

ABSTRACT

O'CONNELL, SUZANNE. Grafted Tomato Performance in Organic Production Systems: Nutrient Uptake, Plant Growth, and Fruit Yield. (Under the direction of Mary M. Peet.)

There are many inherent challenges with growing tomatoes in the Southeast which can be intensified under organic production. Cultivating tomatoes under high tunnel systems may offer a number of benefits and opportunities such as season extension, higher fruit quality, less foliar disease pressure, and protection from extreme weather events. Grafted plants may be uniquely suited to production in organic systems and also high tunnel environments due to their higher stress tolerance, increased crop longevity, more efficient fertilizer use, and soil borne disease resistance. The combination of growing high-value grafted crops under high tunnel structures is an innovative systems approach that can offer new economic opportunities, greater production stability, higher fruit quantity and quality.

A baseline greenhouse study with conventional inputs was conducted in 2007, to evaluate the grafting effect on tomato plant growth and nutrient accumulation expressed in the leaf tissue. Grafting treatments included two scion-hybrid rootstock combinations *Solanum lycopersicum* L 'Trust' or 'German Johnson' grafted on *Solanum lycopersicum* L. x *Solanum habrochaites* S. Knapp & D.M. Spooner 'Maxifort', two self-grafted controls, and two non-grafted controls. Both shoot and root growth, were significantly higher in grafted treatments compared to non-grafted treatments. The leaf tissue nutrient concentrations were greater in grafted plants for: N, P, Ca, Mg, S, Fe, Mn, Zn, Cu, and B compared to non-grafted plants. Self-grafted controls had an intermediate values for selected plant growth and nutrient uptake compared to grafted and non-grafted treatments. Values were not different among scion cultivars.

In 2007 and 2008, a systems comparison study was conducted at The Center for Environmental Farming Systems in Goldsboro, North Carolina. An organic high tunnel system was compared to an organic field system. Three levels of N inputs were applied to each system. Grafting treatments included two heirloom scion-hybrid rootstock combinations: *Solanum lycopersicum* L. 'Cherokee Purple' grafted on *Solanum lycopersicum* L. x *Solanum habrochaites* 'Maxifort' or 'Beaufort', a self-grafted control (2008 only) and a non-grafted control. System type, N level, and grafting effects on nutrient uptake, plant growth, and fruit yield were evaluated.

The high tunnel system produced greater fruit yields for all treatments and hit peak production three weeks earlier compared to the field system. The high tunnel system had a higher incidence of blossom end rot, cat-facing, and cracking but lower incidence of TSWV and insect damage compared to the field system. Mean leaf tissue N concentrations were highly correlated with total harvest weight (>70.4%) in 2008. The N input level effect on yield was not consistent across the two seasons, however, in 2008 both the high and medium N input levels (168 kg ha⁻¹ and 122 kg ha⁻¹, respectively) produced greater total harvest yields compared to the low N level (93 kg ha⁻¹).

Grafted plants had higher mean leaf tissue concentrations for: N, P, K, Mn, Cu, Zn, and B but lower concentrations of Mg and Na compared to non-grafted plants. Grafted plants produced a greater fruit yield compared to non-grafted plants in a low disease pressure environment. Grafted plants in the high tunnel system also displayed greater plant growth compared to the non-grafted plants. Self-grafted plants (2008 only) were not different from non-grafted plants in terms of nutrient uptake, plant growth, or fruit yield. Both the 'Cherokee Purple-Maxifort' and 'Cherokee Purple-Beaufort' grafting treatments produced a

greater number of fruit in the high tunnel system compared to the field system. The greatest yield response was achieved in the high tunnel system with the ‘Cherokee Purple-Maxifort’ grafts.

Grafted Tomato Performance in Organic Production Systems:
Nutrient Uptake, Plant Growth, and Fruit Yield.

by
Suzanne O'Connell

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APPROVED BY:

Frank J. Louws, PhD.

Thomas Rufty, PhD.

Mary M. Peet, PhD.
Chair of Advisory Committee

DEDICATION

This thesis is dedicated to my loving family and friends who have supported me along the way. Steve deserves an extra thank you for living with both me and my project over the last two years! I'd also like to dedicate this work to the 2002 Appleton Farm crew and Mrs. Appleton, who together introduced me to the exciting world of farming.

BIOGRAPHY

Suzanne O'Connell will be graduating in 2008 with a Master of Science in Horticultural Science from North Carolina State University building on a Bachelor of Arts in Environmental and Political Science from Barnard College, Columbia University. In 2008, she was awarded first place at the American Society of Horticultural Science (ASHS) National Graduate Student Poster Competition as well as first place in the ASHS Southern Region Graduate Student Poster Competition in 2007 for entries based on work covered in the following thesis. Her love of food/cooking, being outdoors, and environmental science are the motivating factors for an ever growing interest in the field of sustainable agriculture.

Growing positions she has held include: Assistant Manager and Greenhouse Grower of annual and perennial ornamentals at Cavicchio Greenhouses, Inc. in Sudbury, MA and Farm Apprentice at Appleton Farms, an organic CSA operation in Ipswich, MA. Suzanne also served as a Conservation Agent for the Town of Rockport, MA from 2003-2005, which oversees the administration of and enforcement of all local and state wetlands regulations as well as local and regional conservation planning and protection efforts.

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CHAPTER 1 – INTRODUCTION

Tomato Origin and Popularity

Tomatoes are part of the Solanaceae or nightshade family. Domesticated tomatoes are related to the wild species, *Solanum lycopersicum* L. var. *cerasiforme* (Dunal) Spooner, G.J. Anderson & R.K. Jansen, which is found in the Andean region of South America. The wild taxa is a short-lived perennial plant that prefers well-drained soils and a dry climate (Rick, 1973). Tomatoes are believed to have been domesticated in Mesoamerica (Rick and Fobes, 1975). A ‘founders effect’ developed as the fruit moved away from its native region in South America. The lack of natural insect pollinators and solanaceous weed species in the new regions of cultivation resulted in populations with relatively low genetic variation. This effect has been amplified by cultivation and breeding efforts away from the species center of origin (Edwards, 1990; Rick, 1949, 1958; Rick and Fobes, 1975). Breeding for agricultural traits such as increased fruit uniformity, self-pollination, determinate growth and ripening, pest resistance, and production in high-input systems has further decreased the germplasm base for domesticated varieties (Edwards, 1990; Rick, 1973, 1978; Yeager, 1927).

Domesticated tomatoes are generally grown as an annual crop in temperate regions of the U.S. and have been bred for field or greenhouse production. Tomatoes are highly valued by consumers for their edible fruits. They are the 4th most popular fresh market vegetable in the U.S., trailing only potatoes, lettuce, and onions (Lucier and Plummer,

2004). A 30% increase in domestic consumption since 1985 has been linked to new and improved varieties, better product handling, increased year round availability, and health awareness (Calvin and Cook, 2005).

Tomatoes are the dominant source of carotenoids in the human diet (Dorais et al., 2008) and one medium, fresh tomato (about 5.2 oz) has 35 calories and provides 40% of the U.S. recommended daily allowance vitamin C and 20% of vitamin A (Calvin and Cook, 2005). The enduring popularity of salads and submarine sandwiches, along with a surge in new immigrants whose diets include high levels of fresh vegetables, are credited with sustaining the fresh market demand (Lucier and Plummer, 2004). The average U.S. citizen consumed 8.8 kg of fresh tomatoes in 2003 (Lucier and Plummer, 2004).

Florida and California dominate production in the U.S. but twenty states produce a commercial crop, including North Carolina. In 2007, North Carolina produced over \$24 million dollars worth of tomatoes (fresh and processing combined), accounting for 1.1% of the total U.S. production value (ERS, 2008). Tomatoes are the 5th most valuable horticultural crop in the state behind greenhouse/nursery, sweet potatoes, blueberries, and cucumbers (ERS, 2008). Greenhouse grown tomatoes represent a growing share of the market, capturing 17% of all fresh market tomatoes sold in the U.S. in 2005 (Calvin and Cook, 2005).

Organics

According to the Organic Trade Association (OTA), organic food sales in the U.S. have grown by 17%-21% compared to 2%-4% for the general market for each year since 1997. Organic food sales are predicted to rise another 18% each year until 2010 (OTA, 2004, 2008). Organic foods now represent 2% of the retail food market in the U.S., with a value estimated at \$20 billion dollars in U.S. sales (OTA, 2004, 2008). Between 1997 and 2002, the number of acres certified as organic nearly doubled in the U.S. (Dimitri and Greene, 2002). In 2005, the United States was 4th in the world in terms of certified production on 1.6 million hectares of land (OTA, 2008). The largest markets for organic products are located in the U.S., the European Union, and Japan (OTA, 2008).

A report published by the American Agricultural Economics Association (AAEA) in 2007 found that consumers of organic food are a diverse group, in terms of income and race (Stevens-Garmon et al., 2007). The fastest growing areas in the U.S. for organic markets are in the South and West (Stevens-Garmon et al., 2007). Tomatoes are the most popular organic vegetable purchased by American consumers, who are 3-4 times more likely to purchase an organic tomato than any other type of organic produce (Stevens-Garmon et al., 2007). The organic premium in 2004 for organic tomatoes was approximately 52% above its conventional counterpart (Stevens-Garmon et al., 2007).

Over 28,975 ha of land in the U.S., representing 1.6% of all vegetable production, was devoted to organic vegetable production in 2001 (Fernandez-Cornejo et al., 1998). The majority of USDA certified operations, especially in the Southeast and Northeast

regions, produce diversified crops of vegetables, fruits, herbs, and flowers (Fernandez-Cornejo et al., 1994). The majority of organic farms are small-scale operations, with 78% having less than 4.1 ha (Fernandez-Cornejo et al., 1994). Fresh market tomatoes are the number one crop produced by organic growers (21%) followed by sweet corn (18%), lettuce (11%), strawberries (8%), carrots (7%), onions (7%), and melons (6%) (Fernandez-Cornejo et al., 1994). Typically, prices for organic tomatoes are highest at the beginning of the season and drop as the summer field season progresses (Diver et al., 1999).

Organic farmers sell directly to consumers much more frequently than conventional farmers and retain a higher share of the consumer food dollar (Greene and Kremer, 2003; Walz, 1999; Fernandez-Cornejo et al., 1998). Farmers markets are a popular type of direct marketing that have dramatically increased (79%) between 1994 and 2002 (Kremen et al., 2004). The majority of organic growers sell their products to consumers through direct marketing channels (49%), followed by grocery wholesale/distribution (14%), grocery retailer (10%), and grower cooperative (9%) (Kremen et al., 2004). Organic farmers are disproportionately represented at farmers markets, many of which are located near major urban areas, universities, religious communities, or holistic health centers (Kremen et al., 2004). Highly valued items at farmers markets include specialty produce varieties including heirlooms (Lyson et al., 1995).

There is no consensus on the definition of an ‘heirloom’ variety. One dictionary defines heirloom varieties as, “a horticultural variety that has survived for several

generations usually due to the efforts of private individuals” (Merriam-Webster, 2008). The Southern Exposure Seed Exchange defines an heirloom variety as both open-pollinated and existing before the 1940’s (Rakita, 2008). In ‘Taylors Guide to Heirloom Vegetables’ three criteria are set forward for heirloom varieties: 1) open-pollination, 2) more than 50 years old, and 3) the tomato variety has a history or folklore of its own (Watson, 1996). Heirloom varieties are typically very susceptible to foliar and soil borne diseases making them a challenging crop to produce (Rivard, 2006).

High Tunnel Production

High tunnels are essentially non-heated, plastic covered greenhouses often with roll up sides to allow passive ventilation. High tunnels are tall enough that crops can be managed from within the structure as opposed to low tunnels which do not allow easy access (<1 m tall) (Wittwer and Castilla, 1995). The planting of high-value crops under high tunnels became popular in Asia and the Mediterranean in the 1950’s and Spain, Italy and China lead the way employing this type of production system (Wittwer and Castilla, 1995). Thirteen percent of the world’s tomato production is grown under some sort of protected cultivation (Dorais et al., 2001).

Growers in the U.S. are becoming more and more interested in utilizing high tunnels in their production systems. Climate change models predict that the Southeast region will become wetter and cooler, experience more extreme weather events, and have increased levels of pests and diseases in the near future (Reilly, 2002). Protected

agriculture offers a measure to help offset the predicted decrease in crop yields and productivity in the Southeast, by increasing the grower's control over environmental factors. Some of the associated benefits may include: protection from cold temperatures, reduction of wind damage, a decrease in foliar disease pressure, expansion of suitable production areas, extension of the growing seasons, increased crop yields, improved fruit crop quality, and achieving a more stable market (Wittwer and Castilla, 1995).

Nutrient Uptake and Yield

Mineral nutrients support a variety of structural formations and metabolic functions in the plant such as protein synthesis, photophosphorylation, CO₂ fixation, stomatal movement, starch synthesis, and sugar transport (Marschner, 1986). Total N accumulation from the soil by field crops is often the limiting yield factor (Sinclair et al., 2004). Supplying nutrients, especially N (N) to a tomato crop, commonly grown under plastic mulch and with drip irrigation, is a challenging task in eastern North Carolina. Both conventional and organic soluble fertilizers are limited and expensive and therefore most growers rely primarily on pre-plant fertilizer inputs. Ultisol soils, common in the Southeast U.S. have intensely weathered soil structure and are typically low in nutrients. In addition long, growing seasons support active microbial communities that breakdown organic matter quickly, compounding nutrient deficiencies that limit the health and productivity of the crop as the season progresses.

Complex interactions between roots and shoots and between roots and environmental factors (nutrient availability, soil characteristics, temperature, etc.) influence root growth and development (Jackson and Bloom, 1990; Marschner, 1986). Specific feedback mechanisms are employed by plants to regulate internal concentrations of nutrients over a wide range of external concentrations (Marschner, 1986). In addition, differences in root growth and morphology can be triggered by responses to internal concentrations of mineral nutrients (Marschner, 1986).

During vegetative growth, a large proportion of photosynthates are used for root growth and metabolism. It is thought that "...a larger root system and a high rate of root replacement, ensuring a high proportion of young roots, are advantageous for water and mineral nutrient uptake, particularly in soils with low fertility" (Marschner, 1986). Roots give off exudates including sugars, organic acids, amino acids, and phenolic compounds. These exudates can affect nutrient uptake by increasing the solubility of selected minerals in the soil, protecting roots from drying out, and attracting beneficial rhizosphere microorganisms (Marschner, 1986). Mycorrhizal associations may also affect nutrient uptake, most often by enhancing phosphorus and micronutrient uptake.

Total N accumulation from the soil by field crops is often the yield limiting factor (Sinclair, 2004). Up to 75% of a plant's N content is utilized as building blocks for chloroplasts, which regulate photosynthesis (Marschner, 1986). N compounds are critical for amino acid, peptide, amide, ureide, and amine formation. A deficiency in N can result in low photosynthetic efficiency, slowing of plant growth, senescence of older leaves,

enhanced flower drop, and pre-mature ripening of fruit (Marschner, 1986). Often N stress in tomatoes is expressed as fewer leaves that are both smaller and thicker (Scholberg et al., 2000).

The root system of the domestic tomato is described as being wide-reaching, highly-branched, deep (2 to 3 m), and with a fairly uniform distribution (Weaver and Bruner, 1927). Tomato plants have been shown to utilize only a small amount of the inorganic N, 3% of the available soil pool, in an un-fertilized, soil-based field system (Jackson and Bloom, 1990). The low rooting density of tomato plants combined with their deep and uniform root structure resulted in a low inorganic N recovery from the soil (Jackson and Bloom, 1990).

Breeding for cultivars with lower reliance on synthetic fertilizers and greater stress tolerances could lead to decreased input requirements for growers and a more sustainable farming system (Edwards, 1990). A different approach toward the same goal is the use of herbaceous grafting for increased nutrient uptake efficiency (Leonardi and Giuffrida, 2006; Ruiz, 1996, 2006). If grafted rootstocks are able to take up and utilize higher amounts of N and other nutrients compared to non-grafted plants, then perhaps greater plant growth and/or yields can be achieved with the same or less fertilizer inputs.

Herbaceous Grafting

Grafting was first used commercially during the 1920's in Asia to manage *Fusarium oxysporum* by grafting watermelons onto bottle gourd rootstock (Tateishi, 1927). Since then its application has expanded to include many cucurbit and solanaceous

crops and has been adopted rapidly outside of Asia during the last two decades (Oda, 2002). Either inter-generic or intra-specific pairings of rootstock and scions are employed in herbaceous grafting; solanaceous crops are generally intra-specific selections. Around the world, tomatoes, *Solanum lycopersicum* have been grafted onto *Solanum lycopersicum*, *Solanum pimpinellifolium*, *Solanum habrochaites*, *Solanum torvum*, and *Solanum aethiopicum*.

The reasons for grafting have expanded from increased soilborne disease resistance (*Fusarium oxysporum*, *Meloidogyne* spp., *Monosporascus cannonbolus*, *Ralstonia solanacearum*, TMV, *Pyrenochaeta lycopersici* and *Verticillium* sp.) to include: greater crop yields, higher salinity tolerance, increased heat and cold tolerance, and enhanced drought and flood resistance (Besri, 2005; Black, 2003; Burleigh et al., 2005; Colla et al., 2006; Edelstein et al., 1999; Estan et al., 2005; Jifon et al., 2008; Lee, 1994; Leonardi and Giuffrida, 2006; Rivero, 2003; Romero et al., 1997; Pulgar et al., 2000; Yetisir et al., 2006). Most research has been conducted in Eastern Asia (Korea, China, Japan) and the Mediterranean region (Greece, Spain, Italy, Israel, Morocco) where grafting is widely employed. Many of the mechanisms for disease resistance, appears to be a result of the ability of rootstocks to limit movement from the growing media to the scion (Grimault and Prior, 1994). Improved salt tolerance appears to be a result of less sodium and chloride accumulation in the leaf tissue (Estan et al., 2005). In addition, the use of herbaceous grafting has been suggested to increase nutrient uptake efficiency (Leonardi and Giuffrida, 2006; Ruiz et al., 1996, 2006) but these effects are not well

documented. Research conducted on changes in fruit quality of grafted plants is inconsistent, however grafting has been shown in some instances to increase lycopene, β -carotene, vitamin C, and antioxidant activity with some of these effects being cultivar dependent (Davis et al., 2008; Dorais et al., 2008; Fernandez-Garcia et al., 2004).

A common method of grafting employed for solanaceous crops is the 'Japanese top-grafting' or 'tube-grafting' method. The grafts are made when the seedlings are very small with two pairs of true leaves and stem diameters approximately 2.0 mm round. The advantage of grafting at this stage is the efficient use of greenhouse space and minimal input costs associated with seedling production.

A successful graft union requires the formation of new connections between vascular strands at the callus graft interface via differentiation and lignification (Fernandez-Garcia et al., 2004). The graft is considered functional between 4-8 days after grafting as vascular strands are forming and fully functional after 15 days when several connections are complete. Failure of a graft union to successfully develop may be due to: a lack of cellular recognition, the growing stage of the respective plants, interference of the wounding response or growth regulators, incompatibility toxins, or an unfavorable grafting environment (Andrews and Marques, 1993; Davis et al., 2008).

In the U.S., the use of grafted tomatoes has been adopted primarily by large-scale hydroponic greenhouse growers (Kubota et al., 2008). Over 90% of grafted transplants in North America are being grown in greenhouse operations and over 40 million grafted plants were cultivated in U.S. systems between 2002 and 2006 (Kubota et al., 2008).

Currently commercial production of grafted seedlings is limited to Canadian operations and small-scale farmers who produce their own transplants. The price of grafted tomato seedlings is estimated at \$0.60-\$0.90 compared to \$0.03-\$0.40 for non-grafted tomato seedlings (Kubota et al., 2008). Some of the limitations to grafted seedling production in the U.S. include: cost of skilled labor, production of uniform seedlings and controlled transportation conditions (Kubota et al., 2008).

Review of the Literature

A handful of studies have evaluated the effects of grafting on plant nutrient uptake with linkages to plant growth and yield. A Spanish study conducted with *Cucumis melo* L. grafted onto *Cucurbita maxima* rootstock concluded that increases of N (N), sodium (Na), and potassium (K) were influenced by rootstock genotype, whereas phosphorus (P) was affected by scion variety (Ruiz et al., 1997). A positive correlation was found between increased N concentration and yield and a negative correlation between increased Na concentration and yield. The study concluded that the root genotype affects yield but causes little difference in macronutrient content for grafted melon. The authors suggest that the scion or the scion-rootstock combination is responsible for varied nutrient content (N, Na, K, and P) in the plant shoot (Ruiz et al., 1997).

An Italian research team evaluated the variation of plant growth and macronutrient uptake in grafted tomatoes and eggplants paired with three different rootstocks (Leonardi and Giuffrida, 2006). The tomato cultivar, 'Rita' and the eggplant cultivar, 'Mission Bell'

were grafted onto two intraspecific rootstocks, 'PG3' and 'Energy', as well as the interspecific rootstock 'Beaufort'. The scion-rootstock combination of 'Rita' tomato and 'Beaufort' were the tallest plants with the greatest shoot weight, and the highest yields per plant. The scion-rootstock combination of 'Mission Bell' eggplant grafted on 'Beaufort' resulted in an opposite manner with shorter plants and lower yields. Variability among nutrient concentrations ranged from an additional 100-300% for the grafting combinations (Leonardi and Giuffrida, 2006).

Boron toxicity is a problem associated with the use of recycled municipal sewage water in Israel for crop production (Edelstein et al., 2008). *Cucumis melo* L. 'Arava' grafted onto *Cucurbita rootstock* 'TZ-148' were found to accumulate less boron in their plant tissue (mainly leaf tissue) compared to non-grafted melons. Thus grafted melons were deemed to have a higher tolerance to boron (Edelstein et al., 2008).

A greenhouse study focusing on grafted pepper production in Italy was carried out with two scion varieties, *Capsicum annuum* L., 'Edo' or *Capsicum annuum* L. 'Lux', grafted onto five different commercial rootstocks. The study concluded that total yield, marketable yield, and fruit number were influenced by the rootstock type. Grafted treatments produced ~22-46% more marketable fruit compared to the non-grafted controls. Grafted plants were also approximately 28% taller than their non-grafted counterparts. No differences were detected in fruit quality or nutritional content of the peppers (Colla et al., 2006).

In 1988, researchers at the University of California investigated net N uptake for *Solanum lycopersicum* L. compared to two ascensions (lowland and highland types) of *Solanum habrochaites*, S. Knapp & D.M. Spooner in a hydroponic greenhouse study. It was determined that the kinetics of net NH_4^+ uptake was different among these taxa and that the net uptake capacity was greater for NO_3^- than NH_4^+ uptake in all three taxa (Smart, 1988). However, the proportion of NO_3^- to NH_4^+ absorption was much greater for the wild taxa suggesting that NO_3^- may be a more important source of mineral N for *Solanum habrochaites*, S. Knapp & D.M. Spooner (Smart, 1988).

Leaf Tissue Nutrient Analysis

By monitoring the leaf tissue nutrient concentration on a regular basis, one can make informed decisions about the nutritional management of a crop. The status of essential and beneficial nutrients being taken up by a plant are reflected by their leaf tissue concentration (concentration = amount of nutrient per unit mass of tissue) and the net change in nutrient influx and efflux in the leaf tissue (Marschner, 1986). Management decisions are driven by the desire to maintain nutrient levels above critical concentrations, especially those that are crop limiting, and to correct any deficiencies or toxicities that may exist. Consideration of the growing medium characteristics and surrounding environmental factors are essential in evaluating any actions that would affect the nutritional status of the target crop.

The ‘most recently mature leaf’ (MRML) is the first, fully expanded leaf below the growing point (typically the 4th or 5th leaf down from the growing point). The North Carolina Division of Agricultural and Consumer Science (NCDA&CS) recommends sampling the MRML for nutrient analysis of tomato plants. These leaves represent the most photosynthetically active leaves and typically have the highest nitrate levels.

Adequate ranges for leaf tissue nutrient concentrations change over time as the plant develops, often with a remobilization of nutrients as the plant moves from vegetative to the reproductive stages; this effect is more pronounced with highly mobile nutrients (Marschner, 1986). Generally speaking, the optimal N content for plant growth is between 2%-5% of the plant dry weight; however, these requirements are specific to plant species, development stage, and organ (Marschner, 1986). ‘Plant Tissue Analysis and Interpretation for Vegetable Crops in Florida’ is widely regarded as an industry standard for tomato crops (Hochmuth et al., 1991) (Table 1.1). These standards were used as a basis for comparison for adequate ranges of leaf tissue nutrient concentrations for optimum crop performance throughout this series of experiments.

Summary

Fresh market tomatoes are an important and popular crop in the U.S. with an increasing demand for organically produced crops (ERS, 2008; Lucier and Plummer, 2004; OTA 2004, 2008). Small-scale, organic growers rely on tomatoes for a large amount of their direct market sales (Fernandez-Cornejo et al., 1994; Kremen et al., 2004).

Consumers are willing to pay a price premium for organic tomatoes, heirloom varieties, and tomatoes offered outside the typical regional growing season (Steven-Garmon et al., 2007; Lyson et al., 1995).

There are many inherent challenges with growing tomatoes in the Southeast which can be intensified with organic production methods. Cultivating tomatoes under high tunnel systems may offer a number of benefits and opportunities such as: season extension, higher fruit quality, less foliar disease pressure, and protection from extreme weather events. Yet growing in high tunnels comes with its own challenges, such as a decreased ability to rotate crops, accumulation of fertilizer salts overtime, and increased potential for heat stress during warm weather. Grafted plants may be uniquely suited to production in a high tunnel environment due to their higher stress tolerances, increased crop longevity, more efficient fertilizer use, and soil borne disease resistance (Besri, 2005; Black, 2003; Burleigh et al., 2005; Colla et al., 2006, 2008; Edelstein et al., 1999, 2005, 2008; Estan et al., 2005; Jifon et al., 2008; Lee, 1994; Leonardi and Giuffrida, 2006; Pulgar et al., 2000; Rivero, 2003; Romero et al., 1997; Yetisir et al., 2006, 2007). The combination of growing high-value grafted crops such as organic heirloom tomatoes under high tunnel structures is an innovative systems approach that can provide growers with new economic opportunities, greater production stability, higher fruit quality, and lower pest management and fertilizer inputs.

Table 1.1 Adequate Ranges and Toxicity Values for Nutrient Content of the Most Recently Mature Whole Leaf, including Petiole, in Tomato^z.

