grade, was grown in the typical row and plant spacing of 42 inches between rows and 24 inches between plants, and had been topped at the appropriate time according to contemporary best management practices. Plot A at Spindletop Research Farm contained the KT206 variety, and Plot B at the UK Princeton Research Farm contained the TN90LC variety. KT206 was a common burley tobacco variety. TN90LC was also a common variety, but it had slightly different characteristics, such as being more prone to ground suckers.

3.2.1.2 Treatments

A total of 11 techniques were tested in the field; some of these methods were refined iteratively over this phase of testing. These techniques were chosen either because they had been found in the literature review to possibly cause wilting, or via brainstorming possible methods to disturb the water balance of the plant. A summary of the methods tested can be found in Table 3-1.

From the literature review, it was felt that root pruning had the highest potential for mechanization, so several variations were explored. Root pruning by means of a reciprocating saw with a 12 inch (30.5cm) blade was the main focus of this phase of testing. This was done on either two or four sides of the plant at either 1.5 inches or 3 inches away from the stalk with an angle of 20 degrees from vertical. The distances of the cuts were initially 2 inches and 4 inches but were changed after initial pre-season testing.

In addition to root pruning, other methods were explored. These methods included freezing the lower stalk with R-134a freeze spray (Figure 3-2), drilling a hole and spraying the freeze spray inside, boring out of the pith area (Figure 3-1), and cutting around the plant with a blade.

Table 3-1 - Summary of treatments in phase one

Test Type	Treatment Number	Description
Main Cohorts	1	Root Pruned on two sides at 1.5" (3.8 cm) away from the stalk
	2	Root Pruned on four sides at 1.5" (3.8 cm) away from the stalk
	3	Root Pruned on two sides at 3" (7.6 cm) away from the stalk
	4	Root Pruned on four sides at 3" (7.6 cm) away from the stalk
Additional Small Scale Tests		-Freezing the stalk with Freeze Spray
		-Drilling a hole through the stalk, then adding Freeze Spray
		-Boring out the pith area from the stalk
		-Burning the stalk with propane
		-Boring out the pith then burning with propane
		-Hose clamp around stalk
		-Cutting wedges out of stalk

Figure 3-1 - A custom drill bit fabricated to bore the pith out of a tobacco stalk without damaging the structural integrity

Figure 3-2 - Treating a tobacco stalk with electronics freeze spray

3.2.2 Procedure

Root pruning was conducted on September 14^{th} and 16^{th}, 2009 in Plot B and Plot A, respectively. Baseline photographs and soil moisture content measurements were collected at each location when the treatments were applied. Six days after the treatments, final pictures were taken of each treatment group from the same point of view used for the baseline pictures. During this time, general observations were recorded about blown over plants, wilted plants, and anything else out of the ordinary.

After the photographs and soil moisture content measurements were recorded on day 6, the two plant water status tests were performed; these were leaf breaking angle and leaf water content tests. The two most visually wilted plants in each treatment block were chosen, and two leaves from each plant were tested: the second leaf from the top and the eighth leaf from the top. First, the angle of the leaf would be recorded at rest using the modified protractor. Next, the leaf would be bent downward from a bolt 2.95 inches away from the base of the protractor, until it broke. The leaf angle at the point of breakage was recorded. This method was replicated from work by Yoder, 1985 in a study on wilting tobacco (Figure 3-3).

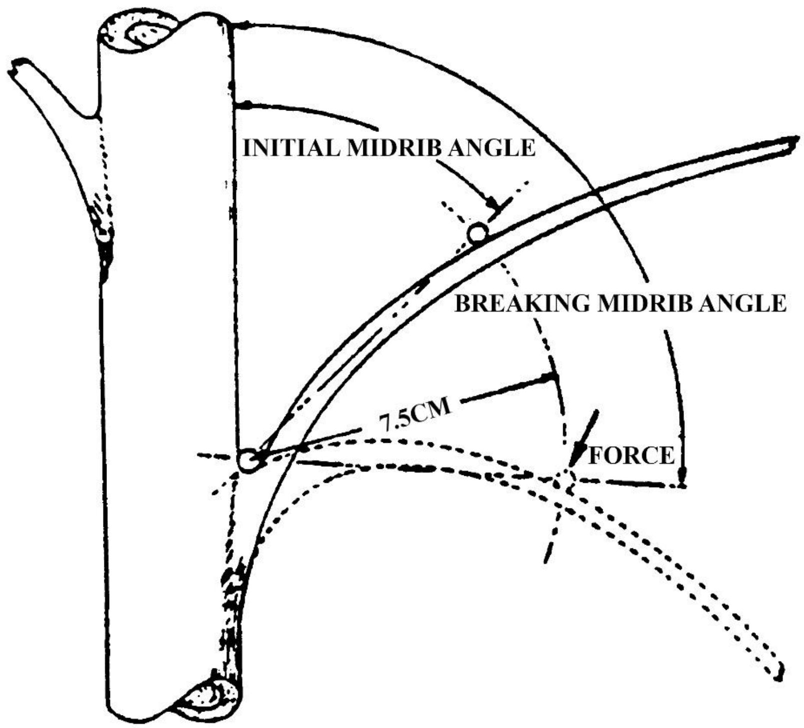


Figure 3-3 - Diagram of leaf breaking angle test, Yoder (1985)

To simplify the testing procedure, after the leaves were broken off the plant for the leaf water content test, they were immediately placed in sealed plastic bags and marked indicating their plot, treatment, and leaf number. In this way, the leaves would not lose any discernible water from when they were broken off the plant. These were transported back to the University of Kentucky to be weighed. A digital scale was tared with the same type of plastic bags used to transport the leaves, and then each sample was weighed and recorded. The leaves could not be removed from the plastic bags and weighed because moisture had transpired out of the leaves and onto the walls of the plastic bags. The leaves were then individually moved to small paper bags, labelled appropriately, and stapled. This method allows the leaves to dry out while keeping every piece that is not able to evaporate inside the paper bag. The bags were then placed inside a drying oven at 75 degrees Celsius for 24 hours (Figure 3-4). The same portable digital scale that was used before was moved next to the drying oven to provide quick and easy access for weighing samples. It was very important that the samples not be left out and allowed to cool down before collecting the final weight; the dry plant matter and the paper bags can possibly reabsorb moisture. To test moisture reabsorption an empty bag was allowed to heat up with the plants for 24 hours; it was the first to be weighed and recorded. After 20 minutes the bag was reweighed and it had gained 0.5g. This source of error was considered small enough to be ignored since the Ohaus scale fluctuated up to ±0.9g on some measurements when checked against lab quality calibration weights. The bags were then taken out and weighed one by one while the oven was still on to minimize cooling and moisture reabsorption.

Figure 3-4 - Paper bags in the drying oven, each with a tobacco leaf inside

In early August to September 2010, a time-lapse camera was utilized in several different locations. Using a single weatherproof camera and tripod setup, a series of pictures were taken using the camera's intervelometer mode at 15 minute increments for a period of about three days. This process was done for several treatments and a control group. In all shots with a treatment group pictured, the camera was placed so that it was possible to see non-treated plants to the sides and/or behind for comparison purposes.

After collection was completed, the resulting pictures were downloaded to a computer where dark or otherwise obstructed shots were deleted. The rest of the good shots were placed in a movie timeline in Adobe Premiere Pro and each given a timestamp with the date and time. These movie clips were then exported individually, running at five frames per second.

3.2.3 Data Collection

In each plot, there were five treatment groups (including a control) and six plants within each treatment replicated in three blocks. In the Plot A tests, six plants were added in each block's tests to try alternative wilting methods. There were three new methods tried, so in each block two plants were tested from each method, allowing testing of six plants total over the whole location. Therefore, in Plot A n = 108 and in Plot B n = 90. Note that after the duration of the test not every plant remained upright, and not every plant that remained upright was used for testing, so the statistical model differs in number of subjects, with n = 76 and n = 53, in plots A and B, respectively. The experiments used a Randomized Complete Block design. The tests were replicated in two different regions of Kentucky and using two different varieties of tobacco – which means we cannot compare either factor in the overall analysis. This was of little concern, since the tests carried out aimed to determine whether the treatments had any effect.

In each of the two locations there were three blocks with all treatments and a control group included. These blocks were all in very close proximity in the field as to exclude soil conditions from affecting the results. Inside each block each of the treatments were applied to six plants, excluding plants that were sick or damaged. In this design each treatment is labelled a plot, and the individual plant is labelled the subplot. Additionally in Plot A, three more treatments were tested to compare with root pruning. The other three aforementioned methods, were: burning the stalk, cutting the stalk with string, and cutting the stalk with blades. Two of each of these per block were performed (totalling six subjects). While these methods had less data points than the first five treatments, there were enough data to indicate whether or not there was an effect.

The variables measured in this phase were: leaf water content, leaf breakage angle, visual analysis, and green weight. The green weight analysis was an effort to scale up the leaf moisture content test and weigh the entire plant. The 'green weight' metric was later abandoned, as it was too cumbersome to cut and weigh all tobacco plants used in data collection, and little value was gained by doing so. Leaf breaking angle is an important factor in determining whether the treatments affected wilting – the more the leaves can bend and flex before they break, the less likely there will be damage to the plant during transportation or hanging. Leaf water content was also thought to be important as it directly relates to how much a plant is wilted and allows quantification of plant wilting. Meticulous pictures were also taken of every plant from the same location about every two days to give a straightforward comparison of the same plants over several days.

3.3 Results and Discussion

3.3.1 Results

The tobacco did not act as predicted to the various treatments applied. The plants were generally far more resilient and unpredictable than anticipated. Positive cases of temporary or permanent wilting were seen, but on an inconsistent basis. Results from the first phase of pre-wilting tests were statistically inconclusive, as were the visual results.

3.3.2 Photographic Observations

Time-lapse Video 3-1, shot at Plot A, shows three different treatments: the two left-most plants were cut at 3-inch distance on four sides, the two middle plants were cut on two sides at a 3-inch distance, and the two right-most were cut at a 1.5-inch distance on two sides. It is clear to see that the four sided cut had the most noticeable effects in this particular test. Moreover, one of the plants that had been cut on four sides reached a state of permanent wilting almost immediately and never recovered. The rest of the plants wilted, then regained turgor in a typical sinusoidal pattern commensurate with the daily heat. It was clear there is a daily cyclical pattern of wilting, reaching its peak at about 6PM.

Video 3-1 - Timelapse video of root-pruned plants: 4-sides at 3", 2-sides at 3", and 2-sides at 1.5"

Video 3-2 - Timelapse video: yellow flag - MAPP gas on stalk, orange - liquid nitrogen poured on plant, blue - liquid nitrogen poured on stalk

Video 3-2 shows special tests conducted at the end of phase one: burning the stalk with MAPP gas, pouring liquid nitrogen all over the plant, and pouring liquid nitrogen directly onto the stalk. MAPP gas used to burn the stalk for 30 seconds evenly around the stalk, liquid nitrogen poured for 10 seconds over the top of the plant, and liquid nitrogen poured from a spout onto the side of the stalk for 10 seconds. No wilting is apparent from any plant during this time-lapse video. The plants with liquid nitrogen poured on top of them developed serious burns and holes in the leaves. The treatment protocol for these early tests was lacking and was improved before later tests.

Video 3-3 - Timelapse video of 2-sided root pruning: yellow flag - 4.5" cut, orange - 3" cut, blue - 1.5" cut, white – burnt

Video 3-3 documents a mixture of two-sided root-pruning and MAPP gas tests. The yellow flag marks 4.5 inch cuts, the orange flag marks 3 inch cuts, the blue flag marks 1.5 inch cuts, and the white flags to the far right mark MAPP gas treatments. This video shows the gradation in intensity the plants in the same conditions can reach after being treated.

The 4.5 inch cut did noticeably wilt the plants, but not very much, and for only a few hours a day. The close two sided cuts are all similar: heavy wilting can be seen, but it is cyclical in nature. Finally, the two plants with their piths killed slowly became intensely wilted and did not exhibit the same cyclical pattern.

3.3.3 Discussion

Results from the pre-wilting test measurements in phase one were inconclusive, as were the visual results. As can be seen by Table 3-2*,* all treatments were very close to one another. It should be noted that while the mean leaf moisture content stayed fairly close between treatments, some individual root pruned plants had moisture contents as low as 65% wet basis. In some cases, like the case of *control* vs. *treatment 4* in leaf moisture content, the control group actually had a lower moisture content than the treated group. Note that this does not mean that *treatment 4* actually increased the leaf moisture content, but that the results are too variable and close together to be significant. Leaf midrib breaking angles varied greatly from location to location, but this is not a sign of wilting either, rather a factor of plant density and variety.

Table 3-2- Summarized results between treatments and locations, phase one

Treatment	**Plot A**		**Plot B**		**Average**	
	Leaf Range (degrees)	*MC (% w.b.)*	*Leaf Range (degrees)*	*MC (% w.b.)*	*Leaf Range (degrees)*	*MC (% w.b.)*
1	42.75	83.49%	20.33	83.86%	31.54	83.68%
2	58.08	82.83%	23.83	83.08%	40.96	82.96%
3	42.67	83.19%	19.25	84.13%	30.96	83.66%
4	49.67	82.96%	16.75	83.07%	33.21	83.02%
Control	45.25	82.35%	18.58	83.70%	31.92	83.03%
Average	47.68	82.97%	19.75	83.57%	33.72	83.27%

Treatment 2 (cut four-sides, 1.5 inches away) showed an increased leaf breaking range in all tests and locations. Other treatments did not show a sizeable difference between the *control* group plants. Average moisture content measurements were very similar for all plants tested. A one-way ANOVA was performed in Microsoft Excel to look for differences between groups. No significant difference was found between any groups for any location ($p > 0.05$).

Lower plant moisture content indicates better wilting. A tobacco plant reaches a sufficiently wilted state when its moisture content is reduced to 75-80% on a mass basis according to Walton et al., 1976 and Burton et al., 1989. The leaf midrib angle breaking test indicates a change in resting leaf position and breaking leaf position, so when referring to the leaf breaking angle, the higher the number the better (for wilting). Practically, this indicates that the leaf is able to move further without breaking.

The only compelling evidence of wilting that was recorded in phase one were pictures of the wilted plants (Figure 3-5). There was undeniable wilting, but it was very inconsistent, and the degree of wilting varied greatly. Also, the methods of data collection could not accurately capture this wilting. Because of the positive photographic evidence, more investigation was done in ways to visually capture the wilting effect and what time it reached its maximum.

Figure 3-5 - Plants from the same vantage point that had been treated with a 4-sided cut on the day of the application, four days after, and seven days after, phase one.

The freeze spray did nothing at all to the plant because the cork layer in the stalk acted as insulation and prevented the plant from being rapidly heated or cooled at such temperatures. Some plants that were frozen with this method were immediately cut down in the middle of the treated area, and the pith and other vascular tissues were not frozen; in fact, the subjects were not very cold at all. No other treatments attempted had any significant results.

3.4 Conclusions

3.4.1 Overview

The findings in this experiment were not as conclusive as hoped, but the results were still useful in determining a better set of test procedures to use. It was predicted that severing some roots would cause some in-field wilting and severing nearly all the roots (in a pyramidal-plane fashion) would make a plant wilt. Tobacco plants seemed to be more robust than expected, even in dry soil. While isolated cases of wilting were seen, overall on the day of data recording there was little to no visible wilting. This allows for the possibility that the treatments performed were not severing the roots equally between all subjects, or that extraneous factors were playing a bigger role than anticipated.

The single biggest factor in the result of wilting, or lack thereof, seemed to be timing of data collection. The time frame of 7 to 8 days turned out to be entirely too long – most plants tested had experienced some regrowth of roots by the 5th day (Figure 3-6). Plants that were allowed to regrow roots seemed to have almost no chance at remaining wilted.

As noted in the results, many of the plants wilted almost immediately and recovered by the time data collection had begun. This indicated that the prediction that the longer elapsed time after the treatment, the more wilted the plant will be was wrong; there was a finite window where wilting will be at its maximum. In addition, there seems to be a daily sinusoidal wilting pattern that all plants (even non-treated control plants) incur.

Figure 3-6 - Healthy root tips appear only five days after root pruning

3.4.2 Impacts on further testing

Changes were made to the experimental design in the next phase so that each treatment could be more easily compared to another. For example, one location would group plants by treatment while the other location would group plants by day; this allowed for more in depth photographic comparisons. Data collection methods were also made more precise, particularly with improvements to the timing and efficiency of data collection.

Timing changes were found to have a much higher than predicted effect on wilting. Because of this, changes were necessary in all facets of testing: application, photography, and data collection. With more consistent and exact timing, it was predicted that the results would become more uniform than they had previously been. The average amount of wilting was also expected to increase since the “maximum” point of wilting would be captured more consistently.

CHAPTER 4 – Large Scale Testing and Refinement

4.1 Introduction

4.1.1 Introduction

Phase one of the project was successful in identifying several possible techniques for affecting and measuring wilting of a tobacco plant. The purpose of phase two of the project was to further refine these methods with the goal of identifying the best candidate for field scale mechanization, which is Objective 1, Task 2 of the project. Both novel and established treatments were tested in this section in order to exhaust all possibilities brought forth. The importance of the timing of treatments and data collection were underestimated in phase one; in phase two, much greater detail was paid to the timing with the possibility of refining schedules further by phase three. The following specific sub-objectives were completed as part of this testing phase.

4.1.2 Sub-Objectives

4.1.2.1 Sub-Objective 1: Select one treatment method to be mechanized.

The best treatment application to be mechanized had to be determined in order to proceed to the next phase of this project. This was accomplished by generating a decision matrix. Examples of factors that were included in this determination were: how often the treatment wilts the plant, how much the treatment wilts the plant, and how easy it would be to mechanize the treatment.

4.1.2.2 Sub-Objective 2: Refine methods and data collection.

Through time-lapse photography it was determined that time of day seemed to be the most influential factor affecting wilting maxima and minima. Knowing this, the data collection had to be conducted within a very narrow window of maximum wilting. Data collection methods were refined and standardized in this phase, and were subsequently able to collect data from a large number of subjects in a short amount of time.

4.2 Experimental Methods & Data Collection

4.2.1 Introduction

Refinements were the focus of testing in the second phase, especially with how and when the data collection occurred. Instead of measuring wilting a week after the treatments were applied, in phase two the data collection was conducted on the same day as the treatment (day 0), the day after treatment (day 1), and a week after the treatment (day 6). Using time-lapse photography it was determined that the time of day played a large role in the plants' state of wilting and that the highest degree of wilting occurred between 5PM and 6PM EST at the test locations in September of 2010.

4.2.1.1 Locations and Subjects

In 2010, Plot B was used again as well as a plot located in Versailles, Kentucky at the UK Woodford County Research Farm (38° 5'22.11"N 84°43'54.42"W). The Woodford Farm location was designated Plot C.

In the previous phase, different numbers of subjects in each location were used since a rigid experimental design had not yet been determined. In this phase of testing, more attention was paid to the experimental design; however, the plot layout was changed from one location to the other due to the layout of the crops that were available. In Woodford County (Plot C) the *treatment* factors were grouped together in the field, while in Princeton, KY (Plot B) the *day* factors were grouped together. For example, when working on the Woodford County test, the data collector must visit every treatment block for samples. When working in Plot B, the data collector would collect from a different block on each day. Because of the destructive nature of testing, there had to be a different plant for each *day* so that no plant was sampled on two separate occasions. The method used in Woodford County was to have the day 0 plants, day 1 plants, day 6 plants and an extra all in one line. In Plot B this was changed to having the five different treatments of a particular *day* of plants to be in a line. This also aided in photographing the subjects more easily.

4.2.1.2 Treatments

Four different treatments were considered in full-scale testing: 2-sided root pruning at 1.5 inches from the stalk, 2-sided root pruning at 3 inches from the stalk, burning the stalk with MAPP gas for 60 seconds, and string trimming the stalk then burning it for 60 seconds with MAPP gas. The root pruning tests were all done at a 20-degree angle from vertical with a cut distance of 12 inches deep. Each treatment was given a number, which would be used for flagging, data collection, etc. (Table 4-1).

Table 4-1 - Summary of treatments performed in phase two

Treatment	*Description*
1	Cut 2-Sides at 1.5 inches (3.8cm)
2	Cut 2-Sides at 3 inches (7.6cm)
3	String trim and burn stalk
4	Burn stalk
5	Control
6	Liquid Nitrogen

In the first phase of testing, the reciprocating saw had a very tough time cutting through the soil, and dulled quickly. The teeth were optimal size for cutting wood, but did not move enough material per stroke to adequately move soil. The saw blades for this phase were altered using a grinder (Figure 4-1) to make teeth that were much bigger than standard. This was tested before the start of the experiment and it performed far better than standard blades, making each plant much easier to treat and require less time.

Figure 4-1 - modified reciprocating saw blade

In Plot B just one repetition of a liquid nitrogen treatment was performed on four subjects. Full-scale testing would have been cost prohibitive as well as time intensive. Additionally, it was recognised that such a treatment would be very hard to mechanize under the project restraints and would not likely be pursued further. Because repetitions and blocks were not used with the liquid nitrogen test no full statistical analysis could be done to test this treatment's significance via analysis of variance.

Attempts to pour the liquid directly onto the plant failed; the contact with the plant tissue was too minimal. Therefore, the liquid nitrogen treatment required a specially made, two-piece Styrofoam fixture to be placed around the stalk of the plant. This device, once attached, became a cup to hold the liquid nitrogen against the plant stalk. The liquid nitrogen was then poured in a slow and steady stream into the cup, keeping it topped off for at least 60 seconds.

4.2.2 Procedure

Each treatment was repeated four times per group (three to be used for testing, and one as a spare), with four blocks (replications) of five different treatments. This is a total of n = 80 plants to be treated per location, including controls. The treated plants were flagged a different color for each treatment, including the control. Numbers written on the flags corresponding to when data collection would occur during the week for any given subject.

The experiments used a Randomized Complete Block design. The tests were replicated in two different regions of Kentucky and using two different varieties of tobacco – which means it was not possible to test whether location or tobacco variety influenced the results. This was of little concern, since the desired goal was a simple determination of whether the treatments had an effect, and not which location in Kentucky, tobacco varieties, tobacco spacing, etc., would respond most favourably.

4.2.3 Data Collection

Data were collected on days zero, one and six at both locations – with day zero being the same day the treatments were applied. All data were collected starting at 5PM and completed no later than 6:30PM. Data collection at Plot C was completed on September 13, 14, and 19, 2010 and data collection at Plot B was done on September 21st, 22nd, and the 27th 2010.

The first pictures were taken in the same manner as the phase one testing; the camera was located at the same position on each data collection day and each plot was identified in the picture. Time lapse was ultimately much more useful than taking one picture per day of each treatment group; however, the time lapse was only collected at one location for the entire seven days. Time-lapse was done at both locations on additional subjects not included in the general population of subjects being recorded.

Just as in the first phase, the leaf breaking angle measurements were the first destructive test performed. Unlike phase one, the first plant in the block was tested on the first day, the second plant on the second day, and so on. The modified protractor was then used to collect the leaf breaking angle range from the second and eighth leaves from the top of each plant. This was done in the same manner as it had been done in phase one. Each leaf was placed into a double sealed plastic bag with the air expelled, and then tagged with the appropriate location, block, treatment, replication, and leaf height.

The samples were then taken to the University of Kentucky Biosystems and Agricultural Research Labs where the moisture content of each leaf was determined. This was done in a similar fashion to phase one, except testing took place immediately after data collection each day.

4.3 Results & Discussion

4.3.1 Introduction

The second phase of testing had similar overall outcomes as the first phase. Individual plants or small groupings of plants would show signs of wilting in the evening, and regain most of their turgor by morning. All tested treatments followed this general pattern of inconsistency, with the exception of the plants treated with liquid nitrogen.

Photographic results from all tests showed some noticeable wilting in the time lapses and photos that seem to corroborate with measurements. Using time lapse photography greatly increased the likelihood that wilting was observed and measured, though it limited the amount of time for data collection each day.

4.3.2 Photographic Observations

Time-lapse video was utilized to be able to pinpoint which treatments appeared to have been working, and at exactly what times. The time-lapse video in Video 4-1 was taken at Plot B on September 25th, 2010 of some of the subjects that were included in the large scale testing. The results shown were fairly typical, especially for this variety (TN90LC) of tobacco. Small changes throughout the week were noticeable when comparing the treatments to the plants on either far side of the video, but nothing remarkable occurred. Wilting was, overall, intermittent and seemed somewhat dependent on weather.

Video 4-1 -Timelapse video of all phase two treatments, Plot B, phase two

Video 4-2 - Timelapse video of liquid nitrogen treated plants, Plot C, phase two

Plants treated with liquid nitrogen held against their stalk for one minute typically exhibited noticeable wilting (Video 4-2). Burning the stalk with MAPP gas often produced very similar results. Both methods did not seem to affect the plant much on the day of the treatment; instead the plants seemed to slowly die over the course of several days. About five days after the treatment, the plants treated with either liquid nitrogen or MAPP gas would be almost completely limp. If the treatment was not successful, the plant would remain indistinguishable from a non-treated plant. Increasing the time the liquid nitrogen or MAPP gases are applied to the stalk could possibly result in more consistent wilting, but at one minute per plant, the treatment was already impractical for application on a field scale.

4.3.3 Statistical Analysis

4.3.3.1 Introduction

Thorough statistical analyses were conducted starting in phase two to be able to more analytically determine if any treatment was causing more wilting than the control group, and if so, if this was happening robustly. Microsoft Excel and IBM SPSS 24.0 were used to perform all statistical operations. Analyses were conducted separately for each location due to factors such as tobacco variety, soil type, date, weather, and row spacing that contributed to differences between locations.

4.3.3.2 Significance Testing

It was determined graphically that there might be significant effects both from *Treatment* and *Day* factors. A two-way ANOVA can be performed to determine if there is an interaction between two factors. If there is not, each main factor can be analyzed using a one-way ANOVA.

A two-way ANOVA was performed to examine the effects of *Treatments* and *Day* on *Leaf Breaking Angle* at both Plot C and Plot B locations. Residual analysis was performed to test for the assumptions of the two-way ANOVA. Outliers were assessed by inspection of a boxplot; normality was assessed using Shapiro-Wilk's normality test, and homogeneity of variances was assessed by Levene's test. At the Plot C location there were no outliers, residuals were normally distributed ($p > 0.05$), and there was no homogeneity of variances ($p = 0.001$). At the Plot B location there were no outliers, residuals were normally distributed ($p > 0.05$), and there was no homogeneity of variances ($p = 0.023$).

The interaction effect between *Treatment* and *Day* was not statistically significant for either location (Plot C, $F(8, 120) = 0.870$, $p = 0.544$, partial $\eta^2 = 0.0062$; Plot B, $F(8, 120) = 1.205$, $p = 0.303$, partial $\eta^2 = 0.084$). *Day* was found to be significant at Plot C and Plot B with $F(2, 120) = 15.516$, $p = 0.000$, partial $\eta^2 = 0.217$ and $F(2, 120) = 4.217$, $p = 0.017$, partial $\eta^2 = 0.074$, respectively. *Treatment* was found to not be significant at either Plot C or Plot B with $F(4, 120) = 0.941$, $p = 0.443$, partial $\eta^2 = 0.035$ and $F(4, 120) = 0.525$, $p = 0.718$, partial $\eta^2 = 0.020$, respectively. All pairwise comparisons were run where reported 95% confidence intervals and *p*-values are Bonferroni-adjusted. No significant pairwise comparisons were found within *Treatment* ($p > 0.05$) at either location. No significant pairwise comparisons were found within *Day* ($p > 0.05$) at Plot B. At Plot C, several significant pairwise comparisons were found within *Day* ($p < 0.05$).

4.3.4 Discussion

The timing refinements made in the second phase of testing regarding both application and data collection were predicted to aide capturing the wilting effect. The visual observations and photographic evidence showed a marked increase in wilting over the first phase of testing overall. This is thought to mostly be due to the changes in timing from the previous phase, especially data collection timing. With the aid of time-lapse photography, a clear cyclical wilting effect can be seen in most root-pruned plants. It was challenging to record all the data between the allotted time frame (5PM-6PM) even with three people working. For future testing containing larger numbers of subject, data collection would have to be altered to be able to capture this effect in such a small time-frame.

Several things stand out when looking at overall trends for each treatment in both locations (Table 4-2). First, the standard error was very high relative to the difference between the outcomes of each treatment, which indicated that there was a high rate of variability in the outcomes of both the *Leaf Range* and the *Moisture Content* tests. This mirrored visual observations: while it was more common for a treated plant to be seen very wilted as compared with a control group plant, there were many treated plants that showed no wilting at all. High standard error was also seen in control groups, which likely indicated how imprecise these tests can be, especially at detecting small amounts of wilting.

Secondly, in both locations the *Leaf Range* increased, on average, in order from the control group to the most closely root-pruned group. However, there seemed to be no apparent pattern when *Moisture Content* was considered.

Lastly, while treatment 6 (*Liquid Nitrogen test group)* was not done at both locations or with a full set of subjects, it appeared to show the strongest signs of wilting through both tests. Visually, these plants were some of the most wilted.

Table 4-2 - Summary of Descriptive Statistics for Phase Two. 'a' denotes a treatment that was not done in both locations or with an equal number of subjects.

