

**SUB-SURFACE DRIP AND OVERHEAD IRRIGATION EFFECTS ON ASPARAGUS  
PRODUCTION UNDER MICHIGAN GROWING CONDITIONS**

**By**

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## **ABSTRACT**

### **SUB-SURFACE DRIP AND OVERHEAD IRRIGATION EFFECTS ON ASPARAGUS PRODUCTION UNDER MICHIGAN GROWING CONDITIONS**

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Asparagus is a perennial crop historically grown without irrigation in western Michigan for the processing market. Shifts to fresh market production, new hybrids, increased incidence of summer drought, and increased disease pressure, justify evaluation of more intensive production practices including irrigation. Field and greenhouse trials evaluating the impact of drought stress and irrigation delivery system (overhead vs. sub-surface drip) on two varieties (Guelph Millennium [GM] vs. Jersey Supreme [JS]) were initiated to guide Michigan asparagus grower's irrigation decisions. Short-term results from these studies indicate a variety of positive plant responses to irrigation treatments. Asparagus yields increased from 6 to 21% with trickle and overhead irrigation treatments in GM and JS during the 2012 field season. With supplemental irrigation, increases in stem number, light interception, fern height, root carbohydrates, cladophyll weight, and dry fern weight occurred. Cultivar responses to irrigation treatments differed depending on drought stress severity and plant growth stage. Increased yields for GM were attributable largely to increased weight per spear, rather than increased spear number as seen in JS. Results from multi-season greenhouse trials with GM demonstrated that prolonged low-level drought stress reduced fern growth and root weight, while short-duration intense drought stress had greater impact on root carbohydrate concentration and short-term yield. Overall, these results suggest that under weather conditions similar to those of 2011-2012 irrigation increases yield and plant health enough to justify the added costs of irrigation for Michigan asparagus growers.

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# **Sub-surface Drip and Overhead Irrigation Effects on Asparagus Production under Michigan Growing Conditions**

## **CHAPTER 1**

### **Introduction**

#### **Asparagus growth and development**

*Asparagus officinalis* is a perennial vegetable crop that is hand-harvested every spring. Root carbohydrates are replenished following harvest, during the summer and fall from photosynthesis occurring in a two-meter tall fern canopy. Herbicides are typically used twice a year to control weed populations and very little tillage occurs after the year of planting. Most Michigan fields are established using crowns—year old root systems grown from seed offsite during the previous year—instead of greenhouse grown seedling transplants.

During the spring of field establishment 70 cm trenches, or furrows, are plowed into the sandy soil. Crowns are set out on the bottom of the furrow with granular fertilizer and covered with soil by eroding the trench walls with a soil blade. As the asparagus develops over the growing season furrows are filled gradually in by grading soil from the berms into the trenches (Zandstra et al. 1992).

Asparagus plants consist of two main sections: the above ground fern, and the subterranean crown consisting of rhizomes and roots. The crown is the heart and central interchange of the plant, producing growth buds for future spears and storing resources for future growth. Asparagus roots can be separated into two categories: the numerous thin, short-term feeder roots that secure nutrients and water and the fleshy storage roots that ensure overwinter plant survival by amassing soluble carbohydrate reserves (Drost 1997).

The fern consists of fibrous stalks, which develop from edible immature spears. As stems mature, branches develop off of growth scales, and plants “fern out” producing many

photosynthetic leaflets as they mature (Drost 1997). The small leaflets that develop off the branches and stems of asparagus are called cladophylls; they are the most critical photosynthetic area of the plant.

The simplest method of gauging asparagus size and health is by observing the fern canopy that develops after the spring harvest season has concluded. Several studies (Guo 2001; Read 2009) have shown that fern weight is correlated with yield the following season. Although in general this rule is true, overall fern size should not be the sole means of predicting asparagus yield (Schaller and Paschold 2009). Survival of the crown is the most critical aspect of producing asparagus because replacing plants in an established stand is generally not feasible and the early loss of plants results in compounding yield losses over multiple years (Sinton et al. 2008; Sterrett et al. 1990).