Time of sampling	Status	%				ppm							
		N	P	K	Ca	Mg	S	Fe	Mn	Zn	B	Cu	Mo
5-leaf stage	Adequate range	3.0	0.3	3.0	1.0	0.30	0.3	40	30	25	20	5	0.2
		5.0	0.6	5.0	2.0	0.50	0.8	100	100	40	40	15	0.6
First flower	Adequate range	2.8	0.2	2.5	1.0	0.30	0.3	40	30	25	20	5	0.2
		4.0	0.4	4.0	2.0	0.50	0.8	100	100	40	40	15	0.6
Early fruit set	toxic (>)								1500	300	250		
	Adequate range	2.5	0.2	2.5	1.0	0.25	0.3	40	30	20	20	5	0.2
		4.0	0.4	4.0	2.0	0.50	0.6	100	100	40	40	10	0.6
First ripe fruit	toxic (>)										250		
	Adequate range	2.0	0.2	2.0	1.0	0.25	0.3	40	30	20	20	5	0.2
		3.5	0.4	4.0	2.0	0.50	0.6	100	100	40	40	10	0.6
During harvest period	Adequate range	2.0	0.2	1.5	1.0	0.25	0.3	40	30	20	20	5	0.2
		3.0	0.4	2.5	2.0	0.50	0.6	100	100	40	40	10	0.6

^z Adapted from Hochmuth et al., 1991.

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CHAPTER 2 –NUTRIENT UPTAKE AND PLANT GROWTH IN A CONTROLLED ENVIRONMENT

Abstract

Grafted herbaceous plants may increase plant growth and nutrient uptake efficiency. A greenhouse study evaluating the tomato cultivars ‘Trust’ (*Solanum lycopersicum* L. ‘Trust’) and ‘German Johnson’ (*Solanum lycopersicum* L. ‘German Johnson’) grafted on ‘Maxifort’ rootstock (*Solanum lycopersicum* L. x *Solanum habrochaites* S. Knapp & D.M. Spooner.), was conducted in 2007 at North Carolina State University to examine the potential effects conferred by grafting and evaluate two scion-rootstock combinations.

The experiment was a 2 x 3 factorial with a completely randomized block design. Six treatments were evaluated: 1) ‘Maxifort-Trust’ grafts, 2) self-grafted ‘Trust’, 3) non-grafted ‘Trust’, 4) ‘Maxifort-German Johnson’ grafts, 5) self-grafted ‘German Johnson’, and 6) non-grafted ‘German Johnson’. Five successive weekly harvests were conducted, representing the 5-9 weeks after grafting. Plant growth parameters (leaf weight, stem weight, leaf area, plant height, and root weight) were measured and leaf tissue nutrient content calculated.

Both shoot and root growth, were greater in grafted treatments compared to non-grafted treatments. The rootstock ‘Maxifort’ further boosted shoot growth and leaf area while the scion selection influenced the shoot weight and root weight to differing degrees.

The leaf tissue nutrient content for grafted plants was greater for N, P, Ca, Mg, S, Fe, Mn, Zn, Cu, and B compared to non-grafted plants. Selected nutrient uptake (Ca, Fe, Mn, Zn, and Cu) was greater for scion grafted on ‘Maxifort’ rootstock compared to self-grafted treatments. No interactions were present between grafting effects and scion selections.

Our results suggest that it is both the scion-rootstock pairing and the physical act of grafting itself which results in enhanced plant growth and leaf tissue nutrient accumulation. The overlapping effects from to grafting and cultivar selections can provide a wide range of plant responses for a variety of potentially beneficial applications.

Introduction & Objectives

Grafting herbaceous plants for commercial fruit and vegetable production can be traced back to the 1920’s in Asia (Sato and Takamatsu, 1930; Tateishi, 1927). Originally, grafting was used as a cultural technique to manage *Fusarium oxysporum* in watermelon crops. The application has since been expanded to many cucurbitaceous and solanaceous crops (Oda, 2002). Reasons for grafting include increased soilborne disease resistance (*Fusarium oxysporum*, *Meloidogyne spp.*, *Monosporascus cannonbolus*, *Ralstonia solanacearum*, TMV, *Pyrenochaeta lycopersici* and *Verticillium albo-atrum*,) as well as: greater crop yields, higher salinity tolerance, increased heat and cold tolerance, and enhanced drought and flood resistance (Besri, 2005; Black, 2003; Burleigh et al., 2005; Colla et al., 2006, 2008; Edelstein et al., 1999, 2005, 2008; Estan et al., 2005; Jifon et al.,

2008; Kato and Lou, 1989; Lee, 1994; Leonardi and Giuffrida, 2006; Matsuzoe et al., 1993; Pulgar et al., 2000; Rivero, 2003; Romero et al., 1997; Siguenza et al., 2005; Yetisir et al., 2006, 2007). In addition, the use of herbaceous grafting has been suggested to increase nutrient uptake efficiency (Leonardi and Giuffrida, 2006; Ruiz et al., 1996, 1997, 1999, 2006), but these effects are not well documented. Most research has been conducted in Eastern Asia (Korea, China, and Japan) as well as the Mediterranean region (Greece, Spain, Italy, Israel, and Morocco) where grafting is widely employed commercially and has focused on cucurbit crops. The objectives of this study were to evaluate the effects of grafting and scion-rootstock combinations on tomato plant growth and nutrient uptake, with an emphasis on nitrogen (N) accumulation in the leaf tissue.

Nitrogen is widely considered the most limiting nutrient for plant growth and productivity (Epstein and Bloom, 2005). Nitrogen is a major contributor to proteins, nucleic acids and other important metabolic compounds critical for the formation and function of chloroplasts (Marschner, 1986). Furthermore N is a mediating factor in maintaining the phyto-hormone balance within a plant that directs shoot and root growth (Marschner, 1986). In addition to the key metabolic roles of N in a plant, the amount of energy put into the manufacturing and application of N fixation is more than half of the total energy consumed in agriculture (Brown et al., 1987). The average high-yielding tomato crop grown in the Southeast U.S. requires 180-200 kg N ha⁻¹ (Maynard and Hochmuth, 1995). Therefore efforts to cultivate crops with lower N inputs and to increase N use efficiency of plants are at the forefront of agricultural research and breeding efforts.

If grafted plants are more efficient with their nutrient uptake perhaps they would be more efficient with their N utilization as well?

Explanations for why rootstocks may influence nutrient uptake include both physiological and biochemical factors. Variability among root vigor may result in a larger total root system that can extend deeper and wider, therefore accessing additional nutrient and water sources (Castle and Krezdorn, 1975) or the nutrient transport mechanisms associated with particular rootstocks may be more efficient or vary in their affinities for selected nutrients (Glass, 2003; Leonardi and Giuffrida, 2006; Ruiz et al., 1996). Differential hormone synthesis (cytokinins, abscisic acid, ethylene, gibberellins, auxins) controlled by root systems could lead to variations in growth and root to shoot ratios (Itai and Birnbaum, 1991; Zijlstra et al., 1994). Lastly, interactions between root exudates and non-infecting rhizosphere micro-organisms may be different among grafted and non-grafted plants resulting in increased or decreased nutrient availability (Bowen and Rovira, 1991).

A handful of studies have evaluated the effects of grafting on nutrient uptake and plant growth. One study evaluating grafted melons found that there were differences in leaf tissue nutrient concentration when *Cucumis melo* L. was grafted onto *Cucurbita maxima* rootstock. Concentrations of nitrogen (N) and sodium (Na) were both higher and lower depending on the scion-rootstock combination, when compared to non-grafted plants (Ruiz et al., 1997). The potassium (K) concentration was consistently lower in the grafted plants and the phosphorus (P) concentration consistently higher in grafted plants

compared to the non-grafts. By comparing the rising or declining nutrient concentration values of the grafted treatments to the non-grafts it was determined that the rootstock genotype was the most influential factor for differences in N, Na, and K expressed in the leaf tissue. A positive correlation was found between N concentration and fruit yield while a negative correlation was evident between Na concentration and fruit yield. Fruit yields were greater for all grafted plants compared to non-grafted plants. Yields were similar for across each rootstock type and were also affected by a scion-rootstock interaction. The authors suggest selecting a desired scion cultivar first, and then evaluating the scion-rootstock interactions in order to maximize yield potential (Ruiz et al., 1997).

Boron (B) toxicity is a problem associated with the use of recycled municipal sewage water in Israel for crop production (Edelstein et al., 2005, 2008). *Cucumis melo* L. ‘Arava’ grafted onto *Cucurbita rootstock* ‘TZ-148’ was found to have lower concentrations of B in the leaf tissue compared to non-grafted melons when exposed to toxic levels B over time. Melons grafted onto squash rootstocks were determined to exclude greater levels of B from the leaf tissue compared to non-grafted plants (Edelstein et al., 2005, 2008).

An Italian research team evaluated the variation of plant growth and macronutrient uptake in grafted solanaceous crops, tomatoes and eggplants, paired with three different rootstocks (Leonardi and Guiffrida, 2006). The tomato cultivar, ‘Rita’ and the eggplant cultivar, ‘Mission Bell’ were grafted onto two intraspecific rootstocks, ‘PG3’ and

‘Energy’, as well as the interspecific rootstock ‘Beaufort’. The scion-rootstock combination of ‘Rita’ tomato and ‘Beaufort’ were the tallest plants with the greatest shoot weight, and the highest yields per plant. The scion-rootstock combination of ‘Mission Bell’ eggplant grafted on ‘Beaufort’ resulted in shorter plants and lower yields. Macronutrient concentrations in the leaf tissue were greater for ‘Rita’ tomato grafted on ‘Beaufort’ hybrid rootstock compared to self-grafted ‘Rita’ for N, K, and Ca (Leonardi and Guiffrida, 2006).

A greenhouse study focusing on grafted pepper production was carried out with two scion varieties, *Capsicum annuum* L., ‘Edo’ or *Capsicum annuum* L. ‘Lux’, grafted onto five different commercial rootstocks (Colla et al., 2008).. The total fruit yield, both marketable weight and fruit number were influenced by the rootstock genotype used in the grafted scion-rootstock combinations. Grafted treatments produced approximately 22-46% more marketable fruit compared to the non-grafted controls. Grafted plants were also approximately 28% taller than their non-grafted counterparts. No differences were detected in fruit quality or nutritional content of the peppers (Colla et al., 2008).

Materials & Methods

In the spring of 2007, a greenhouse experiment was conducted at North Carolina State University’s (NCSU) Southeastern Plant Environment Laboratory. Three tomato cultivars were utilized, ‘Trust’ (*Solanum lycopersicum* L. ‘Trust’), ‘German Johnson’ (*Solanum lycopersicum* L. ‘German Johnson’), and ‘Maxifort’ (*Solanum lycopersicum* L.

x Solanum habrochaites S. Knapp & D.M. Spooner., ‘Maxifort’) (Knapp, 1999). ‘Trust’ is a popular indeterminate hybrid with beefsteak-type fruits that is grown extensively in greenhouses. ‘German Johnson’ is an indeterminate heirloom variety with large pink fruits that is very popular in the Southeastern, U.S. *Solanum lycopersicum* L. is a distant relative of *Solanum lycopersicum* var. *cerasiforme* (Dun.) A. Gray. ‘Maxifort’ is a hybrid rootstock produced by DeRuiter Seeds, a cross between the common cultivated tomato ((*Solanum lycopersicum* L.) and a wild-type, short-lived, perennial plant from the western regions of Ecuador and Peru (*Solanum habrochaites* S. Knapp & D.M. Spooner) (F. Knol, personal communication; Rick et al., 1979). Six treatments were included in this study: 1) ‘Trust’ grafted onto ‘Maxifort’, 2) ‘Trust’ grafted onto ‘Trust’ (self-grafted), 3) non-grafted ‘Trust’, 4) ‘German Johnson’ grafted onto ‘Maxifort’, 5) ‘German Johnson’ grafted onto ‘German Johnson’ (self-grafted), and 6) non-grafted ‘German Johnson’.

Seedling Production

‘German Johnson’ seeds were sown on 22 Jan. 2007, while ‘Trust’ and ‘Maxifort’ varieties were sown on the following day. Flats were filled with a commercial potting media (Redi Earth, ‘Plug and Seedling Mix’, Sun Gro Horticulture: Bellevue, WA) consisting of: 55-65% (by volume) Canadian peat, 35-45% medium grade vermiculite (by volume), 1.0 - 1.5% dolomite lime (by weight), 0.5 – 1.0% starter nutrient charge, plus gypsum (by weight), and 0.05 – 0.10% wetting agent (by weight). All flats were topped with a thin layer of horticultural grade vermiculite (Larrea, 2005). Flats were then placed

in a germination chamber with an underlain seedling heat mat (26-28°C), a 12 h light/12 h dark photoperiod (fluorescent lighting with a Photosynthetic Photon Flux Density (PPFD) of $\sim 80 \mu\text{mol m}^{-2}\text{s}^{-1}$ during light hours), and fitted with a series of overhead fine mist water nozzles (output every 3 minutes for 3 seconds during light hours only) (Shurtleff, J., personal communication, 12 Sept. 2008). When approximately 75% of the seeds had germinated (between 3-7 days), each flat was transported to a controlled environment growth chamber.

The controlled environment growth chamber measured 9 m^2 (NCSU & NC-ARS, 2008). Environmental conditions of the growth chamber were maintained at day/night temperatures of 26/22°C and coordinated with a photoperiod of 12 h light/12 h dark. A combination of T-12, 1500 ma, cool-white fluorescent and 100 W incandescent lamps provided light in the chamber. A 'plexiglas' barrier separated the lamps from the rest of the growing area. The resulting spectral energy distribution was measured 113 cm below the barrier with a spectroradiometer (LI-COR 1800: Lincoln, NE). The PPFD readings ranged from 450-550 $\mu\text{mol m}^{-2}\text{s}^{-1}$ over the course of the experiment (NCSU & NC-ARS, 2008; Shurtleff, J., personal communication, 12 Sept. 2008).

'Maxifort' and 'German Johnson' seedlings were transplanted on 29 Jan. 2007 whereas the 'Trust' variety was transplanted on 2 Feb. 2007. All seedlings were transplanted into propagation flats (DPS 50 - Dillen Products: Middlefield, OH) filled with a commercial potting media (Redi Earth, 'Plug and Seedling Mix', Sun Gro Horticulture: Bellevue, WA). All seedlings were watered twice per day with de-ionized water, once in

the morning and once in the early afternoon. From 5 Feb.-13 Feb. 2007 all seedlings were fertilized with a modified Steiner nutrient solution during the morning watering event followed by de-ionized water in the afternoon (Table 2.1).

Grafting Procedures & Healing Chamber Management

On 14 Feb. 2007 all plants were grafted following the ‘Japanese top-grafting’ or ‘tube-grafting’ method described in the NC Cooperative Extension Bulletin # AG-675 (Rivard and Louws, 2006). Grafted plants were placed in a healing chamber located inside the growth chamber. The healing chamber consisted of a series of wheeled carts with wire shelves approximately 0.9 m off the ground. A series of carts were duct taped together and enveloped with greenhouse grade plastic forming an enclosed, rectangular chamber. A humidifier with an 8.3 liter tank (ReliOn[®] Ultrasonic Humidifier, Model #FHC-502/H-0695-0) was fitted with a 90° PVC elbow and 0.3 m straight length of PVC pipe, in order to facilitate the entry of water vapor into the plastic enclosure. Mist was directed under the grafted seedlings to prevent unnecessary leaf tissue wetness. Non-grafted treatments were transferred to a separate, cooler growth chamber, 22/18°C (day/night temperature) with all other parameters (photoperiod, etc.) the same to synchronize their growth with the recovery of the grafted treatments.

Humidity inside the healing chamber was maintained at high levels for the initial two days (~80-90% RH). Output from the cool mist unit was gradually decreased from high to medium, and then to low until the unit was turned off on the fifth day. To decrease

the amount of light entering the healing chamber, black trash bags were laid on the top of the chamber for the first four days. From the second day on, small vents were created in the top four corners of the chamber to allow for airflow and facilitate the escape of hot air. Six days later, on 20 Feb. 2007, the plastic sides and roof of the healing chamber were completely removed. The grafted seedlings were left in place for an additional six days. Regular seedling care included watering delicately.

All grafted and non-grafted seedlings were transplanted into 605 cm³ pots filled with a gravel-potting media substrate mix comprised of '2 gravel:1 potting media (by volume)' (Redi Earth, 'Plug and Seedling Mix', Sun Gro Horticulture: Bellevue, WA) on 26 Feb. 2007. From this date forward, seedlings were fertilized with a modified Steiner solution (Table 2.1) twice per day, once in the morning and once in the early afternoon. On 31 March 2007 all plants were transplanted into 1277 cm³ pots and tied to bamboo supports.

Experimental Design & Data Collection

The experimental design was a 2 x 3 factorial design arranged as a completely randomized block design. Plants were distributed within five adjacent blocks in the growth chamber. Within each block, groups of five plants from each of the six treatments were randomly arranged on carts. One plant per block was selected arbitrarily to be destructively sampled each week, for five weekly harvests (22 March, 28 March, 4 April, 11 April, and 18 April). The first sampling date marked the 36th day after grafting (DAG),

and the last sampling date the 63 DAG.

Leaf area measurements were taken with a leaf area meter (LI-COR Model LI-3100, Biosciences, Lincoln: NE). The leaf area meter was calibrated at each sampling event with a 50cm² metal plate. Leaf area, leaf weight, stem weight, fruit weight and fruit number were sampled on a weekly basis for five weeks during the late morning. Root weight, plant height, and leaf tissue for nutrient analysis were sampled on the last harvest date only. Stems were cut at a visible graft union or 1.5 cm above the potting media surface. Leaf samples consisted of both leaflets and petioles. Potting media was removed from plant roots by rinsing and then spraying with a jet spray hose attachment over a hardware cloth grate.

All leaf, stem, and root samples were placed in a drying oven for 72 hours at 80°C, after which their dry weights were measured. Leaf tissue samples were ground by a Wiley Mini-Mill stainless steel grinder with a 20 mesh screen (1.0 mm) (Thomas Scientific: Swedesboro, NJ) (Campbell, 1992; Campbell and Plank, 1992) and then sent to the North Carolina Department of Agriculture and Consumer Services (NCDA&CS) Agronomic Division for nutrient analysis. Total N was determined by oxygen combustion with an elemental analyzer (NA1500; CE Elantech Instruments: Milan, Italy) (Campbell, 1992). Total phosphorus (P), potassium (K), Calcium (Ca), Magnesium (Mg), Sulfur (S), Boron (B), Copper (Cu), Iron (Fe), Manganese (Mn), Zinc (Zn) and Sodium (Na) concentrations were determined by EPA Method 200.7 with an ICP spectrophotometer (Optima 3300 DV ICP Emission Spectrometer; Perkin Elmer Corporation: Wellesley,

MA), following open-vessel HNO₃ digestion in a microwave digestion system (CEM Corp., Matthews, NC) (Donohue and Aho, 1992).

Leaf tissue samples were also processed for total N by oxygen combustion with an elemental analyzer on the NCSU campus (N Brew Analyzer; Flash 1112 Series EA, Thermo Finnegan: Milan, Italy). The N readings taken from leaf tissue samples from the on-campus laboratory did not consistently correlate with the NCDA analysis, ranging from a 0.4 to 99% correlation. Therefore the on-campus dataset was not analyzed and all information presented is based on the NCDA&CS nutrient analysis. We hypothesize that the lack of consistent correlation was due to differences in sample processing such as sub-sample selection and grinding of tissue to different fineness levels between the two labs.

Statistical Analysis

Statistical analysis was carried out using a general linear model (Proc GLM) for plant growth parameters measured one time (i.e. height and root weight) (SAS Institute, Cary, NC). Proc GLM and a multivariate analysis of variance (ANOVA) were utilized to protect against high type I errors when evaluating the leaf nutrient concentrations and content (SAS Institute, Cary, NC). Repeated plant growth parameter measurements (i.e. leaf area, leaf weight & stem weight) were transformed to their common logarithm to rectify heteroskedacity and then analyzed with a linear mixed model (Proc MIXED) (SAS Institute, Cary, NC). A non-linear mixed model (Proc NLMIXED) was employed to fit an apparent logistic growth curve for fruit weight and fruit number and also to account for

small number counts (0-3) (SAS Institute, Cary, NC). Pearson correlation coefficients were used to measure associations between random variables (Proc CORR) (SAS Institute, Cary, NC).

Results

Plant Growth: Plant Height & Root Weight

Grafted treatments (both scion grafted onto ‘Maxifort’ rootstock and self-grafted) had higher mean values for plant height and root weight compared to non-grafted treatments (Fig. 2.1). The plant height and root weight of scion grafted onto ‘Maxifort’ rootstock and self-grafted treatments were not significantly different from each other (Fig. 2.1). The response to grafting was not different for ‘Trust’ or ‘German Johnson’ scion grafted onto ‘Maxifort’ rootstock. Both scion-rootstock graft combinations had greater mean values for root weight and height compared to non-grafts (data not shown). No interactions between scion type and the effect of grafting were present however, ‘German Johnson’-‘Maxifort’ grafts had greater values for root weight and height compared to ‘Trust’-‘Maxifort’ grafts (Table 2.2).

Plant Growth Over Time: Stem Weight, Leaf Weight & Leaf Area

Grafted treatments (both scion grafted onto ‘Maxifort’ rootstock and self-grafted) had higher mean values for leaf weight and leaf area compared to non-grafted treatments (Fig. 2.2). The stem weight of scion grafted onto ‘Maxifort’ rootstock and self-grafted

treatments were not significantly different from each other, whereas the leaf weight and leaf area was greater for scion grafted on 'Maxifort' compared to self-grafted treatments (Fig. 2.2). The response to grafting was not different for 'Trust' or 'German Johnson' scion grafted onto 'Maxifort' rootstock. Both scion-rootstock graft combinations had greater stem weight, leaf weight, and leaf area compared to non-grafts. No interactions between scion type and the effect of grafting were present however, 'German Johnson'-'Maxifort' grafts had a greater values for stem weight and leaf weight compared to 'Trust'-'Maxifort' grafts (Table 2.2).

Some general patterns between grafting effect and plant growth stage emerged for many of the plant growth parameters measured over time. The 1st sampling date corresponded with the 'early fruit set' growth stage, 2nd-3rd dates with the '1st ripe fruit', and the 4th-5th dates with the 'harvest period' (Hochmuth et al, 1991). Scion grafted onto 'Maxifort' rootstock had higher stem weight for the last two out of five sampling events compared to the self-grafted and non-grafted treatments (Fig. 2.3A). Scions grafted onto 'Maxifort' had higher leaf weight for the last three out of five sampling events compared to the self-grafted and non-grafted treatments (Fig. 2.3B). Scion grafted onto 'Maxifort' had higher leaf area for the last three out of five sampling events, compared to the non-grafted treatments (Fig. 2.3C). The leaf area of self-grafted treatments was greater compared to the non-grafts on the last sampling date. On a related note the specific leaf area (SLA) (leaf area (cm²)/leaf dry weight (g)) was greater for the self-grafted treatments compared to the non-grafts on the first sampling date only (Fig. 2.4).

Leaf Tissue Nutrient Content

The NCDA&CS nutrient analyses are expressed as a percentage (%) for primary and secondary nutrients (macro-nutrients), as well as sodium and micro-nutrients as parts per million (ppm); all results were kept shown in these units. The following nutrients were analyzed for all leaf tissue samples: N, P, K, Ca, Mg, S, Fe, Mn, Zn, Na, Cu, and B. Mean values for leaf tissue nutrient concentrations are presented in Table 2.3. All mean nutrient values tested were within or above the standard adequate range for tomato at specific growth stages with the exception of P (Hochmuth et al., 1991). Phosphorus was slightly below the recommended range of 0.20-0.40% with a mean value of 0.16 % although no symptoms of P deficiency were observed at any point in this experiment (Hochmuth et al., 1991). The leaf nutrient content of each plant was calculated (leaf tissue nutrient concentration (% or ppm) \times the total dry leaf weight (g)) to attain an estimate of the total amount of nutrient present in the leaf tissue. The differences in leaf tissue nutrient content were then analyzed among treatments for the nutrient uptake comparison. Leaf tissue nutrient content and leaf tissue nutrient concentrations were not highly correlated.

The mean leaf tissue nutrient content was higher in grafted treatments compared to non-grafts for the following nutrients: N, P, K, Ca, Mg, S, Fe, Mn, Zn, Cu, and B. Sodium was the only exception, it did not show a significant difference among treatments. Treatments grafted onto 'Maxifort' rootstock had higher mean leaf tissue content

compared to self-grafted treatments for the following nutrients: Ca, Fe, Mn, Zn, and Cu (Fig. 2.5A & 2.5B).

Both ‘Trust’ and ‘German Johnson’ scion grafted onto ‘Maxifort’ rootstock had similar nutrient accumulation levels for the majority of nutrients including: N, P, K, Mg, S, Mn, and Zn but some differences did exist. The scion variety ‘Trust’ had higher mean values for Ca, Fe, and Cu compared to ‘German Johnson’ (Table 2.4). There were no interactions between scion type and the effect of grafting.

The relationship between leaf tissue N content and root weight was examined to investigate whether the ratio was similar across treatments. The ‘leaf tissue N content : root weight’ ratio was not different among grafted and non-grafted treatments, however there was a scion effect. The ‘Trust-Maxifort’ grafts had a higher ratio of 13:1 compared to 11:1 for the ‘German Johnson-Maxifort’ grafts.

The non-grafted treatments had a higher total number of fruit compared to the grafted treatments ($P=0.0019$). Logistic growth curves were fitted to each grafting treatment over time with a nonlinear model; these growth curves suggested that fruiting on the grafted plants was delayed. Unfortunately the asymptote occurred beyond the region of collected data within the time frame of the study, therefore the analysis proved not to be a reliable estimate of fruit production over time.

Discussion and Conclusions

Plant Growth

Overall grafting increased shoot and root growth compared to non-grafted plants; this was true for both scion-rootstock combinations in the study. The increase in plant height was consistent among the existing literature which reports similar effects for additional grafted tomato combinations as well as grafted peppers (Leonardi and Guiffrida, 2006; Colla et al., 2006, 2008). Differences in leaf weight, stem weight, and leaf area became significantly greater for grafted plants compared to the non-grafted plants, 7-8 weeks after grafting. This suggests that the effects of grafting on plant growth are most evident from the ‘early fruiting stages’ of plant growth forward.

Grafting on ‘Maxifort’ rootstock resulted in an additional boost to leaf weight and leaf area. Interactions between the scion type and grafting were not present for any plant growth parameters indicating that the rootstock variety stimulates growth of the scion. In addition, the ‘German Johnson’-‘Maxifort’ grafts demonstrated greater gains in plant height, root weight, leaf weight, and stem weight compared to the ‘Trust’-‘Maxifort’ grafts indicating that the scion variety also plays a role in the extent to which grafted plants respond in terms of plant growth.