	Plot C		**Plot B**		**Average**	
Treatment	*Leaf Range (degrees)*	*MC (% w.b.)*	*Leaf Range (degrees)*	*MC (% w.b.)*	*Leaf Range (degrees)*	*MC (% w.b.)*
1	90.8 ± 40.9	80.3 ± 4.2	47.1 ± 16.7	87.1 ± 2.6	68.9 ± 38.0	83.7 ± 4.9
2	79.0 ± 30.7	79.8 ± 5.3	47.2 ± 17.2	88.2 ± 2.7	63.1 ± 29.4	84.0 ± 5.9
3	80.0 ± 33.4	80.2 ± 5.4	41.6 ± 13.5	87.6 ± 2.9	60.8 ± 31.8	83.9 ± 5.5
4	75.5 ± 32.1	81.1 ± 5.8	46.6 ± 17.3	87.6 ± 2.9	61.0 ± 29.4	84.3 ± 5.6
5	77.4 ± 34.0	80.8 ± 4.3	44.5 ± 17.9	87.9 ± 2.1	60.9 ± 31.6	84.3 ±4.9
6[a]	-	-	110 ± 80.9	82.2 ± 5.1	110 ± 80.9	82.2 ± 5.1

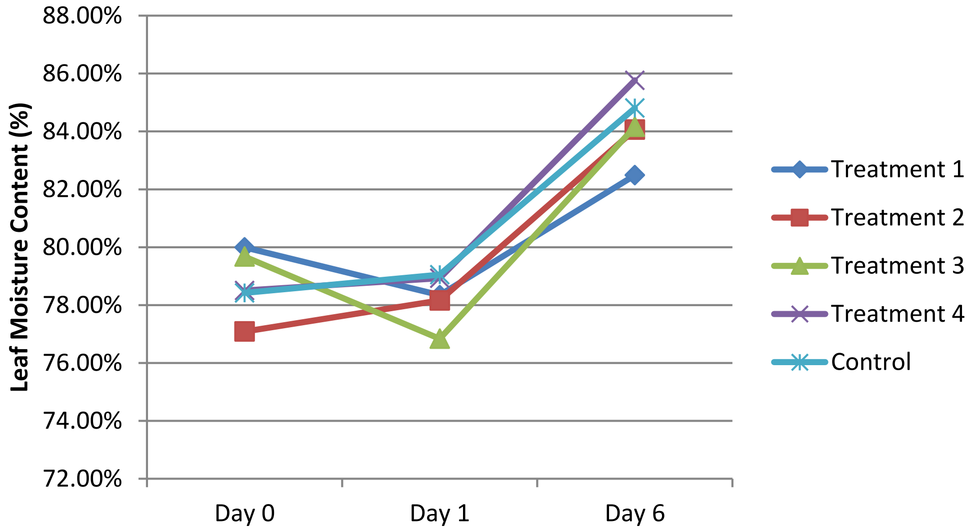


Figure 4-2 - Leaf Moisture Content, Plot C, Phase Two

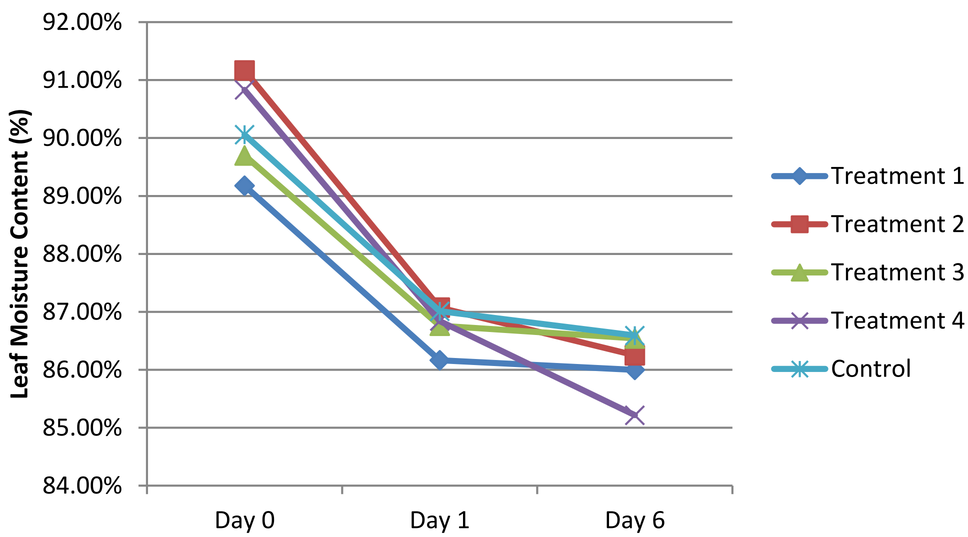


Figure 4-3 - Leaf Moisture Content, Plot B, Phase Two

It was immediately apparent that rather than *Treatment* differences, *Day* had a much more significant impact on *Moisture Content* as an outcome for these treatments on this week (Figure 4-2, Figure 4-3). Looking at the graphs of *Leaf Breaking Range*, the effect of *Day* can still be seen, but it is much less pronounced (Figure 4-4, Figure 4-5).

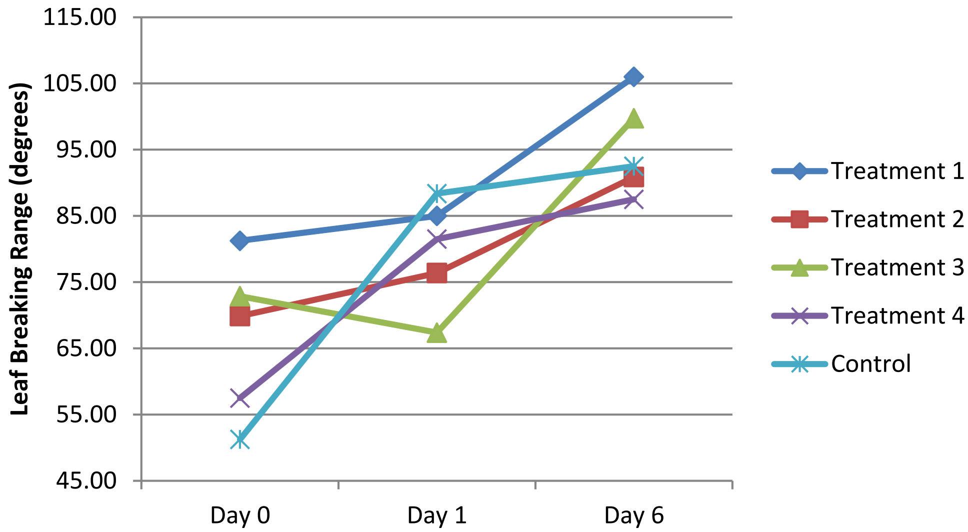


Figure 4-4 - Leaf Breaking Range, Plot C, Phase Two

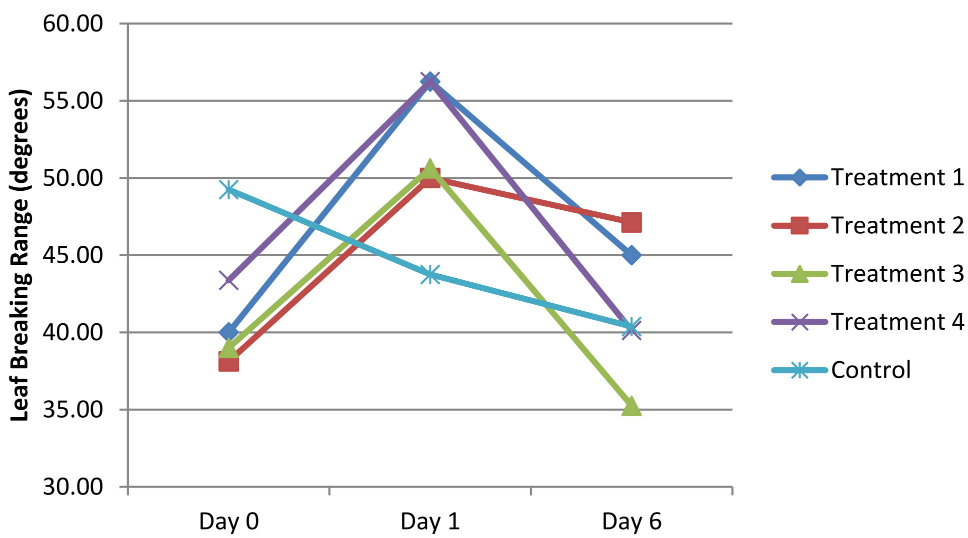


Figure 4-5 - Leaf Breaking Angle, Plot B, Phase Two

The effects between each *Treatment* seem much clearer looking at *Leaf Breaking Range*. Because the *Day* factor still seemed to be a significant influence, its effects were tested for significance in the statistical analysis. It is also noteworthy that the *Control* group did not follow the general pattern of all the other groups at the Plot B location. The *Control* group allowed a comparison of how the tobacco plants were being influenced by external factors like the weather or soil moisture content.

While *Moisture Content* measurements did seem to show something, it needed to be verified that they are a good indicator of plant wilting. *Leaf Breaking Range* did not need to be verified in the same way since reducing leaf damage due to bending and stress was one of the primary desired results of plant wilting. Because of this, a Pearson's correlation test was used to verify that these two continuous variables were indeed related.

Moisture Content vs. *Leaf Breaking Range* was plotted on a scatterplot to determine if there was a linear relationship (Figure 4-6). No linear relationship appeared to exist; *Moisture Content* did not noticeably change with *Leaf Breaking Range*. No outliers were detected at this stage. There was no significant correlation between *Moisture Content* and *Leaf Breaking Angle* as assessed by the Pearson's correlation test ($p > 0.05$). Because no correlation existed, further analysis would only concentrate on *Leaf Breaking Angle.*

Comparing the marginal means of the treatments to each other graphically, it's easy to see in Plots B and C that the *Days* had more influence on the *Leaf Breaking Range* than the treatment type. Results were conflicting overall. For example, Day 0 seemed to show positive results at Plot C, but negative results at Plot B. The estimated marginal means grouped by *Treatments* are shown in Figure 4-7, Figure 4-9, Figure 4-10 and Figure 4-10.

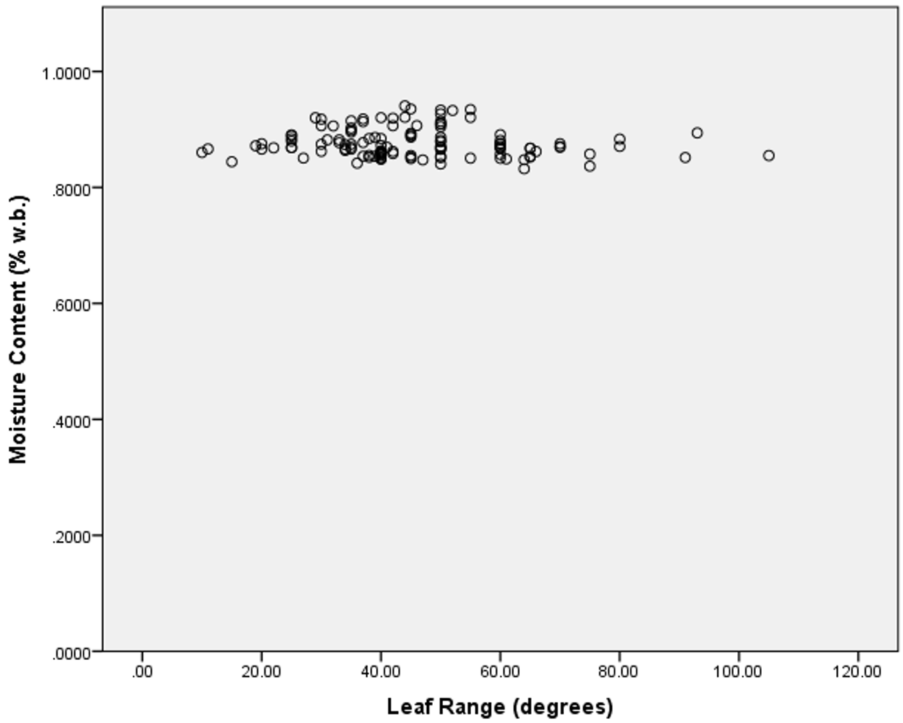


Figure 4-6 Scatter-plot of Leaf Range (degrees) compared against Moisture Content (%, wet basis)

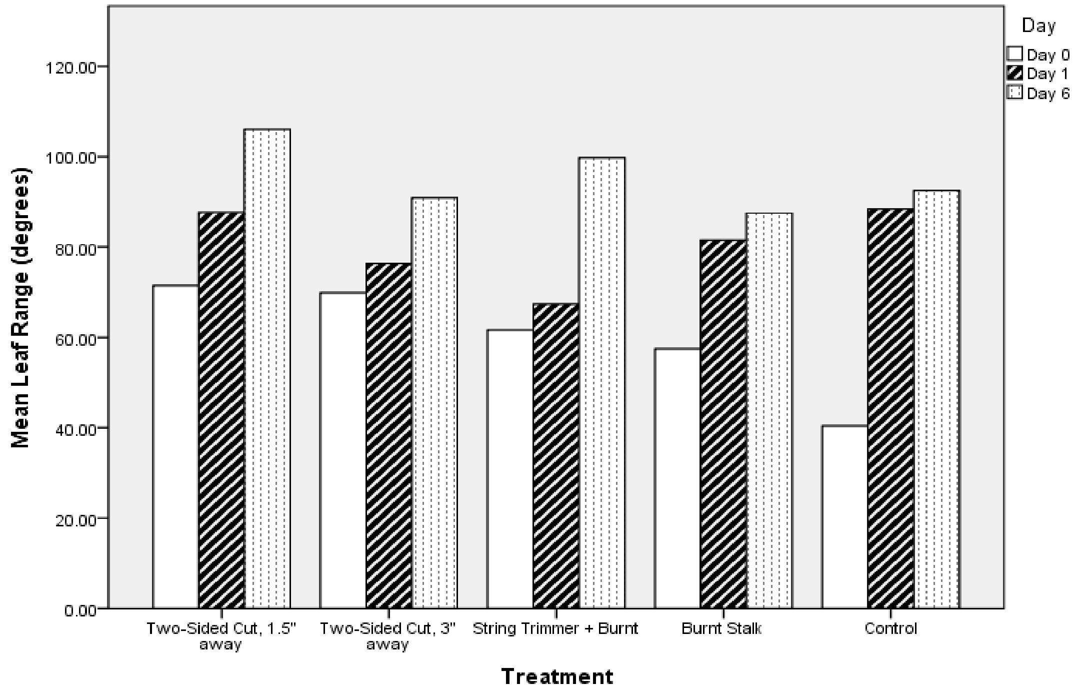


Figure 4-7 - Mean Leaf Range, Plot C, Phase Two

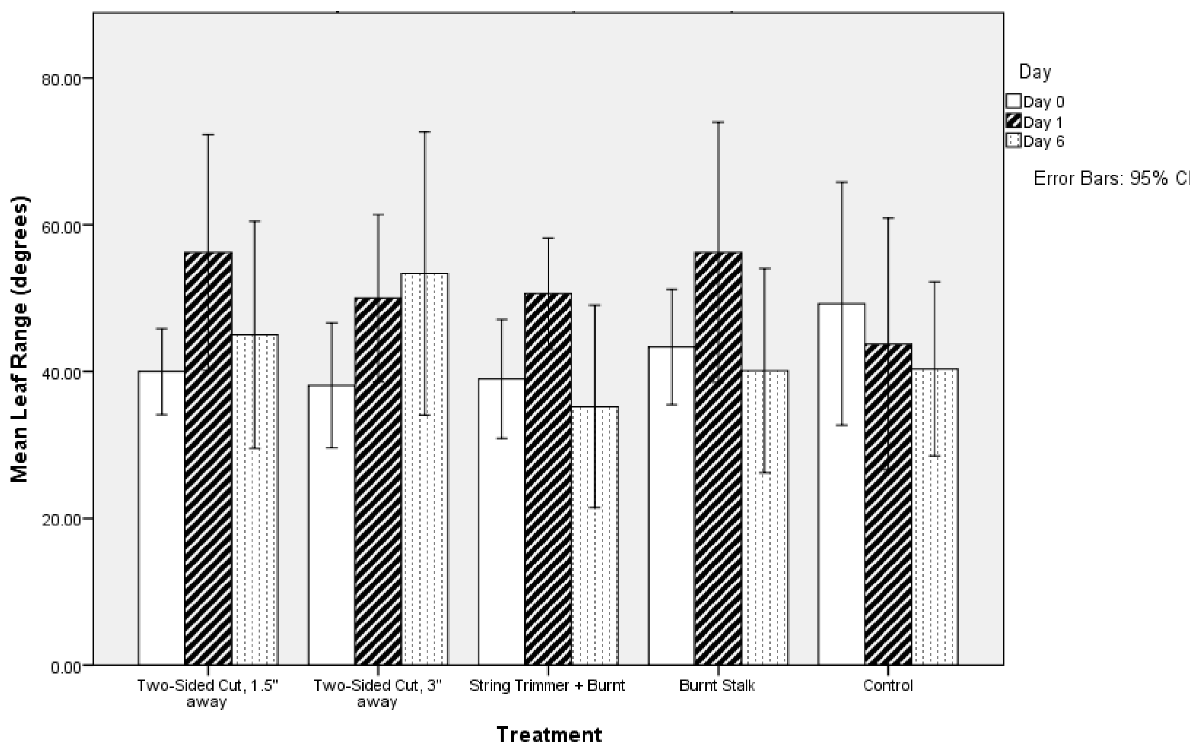


Figure 4-8 - Mean Leaf Range, Plot B, Phase Two

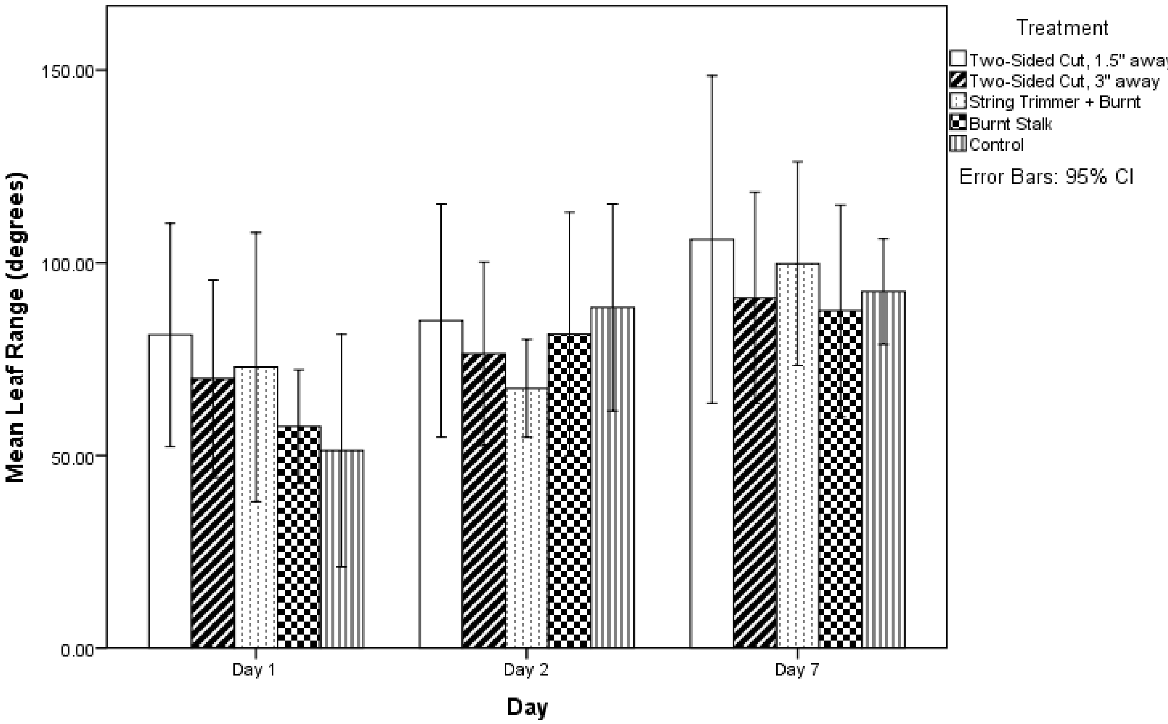


Figure 4-9 - Mean Leaf Range grouped by Days, Plot C, Phase Two

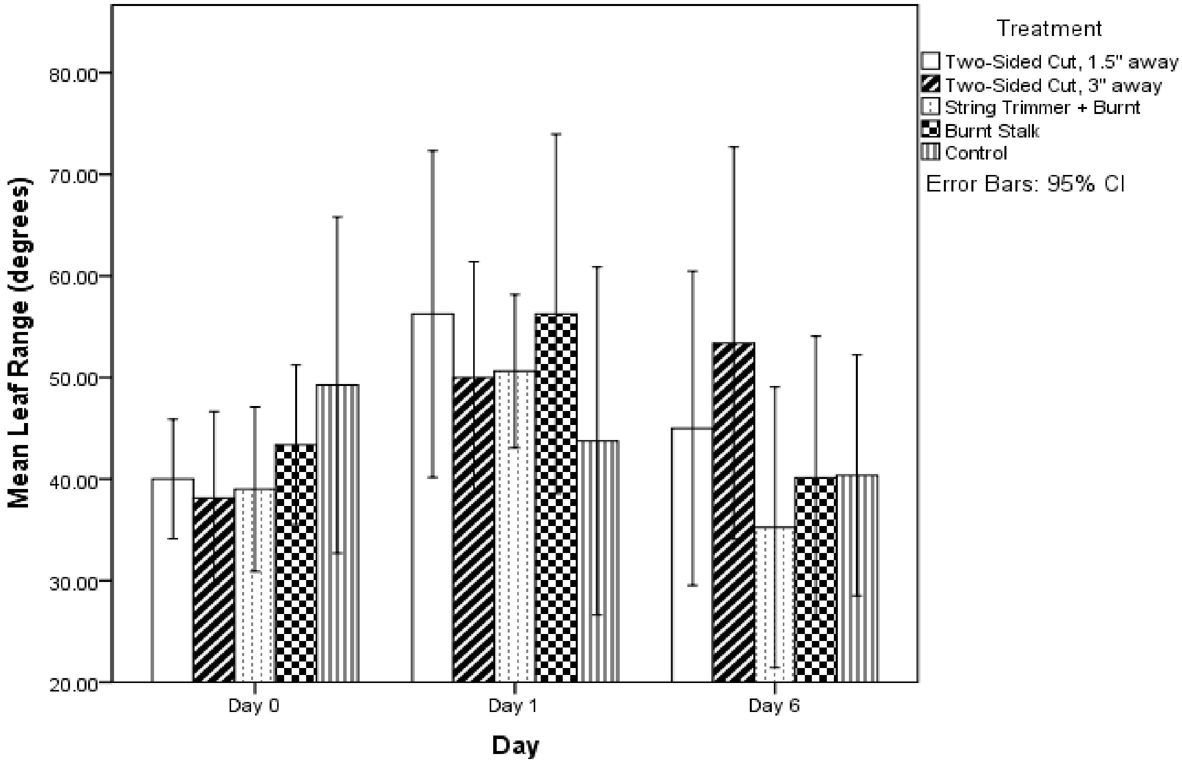


Figure 4-10 - Mean Leaf Range grouped by Days, Plot B, Phase Two

Treatments that destroyed the pith (liquid nitrogen, burning, and string trimming + burning) never had an immediate wilting effect. They did not start to show serious signs of wilting until approximately the fifth day after treatment. If wilting began, it usually resulted in the complete death of the plant. This led to some of the best *Leaf Breaking Range* results; however, results were inconsistent. Results may improve if the data collection window is moved farther out, starting at the 7th day after treatment.

The burning and freezing methods were less dependent on certain ambient conditions, such as soil moisture. It is worth pointing out that it was never the case that a burnt or frozen treated plant would partially wilt, rather they either fully wilted and never recovered or they were unaffected. Because it was hard to consistently get these plants to fully wilt out of the whole population, the results were deemed inconclusive. It seemed that the key to getting tobacco to wilt and stay wilted is for the pith to die and not regenerate. This was the basis for the 'pith boring' treatment; however, upon further investigation, the pith had regenerated. It was likely the tobacco plant could still get enough water via the xylem (the woody layer outside of the pith) that it was able to survive. The pith can be seen in a cross section view in Figure 4-11 after a liquid nitrogen treatment. Freezing plant cells can injure them enough to cause immediate and permanent death (Hincha et al., 1987).

Figure 4-11 - Cross section of tobacco stalk after 7 days since being treated with liquid nitrogen

4.4 Conclusion

4.4.1 Overview

The results from phase two are similar to those from phase one. In both cases, clearly distinguishable signs of wilting were apparent in some plants at varying times. Despite these wilted plants, other plants with the same treatments applied would not achieve near the same results. For example at Plot A during early testing, four out of four plants wilted completely and very evenly by burning the stalks, which can be seen in Figure 4-12. The same test was used at the phase two Plot C full-scale test and the Plot B full-scale test and the results were not nearly as robust. In fact, most of these plants did not wilt. This could be due to ineffectiveness in the treatment itself, or more likely, a difference in plant physiology from location to location or from variety to variety. A slight change in conditions in one location, such as less rainfall, can dramatically change the physiology of the stalk and the root system. It was noted that the woody layer of the stalks in Plot A looked fairly average; Plot C had tobacco stalks with an exceptionally thin woody layer.

Figure 4-12 - Four plants treated with MAPP gas remain wilted in wet soil, Plot C, phase two

Visual results from some of the tests showed very evident wilting in the time lapse videos and in the measured data. Even though wilting was achieved on some plants, if the entire data set is considered, the results are not significant at a 5% confidence level for any of the treatments. There was significance between *Day* groups, but since the control group was also affected this is very likely due to environmental factors. It should be noted that in the phase one testing, data were only collected on the 7^{th} day after the application of the treatments. Data collection started at approximately 10:00AM and lasted several hours. For these reasons, it is hard to compare the phase one data to the phase two data.

4.4.2 Decision Matrix

A decision matrix (Table 4-3) was utilized to determine which treatment would be the best to mechanize. The categories considered were cost, labor requirements, how easy it would be to mechanize, its ability to pre-wilt tobacco, and its potential speed. Scores range from 0 (negative impact) to 100 (positive impact).

Table 4-3 - Decision Matrix, comparing possible treatments to mechanize

Option	*Cost*	*Labor Requirements*	*Ability to Mechanize*	*Ability to Wilt Tobacco*	*Speed*	*Score*
Cut 2-Sides at 1.5 in.	100	95	100	80	90	90
Cut 2-Sides at 3 in.	100	100	100	50	100	78
String trim and burn stalk	40	80	70	45	40	52
Burn stalk	60	85	85	35	55	56
Liquid Nitrogen	1	20	5	90	1	43
	15%	5%	25%	45%	10%	100%

Two-sided cut at close distance was the best option based on results from the second phase of testing. It was fast to apply, simple, and resulted in the greatest mean leaf breaking range (aside from the liquid nitrogen treated plants). The ability to wilt plants was among the best, but still inconsistent. The speed was fast; however, it was recognized that difficulty in controlling the margin of application error may limit the speed of application.

Two-sided cut at the further distance was similar to that of the former treatment in many ways. However, it's mean leaf breaking range was closer to that of the control group than it was of the close distance two-sided cut, so a large portion of points were deducted.

Both the 'string trim and burn stalk' as well as the 'burn stalk' treatments had many shortcomings. They often produced some of the most wilted plants, but also some of the most inconsistent results of this entire project, which is the reason for their low wilting score. They also incurred high application times and labor requirements due to the multiple passes that need to be made to complete a treatment.

Liquid nitrogen was a very strong performer in its ability to pre-wilt tobacco. Of the plants treated during this project, liquid nitrogen produced some of the most wilted plants. Not only that, but many never regained turgor, and their root support system remained intact. This treatment scored extremely low on most other metrics though. The cost was very prohibitive, even for a small number of plants. Great time and labor had to be put into treating each plant, and there was no clear path to mechanizing the process.

CHAPTER 5 – Design and Testing of Root Pruning Implement

5.1 Introduction

5.1.1 Purpose

The primary goal of phase three of the project was to use the results from the first two phases to design a mechanized root-pruning implement (project Objective 2). For mechanized pre-wilting tobacco to become a viable option for farmers, the implement needs to be able to achieve a high rate of application and be deployed in a low-cost manner. Logically, this means a mechanized implement that can be adapted to work with equipment that is already used with these crops. From the results of the first two phases of this project, mechanically pruning the roots via a tow-behind implement was chosen to be the best solution because of its potential to cause wilting as well as the possibility of practical field application. It was noted that field considerations are very important because the weather and field conditions directly determine how fast the plants wilt and how fast they are able to re-grow their roots.

5.1.2 Sub-Objectives

5.1.2.1 Sub-Objective 1: Design a simple low cost implement to root prune tobacco plants.

Because pre-wilting is being done in an effort to reduce costs and further mechanize the harvesting of tobacco crops, this must be a low cost and easily adaptable design. Keeping complexity to a minimum as well as keeping both capital and operating costs low is essential to allowing farmers to adopt this method as widely as possible. Ideally, a consumer could purchase or manufacture this device, integrating it with a wide range of currently capable farm equipment.

5.1.2.2 Sub-Objective 2: Build a model for predicting tobacco wilting based on environmental and treatment data.

It has been well established that plant wilting depends on a plethora of factors and is hard to predict. Building statistical models to predict which plants will wilt, and to what extent, would allow for the ability to judge whether a certain mechanical design is feasible within given parameters.