Photosynthesis levels over the preceding season dictate the yield for the following year's harvest. Most of the above-ground portions of the asparagus plant rapidly green up with chlorophyll from their initial white or yellow coloration, as they are exposed to light. Stem tissue has some photosynthetic capability and stomata, but the cladophylls are the center of the plant's photosynthesis production (Suzuki et al. 2002). Elevated photosynthetic rates were found in higher yielding cultivars, with high ratios of cladophyll to stem (Faville et al. 1999). Guo (2001) illustrated that asparagus partitioning to cladophylls is highly variable between cultivars of asparagus, and that higher yielding types also having more cladophyll weight than lower yielding varieties. Because of their wavy micro-surface, varied width, and small size, physical measurements of cladophyll surface areas are difficult, so using dry weight is considered good scientific practice (Guo 2001). Levels of water use efficiency and total photosynthesis varied with stomatal size and density between asparagus cultivars (Schaller and Paschold 2009).

There have been a number of root studies that have investigated both the size and carbohydrate levels of the asparagus plant throughout the calendar year either in the field or in potted specimens (Haynes 1987; Drost and Wilcox-Lee 1997; Drost and Wilson 2003; Schaller and Paschold 2009; Paschold et al. 2004). The rooting zone of a mature asparagus plant is substantial, typically reaching at least a meter below the surface with the majority of root weight in the top 50 cm near the crowns (Drost 1997). In New Zealand, Wilson et al. (2002) developed a field-based carbohydrate evaluation protocol using refractometers to measure simple sugar levels in root tissue throughout the year. Although this model has not been widely adopted by Michigan growers to schedule harvest length, it remains a practical tool for research into root status.

### **The U.S. asparagus industry**

Michigan has been one of the nation's three largest asparagus producers for the past fifty years. In 2010, there were 146 independent asparagus farms with 4,309 ha in production with a cash value of over \$16 million (USDA-ERS 2010). Michigan harvest occurs from May to June in sandy soils near the Lake Michigan shoreline. Historically asparagus production in the Midwest has focused on making canned and frozen products but there has been a recent shift towards fresh markets. Many mid-sized family fruit and vegetable operations cultivate between twenty to one hundred acres of asparagus to diversify their cropping systems; asparagus extends the working season for harvest laborers and helps growers by generating cash-flow to fund other operations before revenue from fruit production arrives.

Although the past fifteen years have seen increased U.S. asparagus consumption, overall domestic production has decreased. This is primarily due to increased competition from Peru,

which supplied 81% of the asparagus consumed in the U.S. in 2009 (Dartt et al. 2009).

Increasing asparagus imports from Latin America decreased market prices significantly for the first decade of the 2000s. This price drop led to a significant decrease in U.S. production acreage (Read 2009) with the number of new asparagus plantings decreasing and many older fields being plowed out. Established asparagus fields historically lasted 25 years or more, but with the onset of fungal diseases, as well as shifting markets, the duration of profitable production declined to about twelve years. Five or six years are always required for plants to reach full production potential, meaning there are far fewer productive years given declining stand longevity (Dartt et al. 2009).

In traditional production areas prevalence of root pathogens has been a leading factor in asparagus production decline (Hausbeck 2012). Much of the prime land in western Michigan has been cropped in asparagus for decades; growers are forced to replant over previous plantings into soils with elevated populations of fungal pathogens, including *Phytophthora* and *Fusarium* species (Rodriguez 2010). Starting in the 1990's Jersey Giant was the dominant variety planted in Michigan; it produces well in early life, but experiences high plant mortality in later years, probably because of intolerance to these pathogenic fungi.