Our findings both agree and disagree with a similar study investigating the performance of ‘Rita’ tomato grafted onto the hybrid rootstock, ‘Beaufort’ (*Solanum lycopersicum* L. \times *Solanum habrochaites* S. Knapp & D.M. Spooner). This study found that tomato plants grafted onto interspecific rootstocks were taller and had a greater shoot

weight compared to self-grafted plants (Leonardi and Guiffrida, 2006). Our results concur that grafted plants had a greater shoot weight (leaf weight + stem weight) compared to self-grafted plants (data not presented) but did not find that grafted plants were significantly taller than the self-grafted plants. The differences in plant height could be due to either the differences in rootstock or scion variety selection. Our results indicate that a grafting effect, a rootstock effect, and a scion effect all exert influence over both shoot and root growth. In addition the leaf area of a grafted plant is primarily affected by grafting and followed by a rootstock effect but not affected by scion selection.

Many studies have demonstrated that the relationship between shoot weight and root weight during vegetative growth is generally logarithmically linear, although shoots generally continue to grow at a greater rate compared to the root system as a plant moves into the reproductive stage (Russell, 1977). In our study, shoot weight was highly correlated with root weight (84%) as well as plant height (81%), and leaf area (79%) ($P < .0001$). These positive correlations suggest that shoot weight can serve as overall plant growth indicator, which may be particularly helpful in future trials when root weight measurements may be difficult to obtain.

Nutrient Uptake

Overall, the most significant effect on total leaf tissue nutrient accumulation resulting in higher nutrient content levels for: N, P, K, Ca, Mg, S, Fe, Mn, Zn, Cu, and B were due to the grafting effect. Although the grafting effect was the most far reaching,

differences did exist between scion grafted on the 'Maxifort' rootstock and self-grafted treatments. Scion grafted on 'Maxifort' rootstock accumulated greater levels of Ca, Fe, Mn, Zn, and Cu in the leaf tissue compared to self-grafts. Total Zn and Cu leaf tissue content were lower in the non-grafts compared to self-grafted treatments. The nutrient content levels in the 'Trust'- 'Maxifort' grafts and the 'German Johnson'- 'Maxifort' grafts were not different for the majority of nutrients, although the 'Trust'- 'Maxifort' grafts had greater levels of Ca, Fe, and Cu in leaf tissue compared to the German Johnson'- 'Maxifort' grafts.

Nitrogen content in the leaf tissue was highly correlated with leaf weight (86%), stem weight (82%), height (76%), leaf area (81%) and root weight (75%) reaffirming the close relationship between N and plant growth. The 'Trust'- 'Maxifort' grafts had a greater 'N content : root weight' ratio compared to the 'German Johnson'- 'Maxifort' grafts. Previously it was shown that the N content in the leaf tissue was not significantly different between the scion grafted on 'Maxifort' rootstock combinations but at the same time the 'Trust'- 'Maxifort' grafts had a smaller root weight compared to the German Johnson'- 'Maxifort' grafts. This suggests that the 'Trust'- 'Maxifort' grafts were just as efficient at taking up N compared to the 'German Johnson'- 'Maxifort' grafts but with a smaller root mass, indicating that root vigor was not the sole factor determining nutrient accumulation in the grafted treatments.

In conclusion, the grafting effect resulted in both greater shoot and root growth as well as higher nutrient accumulation in the leaf tissue (*exception Na). When the

rootstock 'Maxifort' was used in grafted treatments the shoot weight, leaf area, and selected nutrients (Ca, Fe, Mn, Zn, and Cu) were greater than the self-grafted treatments. In addition, the scion cultivar also had an influence on shoot weight, root weight, and selected nutrients (Ca, Fe, Cu). Our results suggest that it is both the scion-rootstock pairing and the physical act of grafting itself which results in enhanced plant growth and leaf tissue nutrient accumulation. The overlapping effects from to grafting and cultivar selections can provide a wide range of plant responses for a variety of potentially beneficial applications.

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Table 2.1. Modified Steiner Nutrient Solution Analysis^z

Element	Symbol	Source	Total ppm in the solution ^y
Nitrogen	N	Mg(NO ₃) ₂ .6H ₂ O Ca(NO ₃) ₂ .4H ₂ O NH ₄ NO ₃ KNO ₃	106.23
Phosphorus	P	KH ₂ PO ₄ , K ₂ HPO ₃	10.41
Potassium	K	KH ₂ PO ₄ , K ₂ HPO ₄ K ₂ SO ₄ , KNO ₃	111.03
Calcium	Ca	Ca(NO ₃) ₂ .4H ₂ O	54.40
Magnesium	Mg	Mg(NO ₃) ₂ .6H ₂ O	12.40
Iron	Fe	Sequestrene 330	5.00
Sulfur	S	K ₂ SO ₄ , Na ₂ SO ₄	13.19
Manganese	Mn	MnCl ₂ .4H ₂ O	0.113
Boron	B	H ₃ BO ₃	0.24
Zinc	Zn	ZnSO ₄ .7H ₂ O	0.013
Copper	Cu	CuSO ₄ .5H ₂ O	0.005
Cobalt	Co	CoCl ₂ . 6H ₂ O	0.00003
Molybdenum	Mo	MoO ₃ .2H ₂ O	0.005
Sodium	Na	Na ₂ SO ₄	11.04

^z Source: NCSU & NC-ARA, Phytotron procedural manual for controlled-environment research at the Southeastern Plant Environment Laboratory, 2008 (rev.).

^y While usage of the unit "ppm" is common laboratory practice, the correct but less familiar SI unit would be mol m⁻³.

Table 2.2. Mean Plant Growth Parameters, Differences by Scion Cultivar.

	‘German Johnson’	‘Trust’
Mean Height (mm)	839.67 a ^z	743.00 b
Mean Root Weight (g)	2.74 a	2.12 b
Mean Stem Weight (g) ^y	1.03 a	0.78 b
Mean Leaf Weight (g) ^y	1.65 a	1.54 b
Mean Leaf Area (cm ²) ^y	6.92 a	6.94 a

^z Rows marked with the same lowercase letters are not significantly different based on Tukey’s test ($p \leq 0.05$).

^y All mean values are presented as log transformation.

Table 2.3. NCDA&CS, Mean Leaf Tissue Nutrient Concentrations.

Treatment ^z	%N	%P	%K	%Ca	%Mg	%S	Fe ppm	Mn ppm	Zn ppm	Cu ppm	B ppm	Na%
Trust/Max	2.79	0.16	5.47	1.86	0.77	0.35	194.80	249.60	48.06	23.12	81.06	0.10
Trust/Trust	3.11	0.17	5.42	1.61	0.77	0.35	157.20	146.40	38.28	21.34	73.98	0.12
Trust	3.14	0.14	5.80	1.79	0.83	0.32	183.80	178.60	30.34	18.40	69.28	0.17
GJ/Max	2.63	0.15	5.03	1.50	0.68	0.32	138.20	195.60	44.40	18.34	68.74	0.09
GJ/GJ	3.31	0.18	5.03	1.34	0.77	0.33	117.02	143.06	48.24	17.22	72.16	0.12
GJ	3.12	0.16	5.51	1.70	0.98	0.39	176.80	191.00	39.02	17.92	69.10	0.17

^z Trust/Max = 'Trust' grafted on 'Maxifort', Trust/Trust = self-grafted 'Trust', Trust = non-grafted 'Trust', GJ/Max = 'German Johnson' grafted on 'Maxifort', GJ/GJ = self-grafted 'German Johnson', and GJ = non-grafted 'German Johnson'.

Table 2.4. Nutrient Content^z of 'Trust' and 'German Johnson' Scion Grafted on 'Maxifort' Rootstock.

Nutrient	'German Johnson'	'Trust'	MSD ^x
N	28.25 a ^y	27.10 a	4.35
P	1.56 a	1.40 a	0.32
K	48.03 a	49.98 a	5.97
Ca	13.81 b	15.81 a	1.81
Mg	7.27 a	7.06 a	0.88
S	3.12 a	3.09 a	0.45
Fe	1,284.21 b	1,602.40 a	153.93
Mn	1,628.00 a	1,747.40 a	275.39
Zn	409.46 a	361.32 a	48.58
Na	1.06 a	1.13 a	0.14
Cu	164.12 b	191.59 a	22.62
B	652.14 a	681.66 a	94.09

^z Nutrient Content = (Leaf Tissue Concentration (% or ppm)*Leaf Dry Weight (g)).

^x MSD = Mean Significant Difference.

^y Tukey's LSD (≤ 0.05).

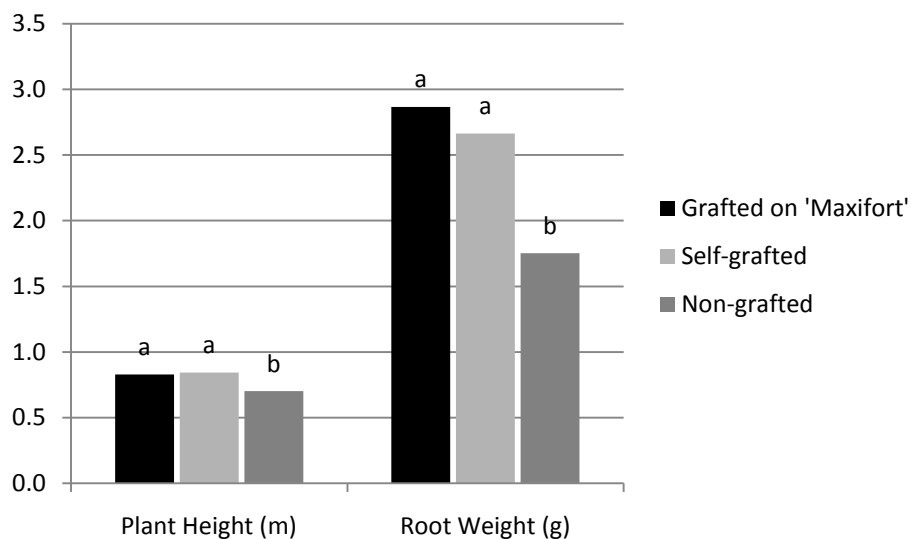


Fig. 2.1. Mean Plant Height and Root Weight, 63 Days After Grafting (DAG).
 Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's LSD ($p \leq 0.05$).

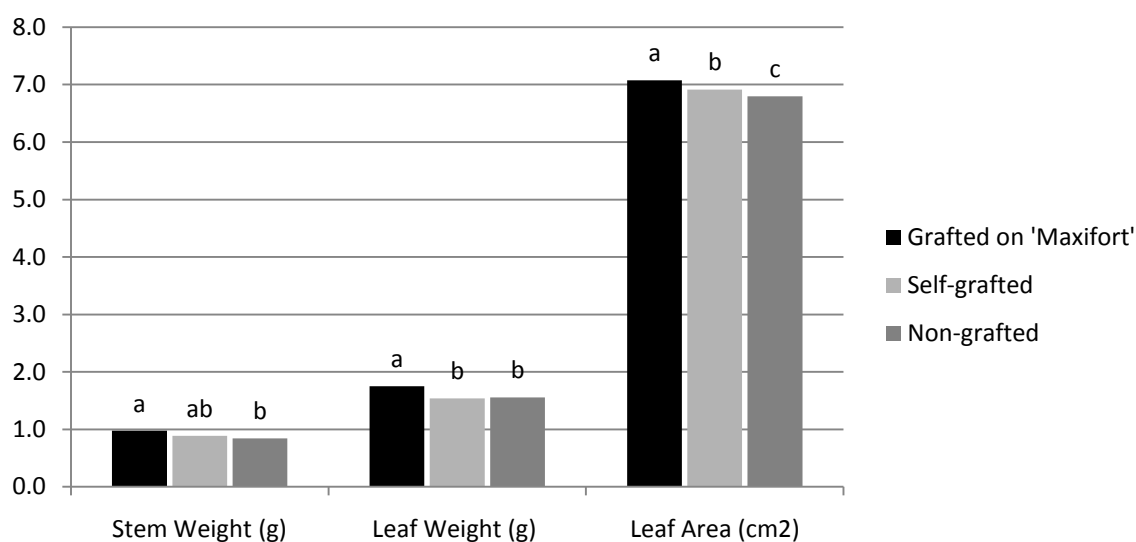


Fig. 2.2. Mean Stem Weight, Leaf Weight, and Leaf Area. All mean values are presented as log transformation. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's LSD ($p \leq 0.05$).

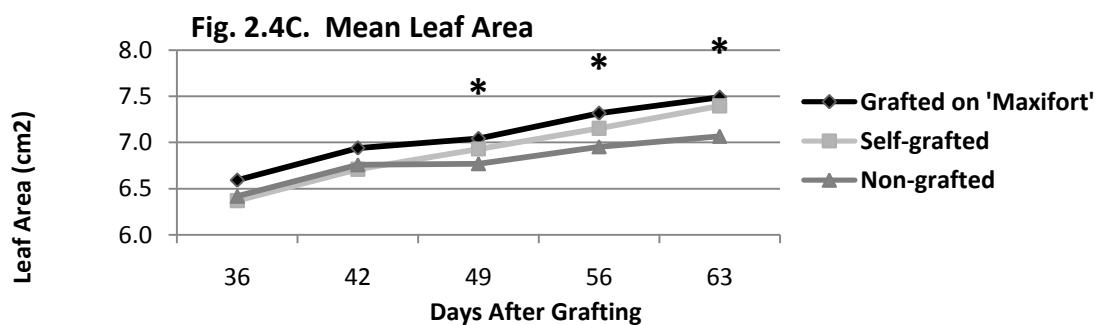
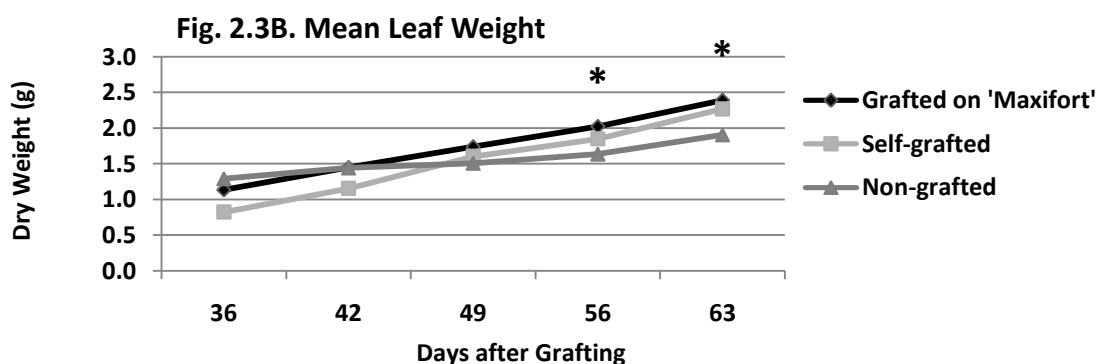
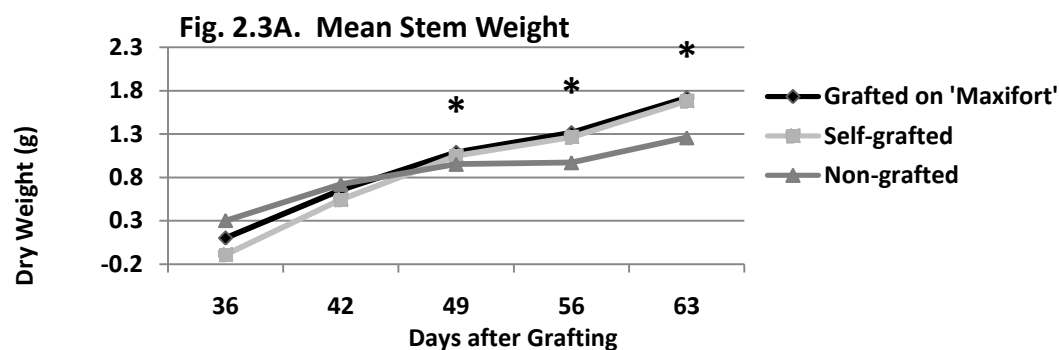


Fig. 2.3A, 2.3B, and 2.4C. Mean Stem Weight, Mean Leaf Weight, and Mean Leaf Area Over Time. All mean values are presented as log transformations. All grafted treatments has Cherokee Purple scion. * Points marked with an asterisk indicate that on the corresponding sample date that 'Cherokee Purple – Maxifort' grafts had higher values compared to the 'Non-grafted treatments' ($p \leq 0.05$).

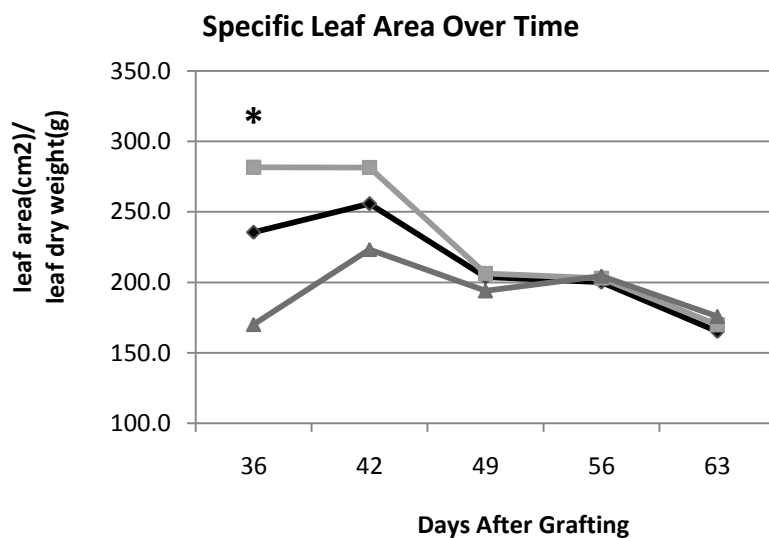


Fig. 2.4. Specific Leaf Area Over Time. All mean values are presented as log transformations. All grafted treatments have Cherokee Purple scion. * Points marked with an asterisk indicate that on the corresponding sample date that 'Self-grafted treatments' were higher compared to the 'Non-grafted treatments' ($p \leq 0.05$).

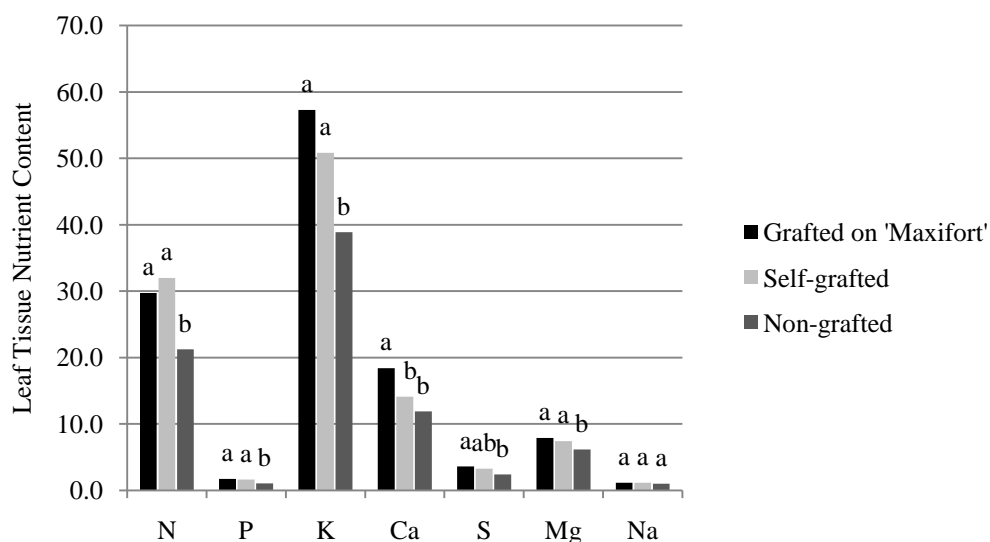


Fig. 2.5A. Mean Leaf Tissue Macro-nutrient Content. Mean values for leaf tissue nutrient content. Leaf tissue nutrient content = (leaf tissue nutrient content (%) x the total dry leaf weight (g)). Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

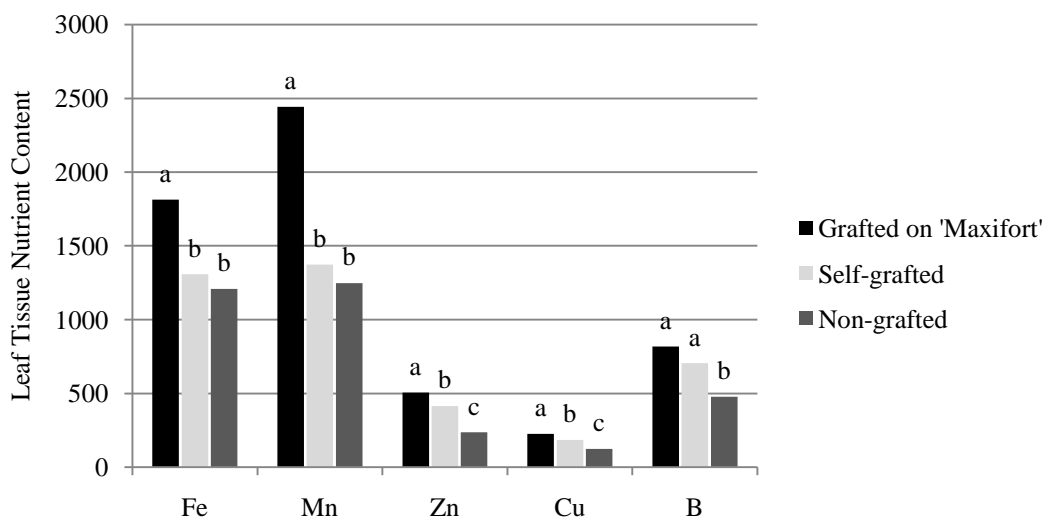


Fig. 2.5B. Mean Leaf Tissue Micro-nutrient Content. Mean values for leaf tissue nutrient content. Leaf tissue nutrient content = (leaf tissue nutrient concentration (ppm) x the total dry leaf weight (g)). Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

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CHAPTER 3 - CEFS: GROWING SYSTEM, NITROGEN INPUT LEVEL, AND GRAFTING EFFECTS ON NUTRIENT UPTAKE, PLANT GROWTH, AND FRUIT YIELD.

Abstract

In 2007 and 2008, a systems comparison study was conducted at ‘The Center for Environmental Farming Systems’ (CEFS) in Goldsboro, North Carolina. An organic high tunnel system was compared to an organic open field system. Three levels of N inputs were applied to each system. Grafting treatments included two heirloom scion-hybrid rootstock combinations: *Solanum lycopersicum* L. ‘Cherokee Purple’ grafted on *Solanum lycopersicum* L. *xSolanum habrochaites* ‘Maxifort’ or ‘Beaufort’, a self-grafted control (2008 only), and a non-grafted control. System type, N input level, and grafting effects on leaf tissue nutrient concentrations, plant growth, and fruit yield were evaluated.

The high tunnel system produced greater fruit yields for all treatments ($p \leq 0.05$) and hit peak production three weeks earlier compared to the field system ($p < 0.0001$). The high tunnel system had a higher incidence of fruit with blossom end rot, cat-facing, and cracking but lower incidence of TSWV and insect damage compared to the field system ($p < 0.05$). The mean leaf tissue N concentration never fell below standard optimum N ranges at any of the N input levels evaluated ($93 - 224 \text{ kg ha}^{-1}$) however a positive correlation between mean leaf tissue N concentrations and total harvest weight ($>70.4\%$) was present in 2008. Grafted plants produced a higher fruit yield compared to non-grafted plants in a low disease pressure

environment ($p \leq 0.0012$). The N input level effect on yield was not consistent across the two seasons, however, in 2008 both the high and medium N input levels (168 kg ha^{-1} and 122 kg ha^{-1} , respectively) produced greater total harvest yields compared to the low N level (93 kg ha^{-1}) ($p < 0.05$). Grafted plants also displayed greater plant growth compared to non-grafted plants ($p \leq 0.05$). Self-grafted plants (2008 only) and non-grafted plants were not different in terms of fruit yield or plant growth ($p \leq 0.05$).

The mean leaf tissue concentrations of N, P, K, Ca, Mg, and Fe were higher in the high tunnel system in 2007, but lower in 2008 compared to the field system ($p < 0.05$). The mean leaf tissue concentrations of Mn, Cu, B, and Na were lower in the high tunnel system compared to the field system across both years ($p < 0.05$). The mean leaf tissue concentrations for grafted plants were higher for N, P, K, Mn, Cu, Zn, and B but lower for Mg and Na compared to non-grafted plants across both years ($p \leq 0.05$). Self-grafted (2008 only) were not different from non-grafted plants for all leaf tissue nutrient concentrations ($p \leq 0.05$).

‘Cherokee Purple’ scion grafted on hybrid rootstocks had greater mean leaf tissue concentrations for N, P, K, Mn, Zn, and B compared to the self-grafted or non-grafted controls ($p \leq 0.05$). ‘Cherokee Purple’ scion grafted on ‘Maxifort’ rootstock had higher mean leaf tissue nutrient concentrations for P, S, Cu and Zn compared to those grafted on ‘Beaufort’ rootstock; the opposite was true for B and Mg.

In conclusion grafting resulted in greater nutrient accumulation in the leaf tissue for the majority of essential nutrients. Although the system and fertilizer level effects on leaf tissue concentration were variable across the two study years, grafting effects were

consistent, indicating that grafting on hybrid rootstock had the strongest influence on nutrient accumulation in the leaf tissue. N input levels above 122 kg ha⁻¹ produced the greatest fruit yield in 2008. Maximum fruit yield was achieved in the high tunnel system with the ‘Cherokee Purple-Maxifort’ grafts ($P < .0001$), indicating that when specific scion-rootstock combinations are paired with the a more controlled growing system fruit yields can be maximized.

Introduction

Fresh market tomatoes are an important and popular crop in the U.S. with an increasing demand for organically produced crops (ERS, 2008; Lucier and Plummer, 2004; OTA 2004, 2008). Small-scale, organic growers rely on tomatoes for a large amount of their direct market sales (Fernandez-Cornejo et al., 1994; Kremen et al., 2004). Consumers are willing to pay a price premium for organic tomatoes, heirloom varieties, and tomatoes offered outside the typical regional growing season (Steven-Garmon et al., 2007; Lyson et al., 1995).

There are many inherent challenges with growing tomatoes in the Southeast which can be intensified with organic production methods. Total N accumulation from the soil by field crops is often the limiting yield factor (Sinclair, 2004). Supplying N to an organic tomato crop, commonly grown under plastic mulch and with drip irrigation, is a challenging task in eastern North Carolina. Soluble materials that meet the United States Department of Agriculture (USDA), National Organic Program (NOP) standards are limited and expensive;

therefore, most growers rely primarily on pre-plant fertilizer inputs. Ultisol soils typical of the Southeast, are intensely weathered soil structures and are generally low in nutrients. In addition, long growing seasons support active microbial communities that breakdown organic matter quickly, compounding nutrient deficiencies that limit the health and productivity of the crop as the season progresses.