5.2 Implement Design & Fabrication

5.2.1 Introduction

In phase three a special-purpose research tobacco crop spraying machine was modified by adding a hydraulically-actuated 28” diameter straight coulter disc that is forced into the ground to cut the soil at an angle near the base of the stalk. The coulter was mounted on a three-wheeled high-clearance tobacco sprayer (Figure 5-1) that had previously been custom designed and built by the Biosystems and Agricultural Engineering Department at the University of Kentucky in 1991 by Duncan, et al. It was designed to be a small, low-cost, specialty crop focused alternative to contemporarily available retail sprayers. It was chosen due to its availability as well as its mounting options.

Figure 5-1 - Tri-wheeled tobacco sprayer used in Phase 3

5.2.2 Sprayer Modifications

Several modifications were necessary to allow the tri-wheeled tobacco sprayer to be used to tow a hydraulically operated implement. Two new 3500PSI relief valves were installed in series between the wheel motors and pump unit to deliver more tractive power (Figure 5-2). This modification was feasible due to an engine upgrade in previous years.

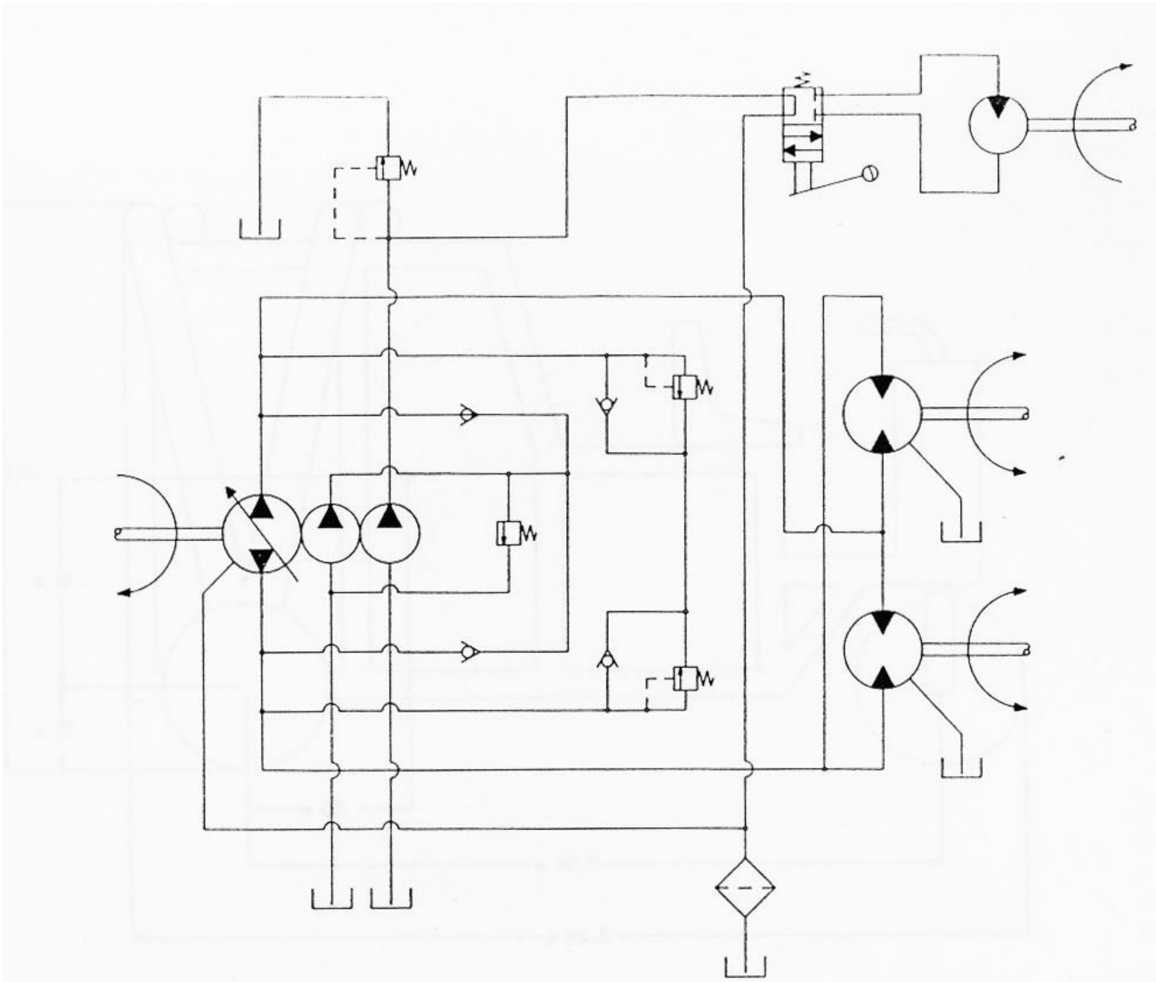


Figure 5-2 - Hydraulic circuit schematic for hydrostatic drive and auxiliary open center circuit (Duncan et al. 1991)

Because the angle of the coulter disc implement is not orthogonal to the ground, there will be a diagonal reactionary force pushing the rear end of the sprayer upwards and towards the right, which will decrease available traction to the left drive wheel. To remedy this, five tractor ballasts of 25kg each were mounted to the left side of the sprayer to increase down force on the drive wheel (Figure 5-3).

Figure 5-3 - Ballasts added to one side of the sprayer to combat uneven loading

5.2.3 Design

All design work was done using Solidworks. The frame of the sprayer and all critical dimensions were first measured and entered as CAD models. Using this information along with the previously determined cut angle and cut depth, exact length and type of steel channel were determined. The sprayer chassis, coulter disc, steel channels, and hydraulics were all then added to a single assembly where cut angles, extension range, and clearances could be determined and measured.

The hydraulically-actuated coulter disc required a chassis that had several mounting options and was appropriately low to the ground. This machine framework included horizontal and vertical 4-inch square steel bars sufficient for mounting in a variety of configurations if needed. The machine also had a large enough engine to be able to overcome the draft forces produced by the coulter disc. Universal brackets were made that allowed the angle and height of the counter disc attachment to be changed in the field if necessary. The final angle used was approximately 25 degrees from vertical.

Several coulter discs were considered with various mounting options. The Bigham Brothers, Inc. (Lubbock, TX) 28-inch stabilizing coulter disc (P/N 808-987) was chosen due to disc size and arm geometry. It was estimated from analyzing the geometry of the coulter arm as it would be used that it would be able to penetrate the ground to nearly 12.5 inches. The critical dimension in this case was the distance between the coulter disc arm and the soil (Figure 5-4). The arm would not be positioned in the opposite manner due to possible damage to the tobacco plant.

Figure 5-4 - CAD model of the possible interaction between the extended coulter arm and the soil

To size the hydraulics, the equation for force that a cylinder could generate was used (Equation 5-1).

$$F = \frac{\pi d^2}{4} P$$

Equation 5-1 - Equation for the force generated by a hydraulic cylinder, pushing away from the rod

In that equation, *P* is the pressure in the cylinder, *d* is the bore diameter, and *F* is the resulting force. With a maximum system pressure of 2250PSI and a cylinder diameter of 2 inches, the resulting maximum cylinder force was 7100 lbs. According to Pitla et al., 2009, the average vertical force of a coulter disc at similar soil depth is 2700 lbs., with a maximum encountered vertical force of 3400 lbs. The same study reported that such a coulter disc would also encounter a horizontal force of 1900 lbs. on average, with a maximum of 2000lbs. Given that the maximum angle from vertical the cylinder could be positioned was 31°, the maximum force on the cylinder was 4800 lbs. If extremely hard soil is encountered or max depth is hit, the machine will be lifted into the air before any mechanical components would fail. A tie-rod cylinder from Maxim (P/N: 219-311) was selected with a retracted length of 30 ¼ inches, an extended length of 50 ¼ inches, a 2 inch bore, and a 6,950 lbs. max column load. The cylinder was placed into CAD to verify that the extended and retracted lengths would be sufficient (Figure 5-5).

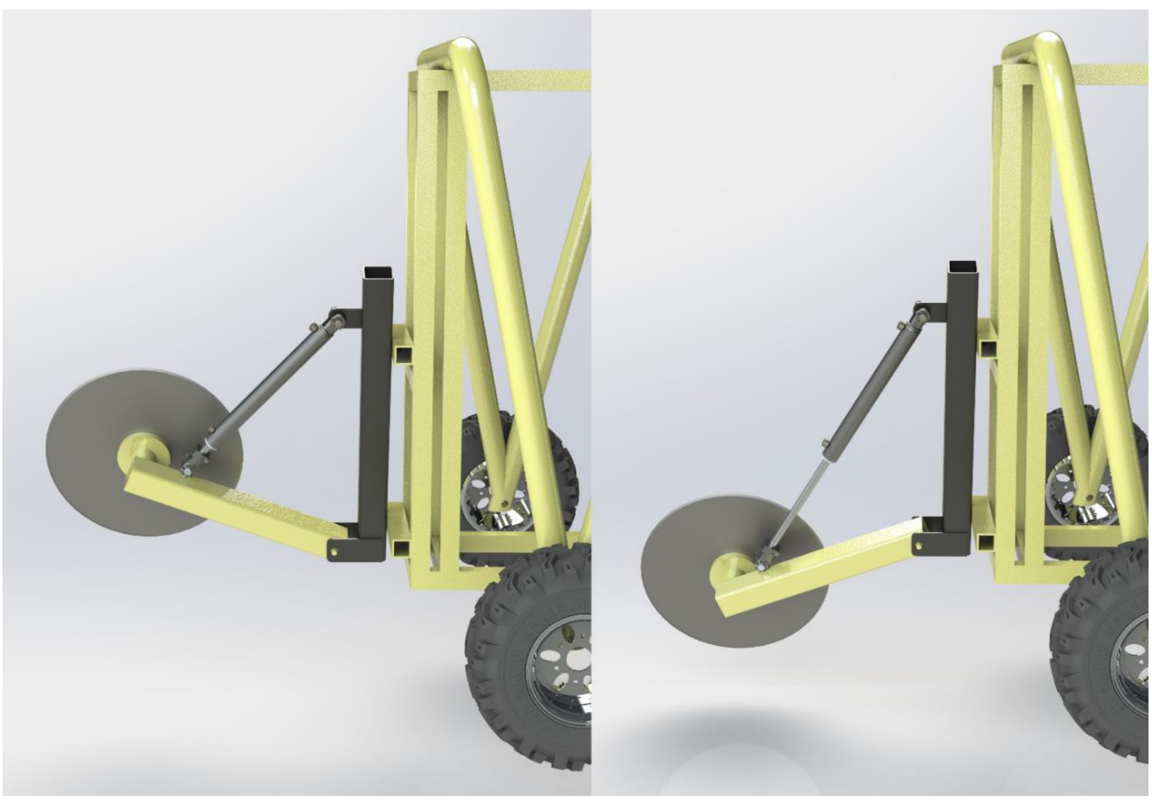


Figure 5-5 - CAD comparison of fully retracted and fully extended coulter implement

Additional modifications were made to add a four-way, three-position, pressure release detent hydraulic valve (Prince Hydraulics, LS3000 Series) to allow the driver to control the height of the coulter. The four-way valve had a built-in pressure relief of 2250 PSI, which would prevent the cylinder from exceeding its maximum column load.

Structural tubing dimensions were obtained solely through use of CAD. The main member that the coulter arm would be mounted to was determined to be 52.5 inches long using 4 x 4 x ¼ inch steel square tubing. This matched the type of tubing used in the preassembled 42-inch coulter arm. Two new 2.5 x 2.5 x ¼ inch square steel tubing members were welded horizontally across the rear of the sprayer as additional mounting points. Several custom mounting plates using 3/8-inch steel would also be made to affix the coulter implement to the sprayer body. These would pair with ¾ by 8 inch bolts. The unit was not welded in case small adjustments in height or angle needed to be made in the field. Dimensions and positions of these components are shown in Figure 5-6.

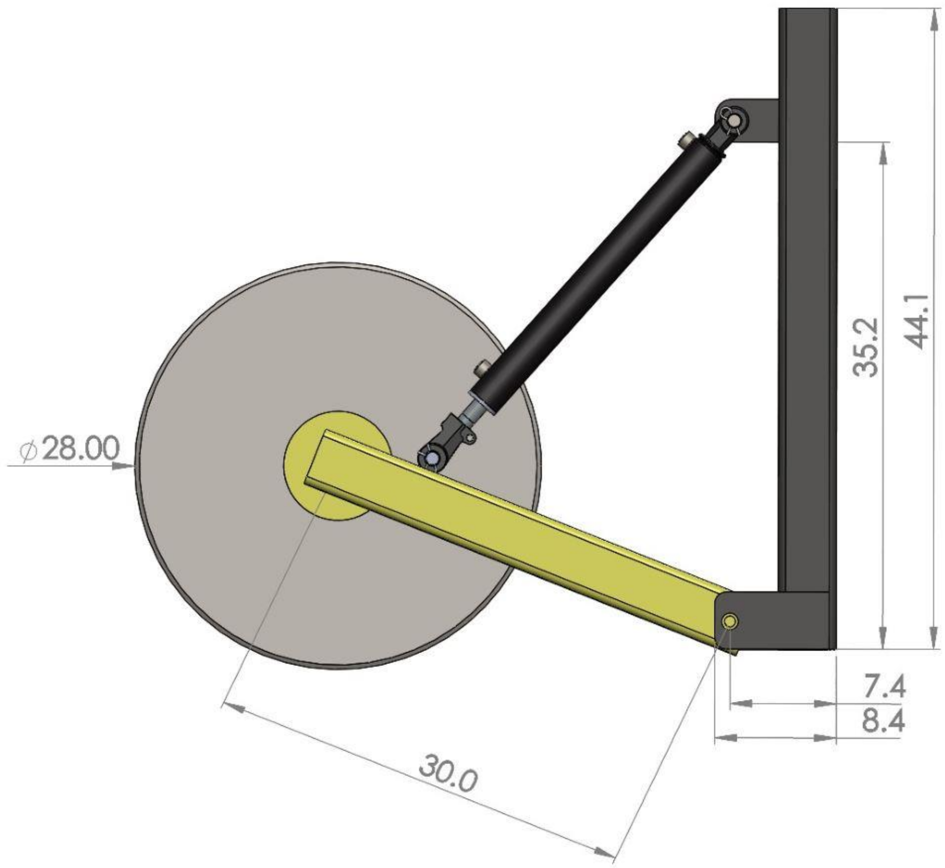


Figure 5-6 - Assembly side view of root-pruning implement with dimensions called out in inches.

5.2.4 Fabrication

All fabrication was performed at the Biosystems and Agricultural Engineering Machine Shop. Sections of 4-inch square tubing and 2.5-inch square tubing were cut down and sanded, per the design. Mounting plates were also cut down and sanded, and bolt holes drilled. Tabs to mount the hydraulics were then welded onto the upright section of square tubing, and the 2.5-inch square structural members were welded onto the body of the sprayer.

The upright section was first clamped onto the chassis and tightened down (Figure 5-7). Next, the lower section of the implement was attached to the upper section via a 1-inch clevis pin. Finally, the hydraulic cylinder was connected between the two sections of the implement to complete installation. The hydraulic lines were then connected.

Figure 5-7 - Coulter disc implement affixed to the sprayer via mounting plates

The machine was then turned on and the angle of the implement and the range were measured. This was compared with design requirements, and all specifications were verified. Because this implement was able to be affixed purely with clamps, slight changes were simple to make. The clamps were loosened slightly, the implement was moved into proper position, and the clamps were again tightened.

5.3 Methods

5.3.1 Introduction

The preliminary work done in phases one and two pointed out several important things about how and when to measure the wilting effect. By using time-lapse photography, it was discovered that not only was wilting usually most apparent on the day of application (day 0), but that the time of day of the maximum amount of wilting was between 5PM and 6PM. Maximum wilting occurred at this time because it marks the end of the brightest and hottest part of the day when evapotranspiration rates are at a maximum; at this time of day the water uptake will be the most limiting factor, depending on how much of the root ball is severed. Considering this, all treatments were applied as quickly as possible at approximately 10AM and all data were recorded as quickly as possible at approximately 5PM.

While in previous phases several treatments were used to determine which treatment was the most viable to wilt plants, in phase three the root-pruning machine was the only treatment applied. The goal was to test if the machine could be used to cause significant wilting by root pruning, and also to see if there was correlation between wilting and various factors related to soil moisture uptake.

5.3.1.1 Locations and Subjects

Two locations were again used in the final phase of testing: UK Spindletop Research Farm (38° 6'32.20"N 84°29'50.91"W), referred to as Plot A, and UK Woodford County Research Farm (38° 5'22.11"N 84°43'54.42"W), referred to as Plot C.

KT206 burley tobacco variety was used in both Plot A and Plot C locations with standard row spacing. All subjects were topped according to standard practices. Plant size was heterogeneous, and there were minimal ground suckers at both locations. Occasional subjects in this phase were prone to crooked lower stalks.

5.3.2 Procedure

The first trial run at Plot C was done on 40 plants to verify that everything was working as expected. Three 100-120 plant replications were then pruned, one at Plot C on September 28th and two at Plot A on October 5th and 7th. An additional late test was done at Plot A on October 10th because of the availability of additional tobacco plants. Tests later in the year are likely to show less wilting due to plant maturity and the lower average temperatures.

Video 5-1 - Demonstration of normal operation, root-pruning one side of a row

The machine was driven all the way down a row of the test tobacco with the coulter disc fully inserted into the ground, first going in one direction to prune the roots on one side of the plants, and then going back in the other direction to prune the roots on the other side of the plants (Video 5-1). Driving this machine proved be to very challenging in terms of making the coulter disc fall at an exact distance away from each stalk. The aim was to have the disc enter the ground approximately 1.5 inches away from the stalk on either side of the plant. This rarely happened exactly as had been planned, but exact cut distance was recorded for each section of ten (10) treated plants. It was a common occurrence to accidentally cut the stalks of standing plants, usually at the starting of a row. These plants were not counted in the overall number of treated plants. Stretches of plants that were still standing and had been root pruned within acceptable boundaries were flagged. All of the plants that were treated correctly were flagged; over 100 plants had data collected on them in each of three replicated tests. Control plants, the plants that were not treated in any way, were also flagged at this time.

5.3.3 Data Collection

Further changes to data collection needed to be made from phases one and two. The third measurement was moved up to day 4 to better capture wilting. The leaf breaking angle test was changed to utilize a fulcrum distance of 3.9 inches instead of 2.9 inches to better capture changes in wilting. A subjective rating method was also added to be able to rate a very large amount of subjects very quickly. Fewer leaf breaking range and leaf moisture content measurements were taken, primarily due to time restrictions. The data collected included:

1. Subjective rating
2. Leaf breaking angle
3. Leaf moisture content
4. Soil moisture content
5. Width of cut
6. Root-ball weight
7. Pictures & time-lapse photography

Immediately after the treatment the width of cut (the distance between cuts made by the coulter on either side of the plant row) was measured at 10 plant intervals along the length of the treated row. Measuring the width of cut at every individual plant was not necessary since the disc could only move in a fairly straight line. Starting at 5PM every plant that was flagged was rated for wilting on a scale of 1 to 3 on days 0, 1, and 4 after the treatment was applied. The criteria for the ratings were as follows:

(1) – Very small amount or no wilting when compared to control

(2) – Wilting apparent, but tip of leaf- had not yet passed below the base of the leaf

(3) – Heavy signs of wilting; tip of leaf-dropped below the base of the leaf

Next, five plants from each category (1, 2, 3, and control) were chosen at random to be sampled for leaf breaking angle and leaf moisture content testing, which was done on days 0 and 1 (for a total of ten subjects).

Using the fifth leaf down from the top of each plant a modified protractor was used to measure the range a leaf could be bent downward before breaking. A force was applied 3.9 inches from the base of the leaf slowly and both initial and final (breaking) angles of the leaf were recorded. The same leaf was then immediately placed into a Ziplock bag and labeled for measuring moisture content. This process was repeated on day 1 as well. Different plants than those tested the first day were used if possible; if there were not enough other plants in that particular category, the same plant was used again for the tests. It was not known for certain if removing one leaf from a plant would affect its wilting either positively or negatively, but no effect either way had been noted in previous phases of testing. For example, if there were only five plants in the (3) category, to have two days of data and consistent number of samples, the same plants would have to be used both days. However, if there were 10 plants to choose from, an untouched plant could be used on both days.

For the second and third replications of the tests, soil moisture content was measured using a Field Scout TDR 300, which reports volumetric moisture content at a depth of 3 inches below the soil surface. These measurements were taken immediately after the leaf samples were bagged. Soil moisture content measurements were taken by sampling the soil moisture content 3 inches deep once every ten (10) plants in a row, in-line halfway between two plants – 10 inches from the stalk. Soil moisture content measurements were also taken in the same location 10 inches from the stalk in the direction perpendicular from the row, so that the measurement was outside of the cut made by the coulter. An example can be seen in Figure

5-8; the area inside the root pruned area is denoted by “1”, and the area outside the row is denoted by “2”. These measurements were for investigating the difference in the moisture content between what the root-pruned plant roots were experiencing and the rest of the field.

Figure 5-8 - Approximate locations of soil moisture content readings. One reading taken inside the cut span, the other taken outside the cut area.

A late test different from the three replicated tests was done to specifically investigate relationships between cut distance, soil moisture content, and the wilted state (as indicated by subjective rating) of the plants. Fewer than 100 plants were used for this test. No leaf samples were collected. Subjective ratings were done in the same fashion as in the replicated tests. The cut width measurement and soil moisture content were taken every seven plants only on day four at 5PM. Instead of taking field averages of soil moisture content, soil moisture content could be related to a numbered plant position in the row. This allowed for correlation between soil moisture content and wilting of plants tied to a specific location. The same types of soil moisture content measurements were taken as before: 3-inch depth; one sample between two plants, the other sample taken outside of the row. To minimize error at each location, four samples were recorded and averaged at every location where soil moisture content was measured.

Root-ball collection was done after all other data collection had been completed from the same set of plants that were used to investigate soil moisture content. Both treated and control subjects were chosen at random. All samples were carefully pried up in half-sphere shape using a shovel (Figure 5-6). An 18-inch circular cutout was made from ¾ inch plywood to aide in keeping excavation volume similar.

Figure 5-9 - Root pruned root-ball being dug up. Samples usually came up in a small 'wedge' shape

The tobacco plant was first cut down at the base and discarded. The wood cutout was then placed over the plant stump, centered on the stalk. Two workers then carefully cut the soil with the shovels and slowly pried the sample up. The root ball was then placed in a black plastic bag and tagged with its ID number and date.

At the laboratory, all samples were gently sprayed with a hose to wash off all soil, leaving just the root system. Root-balls were then placed in paper bags and dried in the same manner as the leaves had been dried previously in the study. Once dry, each sample was weighed.

5.4 Results & Discussion

5.4.1 Treatment Application and Data Collection

Application of the treatments with the machine was very difficult because the steering pivot point of the vehicle was near the rear wheels of the machine and the coulter extended 51-inches behind the rear wheels. When the driver adjusted the lateral position of the machine closer to the stalk of the plant by turning the steering wheel toward it, the coulter disc, which was about fifteen plants behind the front steered wheel, temporarily moved farther from the row. After some distance when the machine was moving straight again, the coulter disc would eventually reach the desired position. Even with equipment such as GPS auto-steer, the margin of error with this machine configuration will likely remain too high to get consistent results without a high loss of tobacco; the range of cut required to likely induce wilting is usually the same as the range of error where plants are actually planted compared to where the row was planned to be. Different mounting techniques or steering configurations would have to be explored before a viable solution could be produced, but enough data were collected to evaluate the technique in this study.

The subjective analysis test proved to be a good way to judge wilting because a large number of plants could be rated in a small time frame easily. Other tests required over an hour to complete for a smaller set of subjects. Visual analysis and time lapse photography proved to mirror the results of previous years. It was rare to see extreme wilting on consecutive plants in a row. Figure 5-10 shows a small section that was treated with the coulter being inserted in the ground between 1 and 1.5 inches from the stalk very evenly on both sides. The plants in this section were planted in a very straight row. The wilting effect due to root pruning was very apparent on all subjects in this run. This was on one of the hottest days, which would have amplified any wilting caused by root pruning.

Figure 5-10 - Some of the most wilted plants recorded (all rated 3) during phase three, Plot A, one day after treatment

5.4.2 Statistical Analysis

5.4.2.1 Introduction

Statistical analyses in phase three of this project were handled in a completely different way than they had been in the former two phases. Prior to this phase, statistics were mostly used to compare treatments to one another to determine the most robust method of pre-wilting. In this phase there was only one treatment, and it needed to be analyzed for its efficiency and magnitude of effect.

Using the collected data, a model was created to predict to the probability of pre-wilting a tobacco plant given certain input parameters. This can be useful to forecast the best time to apply the root pruning treatment or the best time to harvest after the application of the treatment.

Due to the nature of testing, strict time restraints, and variety of data collected, there were varying numbers of subjects included in each trial. Because of the complexity involved in analyzing each subgroup, most statistics done in the subsequent sections will be looking at the pooled results of all subjects tested. The subjects included in this phase are listed in Table 5-1.

Table 5-1 - Date, location, and number of subjects treated, Phase Three

Date	*Location*	*Type*	*Subjects*
9/25/2011	Plot C	Treated	40
9/28/2011	Plot C	Treated	113
9/28/2011	Plot C	Control	10
10/5/2011	Plot A	Treated	120
10/5/2011	Plot A	Control	10
10/7/2011	Plot A	Treated	100
10/7/2011	Plot A	Control	10
10/10/2011	Plot A	Treated	92
10/10/2011	Plot A	Control	33

There were *153* treated plants and *10* control plants at Plot C. At Plot A there were to *312* treated plants and *53* control plants. All subjects had *Subjective Ratings* measured daily, but only a subset of these had their *Leaf Breaking Range*, *Moisture Content, Soil Moisture Content,* and *Root Ball Weight* recorded due to time constraints.

Because of this wide variance, the data were binned by d*istance between cuts* in order to be able to more precisely see when and if the treatment had any effect. The groups that were chosen are as follows: 4 to 6 inches, 6 to 8 inches, 8 to 10 inches, and more than 10 inches. This facilitated all treated plants to be included in the data collection, but kept abnormal treatments from skewing the results.

5.4.2.2 Significance Testing

A two-way ANOVA was utilized to test the significance of the treated group compared to the control group in a manner similar to the technique used in phase two. Unlike phase two, however, all subjects that were treated were compared in one group against the control group. Forty treated subjects were selected at random from the Plot A test dates that were paired with forty randomly selected control plants from the same location.

The two-way ANOVA was performed to examine the effects of *Treatment* and *Day* on *Leaf Breaking Angle* at the Plot A Location. Residual analysis was performed to test for the assumptions of the two-way ANOVA. Outliers were assessed by inspection of a boxplot, normality was assessed using Shapiro-Wilk's normality test, and homogeneity of variances was assessed by Levene's test. Extreme outliers were removed before testing, residuals were normally distributed ($p > 0.05$), and there was no homogeneity of variances ($p = 0.012$).

The interaction effect between *Treatment* and *Day* was statistically significant: $F(1, 80) = 4.804$, $p = 0.031$, partial $\eta^2 = 0.059$. *Day* was found to be not significant with $F(1, 80) = 0.041$, $p = 0.841$, partial $\eta^2 = 0.001$. *Treatment* was found to be significant with $F(1, 80) = 16.610$, $p = 0.000$, partial $\eta^2 = 0.179$. All pairwise comparisons were run where reported 95% confidence intervals and *p*-values are Bonferroni-adjusted. Significant pairwise comparisons were found within *Treatment* ($p > 0.05$). No significant pairwise comparisons were found within *Day* ($p > 0.05$).

5.4.2.3 Model Creation and Validation

A predictive model was created with cut width as an input and a binary assessment of wilting as the output. This is a similar analytical process to survival analysis, except instead of 'time to death', the mechanism will be 'cut width to wilting'. Binomial logistic regression is the most appropriate form of analysis in this situation. It can be described through Equation 5-2:

$$\text{logit}(Y) = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_4 + \varepsilon$$

Equation 5-2 - General equation for logistic binomial regression

where β_0 is the intercept term, β_1 is the slope for X_1, and ε is the error.

A binomial logistic regression was performed to determine the effects of cut width on the binary results of wilting. *Subjective Rating* was averaged over the data collection period for each subject. Subject averages with a rating of >2.0 were considered to be wilted while subjects with a rating of <=2 were considered to be not wilted. The logistic regression model was statistically significant, $\chi^2(1) = 184.762$, $p < 0.0005$. The model explained 47.8% (Nagelkerke R^2) of the variance in wilting and correctly classified 83.3% of cases. Sensitivity was 92.8%, specificity was 68.1%, positive predictive value was 82.43% and negative predictive value was 85.38%.

Model validation was performed to verify the reliability. Exactly 10% of the subjects were chosen using a random number generator and removed from the main cohort and moved to a new dataset. The main cohort was then used to generate a new model, repeating all steps of the binomial logistic regression. Each piece of data in the verification cohort was then plugged into the new model and the output recorded. For the verification cohort, the model predicted the correct result for 84.1% of the cases; the model accurately predicted 83.2% of cases in the main cohort. The original binomial logistic regression had a predictive capability of 83.3%; therefore, the model was deemed valid.

5.4.2.4 Soil Moisture Content Analysis

A one-way ANOVA was conducted to determine if the soil moisture content was different inside the cut area compared with outside the cut area. Subjects were classified into two groups: inside of cut (n = 93), and outside of cut (n = 93). There were no extreme outliers, as assessed by boxplot; data were normally distributed for the 'outside of cut' group, as assessed by Shapiro-Wilk test ($p > 0.05$); data were not normally distributed for the 'inside of cut' group, as assessed by Shapiro-Wilk test ($p < 0.05$). Soil moisture content differences were statistically significantly between the two groups, $F(1, 185) = 200.753$, $p < 0.0005$.

5.4.2.5 Root-ball Analysis

A Pearson's correlation test was used to assess the relationship between root-ball weight and cut width. The data were normally distributed, as assessed by Shapiro-Wilk's test ($p > 0.05$). There was a moderate, positive correlation between root-ball weight and cut width, $r^2 = 0.389$, $p < 0.05$.

A one-way ANOVA was conducted to determine if the root-ball weight for plants that have been root-pruned weigh less than those that have not. Subjects were classified into two groups: treated ($n = 28$), and control ($n = 10$). There were no extreme outliers, as assessed by boxplot. Root-ball weight differences were statistically significant between the two groups, $F(1, 36) = 10.79$, $p < 0.005$.

5.4.3 Photographic Observations

Video 5-2 - Timelapse video: treated plants, cut-width between 4 and 5 inches

Time-lapse video data collection in phase three was similar to that of the previous portions of this project. The tobacco plants wilted in a cyclical pattern with the apex of wilting usually occurring between 5PM and 6PM (Video 5-2). The plants in the video were treated with the root-pruning implement and were found to have a cut distance of between 4 and 5 inches, which is narrower than the average cut distance.

5.4.4 Discussion

5.4.4.1 Main Effects

These results give further evidence that root pruning can cause wilting by reducing root moisture uptake potential and water availability. The average soil moisture content within the cut was significantly different from that measured outside the cut in the first two replications of the soil moisture content tests. During the last root-pruning test, the average volumetric moisture content was found to be at least 5% greater outside of the cut distance than within in all cases. This differential in soil moisture content may be a predictor of the amount of wilting the plants will experience. The higher soil moisture content was due to the elimination of soil moisture uptake by the roots of the tobacco plants outside the cuts where the roots were severed. Inside the cut, the roots took up as much moisture as possible, reducing soil moisture levels significantly, which essentially acted like a fast onset harsh drought. As transpiration through the leaves continued, the moisture content and associated turgidity of the leaves was reduced, thereby contributing to wilting.

The trend generally seemed to be toward a higher leaf breaking range for the root-pruned plants, as would be expected. However, the variation in these results, along with the considerable inconsistency in the phase one and two results, is too great for this measurement to be used as a reliable indicator of a wilted state. The average leaf moisture content was slightly higher in the untreated plants in the first test (when very little wilting occurred as indicated by the subjective rating results). In the second and third tests when many of the plants were in a wilted state, there was very little difference between the average leaf moisture contents of the untreated and root-pruned plants, and the moisture content was actually slightly lower for the untreated plants in replication 2, contrary to what would be expected for leaves in a wilted state.

The effect of *Leaf breaking range* and the interaction between the two were all found to be statistically significant. The effect of *Day* was not found to be statistically significant in this test. This is probably due to the soil and atmospheric conditions not fluctuating. *Leaf breaking range* is a primary endpoint that should be kept in future studies, but further improvements need to be made to increase reliability and decrease error. While it did have a statistically significant result in the last phase of testing, it often did not capture perceived wilting throughout the whole project. The *leaf moisture content* cannot be considered to have any relevance in assessing the wilted state of tobacco leaves based on the results of this study, namely, the lack of a correlation with *leaf breaking range* or *subjective rating*.

The results of these tests indicate that it is possible to cause wilting of burley of tobacco plants by pruning the roots to reduce soil moisture uptake, provided the roots are pruned close enough. In the phase three tests, a substantial amount of wilting occurred when the distance between the cuts on either side of the plant row was less than 8 inches, but very little wilting occurred when the cut distance was close to 10 inches. It would be useful to know more about the magnitude of reduction in roots required to cause wilting, but this study has shown how challenging it is to make such determinations with reasonable certainty. These tests showed that neither leaf breaking angle nor leaf moisture content measurements could be used to reliably assess the wilted state of tobacco leaves. This lack of an efficient objective means of assessing wilting (other than a subjective visual rating), combined with the difficulty of pruning the roots of large numbers of plants in the field with accurate control, makes it extremely difficult to determine an accurate relationship.

While the uncertainty of why some plants wilt and others do not is still present, it is obvious from this study that the root pruning has to be done fairly close to the stalk. In phase three, tests that recorded substantial wilting had a cut distance of less than 8 inches. In Figure 5-11 a row treated with an average of an 8 inch cut distance (right) can be easily discerned from a row of control plants (left). This means an average distance from the center of the row of less than 4 inches, and a distance from the outside of the stalk of less than 3 inches -- assuming a stalk diameter of 2 inches at the ground surface. Even this close root pruning did not produce severe wilting for a portion of the treated plants. The reliability of causing wilting by root pruning is further complicated by the effects that environmental conditions (temperature, solar radiation, rainfall, wind velocity) and root geometry have on the rate of transpiration of the leaves.

Figure 5-11 - Untreated plants (left), coulter disc root-pruned plants (right), Plot A. Plants on the right side had an 8 inch average cut distance.

While the root pruning did not consistently cause severe wilting, a substantial amount of wilting was evident when the root pruning was closer to the tobacco plants. The data were analyzed to investigate whether there was any correlation between the cut distance and the subjective rating. The narrower the cut distance, the smaller the plant's root-ball would become. This would lead to a diminished water-uptake capacity. The results of the analysis (Figure 5-12) showed that there was some trend toward a lower rating (less wilting) for a wider cut width with an R^2 value of 0.50. However, the correlation was found not to be statistically significant. This is believed to be because minimum cut distance to either side of the plant is more impactful than total cut width; a very low minimum cut distance on one side might cut the tap root and more than half the root system while a more evenly distributed cut would allow some portion of the tap root to remain. The rating for each subject was determined by taking the average of days 0, 1, and 4. The number of subjects per cut distance was graphed (Figure 5-13). A bimodal distribution is apparent, with peaks around 7.5 inches and 13.5 inches. This is likely due to the difficulty achieving and maintaining the ideal cut distance.

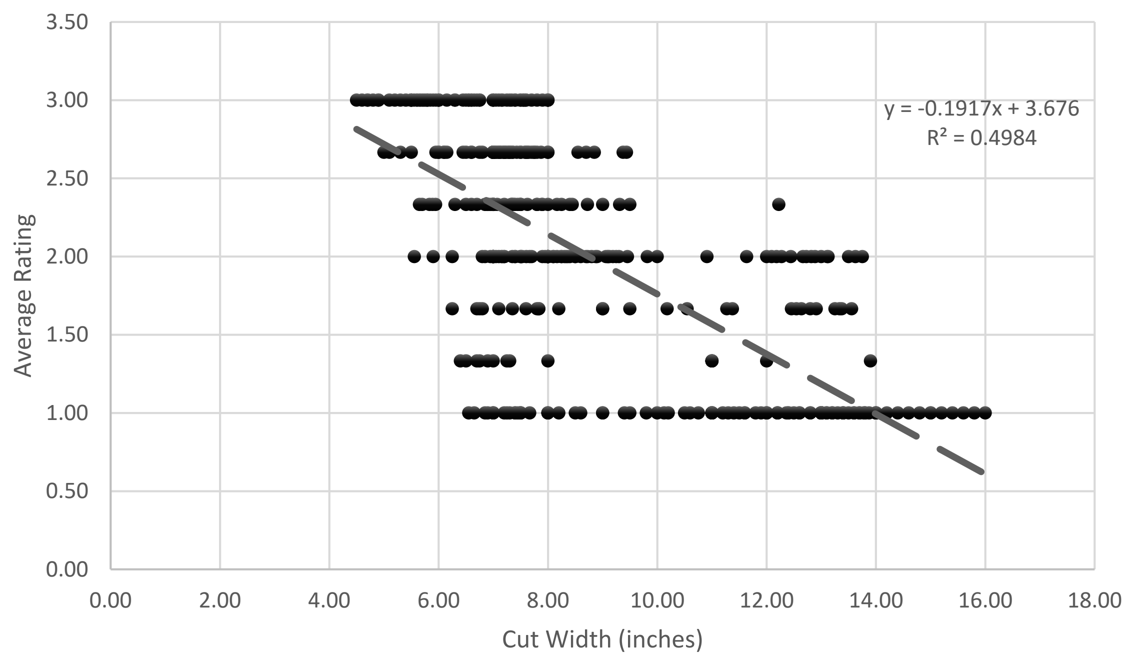


Figure 5-12 - Average Rating vs Cut Width for all subjects in phase three

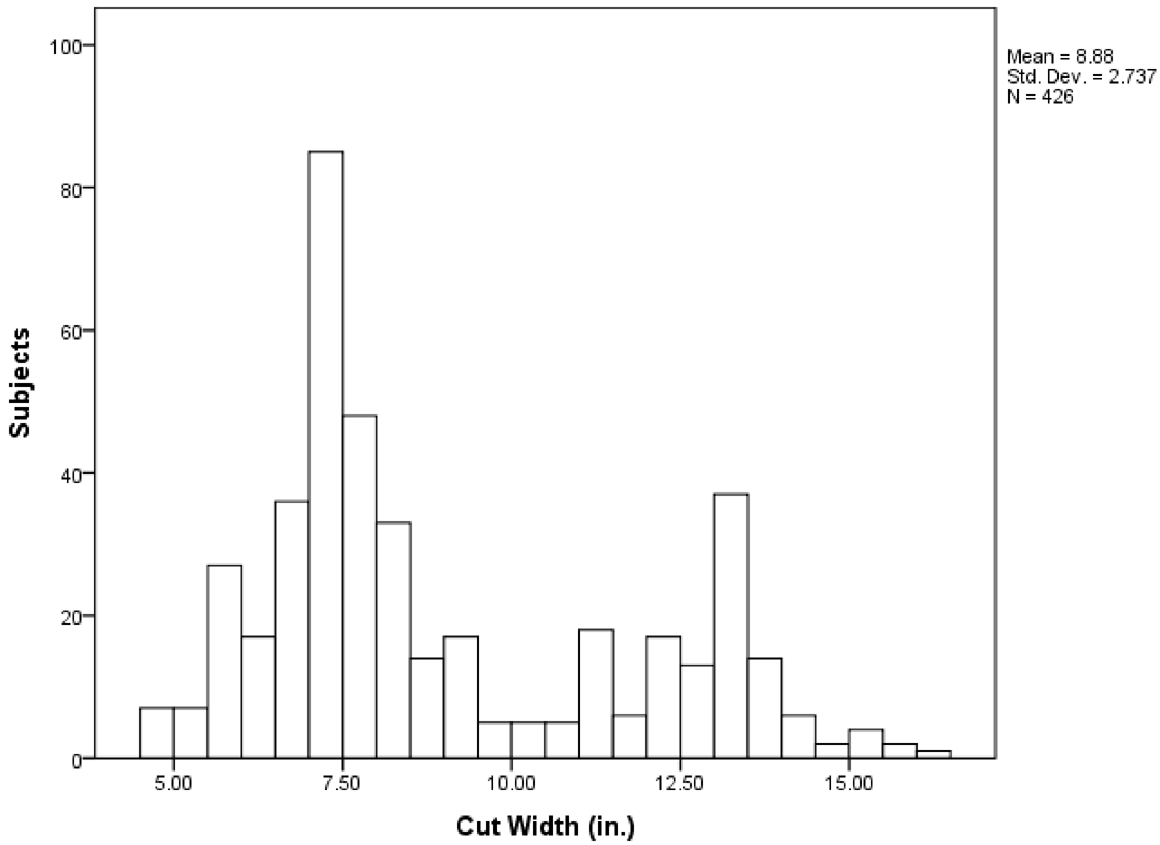


Figure 5-13 - Sum of plants treated, grouped by cut width

All subjects were binned into four categories and one control group (Table 5-2). The data were then dividing by test and entered into SPSS.

Table 5-2 - Total number of measurements taken grouped by cut width and test type

Group	*Subjective Rating*	*Leaf Breaking Angle*	*Leaf MC*	*Soil MC*	*Rootball Weight*
4 to 6 in.	41	16	16	-	4
6 to 8 in.	186	55	55	29	13
8 to 10 in.	69	33	33	27	4
10+ in.	130	17	17	37	6
Control	66	66	66	-	10

Basic statistical analyses for the Plot C location were completed in a similar fashion to phase two, except with the addition of *Subjective Rating*. The results are summarized in Table 5-3, Table 5-4, and Table 5-5.

Table 5-3 - Summary of statistics for Plot C, Phase Three

Cut Width (in.)	*Leaf Range (degrees)*	*Moisture Content (% w.b.)*	*Rating*
4 to 6 in.	62.2 ± 22.3	87.6 ± 1.5	3.0 ± 0.0
6 to 8 in.	62.7 ± 19.3	87.9 ± 1.9	2.1 ± 0.9
8 to 10 in.	58.9 ± 20.2	87.6 ± 2.5	1.2 ± 0.4
10+ in.	40.5 ± 15.8	91.0 ± 2.7	1.0 ± 0.1
Control	48.0 ± 26.1	88.0 ± 2.9	1.0 ± 0.0

Table 5-4 - Summary of results for Plot A, Phase Three

Cut Width (in.)	*Leaf Range (degrees)*	*Moisture Content (% w.b.)*	*Rating*
4 to 6 in.	78.6 ± 28.2	87.6 ± 0.9	2.8 ± 0.4
6 to 8 in.	64.5 ± 25.4	87.8 ± 1.9	2.4 ± 0.7
8 to 10 in.	63.3 ± 23.4	86.0 ± 1.0	2.1 ± 0.5
10+ in.	-	-	1.8 ± 0.4
Control	38.3 ± 15.7	86.9 ± 1.5	1.0 ± 0.0

Table 5-5 - Summary of statistics, all locations, Phase Three

Cut Width (in.)	*Leaf Range (degrees)*	*Moisture Content (% w.b.)*	*Rating*
4 to 6 in.	69.4 ± 25.6	87.6 ± 1.3	2.8 ± 0.4
6 to 8 in.	63.8 ± 23.0	87.8 ± 1.9	2.3 ± 0.7
8 to 10 in.	60.9 ± 21.5	86.9 ± 2.1	2.0 ± 0.6
10+ in.	40.5 ± 15.8	91.0 ± 2.7	1.3 ± 0.4
Control	45.0 ± 23.7	87.7 ± 2.6	1.0 ± 0.0

For nearly every group at all locations, there seemed to be a negative linear relationship between cut width and *Leaf Breaking Range* and *Subjective Rating*. *Moisture Content* did not appear to relate to any other group. Standard Error for all groups was lower in phase three when compared to the results in phase two.

Moisture Content was compared against *Leaf Breaking Angle* by a Pearson's correlation test using IBM SPSS. Results were similar to results from phase two, and no correlation was found. The lack of linear relationship to cut width as well as the lack of correlation to *Leaf Range* shows that *Moisture Content* was an unreliable measure of wilting. Further analysis of *Moisture Content* was not considered.

Leaf Breaking Range results were analyzed for all locations graphically via box and whisker plots in SPSS by group (Figure 5-14). The narrowest cut width resulted in the largest average *Leaf Breaking Range*, followed by the next two most narrow cut width groups. The largest cut width group (greater than 10 inches) did not show better results than the control group graphically.

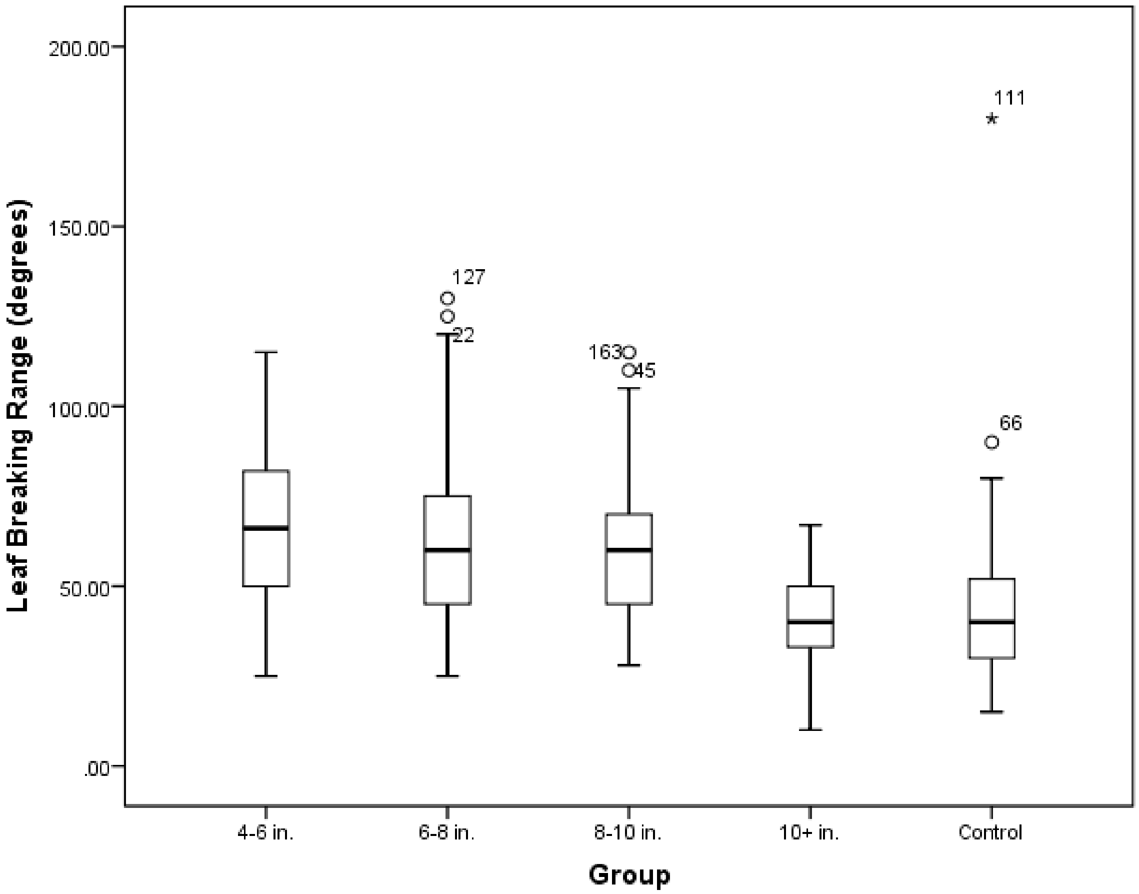


Figure 5-14 - Leaf Breaking Range grouped by cut distance, Phase Three

Subjective Rating results were combined for all locations and entered into SPSS divided into the binned groups. Bar graphs were generated with each level of wilting clustered into each group (Figure 5-15). The percentage of *Some Wilting* and *Very Wilted* start very high at the narrowest binned group and gradually lower to almost 25% by the widest cut width grouping. Similarly, the largest population of *No Wilting* was seen in the widest cut width grouping, while none were seen at the narrowest grouping.

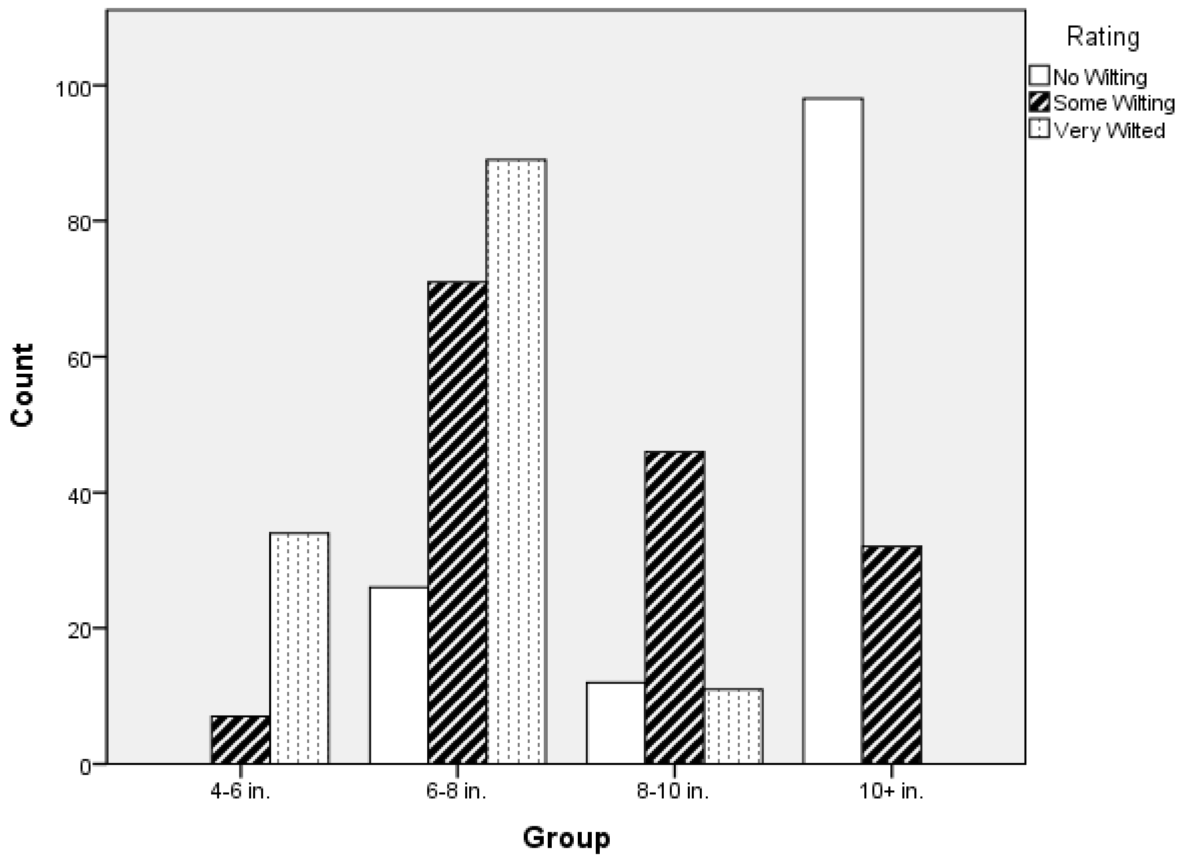


Figure 5-15 - Sum of treated plants, grouped by cut distance and subjective wilting rating

Binomial logistic regression was run on SPSS with all the *Cut Width* and *Subjective Rating* data from all tests. For each additional inch in cut width, there was a resulting 0.545x chance wilting would occur (Table 5-6). Using the variables from Table 5-6, a prediction was graphed (Figure 5-16).

Table 5-6 - Coefficients of model relating cut distance to how wilted a plant appears to be

Variables in the Equation

		B	S.E.	Wald	df	Sig.	Exp(B)	95% C.I.for EXP(B)	
								Lower	Upper
Step 1[a]	Cut Width (in.)	-.607	.056	116.331	1	.000	.545	.488	.609
	Constant	5.963	.517	132.804	1	.000	388.581		

a. Variable(s) entered on step 1: Cut Width (in.).

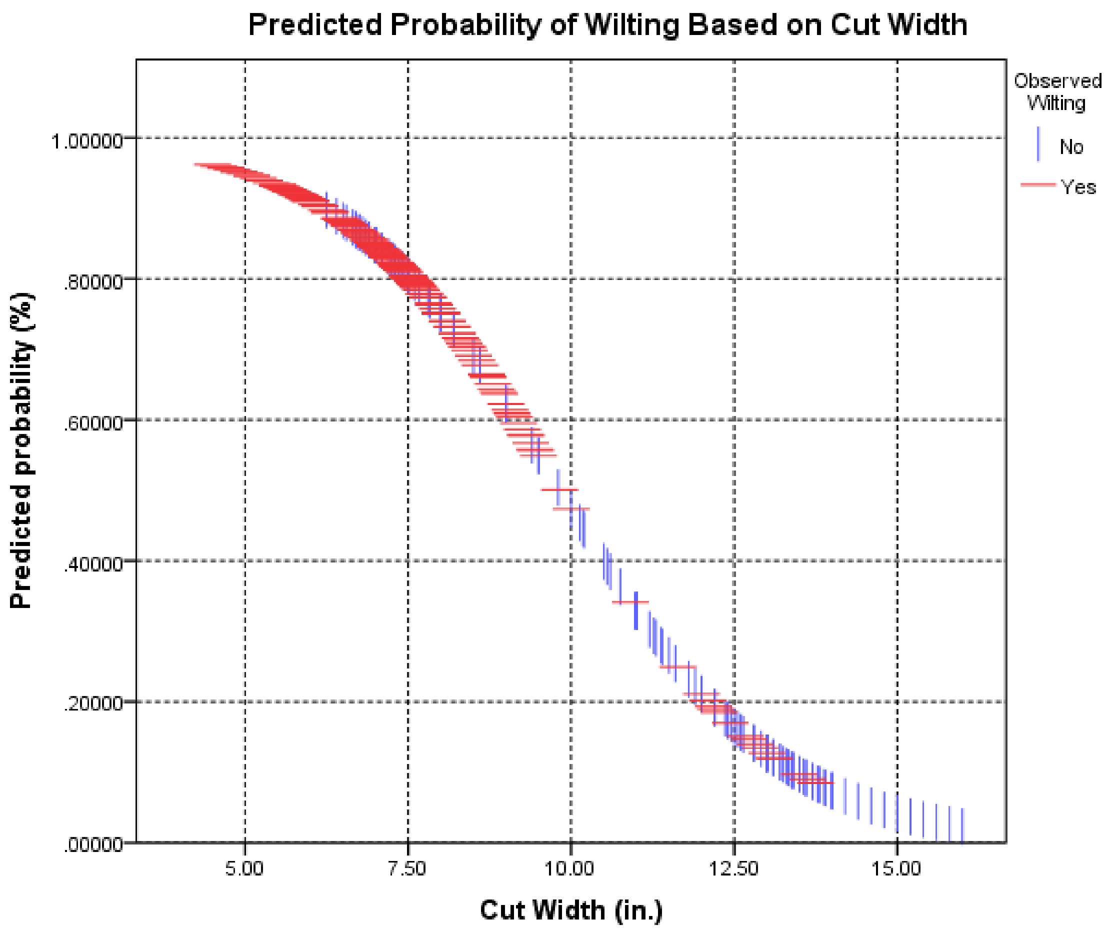


Figure 5-16 - Graph of logistic binomial regression, with observed data fitted

From these analyses, the chance of wilting can be determined for a given cut width. For example, at the 12.5 inch cut width, there is about a 17% chance that a tobacco plant will wilt. At a 7.5 inch cut width, that chance is increased to around 80%. This model might aid future studies by determining minimum design requirements for cut distance or how much treatment variance is acceptable. The model was found to be statistically significant.

5.4.4.2 Soil Moisture Content

Soil moisture content was analyzed to determine if there was a difference in the treated soil area and the untreated soil area. This effect is exaggerated in phase three because there is one continuous trough the whole length of a row created by the coulter disc. A box and whisker plot was generated in SPSS using the Soil Moisture Content data from within the trough and outside of the trough (Figure 5-17). There was a noticeable difference between the soil next to the plant and the soil outside of the cut area.

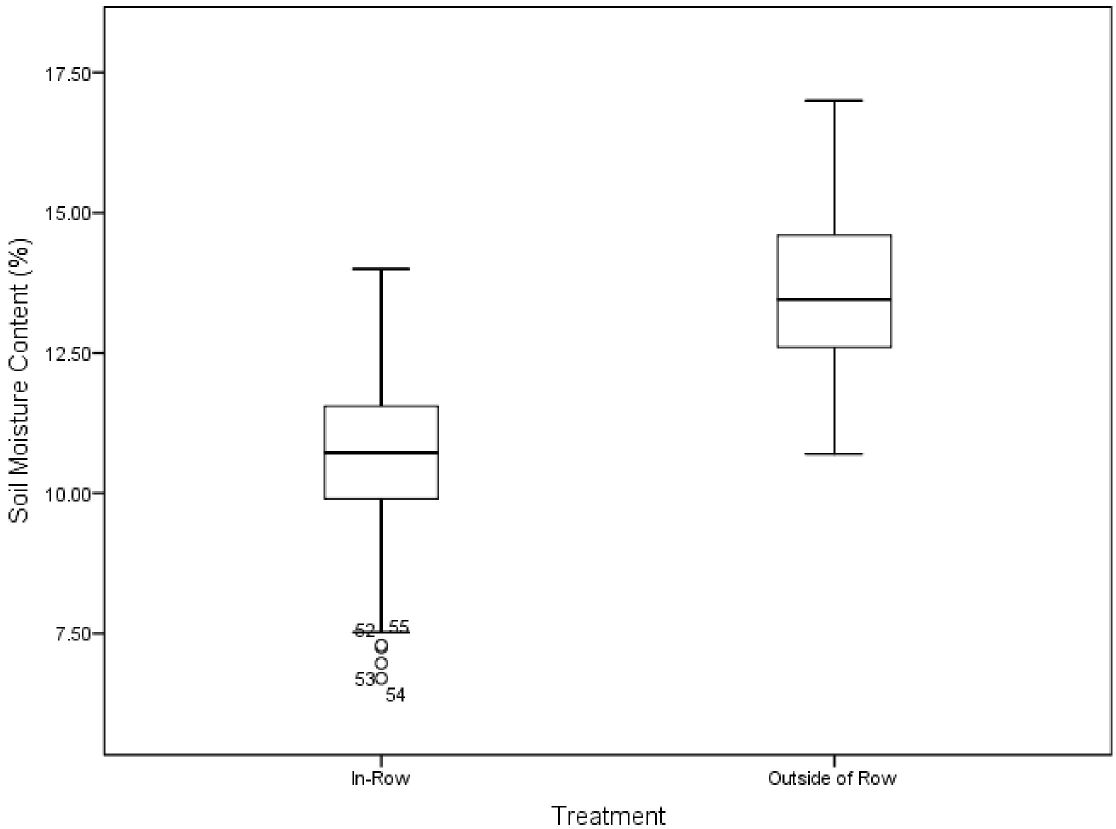


Figure 5-17 - Soil Moisture Content, grouped by location

5.4.4.3 Root-ball weight analysis

Root-ball volume, and therefore weight, are hypothesized to be significantly reduced by the root pruning treatment. Because of the non-uniform growth pattern of roots, it was unknown whether the treatment was producing a significant result, on average.