Grafting of herbaceous plants has been used in commercial fruit and vegetable production in Asia since the 1920's (Tateishi, 1927). Originally grafting was used to manage *Fusarium oxysporum* in watermelon crops but it is now applied to many cucurbit and solanaceous crops (Oda, 2002). The reasons for grafting have also expanded from increased soilborne disease resistance (*Fusarium oxysporum*, *Meloidogyne spp.*, *Monosporascus cannonbolus*, *Ralstonia solanacearum*, TMV, *Pyrenochaeta lycopersici* and *Verticillium albo-atrum*) to greater crop yields, higher salinity tolerance, increased heat and cold tolerance, and enhanced drought and flood resistance (Besri, 2005; Black, 2003; Burleigh et al., 2005; Colla et al., 2006, 2008; Edelstein et al., 1999, 2005, 2008; Eitan et al., 2005; Jifon et al., 2008; Kato and Lou, 1989; Lee, 1994; Leonardi and Giuffrida, 2006; Matsuzoe et al., 1993, 1993; Rivero, 2003; Romero et al., 1997; Siguenza et al., 2005; Pulgar et al., 2000; Yetisir et al., 2006, 2007). In addition, herbaceous grafting may increase nutrient uptake efficiency (Lee, 1994; Ruiz et al., 1996; 2006, Leonardi & Giuffrida, 2006) but these effects are not well documented. Most research has been conducted in Eastern Asia (Korea, China, Japan) and the Mediterranean region on cucurbitaceous crops (Greece, Spain, Italy, Israel,

Morocco) where herbaceous grafting has been widely utilized.

Cultivating tomatoes under high tunnel systems may offer a number of benefits and opportunities such as: season extension, higher fruit quality, less foliar disease pressure, and protection from extreme weather events. Yet growing in high tunnels comes with its' own challenges, such as a decreased ability to rotate crops, accumulation of fertilizer salts and increased potential for heat stress during warm weather. Grafted plants may be uniquely suited to production in a high tunnel environment due to their higher stress tolerances, increased crop longevity, more efficient nutrient and water use, and soil borne disease resistance (Besri, 2005; Black, 2003; Burleigh et al., 2005; Colla et al., 2006, 2008; Edelstein et al., 1999, 2005, 2008; Estan et al., 2005; Jifon et al., 2008; Lee, 1994; Leonardi and Giuffrida, 2006; Pulgar et al., 2000; Rivero, 2003; Romero et al., 1997; Yetisir et al., 2006, 2007). The combination of growing high-value grafted crops such as organic heirloom tomatoes under high tunnel structures is an innovative systems approach that can provide growers with new economic opportunities, greater production stability, higher fruit quality, and lower pest management and fertilizer inputs.

The goals of this research were to evaluate the performance of grafted heirloom tomatoes compared to non-grafted plants under multiple fertilizer regimes in two distinct organic growing systems, high-tunnels versus the open field system. This study attempted to answer the following questions: 1) are there differences in nutrient uptake levels of grafted and/or non-grafted plant between the high tunnel and the open field system? 2) how does the growing system affect plant yield and plant growth? 3) what are the minimum levels of N

inputs required by grafted plants in order to maintain adequate leaf tissue N concentration ranges for plant growth and fruit production over a growing season? 4) how does N input level affect plant yield and plant growth? 5) do grafted tomato plants accumulate more nutrients compared to non-grafted plants in organically managed, soil-based systems? and 6) how does grafting affect plant yield and plant growth?

Materials and Methods

Experimental Design

This systems comparison study was conducted at The Center for Environmental Farming Systems (CEFS)/Cherry Research Farm located in Goldsboro, NC and repeated for two, consecutive years, 2007 and 2008. Plants were cultivated in two separate, 9.14 m x 26.26 m high tunnel units and two, adjacent 9.14 m x 26.26 m field area. Grafting treatments consisted of two rootstock/scion combinations, a non-graft control, and a self-graft control (2008 only). The grafting combinations were: 1) ‘Cherokee Purple’ scion (*Solanum lycopersicum* L. ‘Cherokee Purple’) grafted on ‘Maxifort’ rootstock (*Solanum lycopersicum* L. x*Solanum habrochaites* S. Knapp & D.M. Spooner., ‘Maxifort’), and 2) ‘Cherokee Purple’ scion grafted on ‘Beaufort’ rootstock (*Solanum lycopersicum* L. x*Solanum habrochaites* S. Knapp & D.M. Spooner., ‘Beaufort’). The non-grafted control consisted of ‘Cherokee Purple’ tomato plants. Because of problems with seedling production in 2007, a ‘self-grafted’ control, ‘Cherokee Purple’ grafted back onto itself, was included in 2008 only. Therefore

nine treatments were included in the study in 2007 compared to twelve treatments in 2008. The rootstocks trialed were available commercially in the U.S. (De Ruiter Seeds, Inc.: Lakewood, CO).

The experiment was arranged as a 2 x 3 x 3 (2007) or 4 (2008) factorial with a replicated split-split plot design (Fig. 3.1). Main plots and subplots were randomized for each year separately. The whole plot factor was the growing system type, high tunnel or open field. The sub-plot factor was the fertilizer treatment (different levels of N), and the sub-sub plot factor was the grafting treatment. Each sub-sub plot served as an experimental unit which consisted of six plants per row in 2007 and five plants per row in 2008. Each experimental unit was replicated four times, twice in each of the high tunnels and twice in each of the corresponding fields. Guard rows were planted on the front and back end of each tunnel and field plot.

Field and High Tunnel Description

The project area was located in the CEFS field, 'C2'. On 6 Oct. 2006, 'C2' was cultivated and 2,326 kg ha⁻¹ of calcitic lime applied. On 11 Nov. 2006, 'C2' was fumigated with methyl bromide in order to manage the federally listed noxious weed 'Tropical Spiderwort', *Commelina benghalensis* L. The entire field was planted on 13 Dec. 2006 with a cover crop of winter rye, *Secale cereale*.

The soil type at the project site was a Wickham, sandy loam (WhA), characterized as a deep, well-drained, and a slight to moderately acidic soil (Derrick, 1916). This soil is

typically found near stream terraces in the Piedmont and Coastal Plain regions of NC. The banks of the Little River were located approximately 120 m from our project site beyond a wooded buffer zone.

Two high tunnels (Atlas Greenhouses Inc.: Alapaha, GA) were constructed on site in February, 2007. The high tunnels were a snow-arch design with an inflated double polyethylene film (0.152 mm) roof and twin wall polycarbonate end walls. Doors built into the end walls were sized wide enough to allow the passage of a small tractor for spring tillage. Bows were spaced every 1.2 m. and a ‘1.8 m Z-Lock drop down curtain system’ with a motorized crank was employed. Adjacent field plots were also established at this time.

Seedling Production

Guidelines set forth by the USDA NOP were followed during seedling production. ‘Maxifort’, ‘Beaufort’, and ‘Cherokee Purple’ seeds were sown into flats filled with a mix of “1 potting media : 1 river bottom sand (by volume)” (Sunshine Complete Organic Mix, Sun Gro Horticulture: Bellevue, WA). The potting media was comprised of: 75-85% Canadian sphagnum peat moss (by volume), 25-25% coarse grade perlite (by volume), 2.4 – 2.8% dolomitic lime (by weight), 0.5 – 1.0% organic starter nutrient charge (including poultry manure) plus gypsum (by weight), and 0.05 – 0.10% organic wetting agent (yucca extract) (by weight). All flats were topped with a thin layer of horticultural grade vermiculite (Larrea, 2005). Flats were placed on a heat mat (26-28°C) in a germination chamber. A 12 h light/12 h dark photoperiod (fluorescent lighting with a Photosynthetic Photon Flux Density (PPFD)

of $\sim 80 \mu\text{mol m}^{-2}\text{s}^{-1}$ during light hours) and a series of overhead fine mist water nozzles (output every 3 minutes for 3 seconds during light hours only) was also present (Shurtleff, J., personal communication, 12 Sept. 2008). When approximately 75% of the seeds germinated, each flat was transported to a controlled environment growth chamber.

The controlled environment growth chamber measured 9 m^2 (NCSU & NC-ARS, 2008). Environmental conditions in the growth chamber were maintained at day/night temperatures of $26/22^\circ\text{C}$ and were coordinated with a photoperiod of 12 h light/12 h dark, respectively. A combination of T-12, 1500 ma, cool-white fluorescent and 100 W incandescent lamps provided light in the chamber. A Plexiglas barrier separated the lamps from the growing area. The resulting spectral energy distribution was measured 113 cm below the barrier with a spectroradiometer (LI-COR 1800: Lincoln, NE). The PPFD readings ranged from $450\text{--}550 \mu\text{mol m}^{-2}\text{s}^{-1}$ over the course of the experiment (NCSU & NC-ARS, 2008; Shurtleff, J., personal communication, 12 Sept. 2008).

The seedlings were transplanted into propagation flats (DPS 50 - Dillen Products: Middlefield, OH) filled with a mix of '1 potting media : 1 river bottom sand (by volume)' (Sunshine Professional Organic Potting Mix, Sun Gro Horticulture: Bellevue, WA). Seedlings were watered twice daily, once in the morning and once in the early afternoon, with de-ionized water. A dilution of a soluble organic fertilizer (Omega 6N-6P-6K, Petrik Inc., Woodland, CA) derived from soy protein, casein, blood meal, fish meal, rock phosphate, bone meal and potassium rock was administered to all treatments once per week. During each fertigation event, the nutrient solution was mixed and applied immediately by

hand to each seedling flat. The fertilizer solution was comprised of 10 ml 3.79L⁻¹. Seedlings were grafted using the ‘Japanese top-grafting’ or ‘tube-grafting’ method described in the NC Cooperative Extension Bulletin # AG-675 (Rivard and Louws, 2006).

Approximately two weeks after grafting, all grafted and non-grafted seedlings were transplanted into larger propagation flats (DPS1801 - Dillen Products: Middlefield, OH). Once the seedlings had greater than 2 pairs of true leaves, the strength of the fertilizer solution was doubled to 20 ml 3.79L⁻¹. During each fertigation event, the nutrient solution was mixed and applied immediately by hand to each seedling flat. Seeding, transplanting, and planting dates for both years are listed in Table 3.1.

Cultural Management

Cultural management practices typical for each system were employed. The high tunnel system was approached as a hybrid between a greenhouse and an open field culture. Planting dates in the high tunnel system 20 March 2007 and 18 March 2008, were approximately one month earlier than the field system 19 April 2007 and 17 April 2008, and reflect the typical planting dates of local growers. In 2007, inner floating polypropylene fabric row covers (Agribon+ AG 15: PGI Charlotte, NC) were used to cover seedlings inside the high tunnels when evening temperatures were predicted to drop below 4.4°C. In 2008, the spring weather was milder and inner row covers were not employed based on the same criteria.

Plants in the high tunnel system were pinched to encourage the formation of two leaders, typical of greenhouse production using grafted transplants. These two leaders were trained to a trellis system consisting of vertical strings hanging from horizontal tension cables extending across the width of the high tunnels. The tomato vines were attached to the strings with plastic plant clips, approximately every 25.4 cm. The lower leaves were pruned up to 'one leaf below the first fruit cluster' and suckers were removed on a weekly basis to steer growth towards the main leaders and fruit production. In 2007, suckering was conducted throughout the season. In 2008, in an effort to decrease the amount of sun-scald damage to fruit, suckering was conducted for only 2.5 months, from April to mid-June. The high tunnel drop down curtain system was programmed to lower the sidewall curtains when ambient air temperatures inside the tunnels reached above 18.3°C. The controller is designed to open the curtains incrementally based on the change in temperature after five minutes of idle time. At the end of each season, the high tunnels were sealed for a solarization period of approximately 4 weeks.

In the field system, the stake-and-weave trellis system was utilized. Leaf pruning was conducted up to the first horizontal string in the field and plants were not pinched nor suckered, resulting in a bushier growth form compared to the high tunnel plants. Each system was irrigated on an as needed basis, evaluated twice daily. All planting rows in both systems were 4.1 m long and 1.4 m wide; plants were spaced every 56 cm. Black polypropylene landscape fabric was utilized as a weed barrier in all plots. Drip tape with emitters every 30.5 cm and a flow rate of 19 l h⁻¹ were used for irrigation.

Integrated Pest Management

Integrated pest management practices were utilized for the management of insect pests. Weekly scouting events were conducted and decisions based on established thresholds for organic systems when available. In the early spring of 2007, the high-tunnel system experienced a high population of potato aphids, *Macrosiphum euphorbia*. Two spot applications of insecticidal soap (M-Pede, DowAgroSciences: Indianapolis, IN) were carried out and followed by multiple releases of the aphid midge, *Aphidoletes aphidimyza*, and the parasitic wasp, *Aphidius ervi*.

In both 2007 and 2008, applications of *Bacillus thuringiensis* (*Bt*) were applied to both the high tunnel and field system on two occasions. These sprays targeted tomato hornworms, *Manduca quinquemaculata*, tomato fruitworms, *Helicoverpa zea*, and/or armyworms *Spodoptera sp.* and were alternated between the subspecies *aizawai* (Xentari DF, Valent BioSciences Corp.; Libertyville, IL) and subspecies *kurstaki* (Dipel DF, Valent BioSciences Corp.; Libertyville, IL). In 2008, both the high tunnel and field system experienced high populations of stink bugs, *Acrosternum hilare*. However, no action was taken due as the negative effects of spraying a broad spectrum insecticide to reduce the population were judged to outweigh the benefits from the beneficial insect community present and hand removal was the only control strategy utilized.

A series of bi-culture cover crop strips were planted in succession around the perimeter of each high tunnel and field plot in Oct. 2007 and June 2008 in order to attract and

sustain beneficial insects and pollinators to the project area. These crops consisted of: 1) a crimson clover, *Trifolium incarnatum* and tall fescue ‘Kentucky 31’, *Festuca arundinacea* Schreb. mix, 2) a berseem clover, *Trifolium alexandrinum* L. and arrowleaf clover, *Trifolium vesiculosum* mix, and 3) a buckwheat, *Fagopyrum esculentum* and pearl millet, *Pennisetum glaucum* mix.

Fertilizer Applications and Cover Crops

Three levels of total N inputs (pre-plant + post-plant applications) were evaluated each growing season. In 2007, the three N levels were 112 kg ha⁻¹ (low), 168 kg ha⁻¹ (medium), and 224 kg ha⁻¹ (high). The ‘Vegetable Crop Handbook for Southeastern United States (2007)’ recommends a N application rate of 224 kg ha⁻¹ for soils with low potassium, such as ‘C2’; this recommendation matches the 2007, high N treatment. All three fertilizer levels in 2007 provided more than adequate N as reflected in their sampled leaf concentrations. As a goal of the study was to investigate the critical level of N inputs required by a grafted tomato crop, the three N levels were lowered to 93 kg ha⁻¹ (low), 112 kg ha⁻¹ (medium), 168 kg ha⁻¹ (high) in 2008.

In 2007, a pre-plant application of 22 t ha⁻¹ of compost (‘Leprechaun NOP Compost’ - McGill Environmental Systems: Harrells, NC) and 102 kg ha⁻¹ of feathermeal (Nutrimax ‘Super’ Natural Organic Fertilizer (12N-1P-0K), Nutrimax Inc.: Greensboro, NC) was applied to all plots, approximately 2 weeks before the respective system planting dates. The feathermeal material consisted of bone meal, meat meal, hydrolyzed poultry feathers, and

spray dried blood meal. This pre-plant application of compost and feathermeal was estimated to supply 98.6 kg ha⁻¹ of crop available N (Table 3.2). The remaining balance of N was provided to the crop over a series of post-plant, soluble fertilizer solution applications (Phytamin 801 (6N-1P-K1), California Organic Fertilizers, Inc.: Fresno, CA).

Post-plant fertilizer applications were timed to provide N to the crop at critical times during plant growth (Table 3.3A & 3.3B). The majority of post-plant N was supplied during the growth stages, '1st bloom to 1st harvest' and 'early harvest' to encourage fruit production (Heuvelink (ed.) 2005; Holmes (ed.) 2007). In 2007, post-plant fertilizer was applied over five fertigation events and was dispensed to each plant by hand. In 2008, post-plant fertilizer schedule was modified to only two fertigation events and was dispensed to each plant via the drip irrigation system with the aid of a fertilizer injector set a ratio of 1:64 (Dosatron Liquid Dispenser, Model DI 16-11GPM: Clearwater, FL).

Additions of K and calcium (Ca) were provided to the crop based on NCDA leaf tissue analysis and soil test recommendations. In 2007, K was applied twice to both growing systems on 15 June at 91.5 kg ha⁻¹ (Champion Sulfate of Potash (0N-OP-52K), SQM North America Corp.:Atlanta, GA) and on 28 June at 70.4 kg ha⁻¹ (Diamond Ultrafines Sulfate of Potash (0N-OP-50K), Great Salt Lake Minerals Corporation: Overland Park, KS). Prior to the 2008 season, 145 kg ha⁻¹ of K₂O was incorporated into the high tunnel system on 28 Feb. and the field system on 1 April. Additional K₂O (Royster-Clark Sulfate of Potash (0N-OP-50K), Crop Production Services: Collinsville, IL) was applied as post-plant fertilizer (Champion Sulfate of Potash (0N-OP-52K), to the high tunnel system on 8 July at

91.5 kg ha⁻¹ and to the field on 27 July at 91.5 kg ha⁻¹. In 2007 only, Ca was applied as a foliar spray. The soluble Ca (Fert-all Calcium 12%, Grow More Inc: Gardena, CA) was applied at a rate of 7.8 ml l⁻¹ on 4 June and 26 June in both systems. It was sprayed until complete coverage of the leaf tissue and fruit was achieved (Solo 450 Air Blast Sprayer, Newport News, VA).

Between the 2007 and 2008 season, both systems were planted with a mixed winter cover crop on 26 Sept. 2007. The mix consisted of 33.6 kg ha⁻¹ of hairy vetch, *Vicia villosa*, and 50.4 kg ha⁻¹ of winter rye, *Secale cereale*. Irrigation was provided with overhead sprinklers as needed for each system. The cover crop was sampled just prior to mowing and soil incorporation. The growth of the cover crop in each system was very different over the course of the following 4-5 months. The total cover crop biomass was 43% greater in the high tunnel system (4,572 kg ha⁻¹) compared to growth in the field system (2,590 kg ha⁻¹).

The cover crop in the high tunnel system was estimated to contribute 93 kg ha⁻¹ of crop available N compared to the field cover crop with 53 kg ha⁻¹ for the 2008 growing season. As a result, the target amount of pre-plant N for the low fertilizer treatment was set at 93 kg ha⁻¹ in 2008. In order to bring the field system up to the same pre-plant N input level as the high tunnel system, feathermeal was added to the field but not to the high tunnel system. Fifty-nine kg ha⁻¹ of feathermeal, with an estimated 41.2 kg ha⁻¹ of crop available N, was supplied to the field system (Table 3.2).

Sampling Protocols

Leaf tissue samples consisting of the ‘most recently mature leaves’ (MRML) were collected six or five times over the growing season in 2007 and 2008, respectively (Table 3.4). One leaf was sampled from each plant and then combined with leaves from the same sub-plot to comprise a leaf tissue sample. All leaf tissue samples were sent to the NCDA&CS, Agronomic Division for analysis. Samples were dried for 48 hours at 80°C and then ground by a Wiley Mini-Mill stainless steel grinder with a 20 mesh screen (1.0 mm). (Thomas Scientific: Swedesboro, NJ) (Campbell and Plank, 1992). Total N was determined by oxygen combustion with an elemental analyzer (NA1500; CE Elantech Instruments: Milan, Italy) (Campbell, 1992). Total phosphorus (P), potassium (K), Calcium (Ca), Magnesium (Mg), Sulfur (S), Boron (B), Copper (Cu), Iron (Fe), Manganese (Mn), Zinc (Zn) and Sodium (Na) concentrations were determined by EPA Method 200.7 with an ICP spectrophotometer (Optima 3300 DV ICP Emission Spectrometer; Perkin Elmer Corporation: Wellesley, MA), following open-vessel HNO₃ digestion in a microwave digestion system (CEM Corp., Matthews, NC) (Donohue and Aho, 1992).

In addition to leaf tissue analysis, fruit harvests were conducted twice per week. Fruit was picked from the ‘pink to red’ stages. Fruit was sorted into marketable and non-marketable categories. Qualitative judgements relating to marketable and non-marketable fruit were based on observations from regional direct sales outlets for organic produce. Non-marketable fruit was sorted into categories including: cat-facing, blossom end rot, insect damage, fruit cracking, tomato spotted wilt virus (TSWV), sun-scald (2008 only) and ‘other’.

Both the fruit number and fruit weight was recorded for each category. See Chapter 4 for results relating to yield.

In 2007, plant height measurements from the 2nd and 4th plant in each row were taken on a weekly basis from 26 April 2007 to 2 August 2007 in the high tunnel system. In 2008, plant height measurements were taken on a weekly basis from the 2nd and 4th plant in each row in both the high tunnel system and the field system. Measurements were taken from the high tunnel system from 1 April 2008 to 14 July 2008 and the field system from 12 May 2008 to 28 July 2008.

In both 2007 and 2008, shoot biomass samples were taken from the 2nd plant in each row from the high tunnel system after all harvest information had been collected. Any remaining fruits were removed from the vines and shoots were cut at their graft union or approximately 7.6 cm above the soil line. Samples were dried for 48 hours at 70°C, and then weighed.

Statistical Analysis

Statistical analysis was carried out using a multivariate analysis of variance (MANOVA) to protect against high type I errors and a linear model for repeated measures with Proc Mixed (Proc MIXED) (SAS Institute, Cary, NC). Linear hypotheses were used to test pair wise differences for fixed factors such as grafted versus non-grafted plants, and ‘Beaufort’ versus ‘Maxifort’ rootstock (‘Estimate Statements’, SAS Institute, Cary, NC).

Results

The NCDA plant tissue analysis expresses macronutrients and sodium by the percentage (%) and micronutrients as parts per million (ppm), therefore all conclusions were kept in these same units. The mean nutrient concentration values for each element tested were within or above the adequate range compared to the industry standards with the exception of slightly low Zn in both systems, across both years, on the last sampling date (Hochmuth et al., 1991).

System Effects on Leaf Tissue Nutrient Concentrations

In 2007, the mean leaf tissue nutrient concentrations were higher in the high tunnel system compared to the field system for the following nutrients: N, P, Ca, Mg, Fe, and Zn. The mean leaf tissue nutrient concentrations were lower in the high tunnel system compared to the field system for the following nutrients: K, S, Mn, Cu, B and Na (Fig. 3.2A and 3.2B). An interaction between grafting effect and the system existed for ‘Cherokee Purple-Maxifort’ and ‘Cherokee Purple-Beaufort’ grafts. Both scion-hybrid rootstock graft combinations had higher N and S concentration levels in the leaf tissue in the high tunnel system compared to the field system. These same grafting treatments had lower Na concentrations the high tunnel system compared to the field system. Lastly, the ‘Cherokee Purple-Maxifort’ and non-grafted treatments had higher levels of Mg in the high tunnel system compared to the field system (data not shown).

In 2008, the mean leaf tissue nutrient concentrations were higher in the high tunnel system compared to the field system for the nutrients K and S. The mean leaf tissue nutrient concentrations were lower in the high tunnel system compared to the field for the following nutrients: N, P, Ca, Mg, Fe, Mn, Cu, B, and Na (Fig. 3.3A & 3.3B). Two interactions were present between the growing system and grafting treatments. First, the ‘Cherokee Purple-Beaufort’ grafts had higher K leaf tissue concentrations in the high tunnel system compared to the field system. Second, all grafted and non-grafted treatments had lower Na leaf tissue concentrations in the high tunnel system compared to the field system (data not shown).

System Effects on Fruit Yield

In 2007, the high tunnel system had a greater total cumulative harvest weight (Fig. 3.4A) and number compared to the field system. More specifically, the high tunnel system produced a greater weekly harvest (weight and number) compared to the field system from 9 June to 23 June 2007. The high tunnel system produced a smaller weekly harvest yield weight (Fig. 3.5A) and number compared to the field system from 7 July to 14 July 2007.

In 2008, the high tunnel system had a greater total cumulative harvest yield (weight and number) compared to the field system (Fig. 3.4B). More specifically, the high tunnel system produced a greater weekly harvest (weight and number) from 7 June to 21 June 2008 compared to the field system. The high tunnel system produced a smaller weekly harvest weight (Fig. 3.5B) and number from 8 July to 12 July 2008 compared to the field system.

In 2007, the high tunnel system had higher values compared to the field system for total: harvested fruit weight and number (Fig. 4.10A & 4.10B), culled fruit (weight and number), blossom end rot fruit (weight and number), cat-faced fruit (weight and number), cracked fruit (weight and number), and fruit (weight and number) designated as ‘other’. The high tunnel system had lower values compared to the field system for total marketable fruit (weight), insect damaged fruit (weight and number), and TSWV damaged fruit (weight and number) (Fig. 3.6A & 3.6B).

In 2008, the high tunnel system had higher values compared to the field system for total: harvested fruit weight and number, marketable fruit (weight and number), blossom end rot (weight and number), cat-faced fruit (weight and number), and sun-scald damage (weight and number). The high tunnel system had lower values compared to the field system for total: cracked fruit (weight and number), insect damaged fruit (weight and number), and TSWV damage (weight and number) No differences between growing system type were present for the culled fruit or the ‘other’ category (Fig.3.7A & 3.7B).

N Input Level Effects on Leaf Tissue Concentrations

In 2007, the N input level had an effect on only one nutrient concentration in the leaf tissue, N. The level of N accumulated in the leaf tissue was greater for the treatments which received the high level of N (224 kg ha^{-1}) compared to the low level of N (112 kg ha^{-1}).

In 2008, there were no significant differences among nutrient concentrations in the leaf tissue due to varying levels of N inputs (93, 112, and 168 kg ha⁻¹) (data not shown).

N Input Level Effects on Fruit Yield

During the first season, N input level had little effect on total fruit yield. There were no differences in total yield due to the N input levels (Fig. 3.8A). However, relationships were present between the N input level and the ‘total harvest potential’ yield with the medium and low N input levels retaining higher amounts of un-ripe fruit on the vine at the end of the research study compared to the high N input level (Fig. 3.8 B). In 2007, treatments receiving the lowest N input level, 122 kg ha⁻¹, had a higher incidence of total fruit affected by blossom end rot disorder (weight) compared to the medium N level, 168 kg ha⁻¹. The mean leaf tissue N concentration was highly correlated with the total harvested fruit number and weight, 65.7% and 70.4 %, respectively.