The mean root-ball weight of treated plants was 69.58g, while the mean root-ball weight of control plants was 105.66g (Figure 5-18):

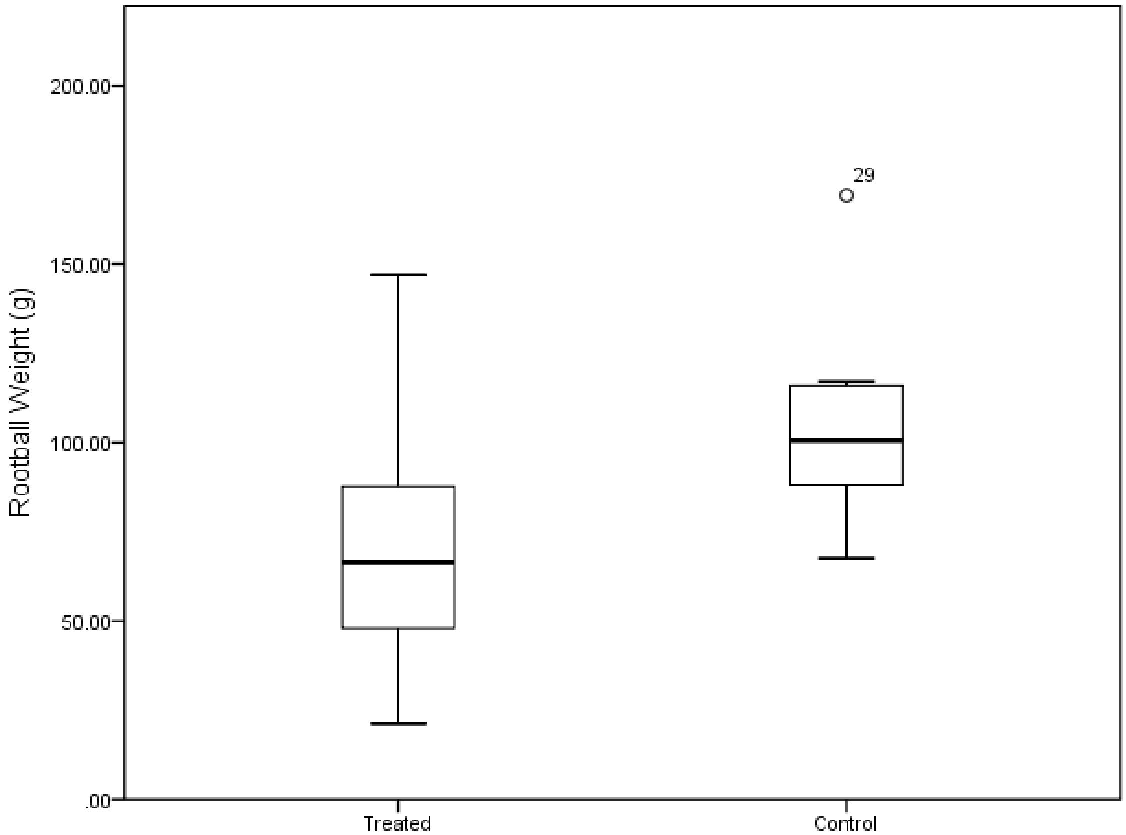


Figure 5-18 - Rootball weight, grouped by treated or control

5.4.5 Conclusion

Results in the final phase of testing remained inconsistent as they have been in previous phases. Some plants showed moderate and extreme wilting, but they were the minority. Differences between the binned groups could be noticed in the leaf breaking range test, and the results were significant. A more reliable method to capture wilting seems to be via subjective rating. This data collection method also allows for quickly collecting hundreds of data points.

According to the logistic binomial regression model that was built, these results are to be expected at the cut distances that were performed. Using the implement to achieve consistent application distances proved very difficult, and hence, yielded inconsistent results. If it were possible to keep the implement cutting closer than 7.5 inches, 80% of similar tobacco plants in similar conditions should show above moderate wilting.

CHAPTER 6 – Conclusion

6.1 Summary

While this project did have a number of unique problems and obstacles, much was learned about what causes tobacco plants to wilt and how to measure that effect. No statistically significant results caused by the treatments were found in the early stages of the project (phase one and two). Individual and small groups of plants occasionally became very wilted, but this did not strongly impact the results. The only statistically significant component found in the early stages of testing was the effect of *Day* on wilting. Leaf moisture content analysis by method of oven drying and weighing the leaves was found to be a poor indicator of wilting. The liquid nitrogen and MAPP gas treated plants occasionally showed the most extreme wilting of any test subjects, but those methods are too costly and time intensive to adapt to a mechanized version.

In the last stage of testing (phase three), the treatment group and the day in which data were collected were both found to have a significant impact on wilting. The subjective rating system was an improvement over previous attempts to quantify wilting, as it was visually driven and allowed data to be taken quickly at the maximum point of wilting. Differences between treated plants and control plants were easier to discern in the final phase; however, results were still intermittent. A model was built to better help predict the cut distance at which any given plant would wilt.

Based on these investigations, the only practical and cost-effective way to root prune tobacco plants in the field is with a rolling coulter. Because of the way the coulter works, there is great risk of damage to the standing tobacco plants in the field, both to the lower leaves on the plant (because such a large diameter disk is required to get sufficient cutting depth) and, of much greater concern, to the entire plant from literally chopping the plant down if the cut distance gets too close. If the roots can be successfully pruned close enough to cause wilting, then there is a potentially increased risk that the tobacco plants will be blown over because of the loosened soil and reduced anchoring from the roots.

6.2 Recommendations for Improvement

The largest source of error in the treatment of plants came from the difficulty of driving the machine accurately in such a small margin of error. There is room for possible innovation using GPS or other methods to appropriately keep a fixed cutting distance from the tobacco stalks. Crop planting practices might also have to be improved; in all locations there were variances in the proximity tobacco stalks were located relative to the row. If the soil is being cut 2 to 3 inches away from the stalk, a two-inch variance can potentially chop down several plants with the current implementation of this machine. Moreover, some plants grew out of the ground crooked further complicating the pruning location.

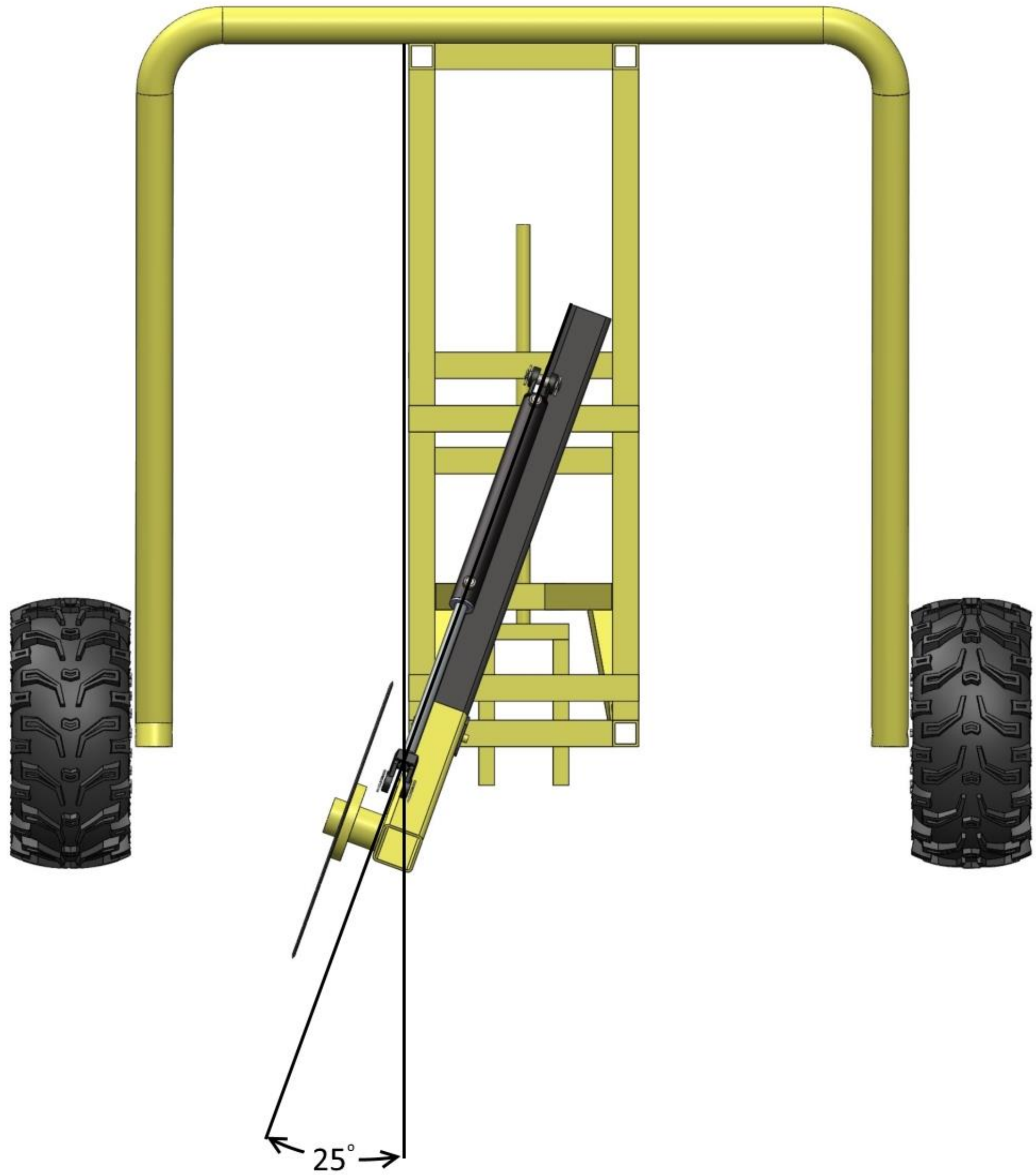
Capturing the wilted state objectively remains a challenge. Moisture content analysis via the methods utilized in this study were shown to be useless at predicting the wilted state with the varieties of tobacco used. Once the leaf breaking range test was extended to 10 cm it did prove to be more useful; however, the standard error was still very high in such a test. Further refinement of this test could decrease its error.

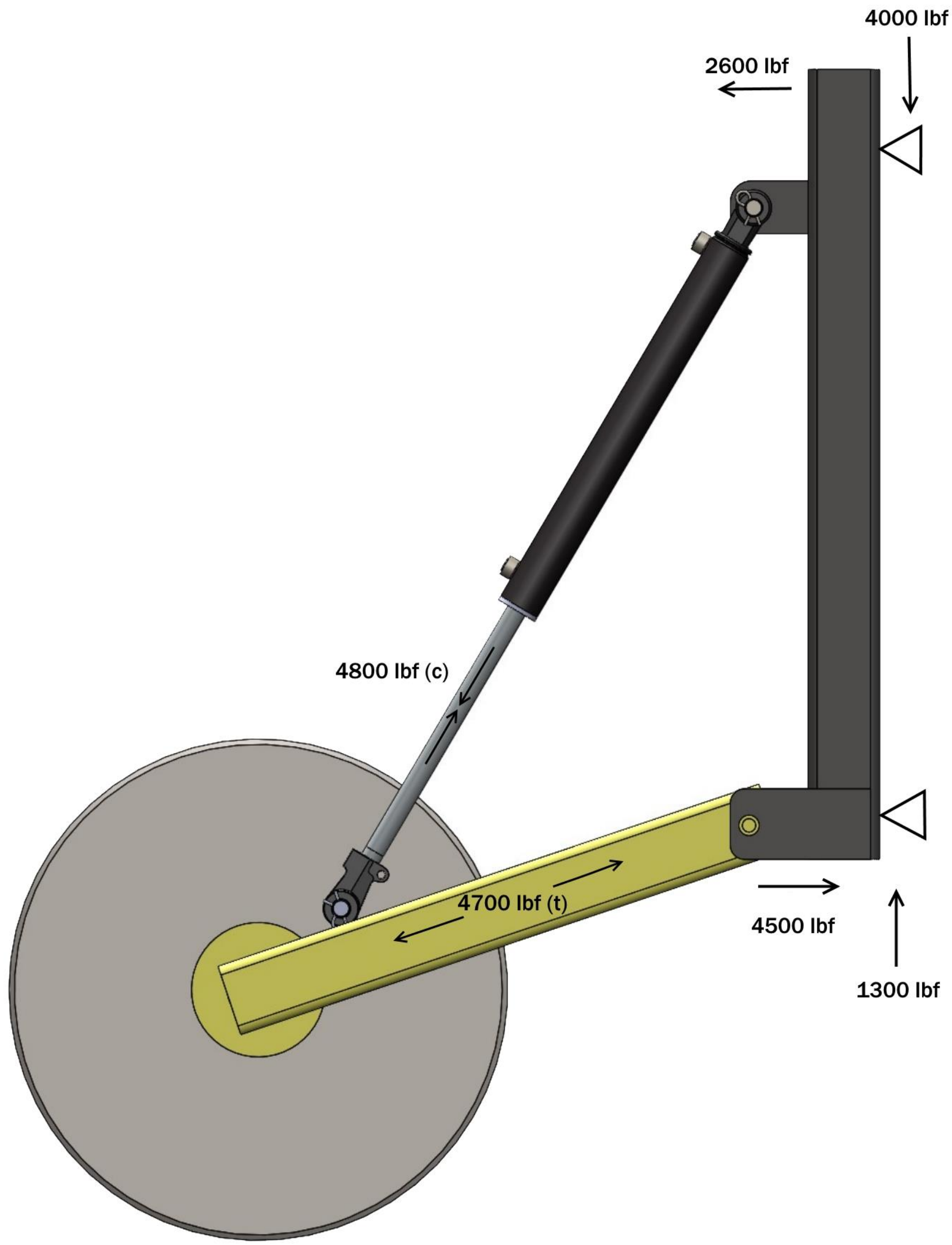
6.3 Future Work

Based on experiences in this study, the conclusion is that it would be extremely difficult to guide a root-pruning coulter with the required accuracy and precision to cause reliable wilting with minimal risk of cutting down plants. While it might be possible achieve considerably better accuracy and precision using innovative GPS guidance systems that track both planting and cut locations, even that would not guarantee that the risk of destroying the plants was minimized because of the tendency that tobacco stalks have of growing to one side or the other from the spot that they were planted. Considering the risk in loss of crop value, both from cutting down plants accidentally and from plants getting blown down, it is not recommended that this method be researched further utilizing the same methods.

Freezing or burning the stalks was seen as too cost prohibitive in this study to recommend; however, if improvements are made such that the treatment time or overall cost were lowered, these methods might be worth investigating further.

Appendix A – Additional Pictures / Renderings




4000 lbf
2600 lbf
4800 lbf (c)
4700 lbf (t)
4500 lbf
1300 lbf

Appendix B - Phase 2 – Plot B Main Effects

```
GET

  FILE='C:\SPSS\2010\PrincetonANOVA.sav'.

DATASET NAME DataSet1 WINDOW=FRONT.

* Chart Builder.

GGRAPH

  /GRAPHDATASET NAME="graphdataset" VARIABLES=Leaf_range MC MISSING=LISTWISE
REPORTMISSING=NO

  /GRAPHSPEC SOURCE=INLINE.

BEGIN GPL

  SOURCE: s=userSource(id("graphdataset"))

  DATA: Leaf_range=col(source(s), name("Leaf_range"))

  DATA: MC=col(source(s), name("MC"))

  GUIDE: axis(dim(1), label("Leaf Range (degrees)"))

  GUIDE: axis(dim(2), label("Moisture Content (% w.b.)"))

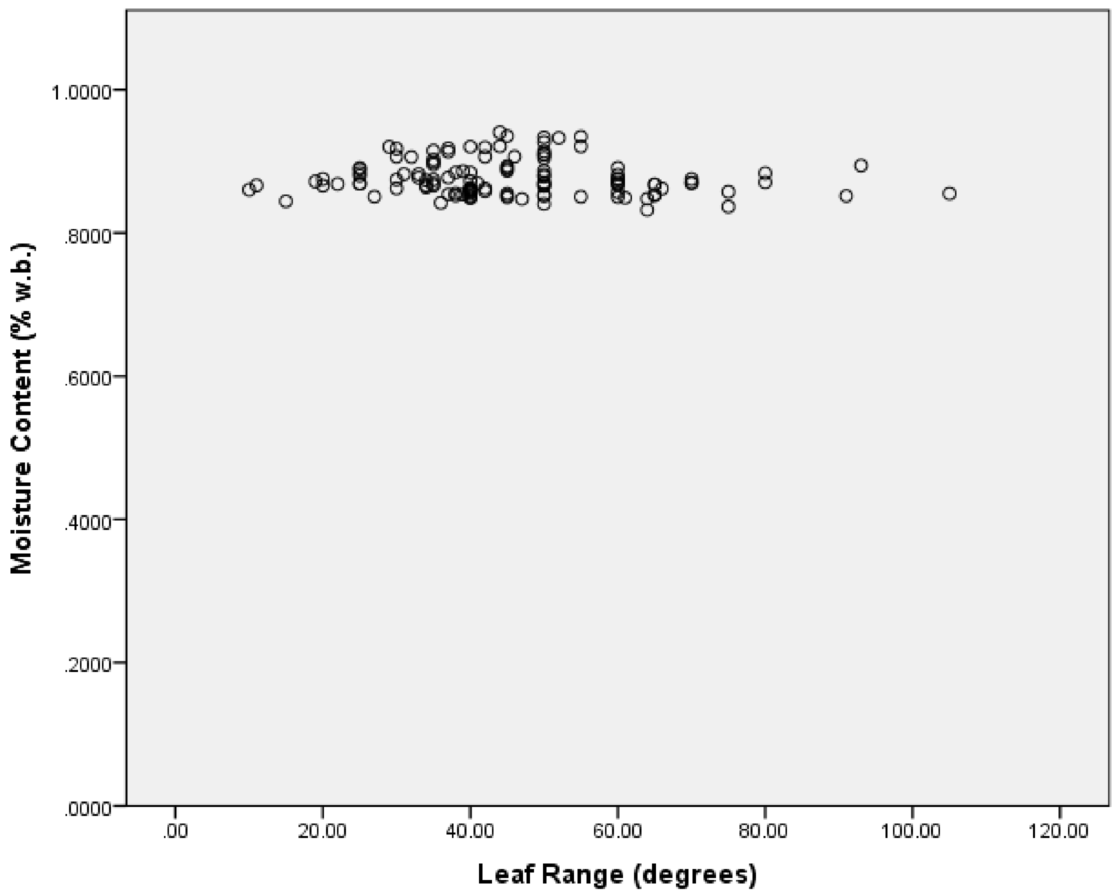
  SCALE: linear(dim(2), min(0))

  ELEMENT: point(position(Leaf_range*MC))

END GPL.
```

GGraph

[DataSet1] C:\SPSS\2010\PrincetonANOVA.sav



```
EXAMINE VARIABLES=MC Leaf_range
 /PLOT NPPLOT
 /STATISTICS NONE
 /CINTERVAL 95
 /MISSING LISTWISE
 /NOTOTAL.
```

Explore

Case Processing Summary

	Cases					
	Valid		Missing		Total	
	N	Percent	N	Percent	N	Percent
Moisture Content (% w.b.)	120	100.0%	0	0.0%	120	100.0%
Leaf Range (degrees)	120	100.0%	0	0.0%	120	100.0%

Tests of Normality

	Kolmogorov-Smirnov[a]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
Moisture Content (% w.b.)	.123	120	.000	.938	120	.000
Leaf Range (degrees)	.131	120	.000	.961	120	.002

a. Lilliefors Significance Correction

Moisture Content (% w.b.)

```
CORRELATIONS
  /VARIABLES=Leaf_range MC
  /PRINT=TWOTAIL NOSIG
  /MISSING=PAIRWISE.
```

Correlations

Correlations

		Leaf Range (degrees)	Moisture Content (% w.b.)
Leaf Range (degrees)	Pearson Correlation	1	-.113
	Sig. (2-tailed)		.220
	N	120	120
Moisture Content (% w.b.)	Pearson Correlation	-.113	1
	Sig. (2-tailed)	.220	
	N	120	120

```
UNIANOVA Leaf_range BY Rep
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /EMMEANS=TABLES(OVERALL)
  /PRINT=ETASQ DESCRIPTIVE
  /CRITERIA=ALPHA(.05)
  /DESIGN=Rep.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Rep	1	Rep 1	30
	2	Rep 2	30
	3	Rep 3	30
	4	Rep 4	30

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Rep	Mean	Std. Deviation	N
Rep 1	39.6000	20.44606	30
Rep 2	47.8000	13.25194	30
Rep 3	44.4333	15.54456	30
Rep 4	49.7000	14.62674	30
Total	45.3833	16.44762	120

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	1764.700[a]	3	588.233	2.243	.087	.055
Intercept	247157.633	1	247157.633	942.244	.000	.890
Rep	1764.700	3	588.233	2.243	.087	.055
Error	30427.667	116	262.307			
Total	279350.000	120				
Corrected Total	32192.367	119				

a. R Squared = .055 (Adjusted R Squared = .030)

Estimated Marginal Means

Grand Mean

Dependent Variable: Leaf Range (degrees)

Mean	Std. Error	95% Confidence Interval	
		Lower Bound	Upper Bound
45.383	1.478	42.455	48.312

```
UNIANOVA Leaf_range BY Rep TRT
 /METHOD=SSTYPE(3)
 /INTERCEPT=INCLUDE
 /EMMEANS=TABLES(OVERALL)
 /PRINT=ETASQ DESCRIPTIVE
 /CRITERIA=ALPHA(.05)
 /DESIGN=Rep TRT Rep*TRT.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Rep	1	Rep 1	30
	2	Rep 2	30
	3	Rep 3	30
	4	Rep 4	30
Treatment	1	Two-Sided Cut, 1.5" away	24
	2	Two-Sided Cut, 3" away	24
	3	String Trimmer + Burnt	24
	4	Burnt Stalk	24
	5	Control	24

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Rep	Treatment	Mean	Std. Deviation	N
Rep 1	Two-Sided Cut, 1.5" away	38.3333	23.17470	6
	Two-Sided Cut, 3" away	51.6667	27.70319	6
	String Trimmer + Burnt	33.6667	20.36337	6
	Burnt Stalk	39.8333	15.89235	6
	Control	34.5000	14.23728	6
	Total	39.6000	20.44606	30
Rep 2	Two-Sided Cut, 1.5" away	47.5000	9.79285	6
	Two-Sided Cut, 3" away	45.1667	18.30209	6
	String Trimmer + Burnt	47.3333	13.47096	6
	Burnt Stalk	51.3333	12.61216	6
	Control	47.6667	14.94880	6
	Total	47.8000	13.25194	30
Rep 3	Two-Sided Cut, 1.5" away	44.6667	14.90861	6
	Two-Sided Cut, 3" away	45.8333	13.19722	6
	String Trimmer + Burnt	41.3333	6.31401	6
	Burnt Stalk	41.8333	11.92337	6
	Control	48.5000	27.94817	6
	Total	44.4333	15.54456	30
Rep 4	Two-Sided Cut, 1.5" away	57.8333	13.93437	6
	Two-Sided Cut, 3" away	46.0000	6.84105	6
	String Trimmer + Burnt	44.1667	8.61201	6
	Burnt Stalk	53.3333	25.60208	6
	Control	47.1667	10.68488	6
	Total	49.7000	14.62674	30
Total	Two-Sided Cut, 1.5" away	47.0833	16.72779	24
	Two-Sided Cut, 3" away	47.1667	17.17092	24
	String Trimmer + Burnt	41.6250	13.45787	24

	Burnt Stalk	46.5833	17.27506	24
	Control	44.4583	17.94431	24
	Total	45.3833	16.44762	120

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	4124.033[a]	19	217.054	.773	.733	.128
Intercept	247157.633	1	247157.633	880.557	.000	.898
Rep	1764.700	3	588.233	2.096	.106	.059
TRT	539.783	4	134.946	.481	.750	.019
Rep * TRT	1819.550	12	151.629	.540	.883	.061
Error	28068.333	100	280.683			
Total	279350.000	120				
Corrected Total	32192.367	119				

a. R Squared = .128 (Adjusted R Squared = -.038)

Estimated Marginal Means

Grand Mean

Dependent Variable: Leaf Range (degrees)

Mean	Std. Error	95% Confidence Interval Lower Bound	Upper Bound
45.383	1.529	42.349	48.418

```
UNIANOVA Leaf_range BY Rep Day
 /METHOD=SSTYPE(3)
 /INTERCEPT=INCLUDE
 /EMMEANS=TABLES(OVERALL)
 /PRINT=ETASQ DESCRIPTIVE
 /CRITERIA=ALPHA(.05)
 /DESIGN=Rep Day Rep*Day.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Rep	1	Rep 1	30
	2	Rep 2	30
	3	Rep 3	30
	4	Rep 4	30
Day	1	Day 1	40
	2	Day 2	40
	3	Day 7	40

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Rep	Day	Mean	Std. Deviation	N
Rep 1	Day 1	38.8000	13.42303	10
	Day 2	45.5000	16.90661	10
	Day 7	34.5000	28.50828	10
	Total	39.6000	20.44606	30
Rep 2	Day 1	38.7000	8.28721	10
	Day 2	57.0000	10.85255	10
	Day 7	47.7000	13.90484	10
	Total	47.8000	13.25194	30
Rep 3	Day 1	47.2000	17.18397	10
	Day 2	43.0000	16.02082	10
	Day 7	43.1000	14.62456	10
	Total	44.4333	15.54456	30
Rep 4	Day 1	43.1000	6.26188	10
	Day 2	60.0000	19.29306	10
	Day 7	46.0000	9.82061	10
	Total	49.7000	14.62674	30
Total	Day 1	41.9500	12.13588	40
	Day 2	51.3750	17.09654	40
	Day 7	42.8250	18.17280	40
	Total	45.3833	16.44762	120

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	5802.167[a]	11	527.470	2.159	.022	.180
Intercept	247157.633	1	247157.633	1011.475	.000	.904
Rep	1764.700	3	588.233	2.407	.071	.063
Day	2169.317	2	1084.658	4.439	.014	.076
Rep * Day	1868.150	6	311.358	1.274	.275	.066
Error	26390.200	108	244.354			
Total	279350.000	120				
Corrected Total	32192.367	119				

a. R Squared = .180 (Adjusted R Squared = .097)

Estimated Marginal Means

Grand Mean

Dependent Variable: Leaf Range (degrees)

Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
45.383	1.427	42.555	48.212

```
UNIANOVA Leaf_range BY Day TRT
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /SAVE=PRED RESID SRESID
  /PLOT=PROFILE(Day*TRT TRT*Day)
  /EMMEANS=TABLES(Day*TRT)
```

```
/PRINT=ETASQ DESCRIPTIVE HOMOGENEITY

/CRITERIA=ALPHA(.05)

/DESIGN=Day TRT Day*TRT.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Day	1	Day 1	40
	2	Day 2	40
	3	Day 7	40
Treatment	1	Two-Sided Cut, 1.5" away	24
	2	Two-Sided Cut, 3" away	24
	3	String Trimmer + Burnt	24
	4	Burnt Stalk	24
	5	Control	24

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Day	Treatment	Mean	Std. Deviation	N
Day 1	Two-Sided Cut, 1.5" away	40.0000	7.03055	8
	Two-Sided Cut, 3" away	38.1250	10.17613	8
	String Trimmer + Burnt	39.0000	9.68061	8
	Burnt Stalk	43.3750	9.39510	8
	Control	49.2500	19.78275	8
	Total	41.9500	12.13588	40
Day 2	Two-Sided Cut, 1.5" away	56.2500	19.22610	8
	Two-Sided Cut, 3" away	50.0000	13.62770	8
	String Trimmer + Burnt	50.6250	9.03861	8
	Burnt Stalk	56.2500	21.17107	8
	Control	43.7500	20.48519	8
	Total	51.3750	17.09654	40
Day 7	Two-Sided Cut, 1.5" away	45.0000	18.49324	8
	Two-Sided Cut, 3" away	53.3750	23.08331	8
	String Trimmer + Burnt	35.2500	16.51623	8
	Burnt Stalk	40.1250	16.66851	8
	Control	40.3750	14.18185	8
	Total	42.8250	18.17280	40
Total	Two-Sided Cut, 1.5" away	47.0833	16.72779	24
	Two-Sided Cut, 3" away	47.1667	17.17092	24
	String Trimmer + Burnt	41.6250	13.45787	24
	Burnt Stalk	46.5833	17.27506	24
	Control	44.4583	17.94431	24
	Total	45.3833	16.44762	120

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Leaf Range (degrees)

F	df1	df2	Sig.
2.024	14	105	.023

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + Day + TRT + Day * TRT

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	5187.617[a]	14	370.544	1.441	.148	.161
Intercept	247157.633	1	247157.633	961.000	.000	.902
Day	2169.317	2	1084.658	4.217	.017	.074
TRT	539.783	4	134.946	.525	.718	.020
Day * TRT	2478.517	8	309.815	1.205	.303	.084
Error	27004.750	105	257.188			
Total	279350.000	120				
Corrected Total	32192.367	119				

a. R Squared = .161 (Adjusted R Squared = .049)