In 2008, both the high and medium N levels, 168 kg ha⁻¹ and 122 kg ha⁻¹ respectively, had greater weekly values for total: harvested fruit (weight), marketable fruit (weight and number), culled fruit (weight and number), and insect damaged fruit (weight and number) compared to the lowest N level, 93 kg ha⁻¹. The high N and medium N input levels resulted in greater total fruit yields (weight and number) compared to the low N input level (Fig. 3.10A). The ‘total harvest potential’ was greater for the high N level compared to the low N level for fruit weight only (Fig. 3.10B).

N Input Level Effects on Plant Growth in the High Tunnel System

In 2007, the N input levels (112, 168, and 224 kg ha⁻¹) did not affect either the shoot weight or plant height (data not shown). However, the mean N content of the shoot sampled at the end of the season (mean leaf N concentration (%) x shoot dry weight (g)) was highly correlated with shoot weight (99.6%) ($p \leq 0.0001$). In 2008, the N input levels (93, 112, and 168 kg ha⁻¹) did not affect either the shoot weight or plant height. However, the mean N content of the shoot sampled at the end of the season was highly correlated with shoot weight (97.9%) ($p \leq 0.0001$).

Grafting Effect on Leaf Tissue Nutrient Concentrations

In 2007, the mean leaf tissue nutrient concentrations were higher in grafted treatments compared to non-grafts for the following nutrients: N, P, K, Mn, Cu, Zn and B. The mean leaf tissue nutrient concentrations were lower in grafted treatments compared to non-grafts for Mg, Fe, and Na (Fig. 3.11A & 3.11B). Differences between the ‘Cherokee Purple-Maxifort’ grafts and ‘Cherokee Purple-Beaufort’ grafts did exist for a select number of nutrients. The ‘Cherokee Purple-Maxifort’ grafts had higher levels of P, K, S, Cu and Zn compared to the ‘Cherokee Purple-Beaufort’ grafts. The ‘Cherokee Purple-Beaufort’ grafts had higher levels of B compared to the ‘Cherokee Purple-Maxifort’ grafts. Only one nutrient, Ca, showed no differences among grafted and non-grafted treatments.

In 2008, the mean leaf tissue nutrient concentrations were higher in grafted treatments compared to non-grafted for the following nutrients: N, P, K, Mn, Cu, Zn, and B. When self-

grafted treatments were removed from the comparison, Ca was also higher in the grafted treatments ('Cherokee Purple-Beaufort' + 'Cherokee Purple-Maxifort') compared to the non-grafts. The mean leaf tissue nutrient concentrations were lower in grafted treatments for Mg and Na (Fig. 3.13A & 3.13B).

Differences between the grafting combinations, 'Cherokee Purple-Beaufort' and 'Cherokee Purple-Maxifort' did exist for a select number of nutrients. The 'Cherokee Purple-Maxifort' grafts had higher levels of P, S, Cu, and Zn compared to the 'Cherokee Purple-Beaufort' grafts. The 'Cherokee Purple-Beaufort' grafts had higher levels of Mg and B compared to the 'Cherokee Purple-Maxifort' grafts. Only one nutrient, Fe, showed no differences among treatments (Fig. 3.13A & 3.13B).

In 2007, a grafting and N fertilizer level interaction was present for all grafting treatments (Fig. 3.12). Both Cherokee Purple-Maxifort' grafts and 'Cherokee Purple-Beaufort' grafts had higher mean leaf tissue nutrient concentrations at all three levels of N inputs. In 2008, there was no interaction between leaf tissue N concentrations and different levels of N fertilizer.

Grafting Effects on Fruit Yield

In 2007, grafted plants ('Maxifort-Cherokee Purple' grafts + 'Beaufort-Cherokee Purple' grafts) had higher values of total: harvested fruit (weight and number), marketable fruit (weight and number), culled fruit (weight and number), fruit with blossom end rot

(weight), cat-faced fruit (weight and number), cracked fruit (weight and number), and insect damaged fruit (weight and number) compared to non-grafted plants (Fig. 3.14A & 3.14B).

The ‘total harvest potential’ was also calculated as the sum of the total harvested fruit in combination with the fruit remaining on the vine at the end of the season. The ‘total harvest potential’ (total harvested fruit + fruit remaining on the vine post-harvest) fruit number and weight was greater for grafted plants compared to non-grafted (data not shown). Grafting did not affect the number or weight of fruit remaining on the vines at the end of the season.

‘Maxifort-Cherokee Purple’ grafts had a higher total: harvested fruit (weight and number), marketable fruit (weight), and incidence of insect damage fruit (weight and number) compared to the ‘Beaufort-Cherokee Purple’ grafts.

In 2008, grafted plants (‘Maxifort-Cherokee Purple’ grafts + ‘Beaufort-Cherokee Purple’ grafts + self-grafted ‘Cherokee Purples’) had higher values of total: harvested fruit (weight), marketable fruit (weight), culled fruit (weight), cracked fruit (weight), insect damaged fruit (weight and number) and sun-scalded fruit (number) compared to non-grafted plants (Fig. 3.15A & 3.15B). When self-grafted treatments were removed from the analysis, plants grafted on hybrid rootstocks also had significantly higher values for total: harvested fruit (number), blossom end rot fruit (number), and cracked fruit (number) compared to the non-grafted plants.

In 2008, the fruit remaining on the vines post-harvest (number and weight) was greater for the grafted plants compared to the self-grafted and non-grafted plants (data not shown). The ‘total harvest potential’ (number and weight) was higher for grafted plants

compared to non-grafted. The ‘Beaufort-Cherokee Purple’ grafts had a higher ‘total harvest potential’ (number and weight) compared to the self-grafted and non-grafted treatments. The ‘Maxifort-Cherokee Purple’ grafts had a higher ‘total harvest potential’ (weight) compared to the non-grafted plants. ‘Maxifort-Cherokee Purple’ grafts had a higher total incidence of insect damaged fruit (weight and number) compared to the ‘Beaufort-Cherokee Purple’ grafts.

Grafting Effects on Plant Growth in the High Tunnel System

Shoot weight and plant height measurements were taken in the high tunnel system across both years. In 2007, ‘Cherokee Purple-Maxifort’ grafts had a greater shoot weight compared to non-grafts. ‘Cherokee Purple-Beaufort’ grafts were taller than both ‘Cherokee Purple-Maxifort’ grafts and non-grafts (Fig. 3.16A). In 2008, ‘Cherokee Purple-Maxifort’ grafts had a greater shoot weight compared to non-grafts. Both the ‘Cherokee Purple-Maxifort’ grafts and the ‘Cherokee Purple-Beaufort’ grafts were taller than the non-grafts (Fig. 3.16B).

Grafting Effects, N Concentration, and Fruit Yield

In 2007, the mean leaf tissue N concentration was highly correlated with the mean total fruit yield (70.9%) (Fig. 3.9A). In 2008, the mean leaf tissue N concentration was highly correlated with the mean total fruit yield (70.2%). These relationships indicate a strong relationship between % N concentration and grafting onto hybrid rootstocks that in

turn boosts fruit yield (Fig. 3.9B).

The mean leaf tissue N concentration was also highly correlated with the ‘total harvest potential’ fruit number and weight, 58.1% and 74.3%, respectively. The mean N content of the shoot (mean leaf N concentration (%) * shoot dry weight (g)) was highly correlated with the total harvested fruit weight (53.7%) and the ‘total harvest potential’ fruit weight (58.3%).

System and Grafting Effects Together

In 2007, all grafted and non-grafted treatments produced a higher total number of fruit in the high tunnels compared to the field system. The ‘Maxifort-Cherokee Purple’ grafts had a higher total harvested fruit weight in the high tunnel system compared to the field system. The ‘Beaufort-Cherokee Purple’ grafts and the non-grafted plants had a lower total marketable fruit weight in the high tunnel system compared to the field system.

In 2008, all grafted and non-grafted treatments produced a higher total weight and number of fruit as well as higher total marketable weight and number of fruit in the high tunnels compared to the field system. The self-grafted and non-grafted plants had higher levels of total fruit weight and number with blossom end rot and total fruit number with cracking in the high tunnel system compared to the field system (Fig. 3.20A).

Discussion and Conclusions

System Effects

The system effect on leaf tissue nutrient concentration showed opposite trends for many macronutrients in 2007 compared to 2008. Although both systems were planted with the same winter cover crop mixture comprised of hairy vetch (*Vicia villosa*) and cereal rye (*Secale cereale*), at the same rate, on the same date, and managed similarly, the growth of the cover crop in each system was nevertheless very different. The growth in the high tunnel system was 43% greater than the growth in the field system leading to a supplemental application of feathermeal in 2008 to the field system only. Possible factors for the differential macronutrient availability that was reflected in the varying leaf tissue concentration trends across the systems by year include:

1) a difference in material decomposition rates (cover crop vs. cover crop and feathermeal mix), 2) a difference in the amount of cover crop material incorporated into each system, and 3) mineralization rates regulated by the microbial community possibly influenced by differing soil moisture levels and soil temperatures.

The high tunnel system produced higher fruit yields compared to the field system. The high tunnel system produced their first peak of fruit approximately three weeks earlier than the field systems in time to capture an early summer market. The categorical breakdown of non-marketable fruit revealed different management challenges in each system type. The high tunnel system had a greater incidence of blossom end rot, cat-facing, and cracking while the field system had greater incidence of TSWV and insect damage compared

to the field system. All grafted and non-grafted treatments grown in the high tunnel system had greater yields (number) compared to the field system. The greatest yield gains were seen with the 'Maxifort-Cherokee Purple' grafts grown in the high tunnel system.

N Input Level Effects

During the first season, N input level had little effect on total fruit yield. In 2008, after the downward adjustment across all N input levels and improvements in post-plant fertilizer application (change from application by hand to application via drip irrigation), and yields effects became more pronounced. The high N and medium N input levels resulted in total fruit yields (weight and number) compared to the low N input level, indicating that N input levels above 122 kg ha⁻¹ can attain comparable yields to applications up to 168 kg ha⁻¹.

The N concentration captured in leaf tissue samples proved to be highly correlated with total fruit yield, reaffirming its use as a tool for crop management and the relationship between plant growth, yield, and N availability ($p < 0.0001$). However, the mean nutrient concentration values for N were within or above the adequate range across all treatments for the entire growing season. This suggests that the reference ranges (Hochmuth et al., 1991) were too broad to identify the optimum range of leaf tissue nutrient concentrations for maximum crop yields within our particular system. In 2008, the mean N content (mean leaf N concentration x shoot dry weight) was also correlated with total fruit yield, but the association was not as strong as the N concentration to total fruit yield relationship.

In both 2007 and 2008, no effects on plant growth among the different N input levels

were present, however the mean N content of the shoot was highly correlated with shoot weight ($p < 0.0001$). This relationship reinforces the assertion that it is the amount of N the plant assimilates rather than the determining factor for plant growth.

Grafting Effects

The majority of patterns for nutrient accumulation in the leaf tissue were consistent across the two experimental years. The mean leaf tissue concentrations were higher for N, P, K, Mn, Cu, Zn and B in grafted plants compared to non-grafted plants in 2007 and 2008. Grafting resulted in a decrease of Mg and Na concentrations in the leaf tissue compared to non-grafted plants. Our results indicated that both ‘Cherokee Purple-Maxifort’ and ‘Cherokee Purple-Beaufort’ grafts have higher salinity tolerances compared to non-grafted and self-grafted ‘Cherokee Purple’ plants. These results are consistent with existing literature on the topic (Estan et. al, 2005).

Different rootstock-scion combinations showed varying leaf tissue nutrient concentrations in some instances indicating that the scion selection influences nutrient uptake likely via systemic feedback controls between the shoot and root systems. Self-grafted treatments were present only in 2008. The self-grafted and non-grafted treatments were not different from each other in terms of nutrient concentration comparisons. This result was a bit unexpected as self-grafted treatments in an earlier greenhouse study demonstrated similar or intermediate levels of response to grafting on commercial rootstocks for selected nutrient uptake and plant growth (see Chapter 2). Differences between the two studies that may

account for the variable performance of the self-grafts include: 1) different scion choices, 2) a shorter study duration, 3) organic versus synthetic fertilizers usage, 4) soil-less potting media versus soil culture, and 5) controlled environmental growth chambers versus a field environment.

The ‘Maxifort-Cherokee Purple’ grafts and the ‘Beaufort-Cherokee Purple’ grafts produced greater total amounts of fruit weight (Fig. 4.12A & 4.12B) and fruit number compared to non-grafts in both growing seasons. This finding is supported by previous work with herbaceous grafted plants in greenhouse and conventional agriculture systems (Besri, 2005; Black, 2003; Burleigh et al. 2005; Colla et al., 2008; Kato and Lou, 1989; Lee, 1994; Matsuzoe, 1993; Romero et al. 1997; Ruiz et al., 2006). In addition, grafted plants had a higher ‘total harvest potential’ (harvested fruit + un-ripe green fruit on the vine at the end of the harvest season) compared to the non-grafted plants indicating that the grafted plants may be capable of a longer, extended harvest season.

Fruit from ‘Maxifort-Cherokee Purple’ grafts were more susceptible to insect damage compared to ‘Beaufort-Cherokee Purple’ grafts and non-grafts. The majority of insect damage in both years was by lepidopterous insects. Perhaps there are differences with phytochemical mediators, canopy characteristics, or fruit appearance among the grafting treatments that increase insect attraction?

Grafted plants in the high tunnel system were taller and had a greater shoot weight compared to non-grafts. The shoot weight effect was greatest with the ‘Maxifort-Cherokee Purple’ grafts indicating that there are differences between rootstock varieties in their

influence on plant growth. An increase in vegetative growth stimulated by grafting may affect biomass allocation and subsequently both total fruit yield and the size of individual fruits (Heuvelink, 2005). It has been reported that an increase in vegetative growth may restrict the assimilate pool available for fruit production and thus limit yield (Heuvelink, 2005). However in our study grafted plants had greater plant growth as well as greater fruit yield suggesting that one factor can work in synergy with the other rather than competition.

Grafting and Nutrient Uptake

‘Cherokee Purple’ scion grafted on two hybrid rootstocks had higher levels of nutrient uptake for the three primary macronutrients (N, P, K) as well as selected essential micronutrients compared to self-grafted and non-grafted plants. These results indicate that regional recommendation levels ($\sim 224 \text{ kg ha}^{-1}$) are relatively high compared to what was necessary for this crop to fall within the standards for adequate nutrient concentrations in the leaf tissue and produce high yields ($+122 \text{ kg N ha}^{-1}$). Because neither the grafted nor non-grafted plants experienced a N deficiency, the minimum N sufficiency limit could not be identified.

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Table 3.1. CEFS Seeding Production Schedule.

<i>Seeding</i>	<i>Transplanting #1</i>	<i>Grafting</i>	<i>Transplanting #2</i>	<i>Planting in High Tunnel</i>	<i>Planting in Field</i>
2007					
5 Feb.	12 Feb.	22 Feb.	9 Mar.	20 Mar.	
8 Mar.	15 Mar.	29 Mar.	12 April		19 April
2008					
31 Jan.	9 Feb.	23 Feb.	8 Mar.	18 Mar.	
6 Mar.	13 Mar.	24 Mar.	3 April		17 April

Table 3.2. Estimated Nitrogen Contributions from Pre-plant Organic Amendments, 2007 & 2008.

<i>Material</i>	<i>Application Rate</i>	<i>Plant Biomass (dry wt.)</i>	<i>% N (estimated availability)</i>	<i>Total N Contribution</i>	<i>Material</i>	<i>Application Rate</i>	<i>Plant Biomass (dry wt.)</i>	<i>% N (estimated availability)</i>	<i>Total N Contribution</i>
2007									
High Tunnel					Field Plot				
Compost	22.6 t ha ⁻¹	-----	2.2 % (70%) ^z	28 kg ha ⁻¹	Compost	22.6 t ha ⁻¹	-----	2.2 % (70%)	28 kg ha ⁻¹
Feathermeal	101.7 kg ha ⁻¹	-----	12 % (70%) ^y	71 kg ha ⁻¹	Feathermeal	101.7 kg ha ⁻¹	-----	12 % (70%)	71 kg ha ⁻¹
				Total 99 kg ha⁻¹					Total 99 kg ha⁻¹
2008									
High Tunnel					Field Plot				
Cover crop	50.4 kg ha ⁻¹ ;	4,202.5	4.0% (50%) ^x ;	93 kg ha ⁻¹	Cover crop	50.4 kg ha ⁻¹ ;	809.1 kg	2.6%	53 kg ha ⁻¹
(<i>Secale</i>	33.6 kg ha ⁻¹	kg ha ⁻¹ ;	5.1% (50%) ^x		(<i>Secale</i>	33.6 kg ha ⁻¹	ha ⁻¹ ;	(50%);	
<i>cereal</i> ; <i>Vicia</i>		368.8 kg			<i>cereal</i> ; <i>Vicia</i>		1,781.1	4.7% (50%)	
<i>villosa</i>)		ha ⁻¹			<i>villosa</i>)		kg ha ⁻¹		
					Feathermeal	58.9 kg ha ⁻¹		12 % (70%)	41 kg ha ⁻¹
				Total 94 kg ha⁻¹					Total 94 kg ha⁻¹

^z Estimates of nitrogen availability coefficient based on NCDA&CS Waste Analysis Report #W07714 & Baldwin and Greenfield, 2006. 'Composting on Organic Farms', NC Cooperative Extension Pub. AG-659W-01

^y Estimates of nitrogen availability coefficient based on NCDA&CS Waste Analysis Report #W07264 & Zublena et al., 1993. 'SoilFacts, Poultry Manure as a Fertilizer Source', NC Cooperative Extension Pub. AG-439-5 (rev.)

^x Estimates of nitrogen availability coefficient based on NCDA&CS, Plant Tissue Analysis Reports #P00880, #P01101 & Baldwin and Creamer, 2006. 'Cover crops for Organic Farms', NC Cooperative Extension Pub. AG-659W-03

Table 3.3A. Fertigation Protocol, CEFS 2007.

Low N <i>crop stage</i>	<i>DAT</i>	<i>N (kg ha⁻¹)</i>	<i>Source</i>	<i>High Tunnel</i>	<i>Field</i>
stand establishment	0-35	98.8	pre-plant (feathermeal + compost)	8-Mar-07	6-Apr-07
pre-bloom	36-49	none	none		
1st bloom to 1st harvest	50-63	5.6	Phytamin 801	4-May-07	1-Jun-07
1st bloom to 1st harvest	64-77	4.4	Phytamin 801	18-May-07	14-Jun-07
early harvest	78-91	1.1	Phytamin 801	1-Jun-07	28-Jun-07
early harvest	92-105	1.1	Phytamin 801	14-Jun-07	12-Jul-07
late harvest	106-119	1.1	Phytamin 801	28-Jun-07	26-Jul-07
Total:		112.3			
Medium N <i>crop stage</i>	<i>DAT</i>	<i>N (kg ha⁻¹)</i>	<i>Source</i>	<i>High Tunnel</i>	<i>Field</i>
stand establishment	0-35	98.8	pre-plant (feathermeal + compost)	8-Mar-07	6-Apr-07
pre-bloom	36-49	none	none		
1st bloom to 1st harvest	50-63	19.0	Phytamin 801	4-May-07	1-Jun-07
1st bloom to 1st harvest	64-77	19.0	Phytamin 801	18-May-07	14-Jun-07
early harvest	78-91	13.4	Phytamin 801	1-Jun-07	28-Jun-07
early harvest	92-105	13.4	Phytamin 801	14-Jun-07	12-Jul-07
late harvest	106-119	4.4	Phytamin 801	28-Jun-07	26-Jul-07
Total:		168.3			
High N <i>crop stage</i>	<i>DAT</i>	<i>N (kg ha⁻¹)</i>	<i>Source</i>	<i>High Tunnel</i>	<i>Field</i>
stand establishment	0-35	98.8	pre-plant (feathermeal + compost)	8-Mar-07	6-Apr-07
pre-bloom	36-49		none		
1st bloom to 1st harvest	50-63	39.2	Phytamin 801	4-May-07	1-Jun-07
1st bloom to 1st harvest	64-77	39.2	Phytamin 801	18-May-07	14-Jun-07
early harvest	78-91	19.0	Phytamin 801	1-Jun-07	28-Jun-07
early harvest	92-105	19.0	Phytamin 801	14-Jun-07	12-Jul-07
late harvest	106-119	8.9	Phytamin 801	28-Jun-07	26-Jul-07
Total:		224.3			

Table 3.3B. Fertigation Protocol, CEFS 2008.

Low N <i>crop stage</i>	<i>DAT</i>	<i>N (kg ha⁻¹)</i>	<i>Source</i>	<i>High Tunnel</i>	<i>Field</i>
stand establishment	0-35	93.0	pre-plant (feathermeal + compost)	28-Feb -08	1-Apr-08
pre-bloom	36-49	none			
1st bloom to 1st harvest	50-63	none			
1st bloom to 1st harvest	64-77	none			
continued harvest	78-91	none			
continued harvest	92-105	none			
continued harvest	106-119	none			
Total:		93.0			
Medium N <i>crop stage</i>	<i>DAT</i>	<i>N (kg ha⁻¹)</i>	<i>Source</i>	<i>High Tunnel</i>	<i>Field</i>
stand establishment	0-35	93.0	pre-plant (feathermeal + compost)	28-Feb -08	1-Apr-08
pre-bloom	36-49	none			
1st bloom to 1st harvest	50-63	none			
1st bloom to 1st harvest	64-77	9.5	Phytamin 801	27-May-08	24-Jun-08
continued harvest	78-91	9.5	Phytamin 801	10-Jun-08	8-Jul-08
continued harvest	92-105	none			
continued harvest	106-119	none			
Total:		112.0			
High N <i>crop stage</i>	<i>DAT</i>	<i>N (kg ha⁻¹)</i>	<i>Source</i>	<i>High Tunnel</i>	<i>Field</i>
stand establishment	0-35	93.0	pre-plant (feathermeal + compost)	28-Feb -08	1-Apr-08
pre-bloom	36-49	none			
1st bloom to 1st harvest	50-63	none			
1st bloom to 1st harvest	64-77	37.5	Phytamin 801	27-May-08	24-Jun-08
continued harvest	78-91	37.5	Phytamin 801	10-Jun-08	8-Jul-08
continued harvest	92-105	none			
continued harvest	106-119	none			
Total:		168.0			

Table 3.4. CEFS Leaf Tissue Sampling Schedule.

	<i>Date 1</i>	<i>Date 2</i>	<i>Date3</i>	<i>Date 4</i>	<i>Date 5</i>	<i>Date 6</i>
High Tunnel						
2007						
	26 April	10 May	24 May	7 June	21 June	16 Aug.
2008						
	17 April	12 May	5 June	26 June	7 Aug.	none
Field						
2007						
	31 May	7 June	21 June	5 July	19 July	16 Aug.
2008						
	12 May	5 June	26 June	25 July	7 Aug.	none

NE High Tunnel 2007	
guard row	guard row
D5^{zy}	D2
D17	D4
D14	D1
D6	D12
D13	D9
D18	D10
D7	C3
D15	C11
D16	C14
D8	C15
D3	C18
D11	C13
C1	C17
C4	C5
C2	C7
C12	C16
C10	C6
C9	C8
guard row	guard row

SE Field Plot 2007	
guard row	guard row
D105	D102
D117	D104
D114	D101
D106	D112
D113	D109
D118	D110
D107	C103
D115	C111
D116	C114
D108	C115
D103	C118
D111	C113
C101	C117
C104	C105
C102	C107
C112	C116
C110	C106
C109	C108
guard row	guard row

NW High Tunnel 2007	
guard row	guard row
B12	B18
B10	B13
B9	B16
B2	B6
B4	B5
B1	B14
A3	B15
A11	B11
A15	B7
A7	B8
A16	B3
A13	B11
A18	A12
A6	A9
A17	A10
A14	A4
A5	A1
A8	A2
guard row	guard row

SW Field Plot 2007	
guard row	guard row
B112	B118
B110	B113
B109	B116
B102	B106
B104	B105
B101	B114
A103	B115
A111	B111
A115	B107
A107	B108
A116	B103
A113	B111
A118	A112
A106	A109
A117	A110
A114	A104
A105	A101
A108	A102
guard row	guard row

**2007 Treatment Code
High Tunnel System**

Treatment Number	Planting Date	Grafting Rootstock ^x	Nitrogen Level
1	18-Mar	Maxifort	Low
2	18-Mar	Beaufort	Low
4	18-Mar	Non-grafted	Low
5	18-Mar	Maxifort	Standard
6	18-Mar	Beaufort	Standard
8	18-Mar	Non-grafted	Standard
9	18-Mar	Maxifort	High
10	18-Mar	Beaufort	High
12	18-Mar	Non-grafted	High

Field System

Treatment Number	Planting Date	Grafting Rootstock	Nitrogen Level
101	17-Apr	Maxifort	Low
102	17-Apr	Beaufort	Low
104	17-Apr	Non-grafted	Low
105	17-Apr	Maxifort	Standard
106	17-Apr	Beaufort	Standard
108	17-Apr	Non-grafted	Standard
109	17-Apr	Maxifort	High
110	17-Apr	Beaufort	High
112	17-Apr	Non-grafted	High

^z Letters A,B,C,D denote one of four replications

^y Bolded treatments were included in this study

^x All treatments had 'Cherokee Purple' scion and either 'Maxifort' rootstock, 'Beaufort' rootstock, or were not grafted 'Non-grafted' as listed above.

Fig 3.1A. CEFS Experimental Plot Plan, 2007.

NE High Tunnel 2008		SE Field Plot 2008	
guard row	guard row	guard row	guard row
D7^{zy}	D11	D107	D111
D6	D14	D106	D114
D8	D12	D108	D112
D5	D18	D105	D118
D2	D16	D102	D116
D3	D9	D103	D109
D1	D13	D101	D113
D4	D17	D104	D117
C12	D15	C112	D115
C15	D10	C115	D110
C17	C7	C117	C107
C18	C6	C118	C106
C9	C8	C109	C108
C13	C5	C113	C105
C16	C4	C116	C104
C11	C3	C111	C103
C10	C1	C110	C101
C14	C2	C114	C102
guard row	guard row	guard row	guard row

2008 Treatment Code			
High Tunnel System			
Treatment Number	Planting Date	Grafting Rootstock ^x	Nitrogen Level
1	18-Mar	Maxifort	Low
2	18-Mar	Beaufort	Low
3	18-Mar	Self-grafted	Low
4	18-Mar	Non-grafted	Low
5	18-Mar	Maxifort	Standard
6	18-Mar	Beaufort	Standard
7	18-Mar	Self-grafted	Standard
8	18-Mar	Non-grafted	Standard
9	18-Mar	Maxifort	High
10	18-Mar	Beaufort	High
11	18-Mar	Self-grafted	High
12	18-Mar	Non-grafted	High

NW High Tunnel 2008		SW Field Plot 2008	
guard row	guard row	guard row	guard row
B9	B3	B109	B103
B12	B4	B112	B104
B11	B1	B111	B101
B17	B2	B117	B102
B10	B5	B110	B105
B14	B6	B114	B106
B16	B7	B116	B107
B15	B8	B115	B108
B18	A18	B118	A118
B13	A10	B113	A110
A3	A11	A103	A111
A1	A12	A101	A112
A4	A17	A104	A117
A2	A14	A102	A114
A6	A9	A106	A109
A7	A16	A107	A116
A8	A15	A108	A115
A5	A13	A105	A113
guard row	guard row	guard row	guard row

Field System			
Treatment Number	Planting Date	Grafting Rootstock	Nitrogen Level
101	17-Apr	Maxifort	Low
102	17-Apr	Beaufort	Low
103	17-Apr	Self-grafted	Low
104	17-Apr	Non-grafted	Low
105	17-Apr	Maxifort	Standard
106	17-Apr	Beaufort	Standard
107	17-Apr	Self-grafted	Standard
108	17-Apr	Non-grafted	Standard
109	17-Apr	Maxifort	High
110	17-Apr	Beaufort	High
111	17-Apr	Self-grafted	High
112	17-Apr	Non-grafted	High

Fig 3.1B. CEFS Experimental Plot Plan, 2008.