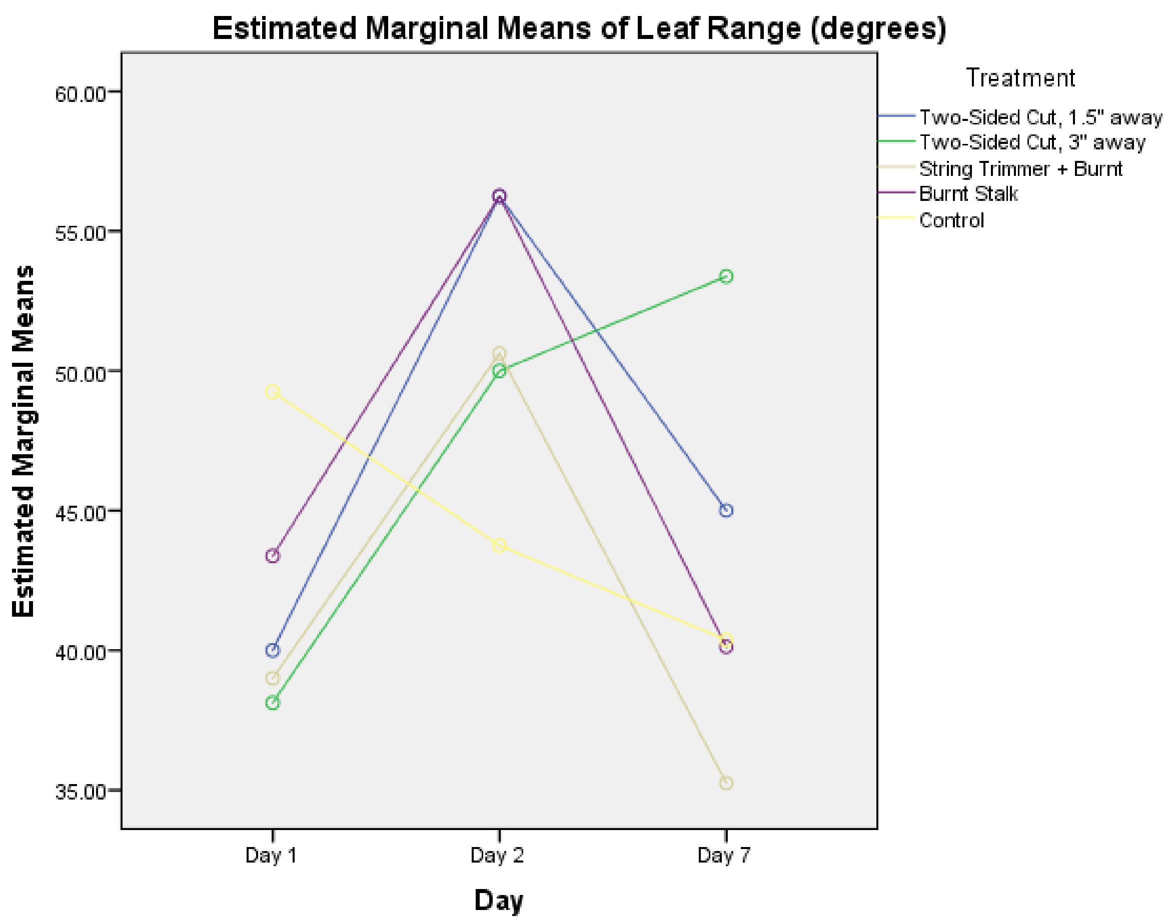
Estimated Marginal Means

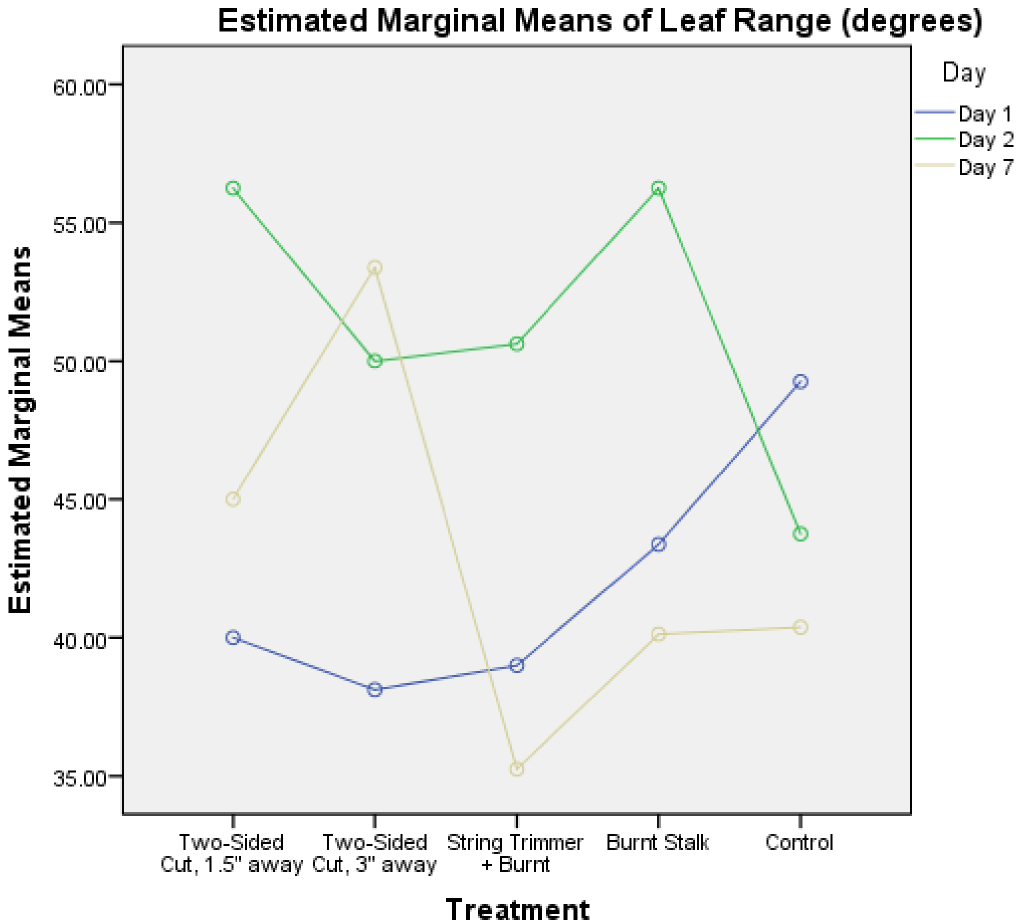
Day * Treatment

Dependent Variable: Leaf Range (degrees)

Day	Treatment	Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Day 1	Two-Sided Cut, 1.5" away	40.000	5.670	28.758	51.242
	Two-Sided Cut, 3" away	38.125	5.670	26.883	49.367
	String Trimmer + Burnt	39.000	5.670	27.758	50.242
	Burnt Stalk	43.375	5.670	32.133	54.617
	Control	49.250	5.670	38.008	60.492
Day 2	Two-Sided Cut, 1.5" away	56.250	5.670	45.008	67.492
	Two-Sided Cut, 3" away	50.000	5.670	38.758	61.242
	String Trimmer + Burnt	50.625	5.670	39.383	61.867
	Burnt Stalk	56.250	5.670	45.008	67.492
	Control	43.750	5.670	32.508	54.992
Day 7	Two-Sided Cut, 1.5" away	45.000	5.670	33.758	56.242
	Two-Sided Cut, 3" away	53.375	5.670	42.133	64.617
	String Trimmer + Burnt	35.250	5.670	24.008	46.492
	Burnt Stalk	40.125	5.670	28.883	51.367
	Control	40.375	5.670	29.133	51.617

Profile Plots






```
SORT CASES  BY TRT Day.

SPLIT FILE LAYERED BY TRT Day.

EXAMINE VARIABLES=RES_1

  /PLOT BOXPLOT NPPLOT

  /COMPARE GROUPS

  /STATISTICS NONE

  /CINTERVAL 95

  /MISSING LISTWISE

  /NOTOTAL
```

Explore

```
* Chart Builder.

GGRAPH

  /GRAPHDATASET NAME="graphdataset" VARIABLES=PRE_1 SRE_1 MISSING=LISTWISE
REPORTMISSING=NO

  /GRAPHSPEC SOURCE=INLINE.

BEGIN GPL

  SOURCE: s=userSource(id("graphdataset"))

  DATA: PRE_1=col(source(s), name("PRE_1"))

  DATA: SRE_1=col(source(s), name("SRE_1"))

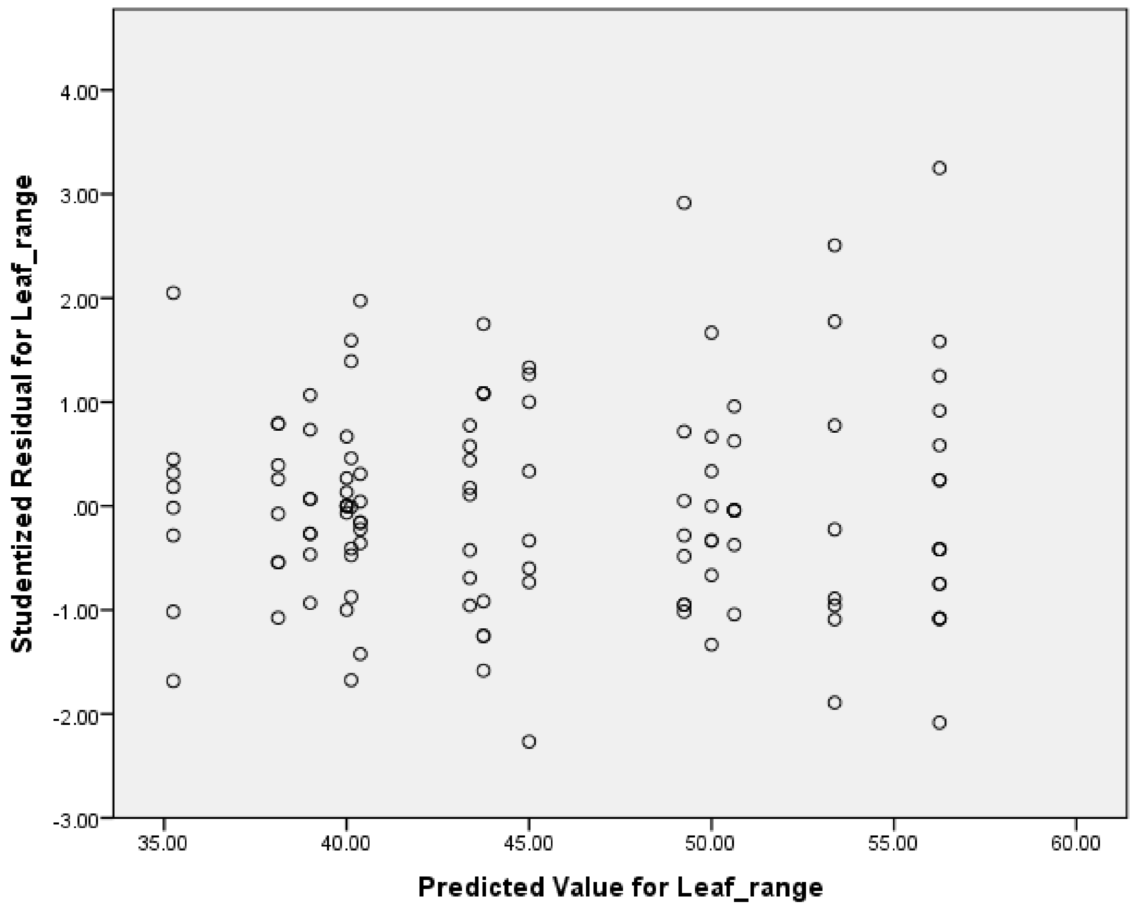
  GUIDE: axis(dim(1), label("Predicted Value for Leaf_range"))

  GUIDE: axis(dim(2), label("Studentized Residual for Leaf_range"))

  ELEMENT: point(position(PRE_1*SRE_1))

END GPL.
```

GGraph




```
UNIANOVA Leaf_range BY Day TRT
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /PLOT=PROFILE(Day*TRT TRT*Day)
  /EMMEANS=TABLES(Day*TRT) COMPARE(TRT) ADJ(BONFERRONI)
  /EMMEANS=TABLES(Day*TRT) COMPARE(Day) ADJ(BONFERRONI)
  /PRINT=ETASQ DESCRIPTIVE HOMOGENEITY
  /CRITERIA=ALPHA(.05)
  /DESIGN=Day TRT Day*TRT.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Day	1	Day 1	40
	2	Day 2	40
	3	Day 7	40
Treatment	1	Two-Sided Cut, 1.5" away	24
	2	Two-Sided Cut, 3" away	24
	3	String Trimmer + Burnt	24
	4	Burnt Stalk	24
	5	Control	24

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Day	Treatment	Mean	Std. Deviation	N
Day 1	Two-Sided Cut, 1.5" away	40.0000	7.03055	8
	Two-Sided Cut, 3" away	38.1250	10.17613	8
	String Trimmer + Burnt	39.0000	9.68061	8
	Burnt Stalk	43.3750	9.39510	8
	Control	49.2500	19.78275	8
	Total	41.9500	12.13588	40
Day 2	Two-Sided Cut, 1.5" away	56.2500	19.22610	8
	Two-Sided Cut, 3" away	50.0000	13.62770	8
	String Trimmer + Burnt	50.6250	9.03861	8
	Burnt Stalk	56.2500	21.17107	8
	Control	43.7500	20.48519	8
	Total	51.3750	17.09654	40
Day 7	Two-Sided Cut, 1.5" away	45.0000	18.49324	8
	Two-Sided Cut, 3" away	53.3750	23.08331	8
	String Trimmer + Burnt	35.2500	16.51623	8
	Burnt Stalk	40.1250	16.66851	8
	Control	40.3750	14.18185	8
	Total	42.8250	18.17280	40
Total	Two-Sided Cut, 1.5" away	47.0833	16.72779	24
	Two-Sided Cut, 3" away	47.1667	17.17092	24
	String Trimmer + Burnt	41.6250	13.45787	24
	Burnt Stalk	46.5833	17.27506	24
	Control	44.4583	17.94431	24
	Total	45.3833	16.44762	120

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Leaf Range (degrees)

F	df1	df2	Sig.
2.024	14	105	.023

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + Day + TRT + Day * TRT

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	5187.617[a]	14	370.544	1.441	.148	.161
Intercept	247157.633	1	247157.633	961.000	.000	.902
Day	2169.317	2	1084.658	4.217	.017	.074
TRT	539.783	4	134.946	.525	.718	.020
Day * TRT	2478.517	8	309.815	1.205	.303	.084
Error	27004.750	105	257.188			
Total	279350.000	120				
Corrected Total	32192.367	119				

a. R Squared = .161 (Adjusted R Squared = .049)

Estimated Marginal Means

1. Day * Treatment

Estimates

Dependent Variable: Leaf Range (degrees)

Day	Treatment	Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Day 1	Two-Sided Cut, 1.5" away	40.000	5.670	28.758	51.242
	Two-Sided Cut, 3" away	38.125	5.670	26.883	49.367
	String Trimmer + Burnt	39.000	5.670	27.758	50.242
	Burnt Stalk	43.375	5.670	32.133	54.617
	Control	49.250	5.670	38.008	60.492
Day 2	Two-Sided Cut, 1.5" away	56.250	5.670	45.008	67.492
	Two-Sided Cut, 3" away	50.000	5.670	38.758	61.242
	String Trimmer + Burnt	50.625	5.670	39.383	61.867
	Burnt Stalk	56.250	5.670	45.008	67.492
	Control	43.750	5.670	32.508	54.992
Day 7	Two-Sided Cut, 1.5" away	45.000	5.670	33.758	56.242
	Two-Sided Cut, 3" away	53.375	5.670	42.133	64.617
	String Trimmer + Burnt	35.250	5.670	24.008	46.492
	Burnt Stalk	40.125	5.670	28.883	51.367
	Control	40.375	5.670	29.133	51.617

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Day		Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Day 1	Contrast	659.650	4	164.913	.641	.634	.024
	Error	27004.750	105	257.188			
Day 2	Contrast	865.000	4	216.250	.841	.502	.031
	Error	27004.750	105	257.188			
Day 7	Contrast	1493.650	4	373.412	1.452	.222	.052
	Error	27004.750	105	257.188			

Each F tests the simple effects of Treatment within each level combination of the other effects shown. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.

2. Day * Treatment

Estimates

Dependent Variable: Leaf Range (degrees)

Day	Treatment	Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Day 1	Two-Sided Cut, 1.5" away	40.000	5.670	28.758	51.242
	Two-Sided Cut, 3" away	38.125	5.670	26.883	49.367
	String Trimmer + Burnt	39.000	5.670	27.758	50.242
	Burnt Stalk	43.375	5.670	32.133	54.617
	Control	49.250	5.670	38.008	60.492
Day 2	Two-Sided Cut, 1.5" away	56.250	5.670	45.008	67.492
	Two-Sided Cut, 3" away	50.000	5.670	38.758	61.242
	String Trimmer + Burnt	50.625	5.670	39.383	61.867
	Burnt Stalk	56.250	5.670	45.008	67.492
	Control	43.750	5.670	32.508	54.992
Day 7	Two-Sided Cut, 1.5" away	45.000	5.670	33.758	56.242
	Two-Sided Cut, 3" away	53.375	5.670	42.133	64.617
	String Trimmer + Burnt	35.250	5.670	24.008	46.492
	Burnt Stalk	40.125	5.670	28.883	51.367
	Control	40.375	5.670	29.133	51.617

Pairwise Comparisons

Dependent Variable: Leaf Range (degrees)

Treatment	(I) Day	(J) Day	Mean Difference (I-J)	Std. Error	Sig.[a]	95% Confidence Interval for Difference[a]	
						Lower Bound	Upper Bound
Two-Sided Cut, 1.5" away	Day 1	Day 2	-16.250	8.019	.136	-35.759	3.259
		Day 7	-5.000	8.019	1.000	-24.509	14.509
	Day 2	Day 1	16.250	8.019	.136	-3.259	35.759
		Day 7	11.250	8.019	.491	-8.259	30.759
	Day 7	Day 1	5.000	8.019	1.000	-14.509	24.509
		Day 2	-11.250	8.019	.491	-30.759	8.259
Two-Sided Cut, 3" away	Day 1	Day 2	-11.875	8.019	.425	-31.384	7.634
		Day 7	-15.250	8.019	.180	-34.759	4.259
	Day 2	Day 1	11.875	8.019	.425	-7.634	31.384
		Day 7	-3.375	8.019	1.000	-22.884	16.134
	Day 7	Day 1	15.250	8.019	.180	-4.259	34.759
		Day 2	3.375	8.019	1.000	-16.134	22.884
String Trimmer + Burnt	Day 1	Day 2	-11.625	8.019	.450	-31.134	7.884
		Day 7	3.750	8.019	1.000	-15.759	23.259
	Day 2	Day 1	11.625	8.019	.450	-7.884	31.134
		Day 7	15.375	8.019	.174	-4.134	34.884
	Day 7	Day 1	-3.750	8.019	1.000	-23.259	15.759
		Day 2	-15.375	8.019	.174	-34.884	4.134
Burnt Stalk	Day 1	Day 2	-12.875	8.019	.334	-32.384	6.634
		Day 7	3.250	8.019	1.000	-16.259	22.759
	Day 2	Day 1	12.875	8.019	.334	-6.634	32.384
		Day 7	16.125	8.019	.141	-3.384	35.634
	Day 7	Day 1	-3.250	8.019	1.000	-22.759	16.259
		Day 2	-16.125	8.019	.141	-35.634	3.384

Control	Day 1	Day 2	5.500	8.019	1.000	-14.009	25.009
		Day 7	8.875	8.019	.813	-10.634	28.384
	Day 2	Day 1	-5.500	8.019	1.000	-25.009	14.009
		Day 7	3.375	8.019	1.000	-16.134	22.884
	Day 7	Day 1	-8.875	8.019	.813	-28.384	10.634
		Day 2	-3.375	8.019	1.000	-22.884	16.134

Based on estimated marginal means

a. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Treatment		Sum of Squares	df	Mean Square	F	Sig.
Two-Sided Cut, 1.5" away	Contrast	1108.333	2	554.167	2.155	.121
	Error	27004.750	105	257.188		
Two-Sided Cut, 3" away	Contrast	1026.583	2	513.292	1.996	.141
	Error	27004.750	105	257.188		
String Trimmer + Burnt	Contrast	1028.250	2	514.125	1.999	.141
	Error	27004.750	105	257.188		
Burnt Stalk	Contrast	1163.583	2	581.792	2.262	.109
	Error	27004.750	105	257.188		
Control	Contrast	321.083	2	160.542	.624	.538
	Error	27004.750	105	257.188		

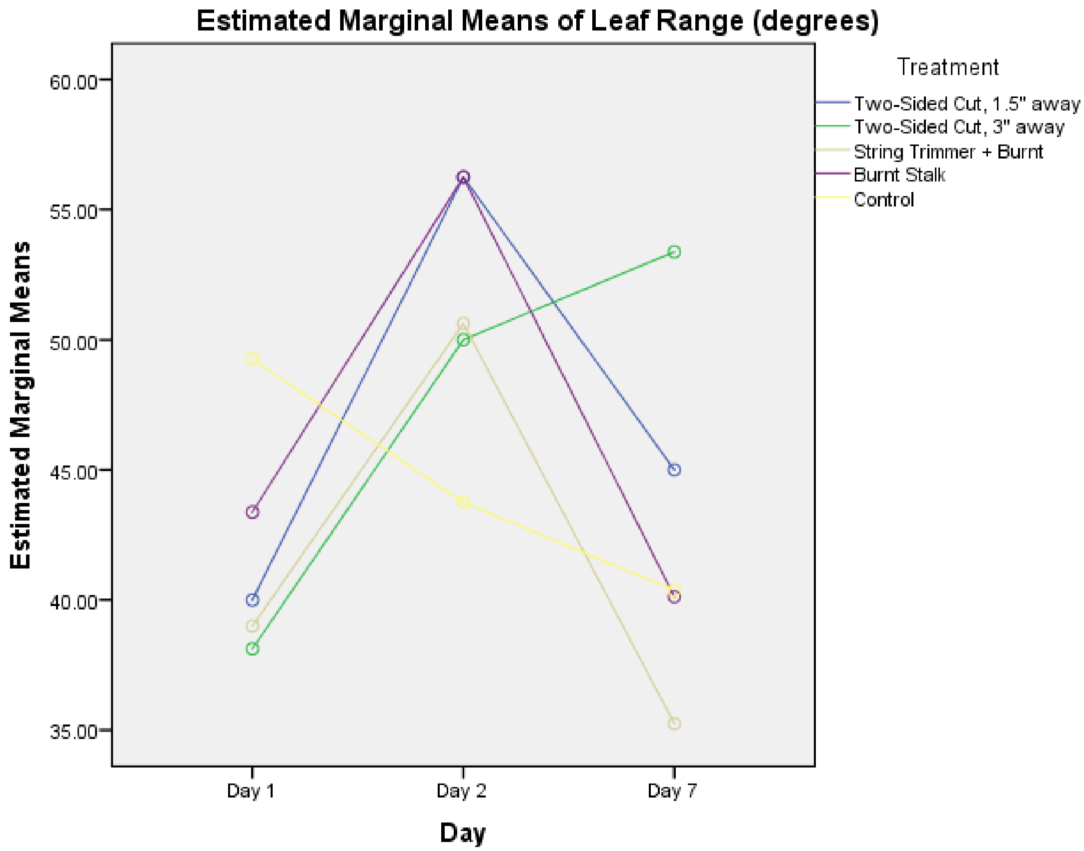
Univariate Tests

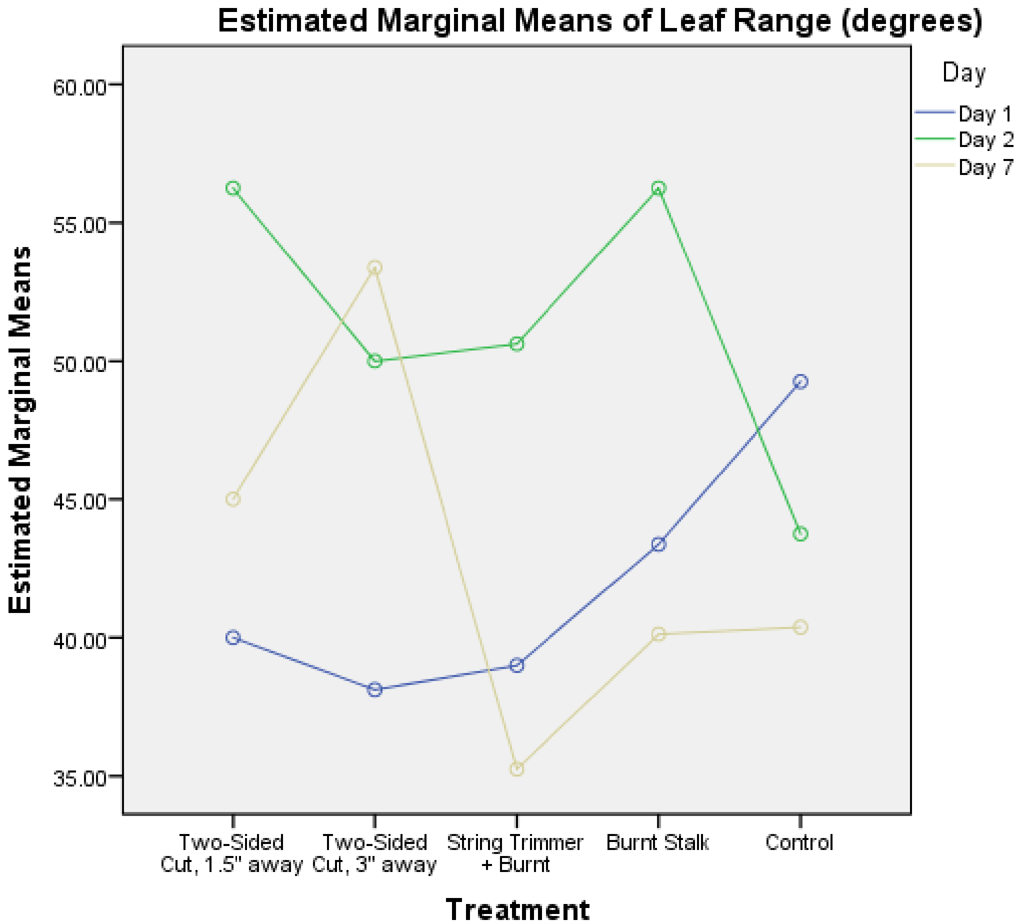
Dependent Variable: Leaf Range (degrees)

Treatment		Partial Eta Squared
Two-Sided Cut, 1.5" away	Contrast	.039
	Error	
Two-Sided Cut, 3" away	Contrast	.037
	Error	
String Trimmer + Burnt	Contrast	.037
	Error	
Burnt Stalk	Contrast	.041
	Error	
Control	Contrast	.012
	Error	

Each F tests the simple effects of Day within each level combination of the other effects shown. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.

Profile Plots






```
* Chart Builder.

GGRAPH

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95)[name="MEAN_Leaf_range"

    LOW="MEAN_Leaf_range_LOW" HIGH="MEAN_Leaf_range_HIGH"] Day MISSING=LISTWISE
REPORTMISSING=NO

  /GRAPHSPEC SOURCE=INLINE.

BEGIN GPL

  SOURCE: s=userSource(id("graphdataset"))

  DATA: TRT=col(source(s), name("TRT"), unit.category())
```

```
 DATA: MEAN_Leaf_range=col(source(s), name("MEAN_Leaf_range"))
 DATA: Day=col(source(s), name("Day"), unit.category())
 DATA: LOW=col(source(s), name("MEAN_Leaf_range_LOW"))
 DATA: HIGH=col(source(s), name("MEAN_Leaf_range_HIGH"))
 COORD: rect(dim(1,2), cluster(3,0))
 GUIDE: axis(dim(3), label("Treatment"))
 GUIDE: axis(dim(2), label("Mean Leaf Range (degrees)"))
 GUIDE: legend(aesthetic(aesthetic.color.interior), label("Day"))
 GUIDE: text.footnote(label("Error Bars: 95% CI"))
 SCALE: cat(dim(3), include("1", "2", "3", "4", "5"))
 SCALE: linear(dim(2), min(0))
 SCALE: cat(aesthetic(aesthetic.color.interior), include("1", "2", "3"))
 SCALE: cat(dim(1), include("1", "2", "3"))
 ELEMENT: interval(position(Day*MEAN_Leaf_range*TRT), color.interior(Day),
  shape.interior(shape.square))
 ELEMENT: interval(position(region.spread.range(Day*(LOW+HIGH)*TRT)),
shape.interior(shape.ibeam))
END GPL.
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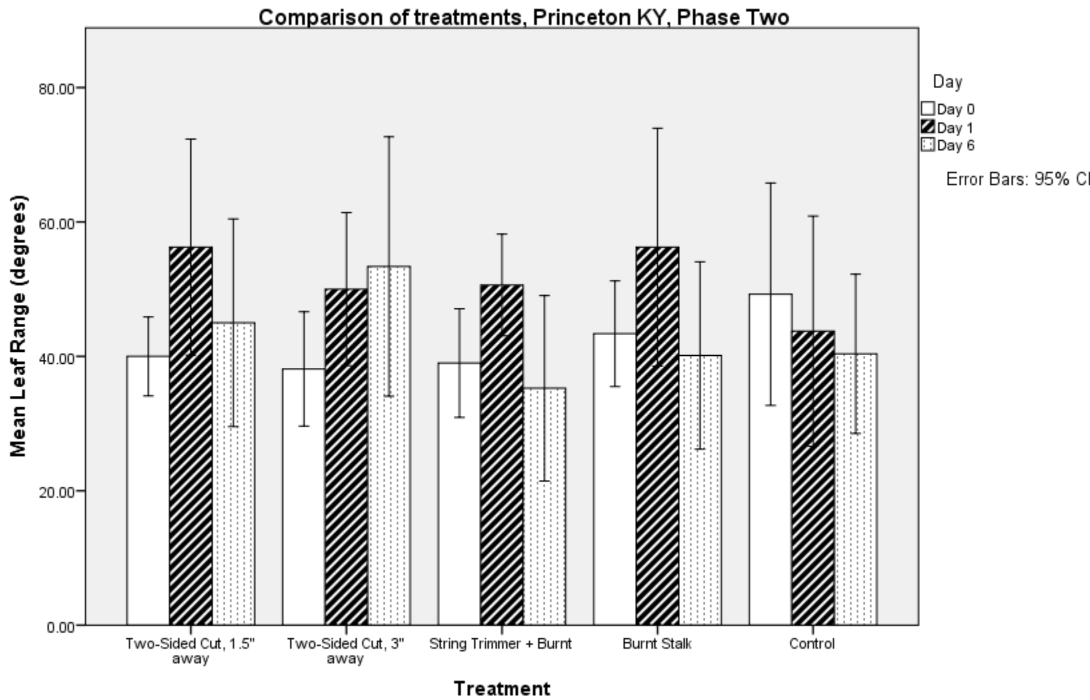
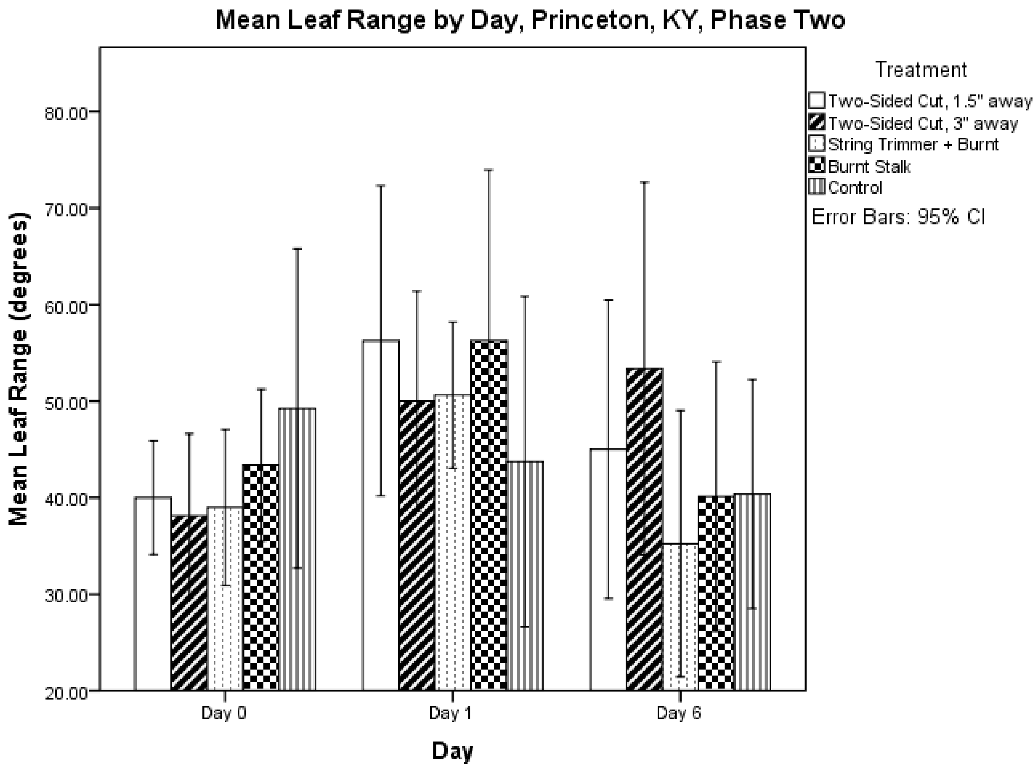
GGraph



GGraph

[DataSet4] E:\Google Drive\Thesis\Statistics\2010\PrincetonANOVA.sav



Mean Leaf Range by Day, Princeton, KY, Phase Two
Mean Leaf Range (degrees)
80.00
70.00
60.00
50.00
40.00
30.00
20.00
Day 0
Day 1
Day 6
Day
Treatment
Two-Sided Cut, 1.5" away
Two-Sided Cut, 3" away
String Trimmer + Burnt
Burnt Stalk
Control
Error Bars: 95% CI

Appendix C - Phase 2 – Plot C Main Effects

```
UNIANOVA Leaf_range BY TRT Day
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /SAVE=PRED RESID SRESID
  /PLOT=PROFILE(Day*TRT TRT*Day)
  /EMMEANS=TABLES(TRT*Day)
  /PRINT=ETASQ DESCRIPTIVE HOMOGENEITY
  /CRITERIA=ALPHA(.05)
  /DESIGN=TRT Day TRT*Day.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Treatment	1	Two-Sided Cut, 1.5" away	24
	2	Two-Sided Cut, 3" away	24
	3	Four-Sided Cut, 1.5" away	24
	4	Four-Sided Cut, 3" away	24
	5	Control	24
Day	1	Day 1	40
	2	Day 2	40
	3	Day 7	40

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Deviation	N
Two-Sided Cut, 1.5" away	Day 1	71.5000	16.50974	8
	Day 2	87.6250	31.83411	8
	Day 7	106.0000	50.85273	8
	Total	88.3750	37.22530	24
Two-Sided Cut, 3" away	Day 1	69.8750	30.67077	8
	Day 2	76.3750	28.45014	8
	Day 7	90.8750	32.76077	8
	Total	79.0417	30.65443	24
Four-Sided Cut, 1.5" away	Day 1	61.6250	19.68275	8
	Day 2	67.3750	15.19340	8
	Day 7	99.7500	31.62165	8
	Total	76.2500	28.04228	24
Four-Sided Cut, 3" away	Day 1	57.5000	17.63114	8
	Day 2	81.5000	37.77376	8
	Day 7	87.5000	32.88074	8
	Total	75.5000	32.14370	24
Control	Day 1	40.3750	20.44461	8
	Day 2	88.3750	32.19111	8
	Day 7	92.5000	16.33576	8
	Total	73.7500	33.28565	24
Total	Day 1	60.1750	23.36762	40
	Day 2	80.2500	29.54332	40
	Day 7	95.3250	33.52411	40
	Total	78.5833	32.28590	120

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Leaf Range (degrees)

F	df1	df2	Sig.
2.829	14	105	.001

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + TRT + Day + TRT * Day

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	34068.667[a]	14	2433.476	2.840	.001	.275
Intercept	741040.833	1	741040.833	864.793	.000	.892
TRT	3225.583	4	806.396	.941	.443	.035
Day	24877.117	2	12438.558	14.516	.000	.217
TRT * Day	5965.967	8	745.746	.870	.544	.062
Error	89974.500	105	856.900			
Total	865084.000	120				
Corrected Total	124043.167	119				

a. R Squared = .275 (Adjusted R Squared = .178)

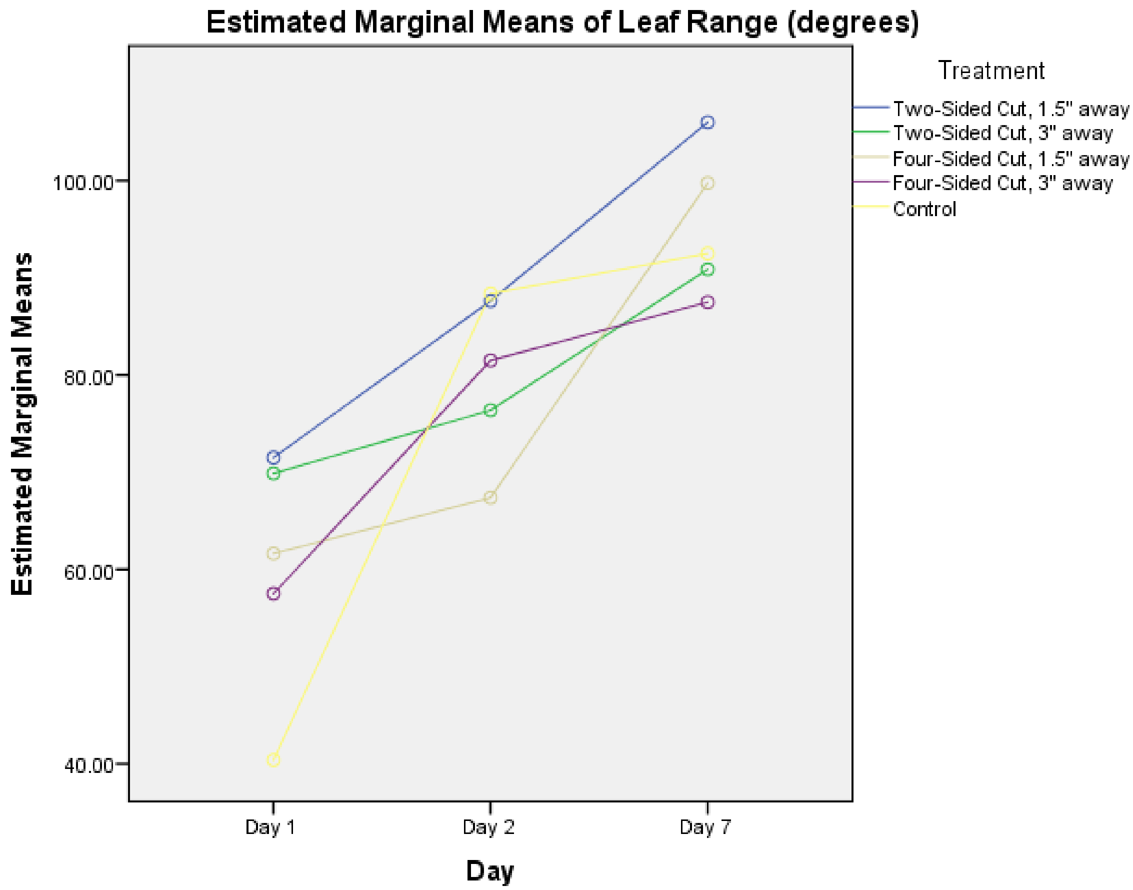
Estimated Marginal Means

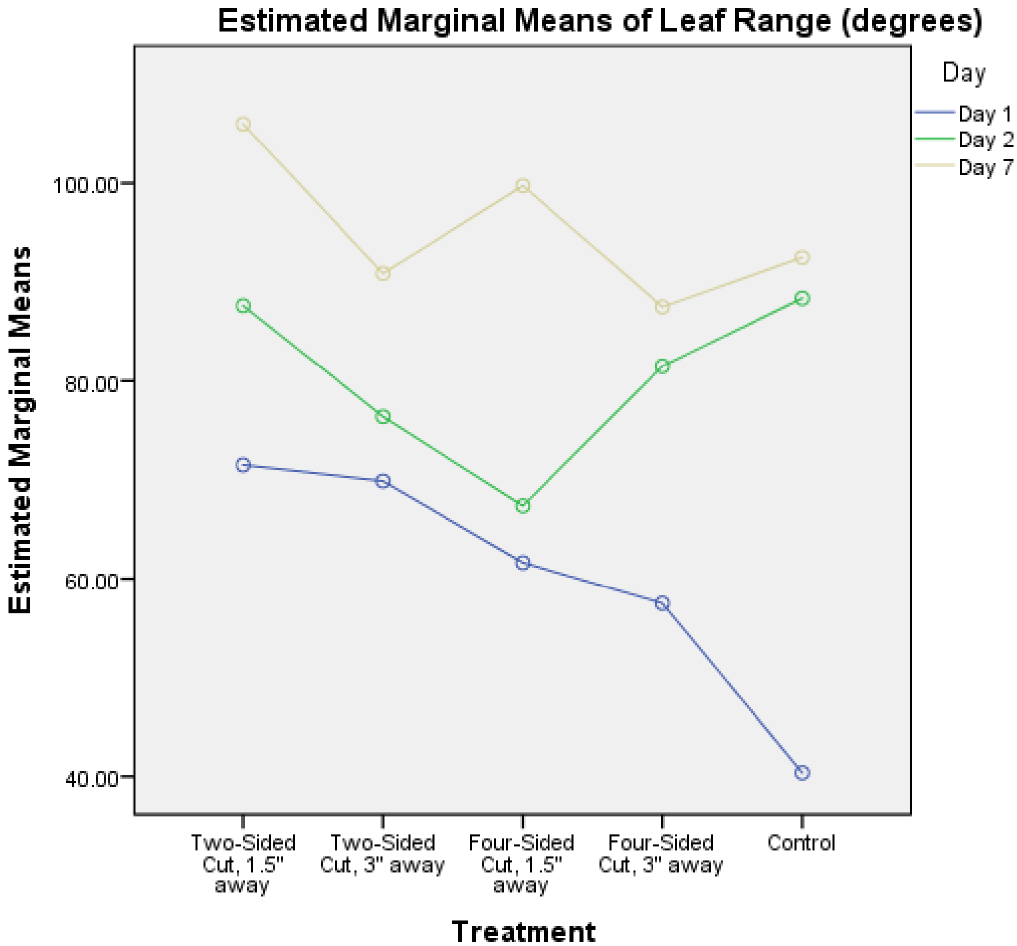
Treatment * Day

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Two-Sided Cut, 1.5" away	Day 1	71.500	10.350	50.979	92.021
	Day 2	87.625	10.350	67.104	108.146
	Day 7	106.000	10.350	85.479	126.521
Two-Sided Cut, 3" away	Day 1	69.875	10.350	49.354	90.396
	Day 2	76.375	10.350	55.854	96.896
	Day 7	90.875	10.350	70.354	111.396
Four-Sided Cut, 1.5" away	Day 1	61.625	10.350	41.104	82.146
	Day 2	67.375	10.350	46.854	87.896
	Day 7	99.750	10.350	79.229	120.271
Four-Sided Cut, 3" away	Day 1	57.500	10.350	36.979	78.021
	Day 2	81.500	10.350	60.979	102.021
	Day 7	87.500	10.350	66.979	108.021
Control	Day 1	40.375	10.350	19.854	60.896
	Day 2	88.375	10.350	67.854	108.896
	Day 7	92.500	10.350	71.979	113.021

Profile Plots






```
SORT CASES  BY Day TRT.

SPLIT FILE LAYERED BY Day TRT.

EXAMINE VARIABLES=RES_1

  /PLOT BOXPLOT NPPLOT

  /COMPARE GROUPS

  /STATISTICS NONE

  /CINTERVAL 95

  /MISSING LISTWISE

  /NOTOTAL
```

Explore

Tests of Normality

Day	Treatment		Kolmogorov-Smirnov[a] Statistic	df	Sig.
Day 1	Two-Sided Cut, 1.5" away	Residual for Leaf_range	.190	8	.200*
	Two-Sided Cut, 3" away	Residual for Leaf_range	.204	8	.200*
	Four-Sided Cut, 1.5" away	Residual for Leaf_range	.193	8	.200*
	Four-Sided Cut, 3" away	Residual for Leaf_range	.170	8	.200*
	Control	Residual for Leaf_range	.171	8	.200*
Day 2	Two-Sided Cut, 1.5" away	Residual for Leaf_range	.213	8	.200*
	Two-Sided Cut, 3" away	Residual for Leaf_range	.218	8	.200*
	Four-Sided Cut, 1.5" away	Residual for Leaf_range	.133	8	.200*
	Four-Sided Cut, 3" away	Residual for Leaf_range	.274	8	.077
	Control	Residual for Leaf_range	.195	8	.200*
Day 7	Two-Sided Cut, 1.5" away	Residual for Leaf_range	.241	8	.191
	Two-Sided Cut, 3" away	Residual for Leaf_range	.261	8	.117
	Four-Sided Cut, 1.5" away	Residual for Leaf_range	.163	8	.200*
	Four-Sided Cut, 3" away	Residual for Leaf_range	.128	8	.200*
	Control	Residual for Leaf_range	.188	8	.200*

*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

Residual for Leaf_range

```
SPLIT FILE OFF.

DATASET ACTIVATE DataSet4.

UNIANOVA Leaf_range BY TRT Day

  /METHOD=SSTYPE(3)

  /INTERCEPT=INCLUDE

  /PLOT=PROFILE(Day*TRT TRT*Day)

  /EMMEANS=TABLES(TRT*Day) COMPARE(TRT) ADJ(BONFERRONI)

  /EMMEANS=TABLES(TRT*Day) COMPARE(Day) ADJ(BONFERRONI)

  /PRINT=ETASQ DESCRIPTIVE HOMOGENEITY

  /CRITERIA=ALPHA(.05)

  /DESIGN=TRT Day TRT*Day.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Treatment	1	Two-Sided Cut, 1.5" away	24
	2	Two-Sided Cut, 3" away	24
	3	Four-Sided Cut, 1.5" away	24
	4	Four-Sided Cut, 3" away	24
	5	Control	24
Day	1	Day 1	40
	2	Day 2	40
	3	Day 7	40

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Deviation	N
Two-Sided Cut, 1.5" away	Day 1	71.5000	16.50974	8
	Day 2	87.6250	31.83411	8
	Day 7	106.0000	50.85273	8
	Total	88.3750	37.22530	24
Two-Sided Cut, 3" away	Day 1	69.8750	30.67077	8
	Day 2	76.3750	28.45014	8
	Day 7	90.8750	32.76077	8
	Total	79.0417	30.65443	24
Four-Sided Cut, 1.5" away	Day 1	61.6250	19.68275	8
	Day 2	67.3750	15.19340	8
	Day 7	99.7500	31.62165	8
	Total	76.2500	28.04228	24
Four-Sided Cut, 3" away	Day 1	57.5000	17.63114	8
	Day 2	81.5000	37.77376	8
	Day 7	87.5000	32.88074	8
	Total	75.5000	32.14370	24
Control	Day 1	40.3750	20.44461	8
	Day 2	88.3750	32.19111	8
	Day 7	92.5000	16.33576	8
	Total	73.7500	33.28565	24
Total	Day 1	60.1750	23.36762	40
	Day 2	80.2500	29.54332	40
	Day 7	95.3250	33.52411	40
	Total	78.5833	32.28590	120

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Leaf Range (degrees)

F	df1	df2	Sig.
2.829	14	105	.001

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + TRT + Day + TRT * Day

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	34068.667[a]	14	2433.476	2.840	.001	.275
Intercept	741040.833	1	741040.833	864.793	.000	.892
TRT	3225.583	4	806.396	.941	.443	.035
Day	24877.117	2	12438.558	14.516	.000	.217
TRT * Day	5965.967	8	745.746	.870	.544	.062
Error	89974.500	105	856.900			
Total	865084.000	120				
Corrected Total	124043.167	119				

a. R Squared = .275 (Adjusted R Squared = .178)

Estimated Marginal Means

1. Treatment * Day

Estimates

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
Two-Sided Cut, 1.5" away	Day 1	71.500	10.350	50.979	92.021
	Day 2	87.625	10.350	67.104	108.146
	Day 7	106.000	10.350	85.479	126.521
Two-Sided Cut, 3" away	Day 1	69.875	10.350	49.354	90.396
	Day 2	76.375	10.350	55.854	96.896
	Day 7	90.875	10.350	70.354	111.396
Four-Sided Cut, 1.5" away	Day 1	61.625	10.350	41.104	82.146
	Day 2	67.375	10.350	46.854	87.896
	Day 7	99.750	10.350	79.229	120.271
Four-Sided Cut, 3" away	Day 1	57.500	10.350	36.979	78.021
	Day 2	81.500	10.350	60.979	102.021
	Day 7	87.500	10.350	66.979	108.021
Control	Day 1	40.375	10.350	19.854	60.896
	Day 2	88.375	10.350	67.854	108.896
	Day 7	92.500	10.350	71.979	113.021

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Day		Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Day 1	Contrast	4989.150	4	1247.288	1.456	.221	.053
	Error	89974.500	105	856.900			
Day 2	Contrast	2422.000	4	605.500	.707	.589	.026
	Error	89974.500	105	856.900			
Day 7	Contrast	1780.400	4	445.100	.519	.722	.019
	Error	89974.500	105	856.900			

Each F tests the simple effects of Treatment within each level combination of the other effects shown. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.

2. Treatment * Day

Estimates

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
Two-Sided Cut, 1.5" away	Day 1	71.500	10.350	50.979	92.021
	Day 2	87.625	10.350	67.104	108.146
	Day 7	106.000	10.350	85.479	126.521
Two-Sided Cut, 3" away	Day 1	69.875	10.350	49.354	90.396
	Day 2	76.375	10.350	55.854	96.896
	Day 7	90.875	10.350	70.354	111.396
Four-Sided Cut, 1.5" away	Day 1	61.625	10.350	41.104	82.146
	Day 2	67.375	10.350	46.854	87.896
	Day 7	99.750	10.350	79.229	120.271
Four-Sided Cut, 3" away	Day 1	57.500	10.350	36.979	78.021
	Day 2	81.500	10.350	60.979	102.021
	Day 7	87.500	10.350	66.979	108.021
Control	Day 1	40.375	10.350	19.854	60.896
	Day 2	88.375	10.350	67.854	108.896
	Day 7	92.500	10.350	71.979	113.021

Pairwise Comparisons

Dependent Variable: Leaf Range (degrees)

Treatment	(I) Day	(J) Day	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b] Lower Bound	Upper Bound
Two-Sided Cut, 1.5" away	Day 1	Day 2	-16.125	14.636	.819	-51.735	19.485
		Day 7	-34.500	14.636	.061	-70.110	1.110
	Day 2	Day 1	16.125	14.636	.819	-19.485	51.735
		Day 7	-18.375	14.636	.636	-53.985	17.235
	Day 7	Day 1	34.500	14.636	.061	-1.110	70.110
		Day 2	18.375	14.636	.636	-17.235	53.985
Two-Sided Cut, 3" away	Day 1	Day 2	-6.500	14.636	1.000	-42.110	29.110
		Day 7	-21.000	14.636	.463	-56.610	14.610
	Day 2	Day 1	6.500	14.636	1.000	-29.110	42.110
		Day 7	-14.500	14.636	.972	-50.110	21.110
	Day 7	Day 1	21.000	14.636	.463	-14.610	56.610
		Day 2	14.500	14.636	.972	-21.110	50.110
Four-Sided Cut, 1.5" away	Day 1	Day 2	-5.750	14.636	1.000	-41.360	29.860
		Day 7	-38.125*	14.636	.032	-73.735	-2.515
	Day 2	Day 1	5.750	14.636	1.000	-29.860	41.360
		Day 7	-32.375	14.636	.087	-67.985	3.235
	Day 7	Day 1	38.125*	14.636	.032	2.515	73.735
		Day 2	32.375	14.636	.087	-3.235	67.985
Four-Sided Cut, 3" away	Day 1	Day 2	-24.000	14.636	.312	-59.610	11.610
		Day 7	-30.000	14.636	.129	-65.610	5.610
	Day 2	Day 1	24.000	14.636	.312	-11.610	59.610
		Day 7	-6.000	14.636	1.000	-41.610	29.610
	Day 7	Day 1	30.000	14.636	.129	-5.610	65.610
		Day 2	6.000	14.636	1.000	-29.610	41.610
Control	Day 1	Day 2	-48.000*	14.636	.004	-83.610	-12.390

		Day 7	-52.125*	14.636	.002	-87.735	-16.515
	Day 2	Day 1	48.000*	14.636	.004	12.390	83.610
		Day 7	-4.125	14.636	1.000	-39.735	31.485
	Day 7	Day 1	52.125*	14.636	.002	16.515	87.735
		Day 2	4.125	14.636	1.000	-31.485	39.735

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Treatment		Sum of Squares	df	Mean Square	F	Sig.
Two-Sided Cut, 1.5" away	Contrast	4767.750	2	2383.875	2.782	.066
	Error	89974.500	105	856.900		
Two-Sided Cut, 3" away	Contrast	1849.333	2	924.667	1.079	.344
	Error	89974.500	105	856.900		
Four-Sided Cut, 1.5" away	Contrast	6759.250	2	3379.625	3.944	.022
	Error	89974.500	105	856.900		
Four-Sided Cut, 3" away	Contrast	4032.000	2	2016.000	2.353	.100
	Error	89974.500	105	856.900		
Control	Contrast	13434.750	2	6717.375	7.839	.001
	Error	89974.500	105	856.900		

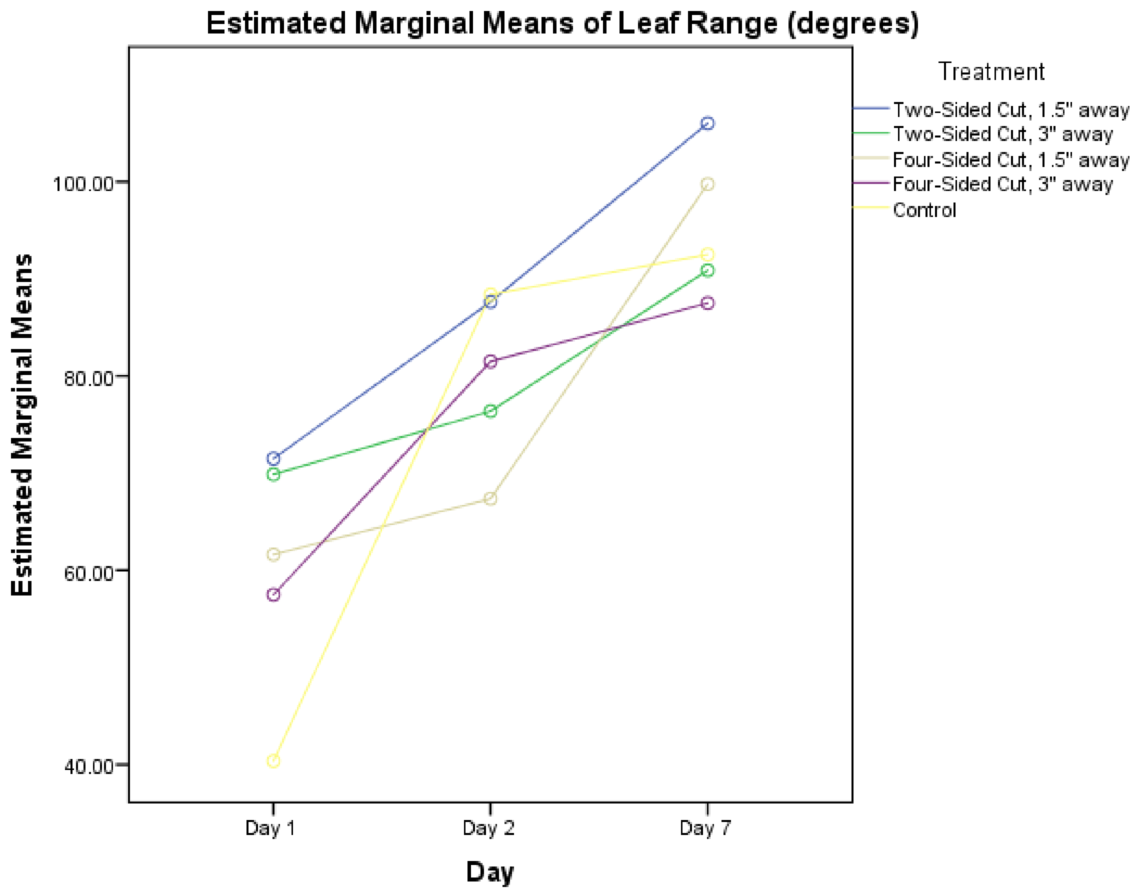
Univariate Tests

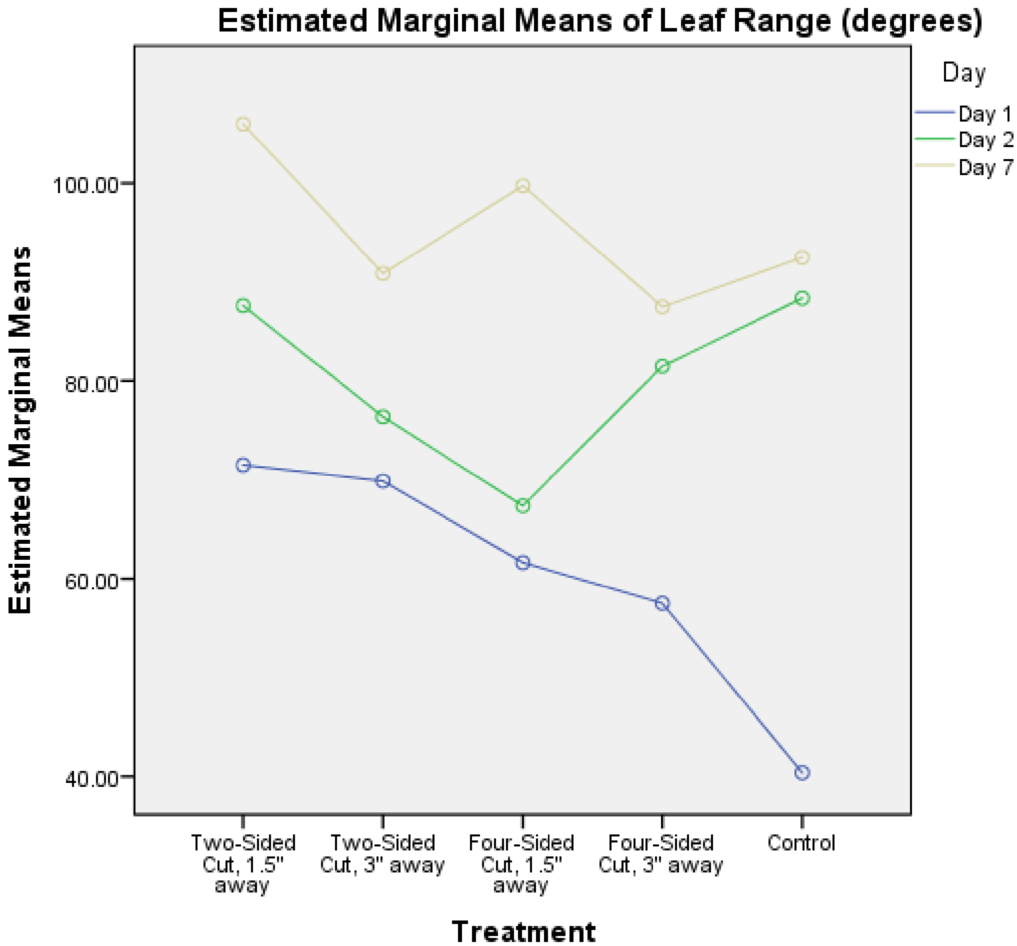
Dependent Variable: Leaf Range (degrees)

Treatment		Partial Eta Squared
Two-Sided Cut, 1.5" away	Contrast	.050
	Error	
Two-Sided Cut, 3" away	Contrast	.020
	Error	
Four-Sided Cut, 1.5" away	Contrast	.070
	Error	
Four-Sided Cut, 3" away	Contrast	.043
	Error	
Control	Contrast	.130
	Error	

Each F tests the simple effects of Day within each level combination of the other effects shown. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.