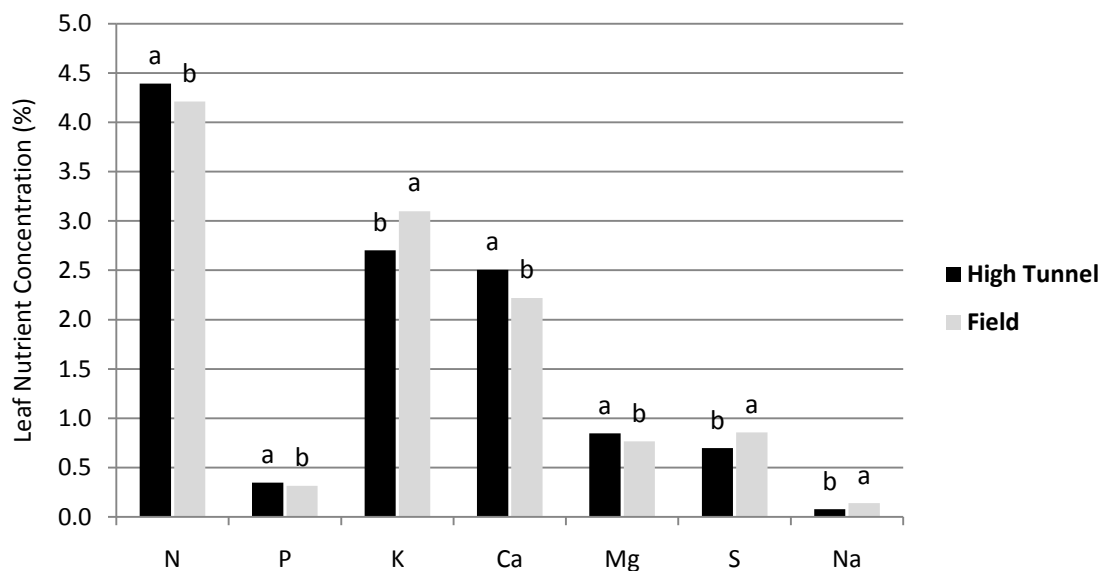


Fig. 3.2 A. System Effect on Mean Leaf Tissue Macro-nutrient Concentrations, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

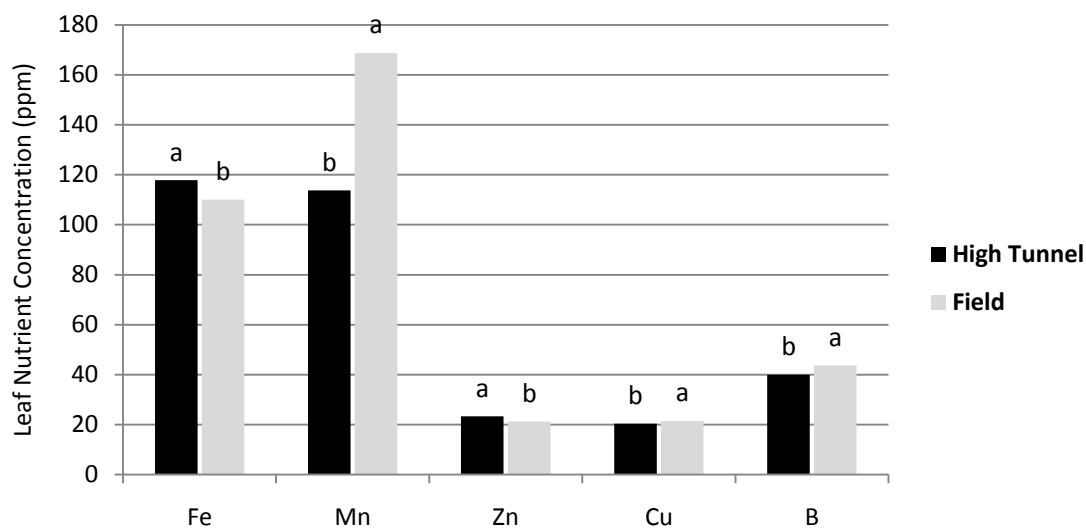


Fig. 3.2B. System Effect on Mean Leaf Tissue Micro-nutrient Concentrations, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

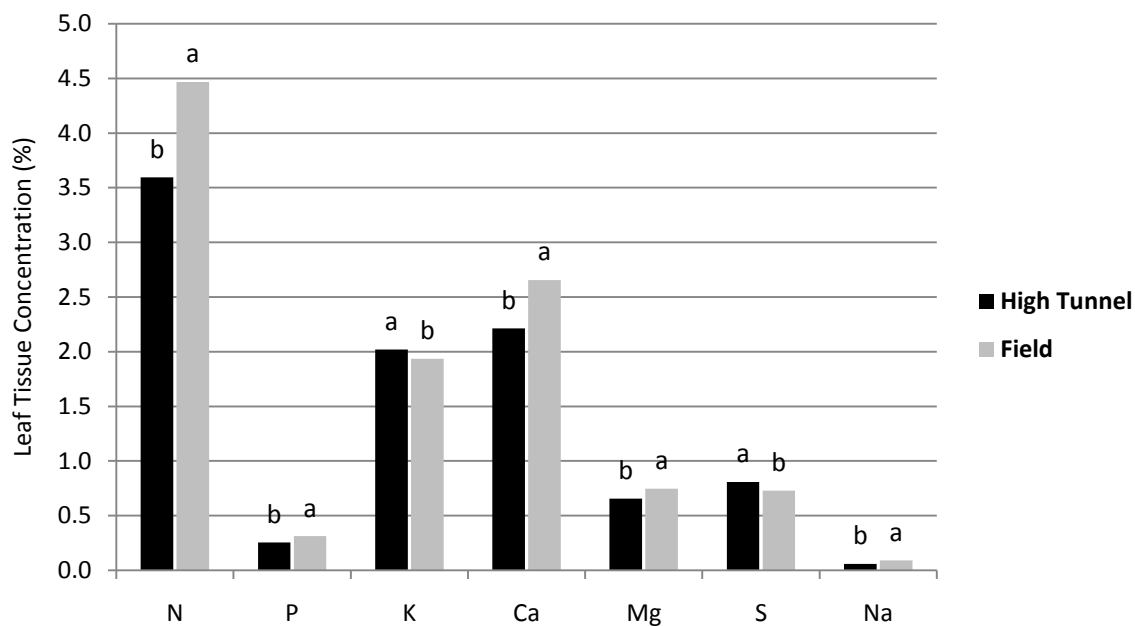


Fig. 3.3A. System Effect on Mean Leaf Tissue Macro-nutrient Concentrations, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

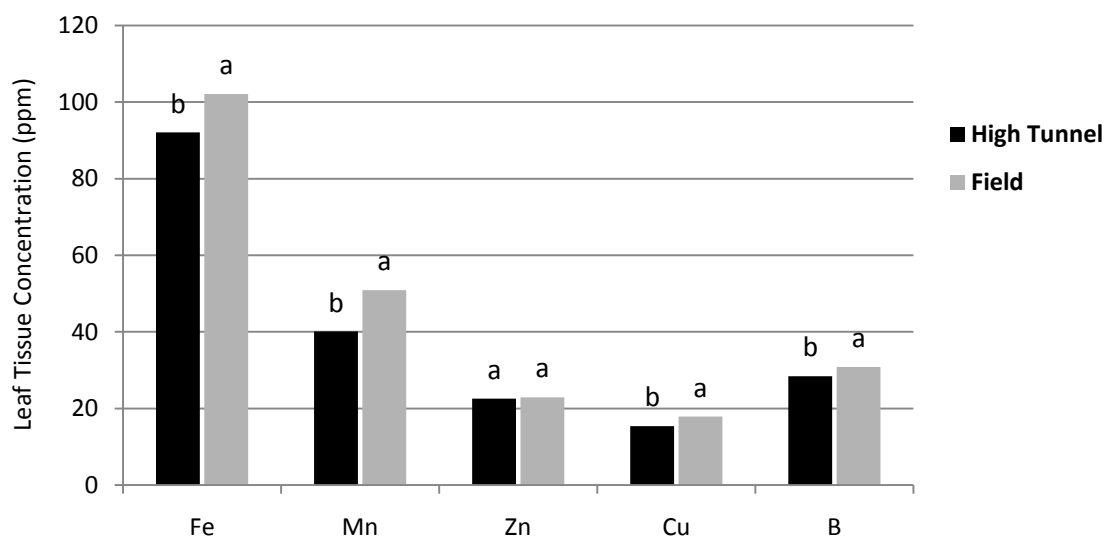


Fig. 3.3B. System Effect on Mean Leaf Tissue Micro-nutrient Concentrations, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

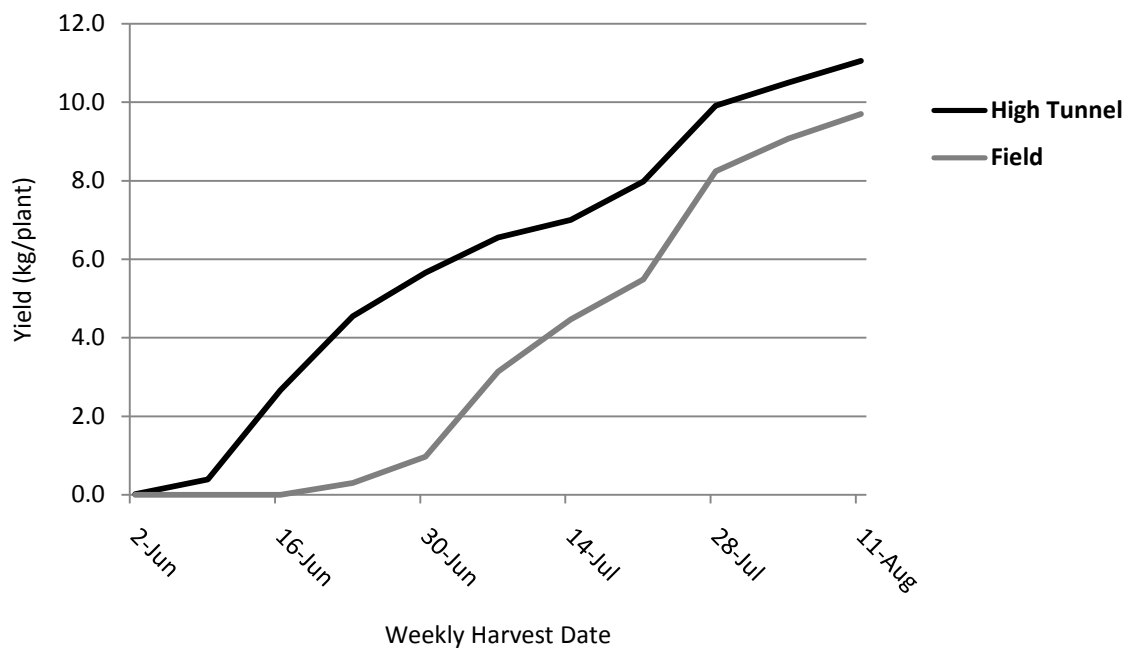


Fig. 3.4A. System Effect on Total Cumulative Fruit Yield, 2007.

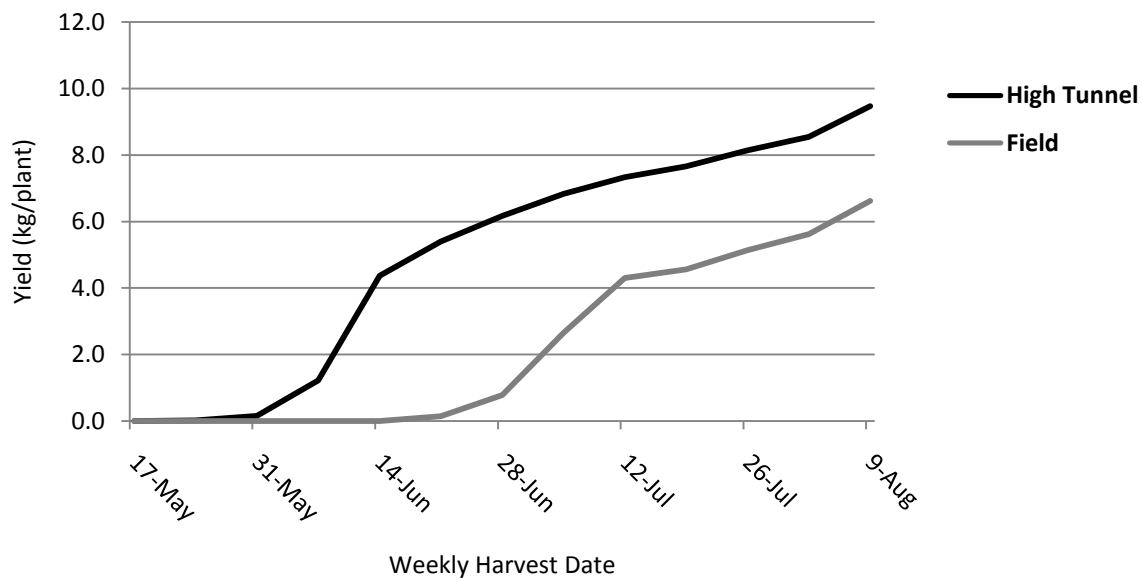


Fig.3.4B. System Effect on Total Cumulative Fruit Yield, 2008.

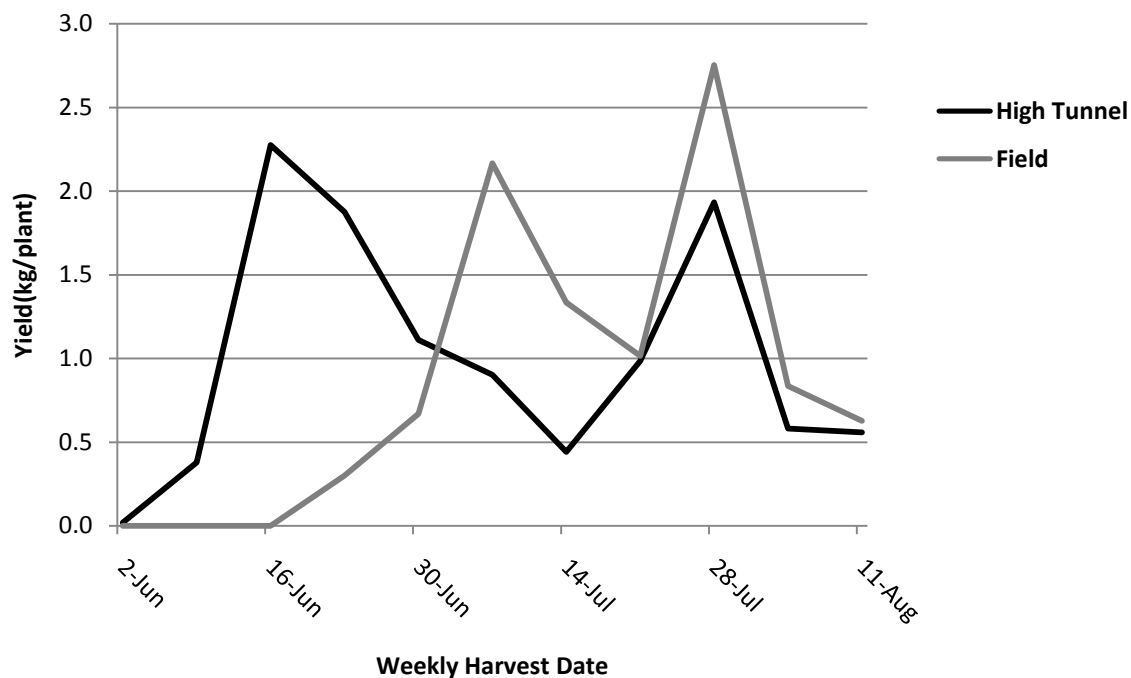


Fig. 3.5A. System Effect on Mean Fruit Yield Over Time, 2007.

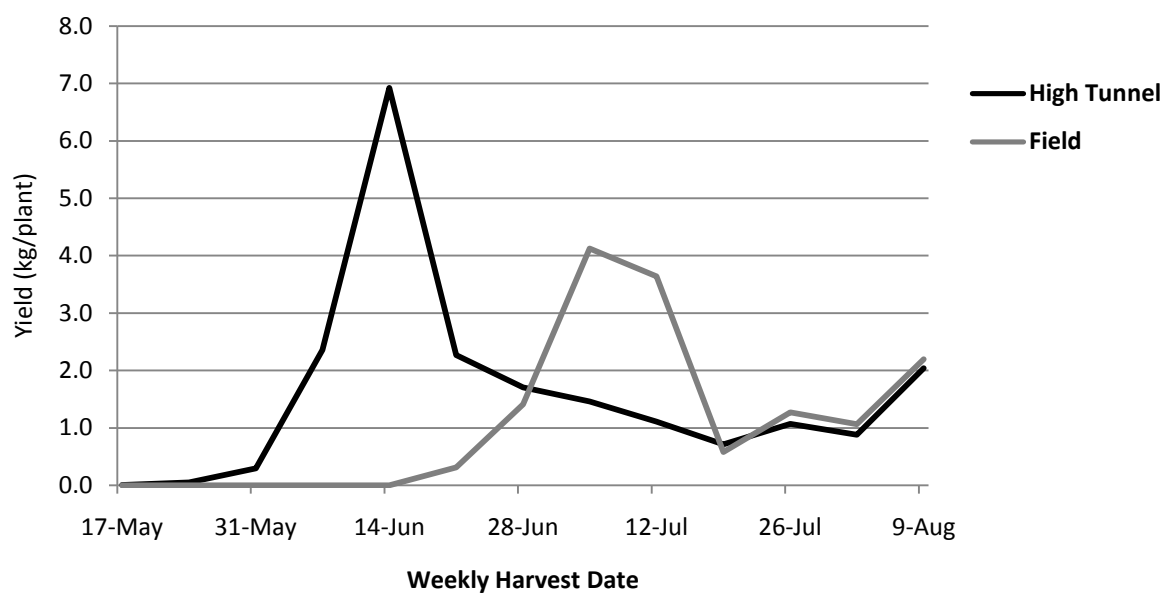


Fig. 3.5B. System Effect on Mean Fruit Yield Over Time, 2008.

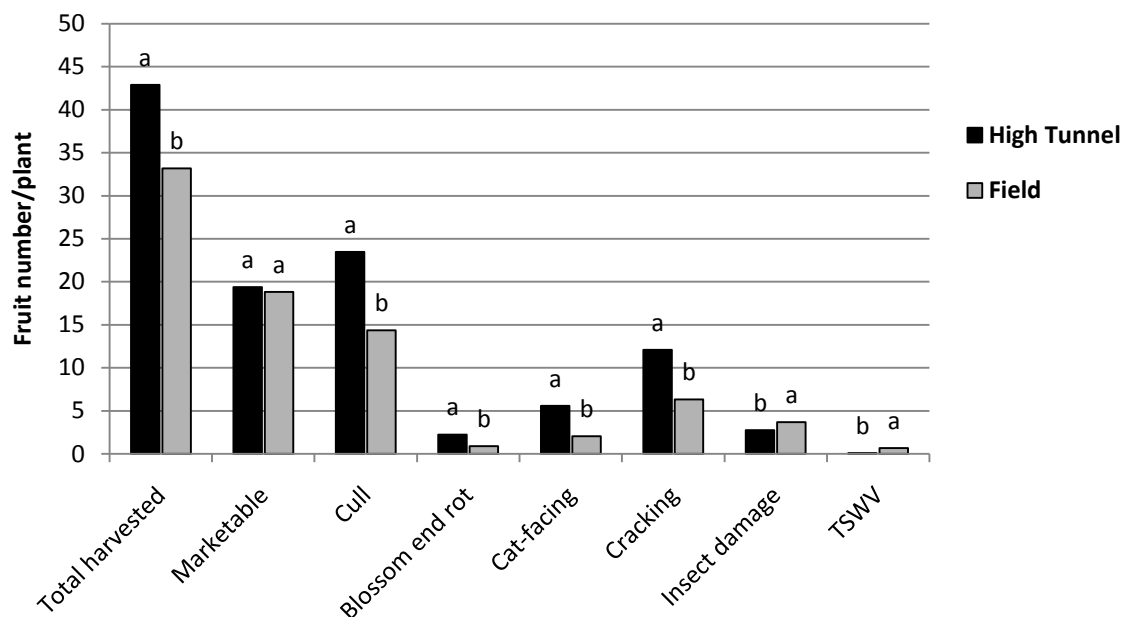


Fig. 3.6A. System Effect on Mean Fruit Number, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

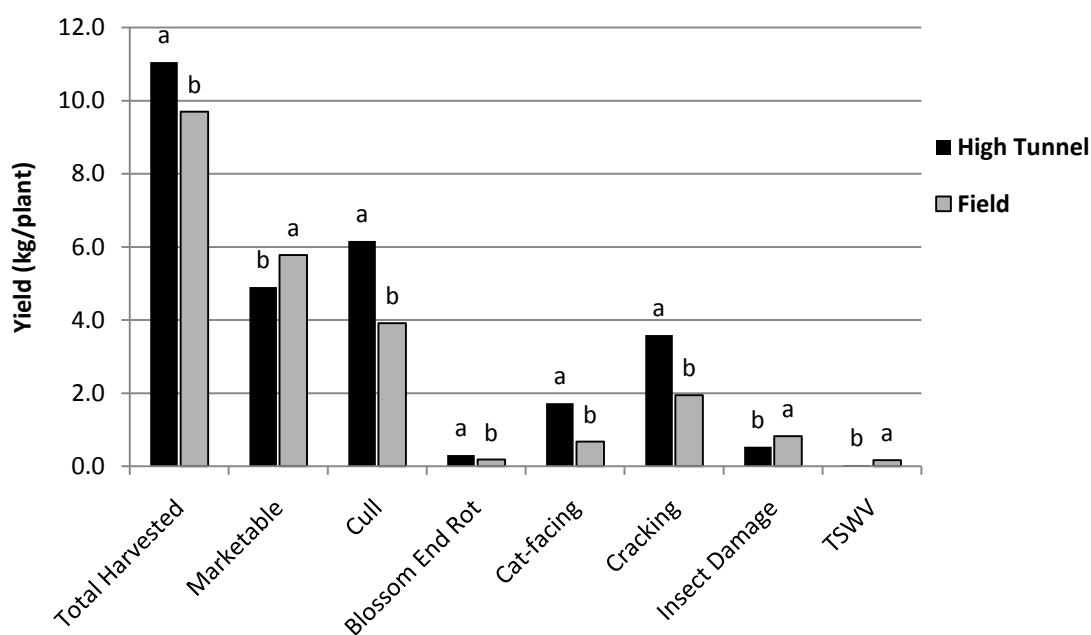


Fig. 3.6B. System Effect on Mean Fruit Weight, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

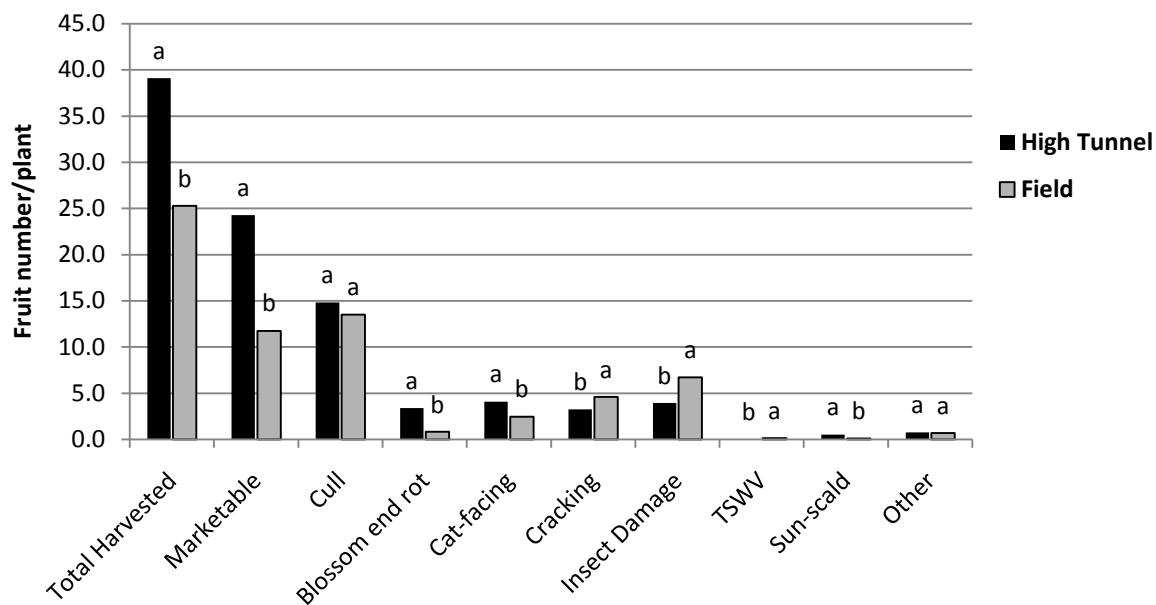


Fig. 3.7A. System Effect on Mean Fruit Number, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

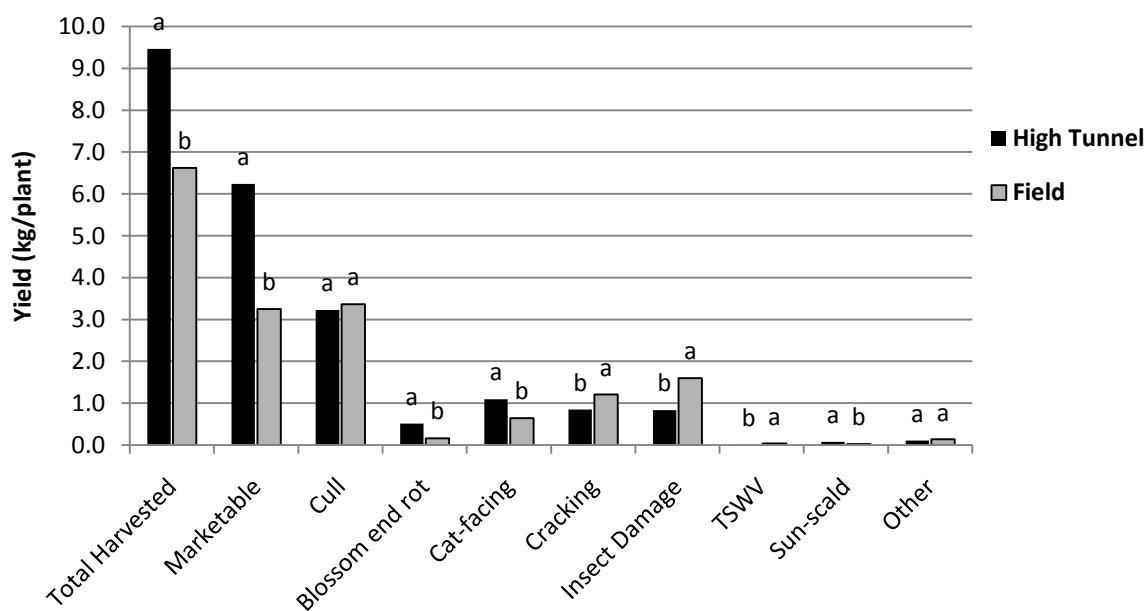


Fig. 3.7B. System Effect on Mean Fruit Weight, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

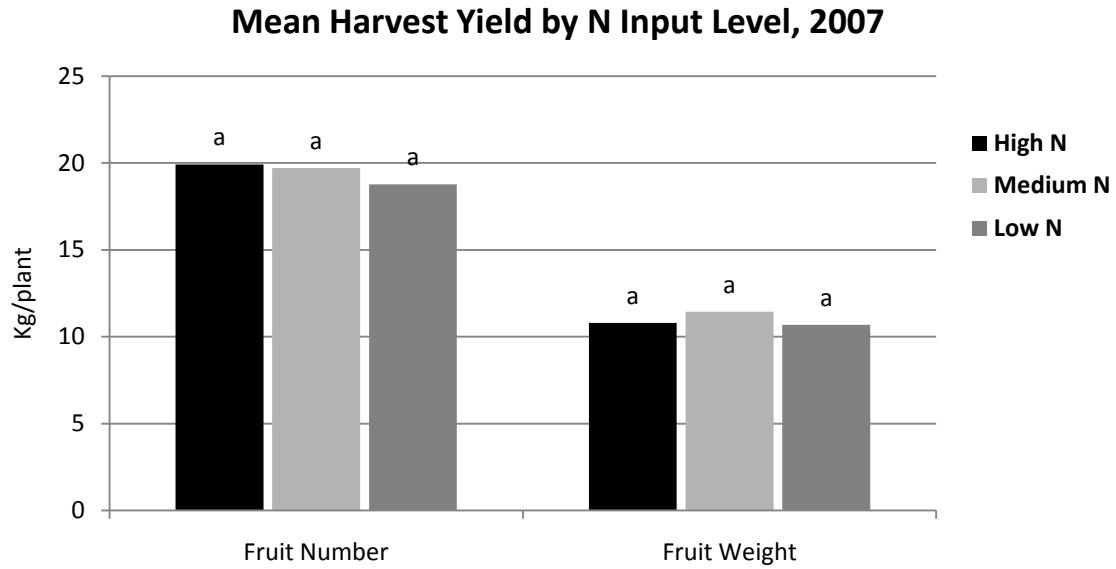


Fig. 3.8A. Mean Total Harvest Yield by N Input Level, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

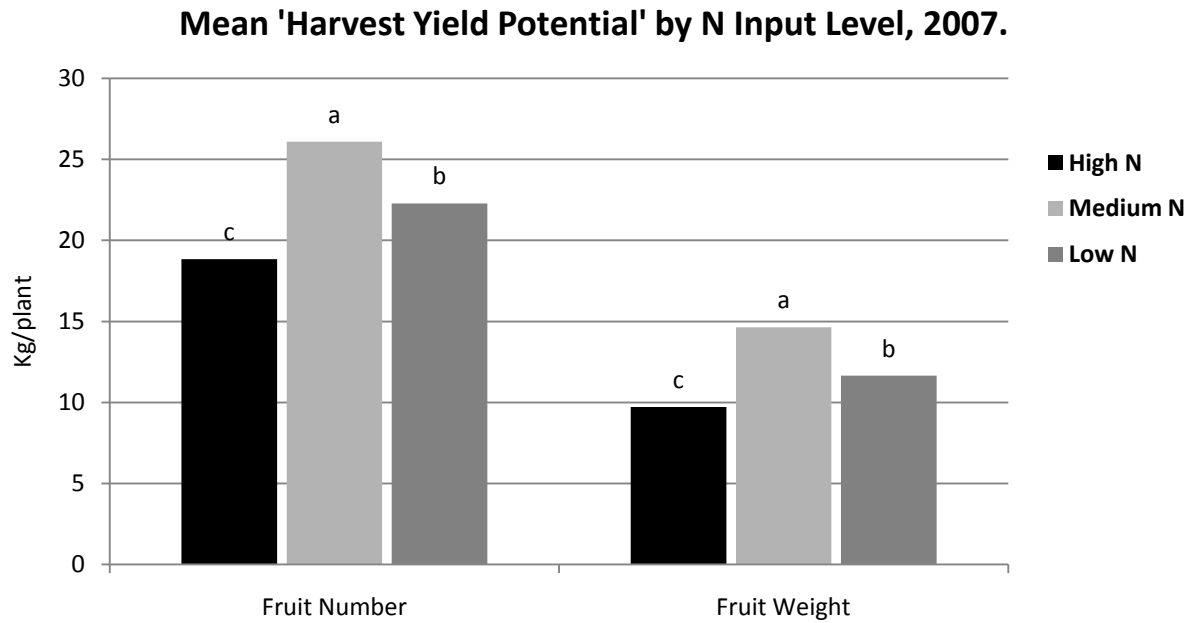


Fig.3.8B. Mean 'Harvest Yield Potential' by N Input Level, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

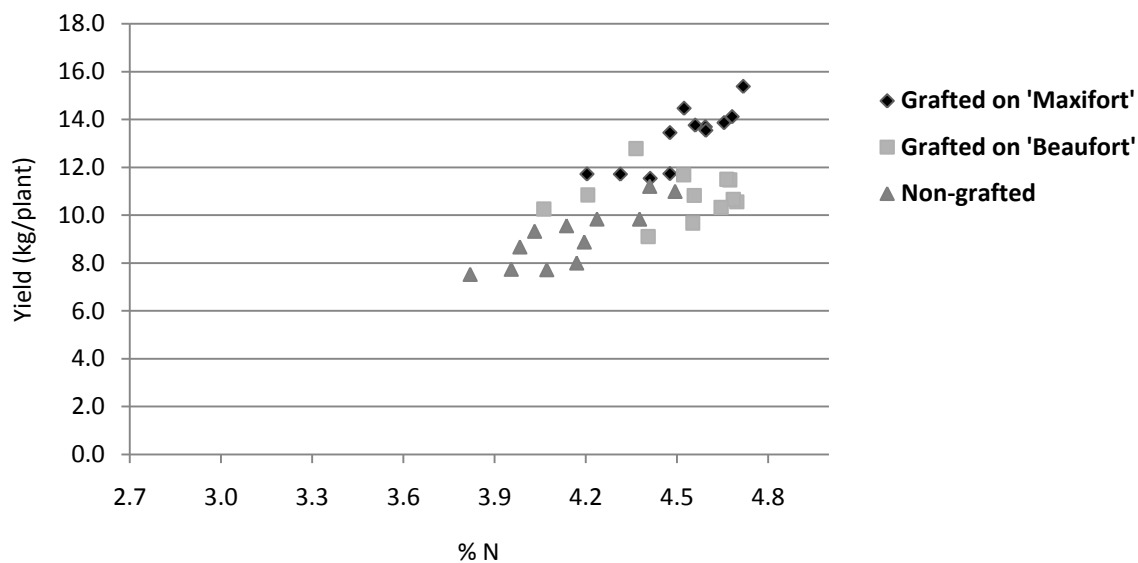


Fig. 3.9A. Mean Nitrogen Concentration of Grafting Treatments across both Growing Systems by Total Cumulative Fruit Yield, 2007.

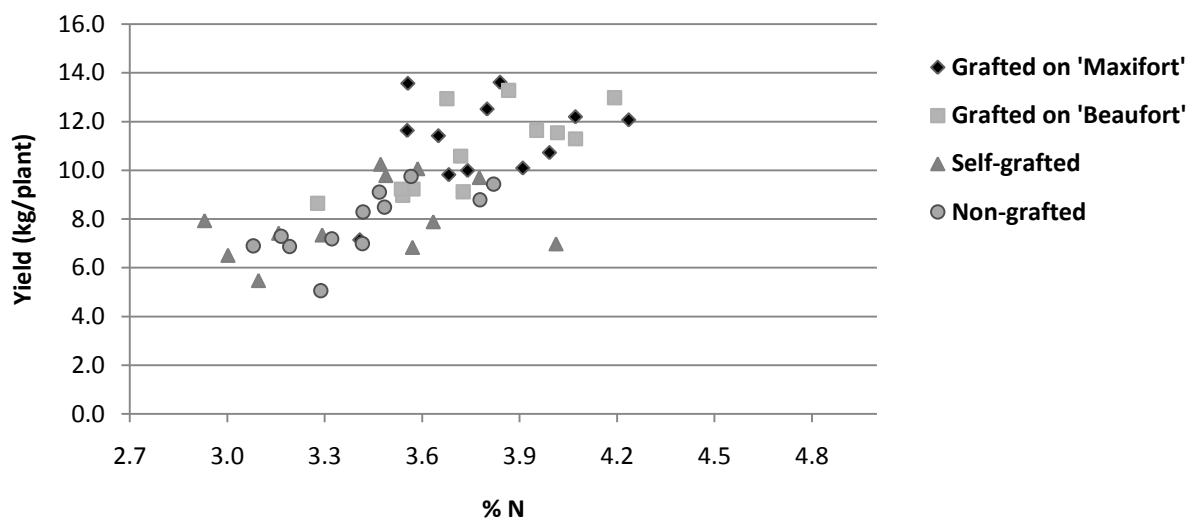


Fig. 3.9B. Mean Nitrogen Concentration of Grafting Treatments across both Growing Systems by Total Cumulative Fruit Yield, 2008.

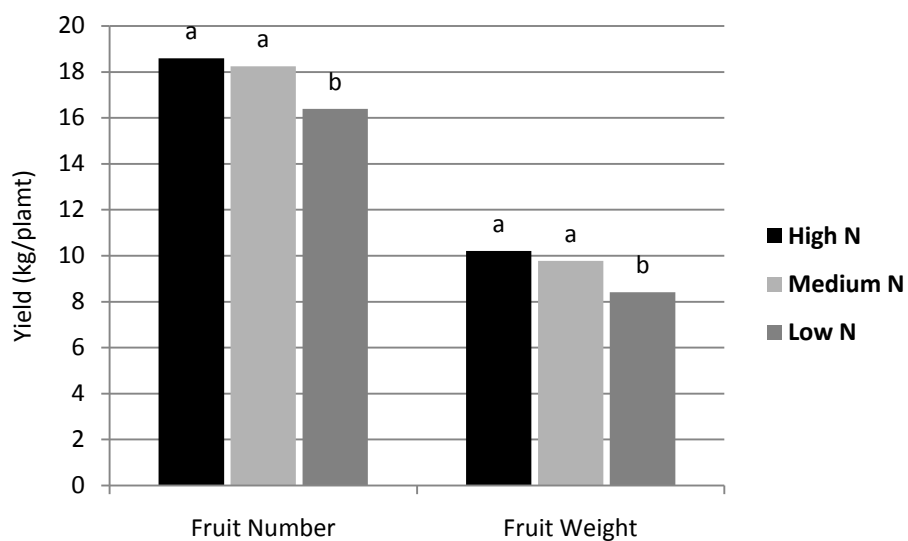


Fig. 3.10. Total Harvested Fruit by Nitrogen Input Level, 2008. N Input Levels: High N (186 kg ha^{-1}), Medium N (122 kg ha^{-1}), Low N (93 kg ha^{-1}). Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

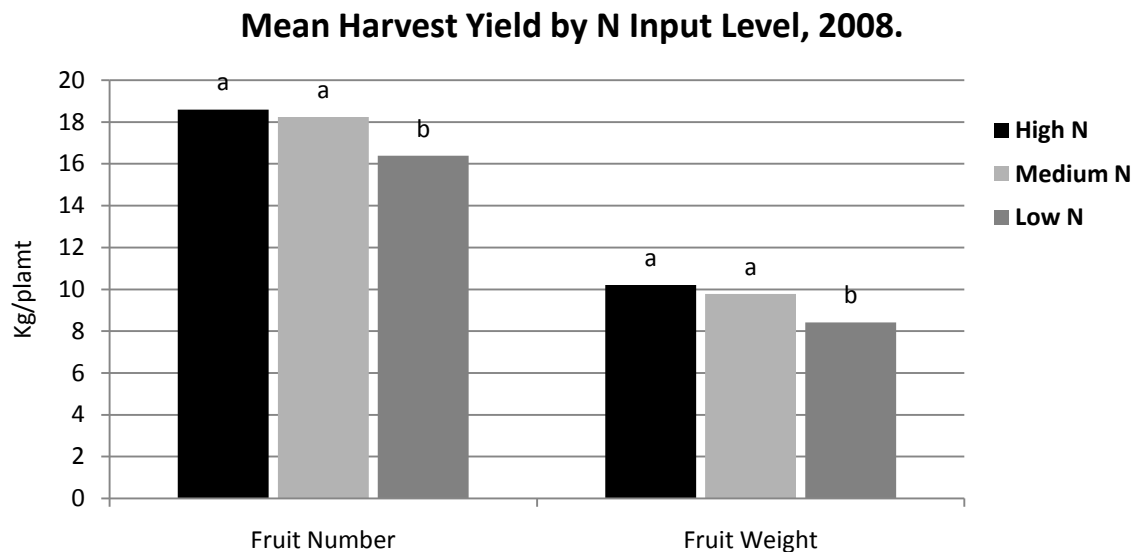


Fig.3.11A. Mean Harvest Yield by N Input Level, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

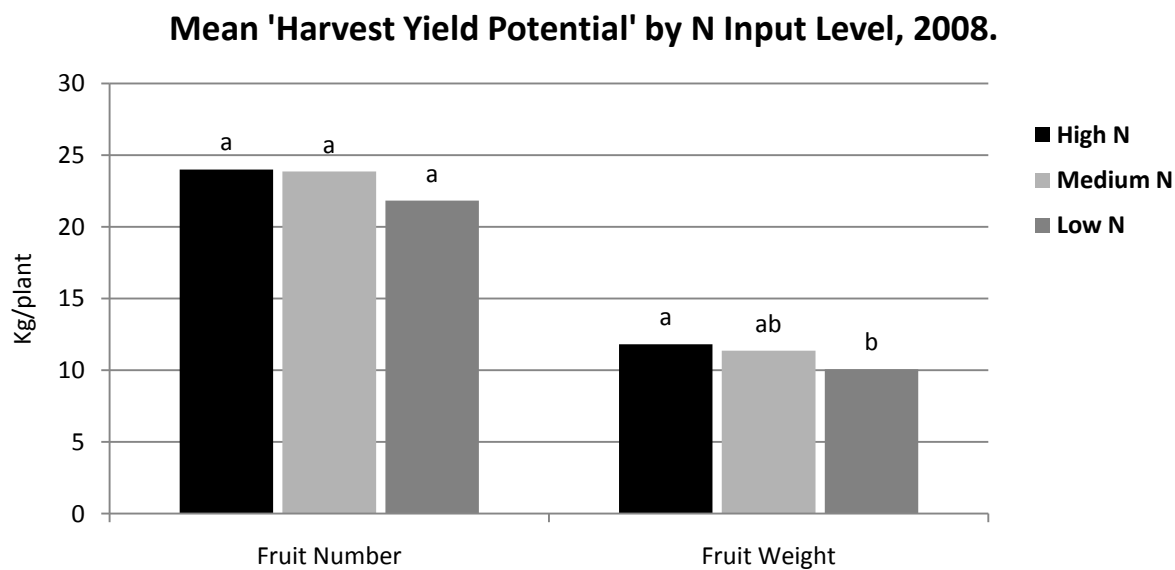


Fig. 3.11B. Mean 'Harvest Yield Potential' by N Input Level, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

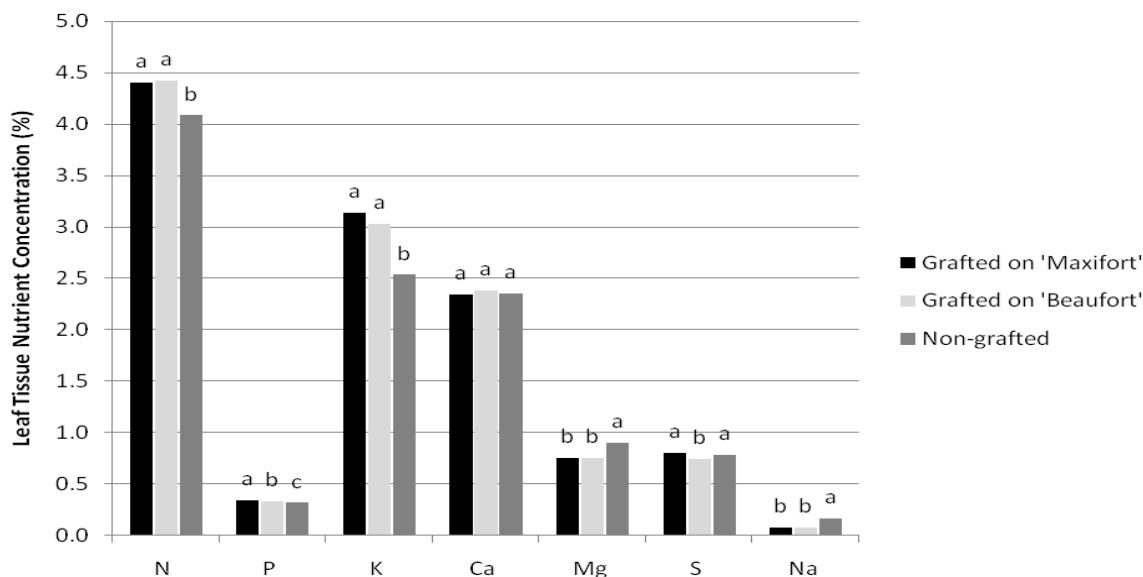


Fig. 3.12A. Grafting Effect on Mean Leaf Tissue Macro-nutrient Concentrations, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

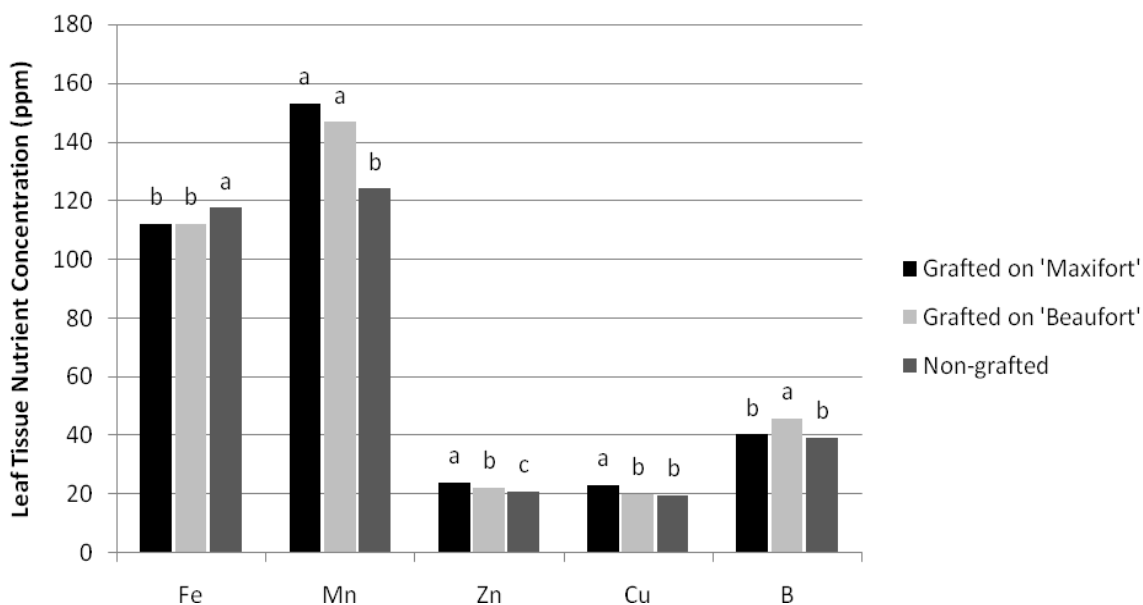


Fig. 3.12B. Grafting Effect on Mean Leaf Tissue Micro-nutrient Concentrations, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

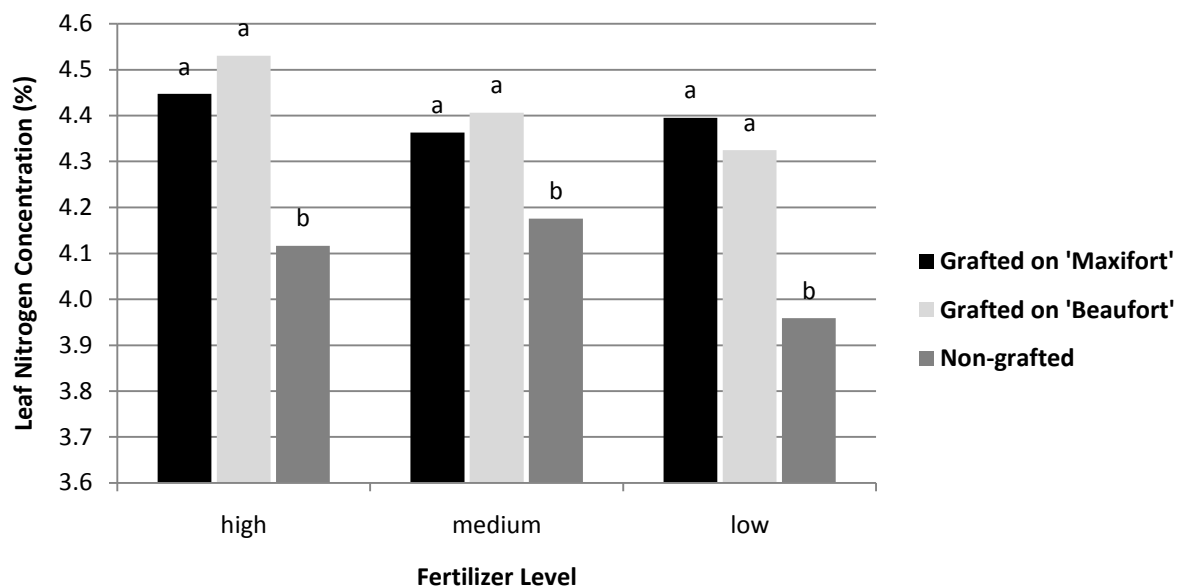


Fig. 3.13. Interaction between the Grafting Effect and Nitrogen Input Level, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

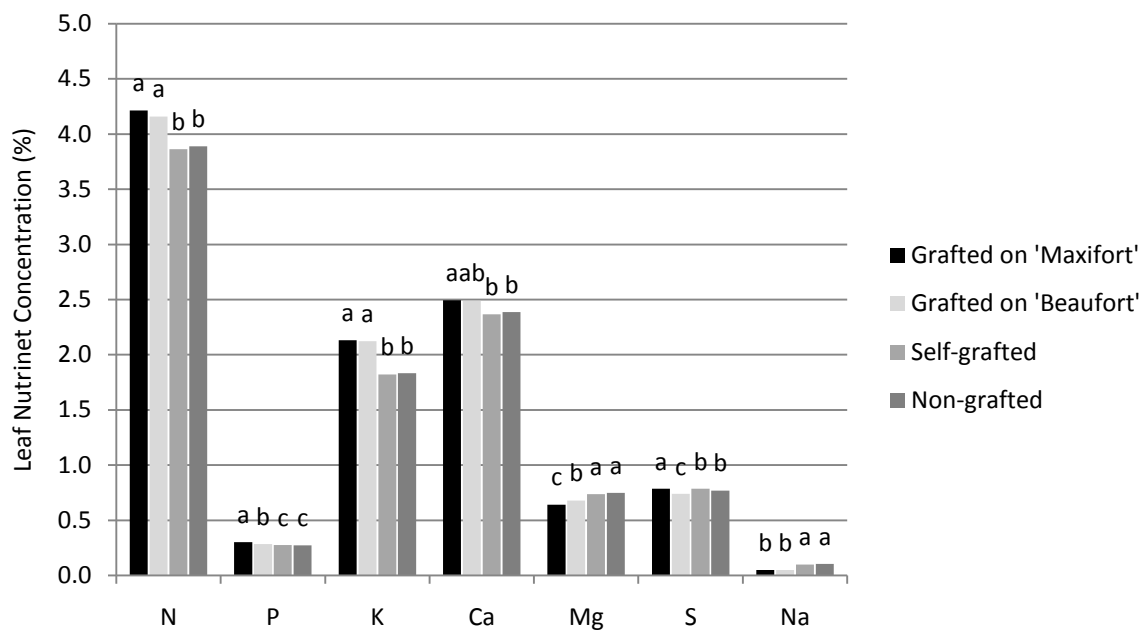


Fig. 3.14A. Grafting Effect of Mean Leaf Tissue Macro-nutrient Concentrations, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$)