Profile Plots





* Chart Builder.

GGRAPH

/GRAPHDATASET NAME="graphdataset" VARIABLES=TRT MEANCI(Leaf_range, 95)[name="MEAN_Leaf_range" LOW="MEAN_Leaf_range_LOW" HIGH="MEAN_Leaf_range_HIGH"] Day MISSING=LISTWISE REPORTMISSING=NO

/GRAPHSPEC SOURCE=INLINE.

BEGIN GPL

SOURCE: s=userSource(id("graphdataset"))

DATA: TRT=col(source(s), name("TRT"), unit.category())

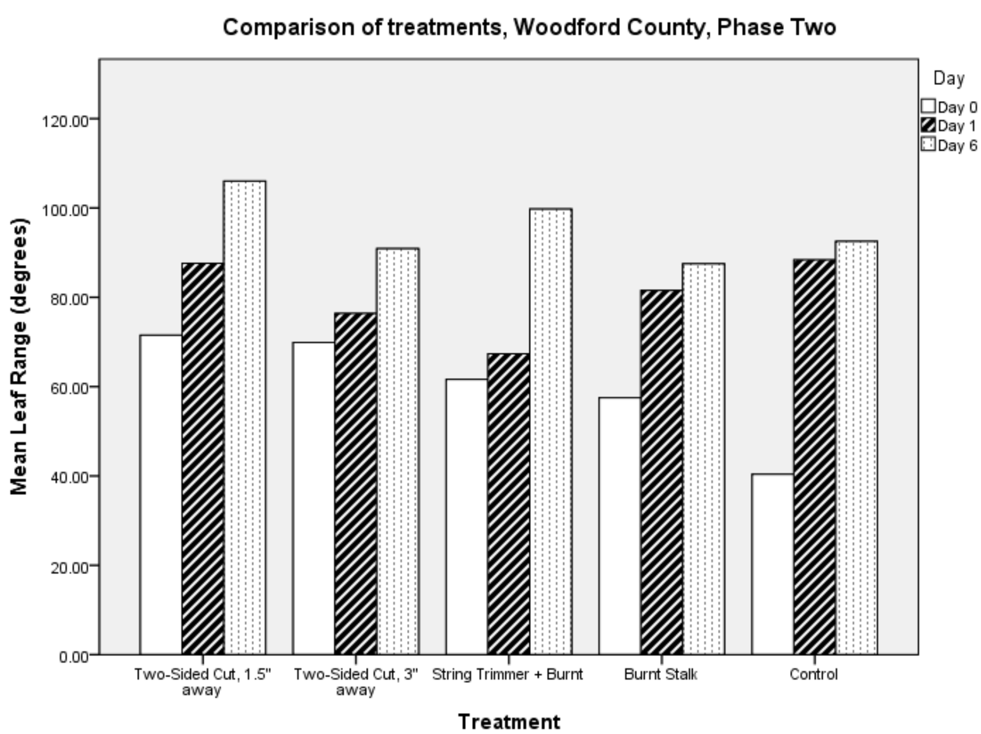
DATA: MEAN_Leaf_range=col(source(s), name("MEAN_Leaf_range"))

DATA: Day=col(source(s), name("Day"), unit.category())

COORD: rect(dim(1,2), cluster(3,0))

```
 GUIDE: axis(dim(3), label("Treatment"))
 GUIDE: axis(dim(2), label("Mean Leaf Range (degrees)"))
 GUIDE: legend(aesthetic(aesthetic.color.interior), label("Day"))
 SCALE: cat(dim(3), include("1", "2", "3", "4", "5"))
 SCALE: linear(dim(2), min(0))
 SCALE: cat(aesthetic(aesthetic.color.interior), include("1", "2", "3"))
 SCALE: cat(dim(1), include("1", "2", "3"))
 ELEMENT: interval(position(Day*MEAN_Leaf_range*TRT), color.interior(Day),
  shape.interior(shape.square))
END GPL.
```

GGraph



* Chart Builder.

GGRAPH

```
  /GRAPHDATASET NAME="graphdataset" VARIABLES=Day MEANCI(Leaf_range,
95)[name="MEAN_Leaf_range"

    LOW="MEAN_Leaf_range_LOW" HIGH="MEAN_Leaf_range_HIGH"] TRT MISSING=LISTWISE
REPORTMISSING=NO

  /GRAPHSPEC SOURCE=INLINE.

BEGIN GPL

  SOURCE: s=userSource(id("graphdataset"))

  DATA: Day=col(source(s), name("Day"), unit.category())

  DATA: MEAN_Leaf_range=col(source(s), name("MEAN_Leaf_range"))

  DATA: TRT=col(source(s), name("TRT"), unit.category())

  DATA: LOW=col(source(s), name("MEAN_Leaf_range_LOW"))

  DATA: HIGH=col(source(s), name("MEAN_Leaf_range_HIGH"))

  COORD: rect(dim(1,2), cluster(3,0))

  GUIDE: axis(dim(3), label("Day"))

  GUIDE: axis(dim(2), label("Mean Leaf Range (degrees)"))

  GUIDE: legend(aesthetic(aesthetic.color.interior), label("Treatment"))

  GUIDE: text.footnote(label("Error Bars: 95% CI"))

  SCALE: cat(dim(3), include("1", "2", "3"))

  SCALE: linear(dim(2), min(0))

  SCALE: cat(aesthetic(aesthetic.color.interior), include("1", "2", "3", "4", "5"))

  SCALE: cat(dim(1), include("1", "2", "3", "4", "5"))

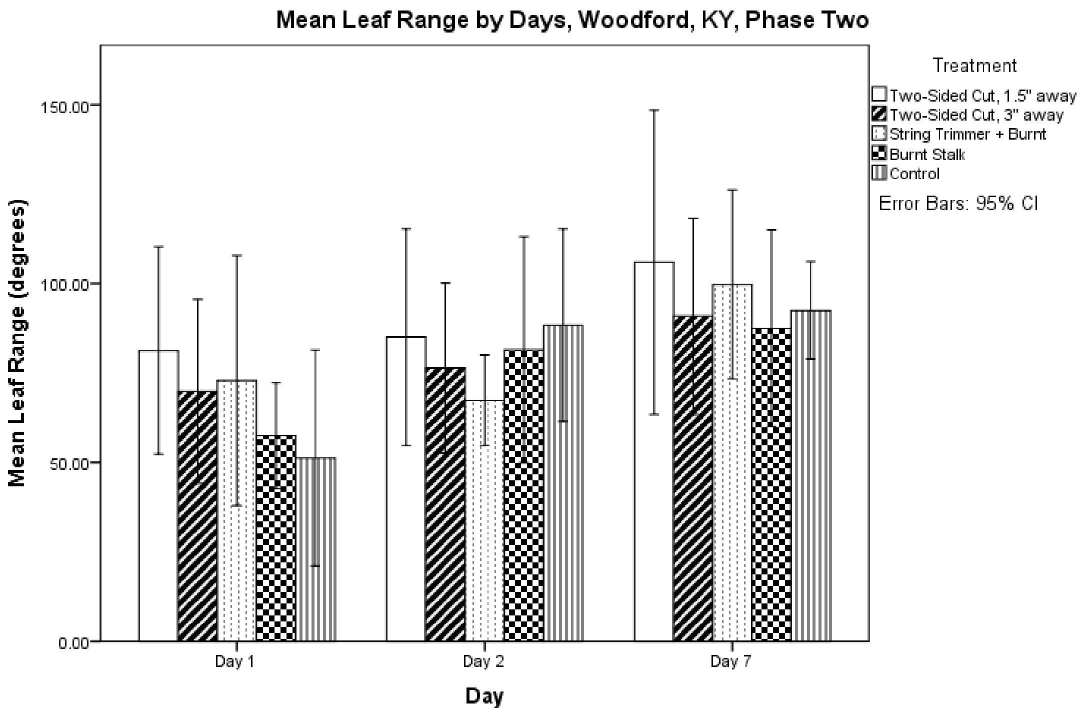
  ELEMENT: interval(position(TRT*MEAN_Leaf_range*Day), color.interior(TRT),

    shape.interior(shape.square))

  ELEMENT: interval(position(region.spread.range(TRT*(LOW+HIGH)*Day)),
shape.interior(shape.ibeam))

END GPL.
```

GGraph



Mean Leaf Range by Days, Woodford, KY, Phase Two
Mean Leaf Range (degrees)
150.00
100.00
50.00
0.00
Day 1
Day 2
Day 7
Day
Treatment
Two-Sided Cut, 1.5" away
Two-Sided Cut, 3" away
String Trimmer + Burnt
Burnt Stalk
Control
Error Bars: 95% CI

Appendix D - Phase 3 - Main Effects

```
* Chart Builder.

GGRAPH

  /GRAPHDATASET NAME="graphdataset" VARIABLES=leafAngle MC MISSING=LISTWISE
REPORTMISSING=NO

  /GRAPHSPEC SOURCE=INLINE.

BEGIN GPL

  SOURCE: s=userSource(id("graphdataset"))

  DATA: leafAngle=col(source(s), name("leafAngle"))

  DATA: MC=col(source(s), name("MC"))

  GUIDE: axis(dim(1), label("Leaf Range (degrees)"))

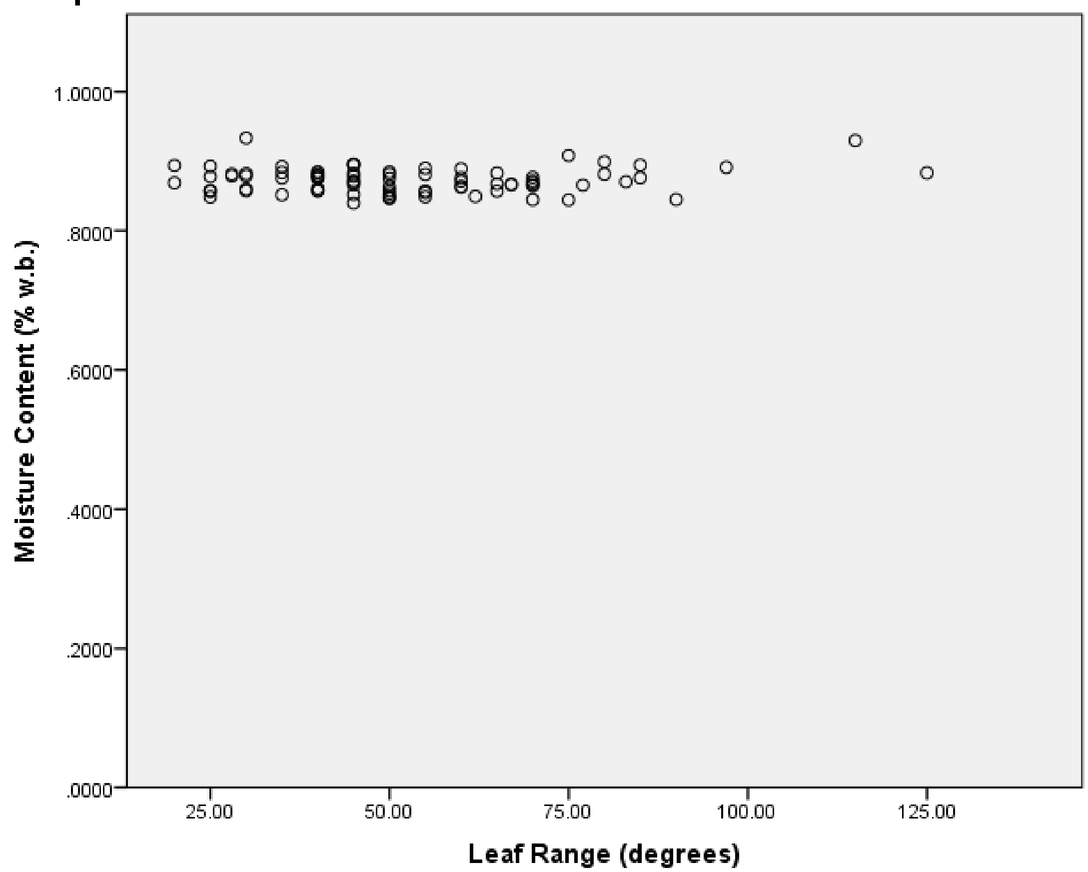
  GUIDE: axis(dim(2), label("Moisture Content (% w.b.)"))

  SCALE: linear(dim(2), min(0))

  ELEMENT: point(position(leafAngle*MC))

END GPL.
```

GGraph




```
EXAMINE VARIABLES=leafAngle MC cutDist
  /PLOT NPPLOT
  /STATISTICS NONE
  /CINTERVAL 95
  /MISSING LISTWISE
  /NOTOTAL.
```

Explore

Case Processing Summary

	Cases					
	Valid		Missing		Total	
	N	Percent	N	Percent	N	Percent
Leaf Range (degrees)	40	40.8%	58	59.2%	98	100.0%
Moisture Content (% w.b.)	40	40.8%	58	59.2%	98	100.0%
Cut Distance (in.)	40	40.8%	58	59.2%	98	100.0%

Tests of Normality

	Kolmogorov-Smirnov[a]			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
Leaf Range (degrees)	.118	40	.174	.944	40	.047
Moisture Content (% w.b.)	.095	40	.200*	.966	40	.264
Cut Distance (in.)	.143	40	.038	.952	40	.088

*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

```
CORRELATIONS
 /VARIABLES=leafAngle MC
 /PRINT=TWOTAIL NOSIG
 /MISSING=PAIRWISE.
```

Correlations

Correlations

		Leaf Range (degrees)	Moisture Content (% w.b.)
Leaf Range (degrees)	Pearson Correlation	1	.103
	Sig. (2-tailed)		.365
	N	80	80
Moisture Content (% w.b.)	Pearson Correlation	.103	1
	Sig. (2-tailed)	.365	
	N	80	80

```
UNIANOVA leafAngle BY TRT Day
  /METHOD=SSTYPE(3)
  /INTERCEPT=INCLUDE
  /SAVE=PRED RESID SRESID
  /PLOT=PROFILE(TRT*Day Day*TRT)
  /EMMEANS=TABLES(TRT*Day)
  /PRINT=ETASQ DESCRIPTIVE HOMOGENEITY
  /CRITERIA=ALPHA(.05)
  /DESIGN=TRT Day TRT*Day.
```

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Treatment	0	Root Pruned	40
	1	Control	40
Day	0	Day 0	40
	1	Day 1	40

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Deviation	N
Root Pruned	Day 0	57.2000	13.43836	20
	Day 1	65.6000	26.57937	20
	Total	61.4000	21.21900	40
Control	Day 0	49.2500	19.00658	20
	Day 1	39.1500	13.28741	20
	Total	44.2000	16.97540	40
Total	Day 0	53.2250	16.73854	40
	Day 1	52.3750	24.68955	40
	Total	52.8000	20.96253	80

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Leaf Range (degrees)

F	df1	df2	Sig.
3.906	3	76	.012

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + TRT + Day + TRT * Day

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	7642.500[a]	3	2547.500	7.152	.000	.220
Intercept	223027.200	1	223027.200	626.104	.000	.892
TRT	5916.800	1	5916.800	16.610	.000	.179
Day	14.450	1	14.450	.041	.841	.001
TRT * Day	1711.250	1	1711.250	4.804	.031	.059
Error	27072.300	76	356.214			
Total	257742.000	80				
Corrected Total	34714.800	79				

a. R Squared = .220 (Adjusted R Squared = .189)

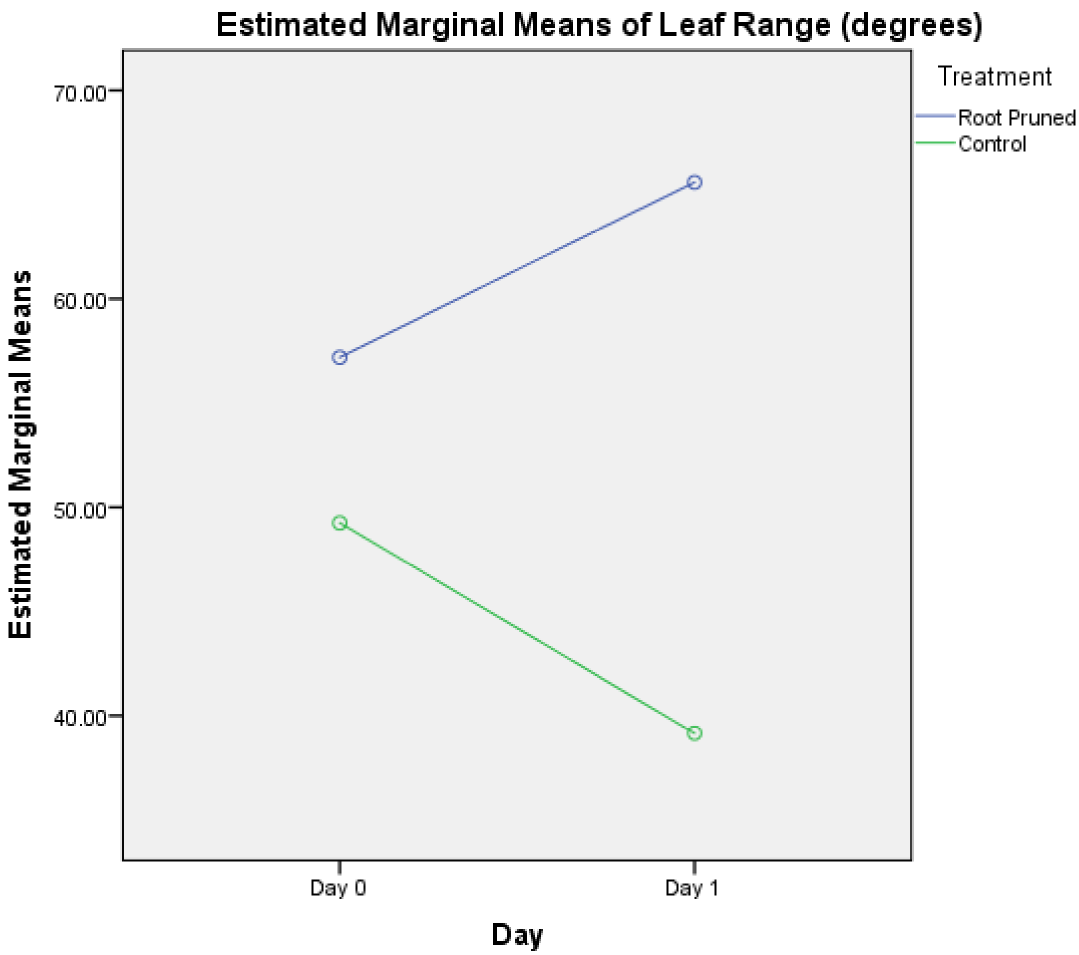
Estimated Marginal Means

Treatment * Day

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Root Pruned	Day 0	57.200	4.220	48.795	65.605
	Day 1	65.600	4.220	57.195	74.005
Control	Day 0	49.250	4.220	40.845	57.655
	Day 1	39.150	4.220	30.745	47.555

Profile Plots




```
SORT CASES  BY Day TRT.

SPLIT FILE LAYERED BY Day TRT.

EXAMINE VARIABLES=RES_1

  /PLOT BOXPLOT NPPLOT

  /COMPARE GROUPS

  /STATISTICS NONE

  /CINTERVAL 95

  /MISSING LISTWISE

  /NOTOTAL
```

Explore

Warnings

There are no valid cases in split file Day=., Treatment=.. Statistics cannot be computed.

Case Processing Summary

			Cases				
			Valid		Missing		Total
Day	Treatment		N	Percent	N	Percent	N
Day 0	Root Pruned	Residual for leafAngle	20	100.0%	0	0.0%	20
	Control	Residual for leafAngle	20	100.0%	0	0.0%	20
Day 1	Root Pruned	Residual for leafAngle	20	100.0%	0	0.0%	20
	Control	Residual for leafAngle	20	100.0%	0	0.0%	20

Case Processing Summary

			Cases
			Total
Day	Treatment		Percent
Day 0	Root Pruned	Residual for leafAngle	100.0%
	Control	Residual for leafAngle	100.0%
Day 1	Root Pruned	Residual for leafAngle	100.0%
	Control	Residual for leafAngle	100.0%

Tests of Normality

			Kolmogorov-Smirnov[a]			Shapiro-Wilk	
Day	Treatment		Statistic	df	Sig.	Statistic	df
Day 0	Root Pruned	Residual for leafAngle	.133	20	.200*	.949	20
	Control	Residual for leafAngle	.234	20	.005	.915	20
Day 1	Root Pruned	Residual for leafAngle	.105	20	.200*	.949	20
	Control	Residual for leafAngle	.154	20	.200*	.955	20

Tests of Normality

			Shapiro-Wilk[a]
Day	Treatment		Sig.
Day 0	Root Pruned	Residual for leafAngle	.359
	Control	Residual for leafAngle	.081
Day 1	Root Pruned	Residual for leafAngle	.347
	Control	Residual for leafAngle	.457

*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

Univariate Analysis of Variance

Between-Subjects Factors

		Value Label	N
Treatment	0	Root Pruned	40
	1	Control	40
Day	0	Day 0	40
	1	Day 1	40

Descriptive Statistics

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Deviation	N
Root Pruned	Day 0	57.2000	13.43836	20
	Day 1	65.6000	26.57937	20
	Total	61.4000	21.21900	40
Control	Day 0	49.2500	19.00658	20
	Day 1	39.1500	13.28741	20
	Total	44.2000	16.97540	40
Total	Day 0	53.2250	16.73854	40
	Day 1	52.3750	24.68955	40
	Total	52.8000	20.96253	80

Levene's Test of Equality of Error Variances[a]

Dependent Variable: Leaf Range (degrees)

F	df1	df2	Sig.
3.906	3	76	.012

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.[a]

a. Design: Intercept + TRT + Day + TRT * Day

Tests of Between-Subjects Effects

Dependent Variable: Leaf Range (degrees)

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Corrected Model	7642.500[a]	3	2547.500	7.152	.000	.220
Intercept	223027.200	1	223027.200	626.104	.000	.892
TRT	5916.800	1	5916.800	16.610	.000	.179
Day	14.450	1	14.450	.041	.841	.001
TRT * Day	1711.250	1	1711.250	4.804	.031	.059
Error	27072.300	76	356.214			
Total	257742.000	80				
Corrected Total	34714.800	79				

a. R Squared = .220 (Adjusted R Squared = .189)

Estimated Marginal Means

1. Treatment * Day

Estimates

Dependent Variable: Leaf Range (degrees)

				95% Confidence Interval	
Treatment	Day	Mean	Std. Error	Lower Bound	Upper Bound
Root Pruned	Day 0	57.200	4.220	48.795	65.605
	Day 1	65.600	4.220	57.195	74.005
Control	Day 0	49.250	4.220	40.845	57.655
	Day 1	39.150	4.220	30.745	47.555

Pairwise Comparisons

Dependent Variable: Leaf Range (degrees)

Day	(I) Treatment	(J) Treatment	Mean Difference (I-J)	Std. Error	Sig.[b]	95% Confidence Interval for Difference[b] Lower Bound
Day 0	Root Pruned	Control	7.950	5.968	.187	-3.937
	Control	Root Pruned	-7.950	5.968	.187	-19.837
Day 1	Root Pruned	Control	26.450*	5.968	.000	14.563
	Control	Root Pruned	-26.450*	5.968	.000	-38.337

Pairwise Comparisons

Dependent Variable: Leaf Range (degrees)

Day	(I) Treatment	(J) Treatment	95% Confidence Interval for Difference Upper Bound
Day 0	Root Pruned	Control	19.837
	Control	Root Pruned	3.937
Day 1	Root Pruned	Control	38.337
	Control	Root Pruned	-14.563

Based on estimated marginal means

*. The mean difference is significant at the .05 level.

b. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Day		Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared
Day 0	Contrast	632.025	1	632.025	1.774	.187	.023
	Error	27072.300	76	356.214			
Day 1	Contrast	6996.025	1	6996.025	19.640	.000	.205
	Error	27072.300	76	356.214			

Each F tests the simple effects of Treatment within each level combination of the other effects shown. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.

2. Treatment * Day

Estimates

Dependent Variable: Leaf Range (degrees)

Treatment	Day	Mean	Std. Error	95% Confidence Interval Lower Bound	95% Confidence Interval Upper Bound
Root Pruned	Day 0	57.200	4.220	48.795	65.605
	Day 1	65.600	4.220	57.195	74.005
Control	Day 0	49.250	4.220	40.845	57.655
	Day 1	39.150	4.220	30.745	47.555

Pairwise Comparisons

Dependent Variable: Leaf Range (degrees)

Treatment	(I) Day	(J) Day	Mean Difference (I-J)	Std. Error	Sig.[a]	95% Confidence Interval for Difference[a] Lower Bound
Root Pruned	Day 0	Day 1	-8.400	5.968	.163	-20.287
	Day 1	Day 0	8.400	5.968	.163	-3.487
Control	Day 0	Day 1	10.100	5.968	.095	-1.787
	Day 1	Day 0	-10.100	5.968	.095	-21.987

Pairwise Comparisons

Dependent Variable: Leaf Range (degrees)

Treatment	(I) Day	(J) Day	95% Confidence Interval for Difference Upper Bound
Root Pruned	Day 0	Day 1	3.487
	Day 1	Day 0	20.287
Control	Day 0	Day 1	21.987
	Day 1	Day 0	1.787

Based on estimated marginal means

a. Adjustment for multiple comparisons: Bonferroni.

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Treatment		Sum of Squares	df	Mean Square	F	Sig.
Root Pruned	Contrast	705.600	1	705.600	1.981	.163
	Error	27072.300	76	356.214		
Control	Contrast	1020.100	1	1020.100	2.864	.095
	Error	27072.300	76	356.214		

Univariate Tests

Dependent Variable: Leaf Range (degrees)

Treatment		Partial Eta Squared
Root Pruned	Contrast	.025
	Error	
Control	Contrast	.036
	Error	

Each F tests the simple effects of Day within each level combination of the other effects shown. These tests are based on the linearly independent pairwise comparisons among the estimated marginal means.

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Vita

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EDUCATION

University of Kentucky, College of Biosystems and Agricultural Engineering
MS in Engineering — Anticipated
2018
Thesis: Pre-wilting Burley Tobacco to Enhance Manual and Mechanical Harvesting and Housing

University of Kentucky, College of Biosystems and Agricultural Engineering
BS in Engineering — 2009

CERTIFICATIONS AND LICENSURE

Engineer in Training, State of Kentucky — 2010

TEACHING EXPERIENCE

University of Kentucky, College of Biosystems and Agricultural Engineering
Teaching Assistant - Special Problems in Engineering — 2011
Developed and graded homework, quizzes, and tests. Presented some material.

RESEARCH EXPERIENCE

University of Kentucky, College of Biosystems and Agricultural Engineering
Graduate Research Assistant — 2009 – 2011
Assisted in research, experimental design, data collection, construction of several probjects

PUBLICATIONS AND TECHNICAL PRESENTATIONS

"Evaluation of Methods for Pre-Wilting Burley Tobacco Plants in the Field"
American Society of Agricultural and Biological Engineers. 1110976, 2011 Aug. — 2011

"Evaluation of Methods for Pre-Wilting Burley Tobacco Plants in the Field"
Presented at the 2011 international American Society of Agricultural and Biological Engineers (ASABE) conference in Louisville, KY — *2011*

"Evaluation of Methods for Wilting Standing Burley Tobacco Plants in the Field"
Presented at the 2012 international Tobacco Workers' Conference (TWC) in Williamsburg, VA — 2012

PROFESSIONAL EXPERIENCE

Kimball Intl.
Product Engineer — 2014 – Current
Design new and special application products in both CAD and BOM environments