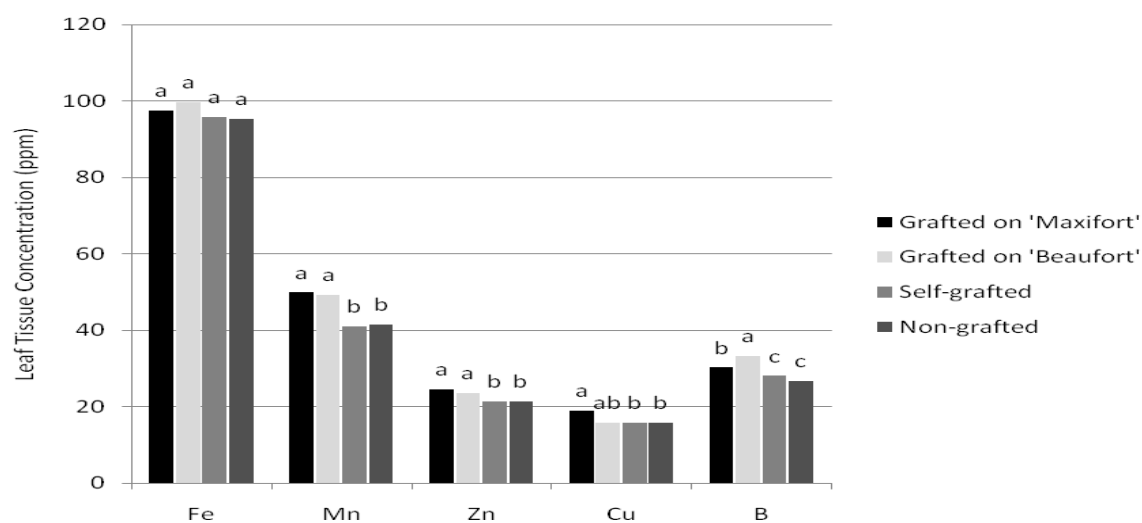


Fig. 3.14B. Grafting Effect on Mean Leaf Tissue Micro-nutrient Concentrations, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

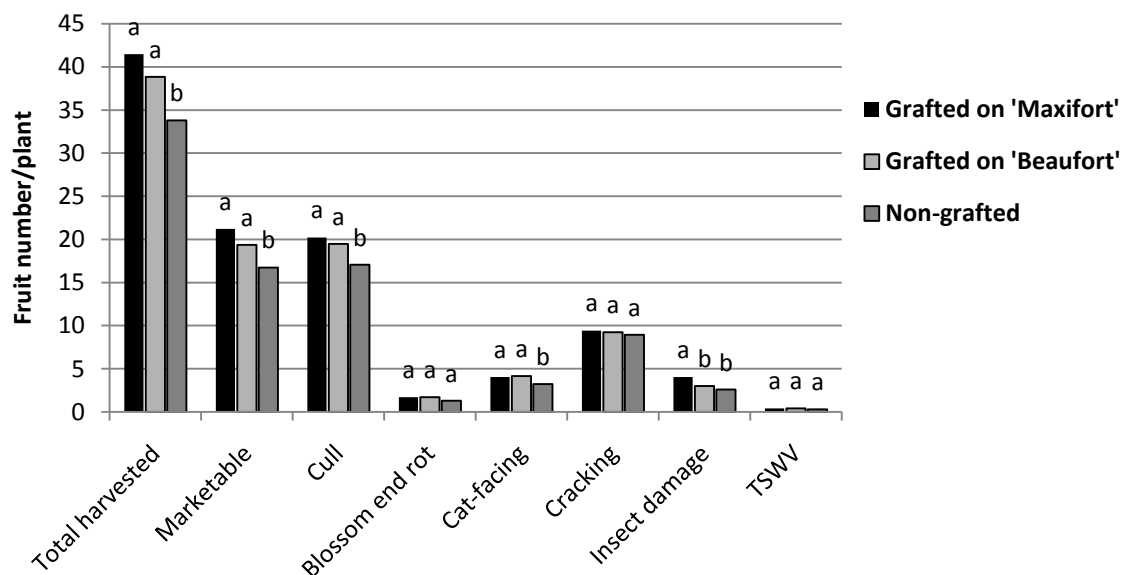


Fig. 3.15A. Grafting Effect on Mean Fruit Number, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

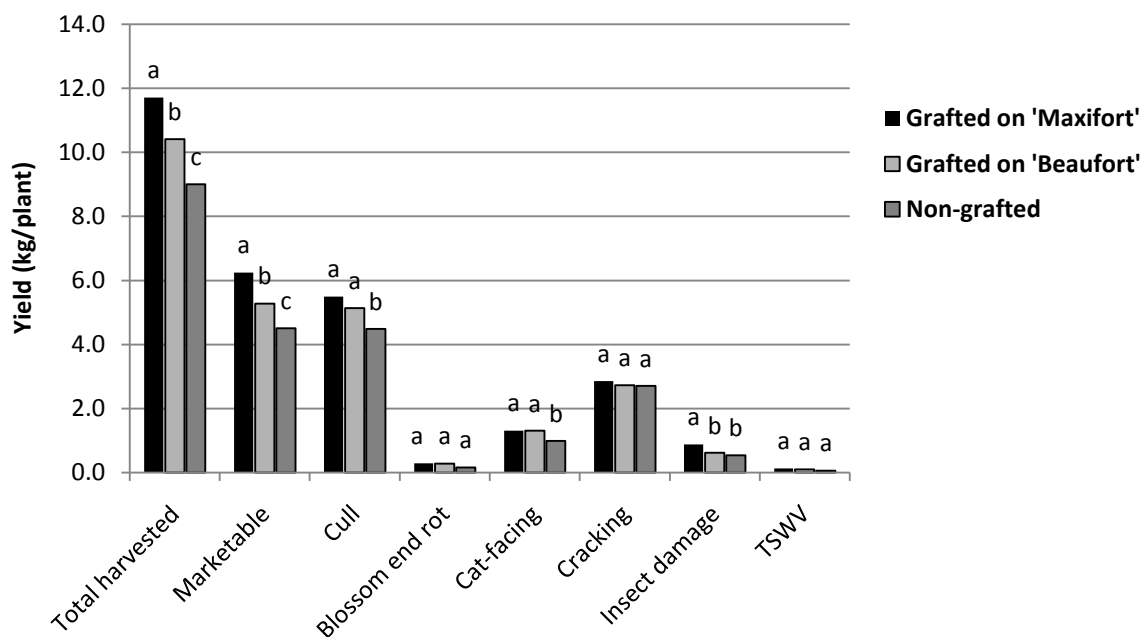


Fig. 3.15B. Grafting Effect on Mean Fruit Weight, 2007. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

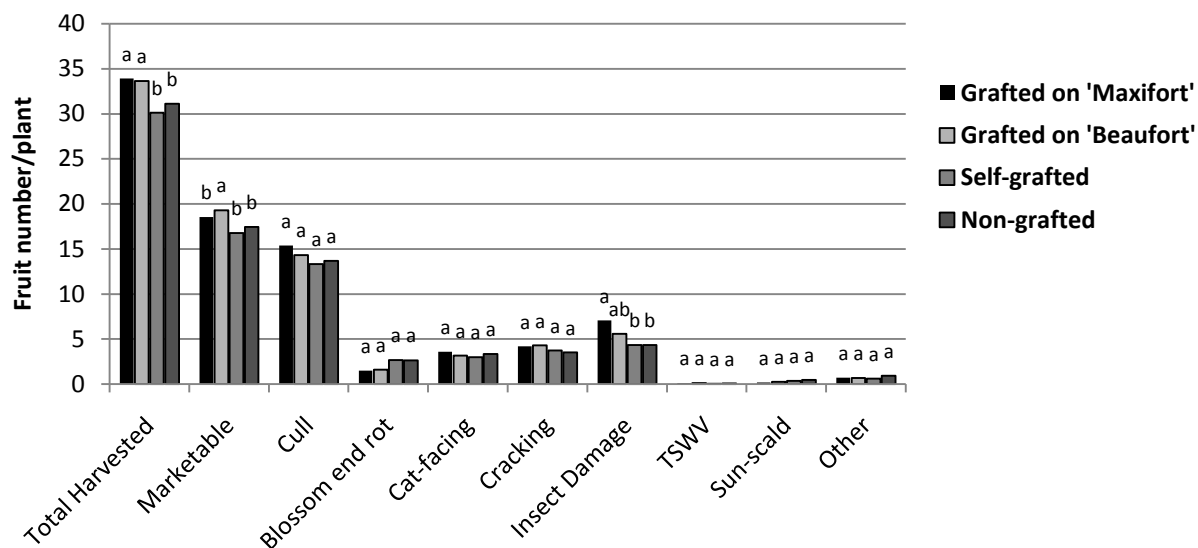


Fig. 3.16A. Grafting Effect on Mean Fruit Number, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

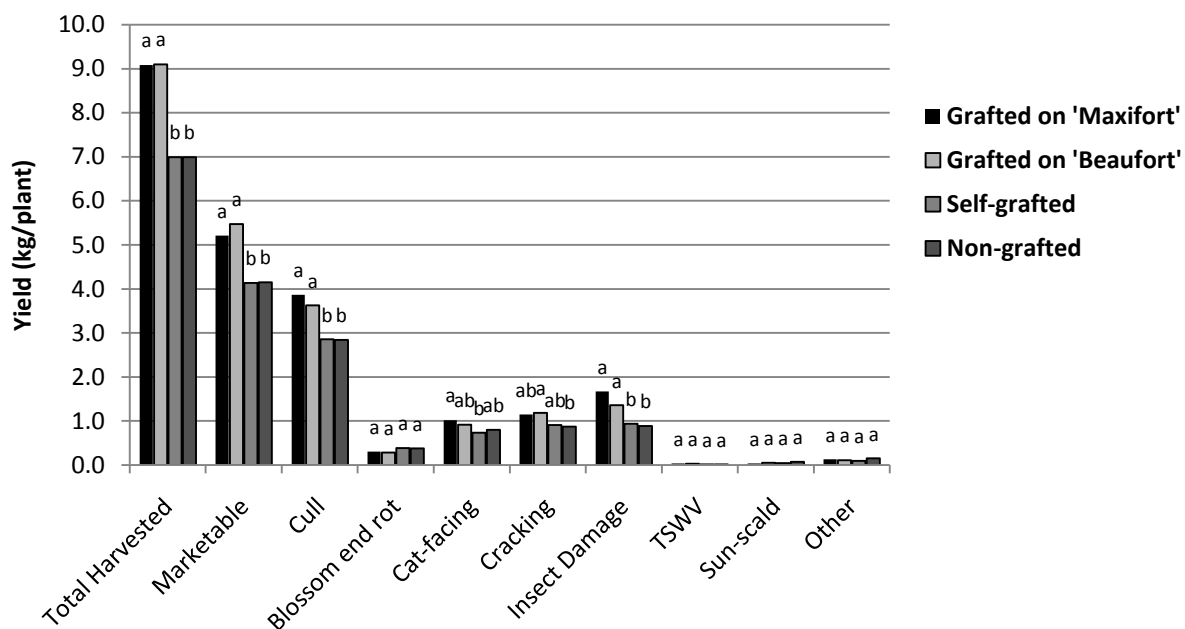


Fig. 3.16B. Grafting Effect on Mean Fruit Weight, 2008. Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

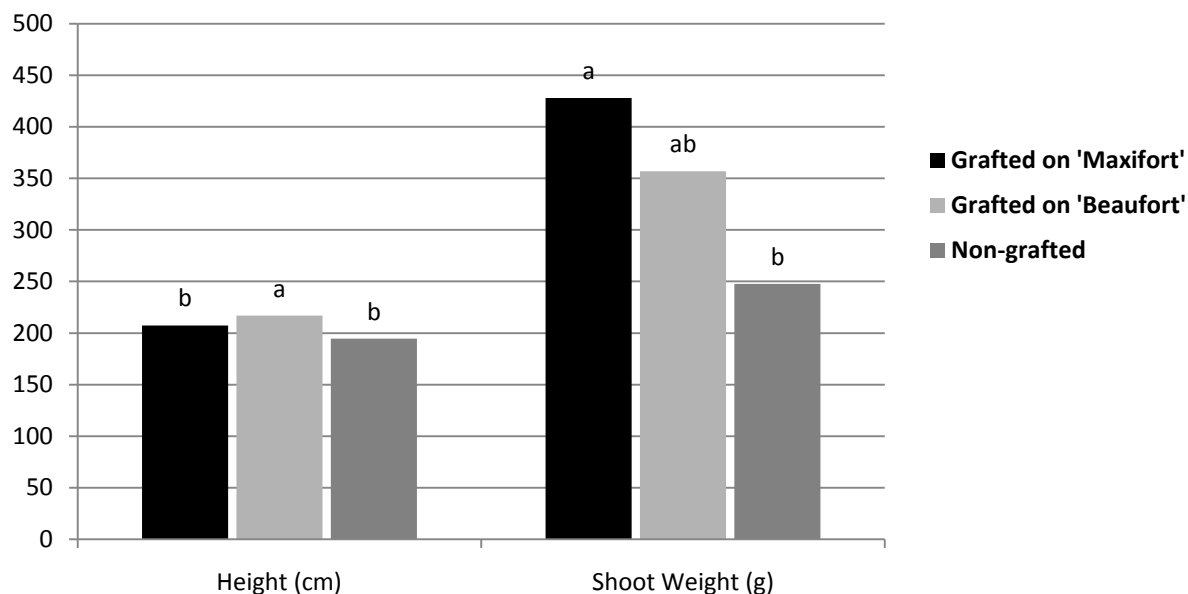


Fig. 3.17A. Grafting Effect on Plant Growth in the High Tunnel System, 2007.

Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

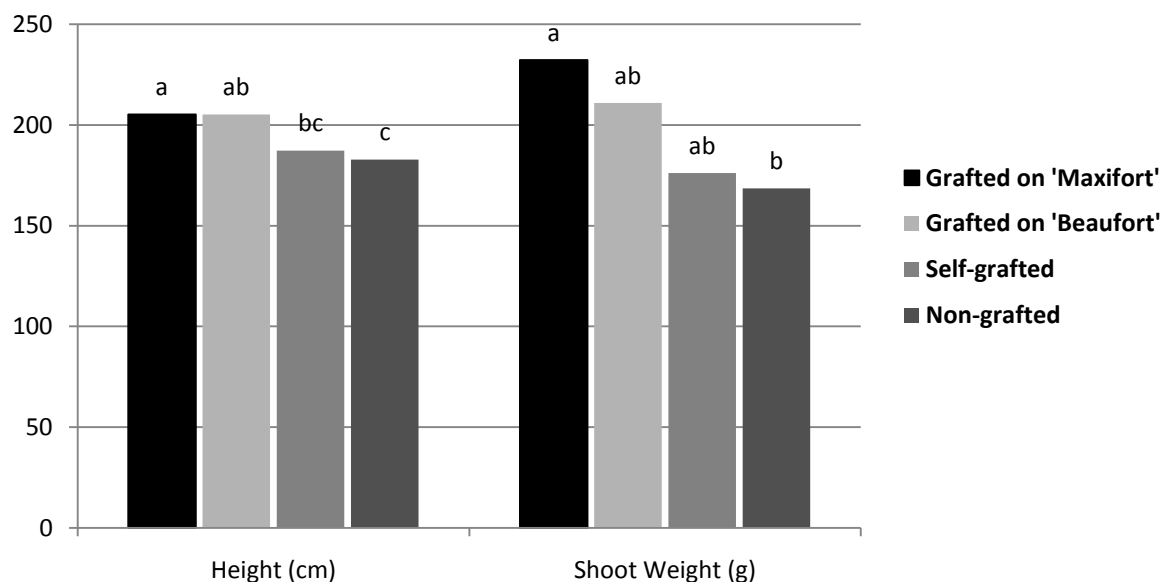


Fig. 3.17B. Grafting Effect on Plant Growth in the High Tunnel System, 2008.

Bars within each data type marked with the same lowercase letters are not significantly different based on Tukey's test ($p \leq 0.05$).

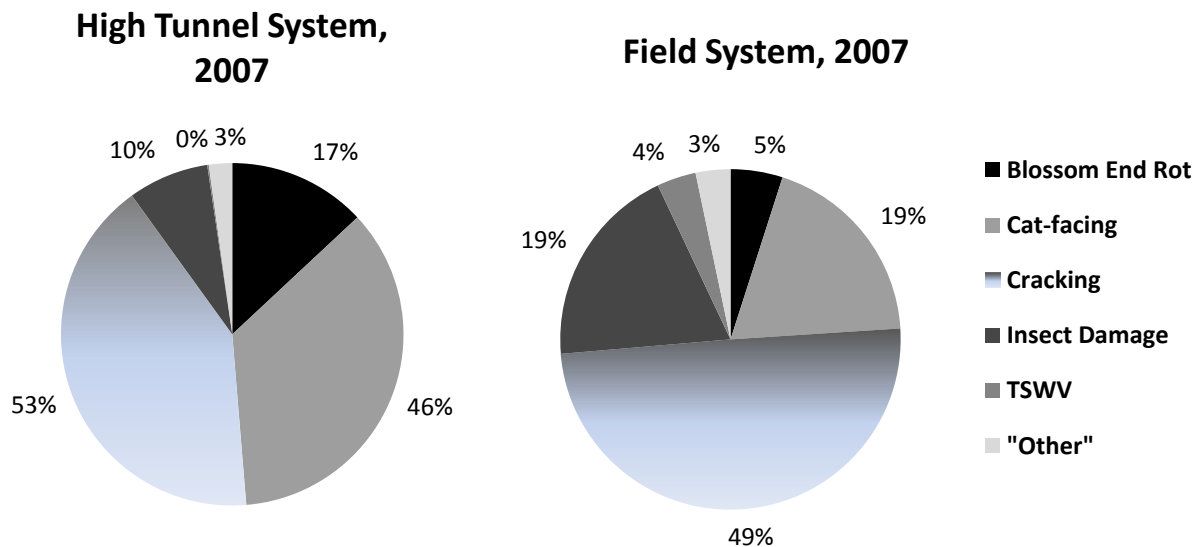


Fig. 3.18A. Categorical Breakdown of Non-marketable Fruit: High Tunnel and Field System, 2007.

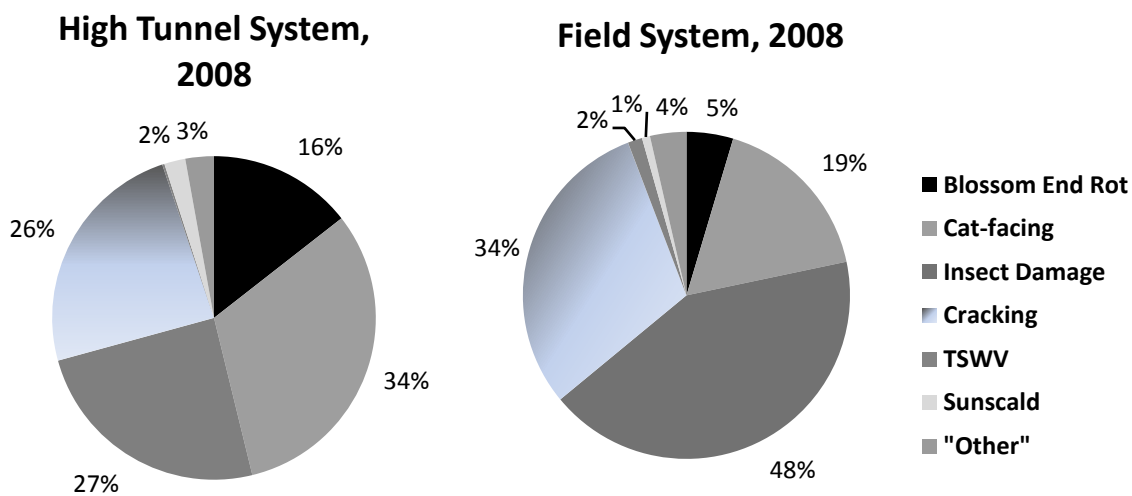


Fig. 3.18B. Categorical Breakdown of Non-marketable Fruit: High Tunnel and Field System, 2008.

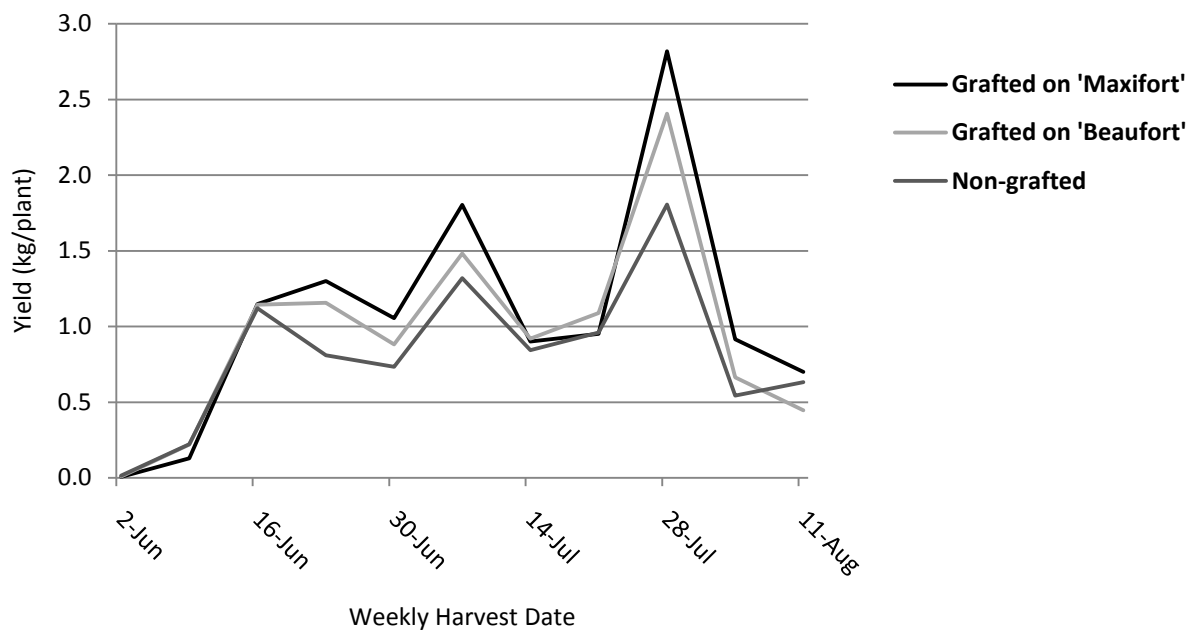


Fig. 3.19A. Grafting Effect on Mean Fruit Yield Over Time, 2007.

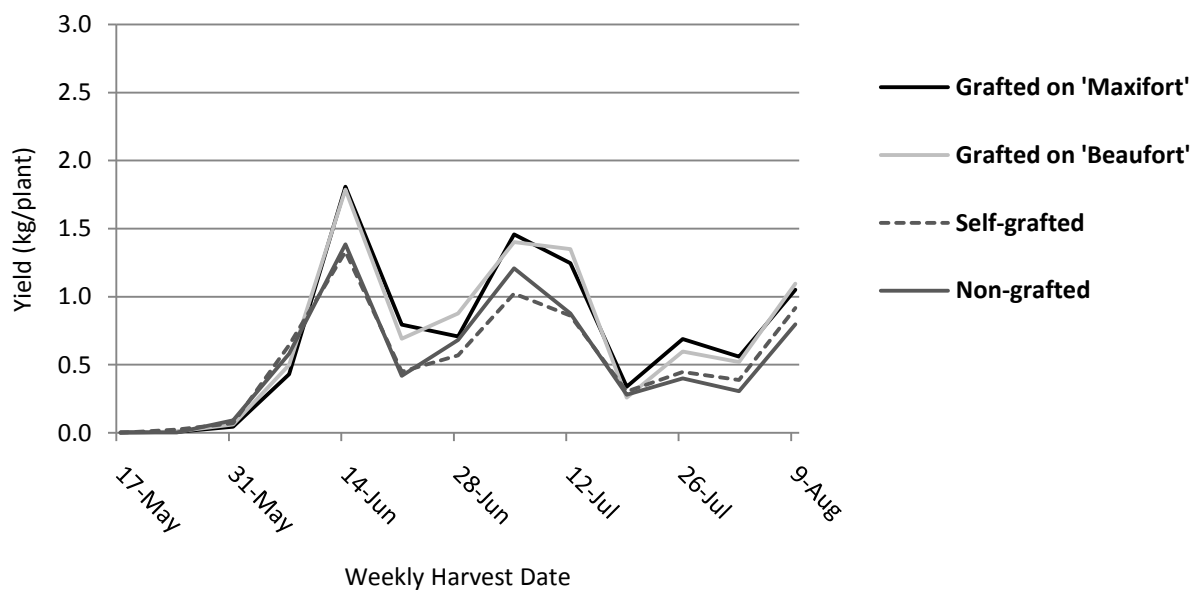


Fig. 3.19B. Grafting Effect on Mean Fruit Yield Over Time, 2008.

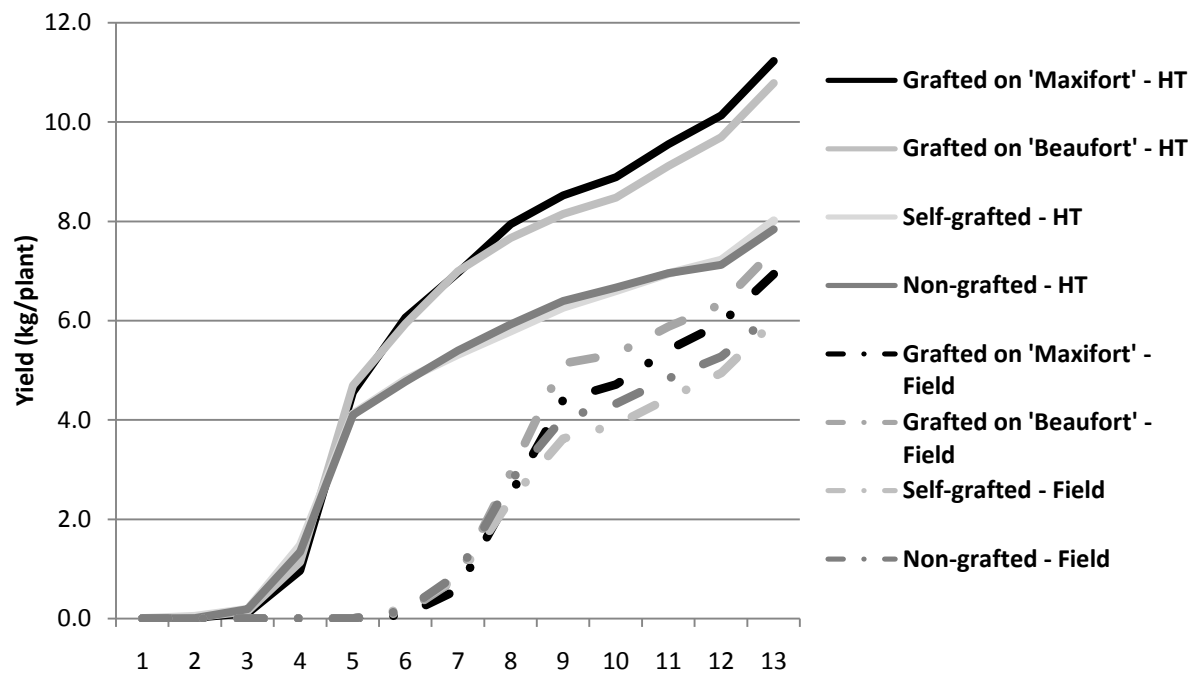


Fig. 3.20. Total Cumulative Harvest Yield Over Time: System * Graft, 2008.

‘Cherokee Purple’ is the scion cultivar for all treatments. High Tunnel system is abbreviated as HT.

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