Growers are now faced with a narrow production window to recuperate the large sunk costs from the first several years of establishment and cultivation before the stand declines and production erodes. Asparagus decline and low prices have led to a decrease of Michigan production acreage from 6,880 hectares in 1990 to under 4,451 hectares currently, and fields are being replanted at much higher cost than the \$5,000 ha establishment costs of even ten years ago (Ball et al. 2001).

Asparagus establishment costs have risen dramatically in the past several years, as new hybrids planted on fumigated soils at higher densities have become the norm. In the nineties Michigan State University Extension recommendations were for 29,640 crowns ha. without fumigation (Zandstra et al. 1992). Now growers are typically planting at least 37,050 crowns ha to raise long-term harvest labor efficiency. Seed costs for modern hybrid varieties have doubled to \$2,250 ha from several years ago and seed costs of these hybrids are twenty times greater than the traditional Mary Washington varieties. At current planting densities, the cost of hybrid asparagus crowns reached \$7,500 ha in 2013 (Nourse 2013). In addition, soil fumigation increases startup input costs today. With the EPA phase-out of several fumigants, fumigation costs have risen from approximately \$750-900 ha to at least \$1,250 ha while treatment efficacy has declined (Hausbeck 2012). Given these rising input costs, growers have increased incentive to intensify production systems through additional practices including irrigation.

### **Irrigation for asparagus production**

Although irrigation has been a mainstay in many significant asparagus production regions around the world, unlike Michigan they tend to be naturally arid. While the asparagus plant is drought hardy (Krug 1998), increases in productivity are necessary to ensure adequate return on the large financial investment of establishing asparagus fields. In Michigan, an annual yield increase of approximately 9% from a baseline yield of 3,359 kg ha has been estimated to justify the installation of irrigation in the first seven years (Harsh 2010). However, such estimates are based on a wide range of assumptions, and very few studies have been conducted to evaluate the potential biological and economic benefits of irrigation in asparagus production for Michigan.

Worldwide, irrigation is typically applied to asparagus fields by one of three methods: 1) furrow irrigation—the traditional practice in the American west; 2) center pivot sprinkler systems; and 3) trickle irrigation, either through tubing buried at crown level or placed on the soil surface. Each system has its own pros and cons. Furrow irrigation—the oldest system—floods production areas periodically using networks of ditches to disperse river water. Due to low water use efficiency, hilly-topography, coarse soil types, and water withdrawal restrictions, furrow irrigation isn't used in Michigan.

Center pivot systems are relatively common in Michigan but have traditionally been installed only on annual vegetable crops with high value and drought-susceptibility like carrots or cucurbits. Given Michigan's high relative humidity and modern spray nozzles these systems can be relatively water efficient, reaching levels of 80% (Kelley 2013; Nelson 2013). Center pivot has the additional benefit of being able to activate residual herbicides, move mobile N fertilizers into soils, and establish cover crops in the fall without reliance on rainfall. Center pivot systems have a working life far longer than asparagus; once sunk costs are recouped with asparagus production, the equipment can continue to be used for another twenty years. On the other hand, center pivot systems may induce higher weed germination and growth by moistening the soil surface. Overhead systems like center pivot can also exacerbate foliar diseases by increasing periods of leaf wetness. Center pivot systems don't achieve reasonable economies of scale on fields less than 15 hectares or in irregularly shaped fields, and they require large water sources and fuel costs for delivery (3,000 L per minute for a 35 ha system) (Neiberger and Waters 2009; Kelley 2013).

Trickle irrigation is common in many intensively produced vegetables in Michigan, but is rarely used in asparagus. In arid asparagus production regions, trickle tubing is placed on the

soil surface and rolled up annually (e.g. Peru), or experimentally, buried below the soil surface and left in place for the lifetime of the asparagus stand (Sinton and Wilson 2008; Ley and Agenbroad 1989). In such sub-surface drip systems, thick-walled tubing is usually buried in every asparagus row. Tubing with emitters every 46 cm are typically placed at crown level at the time of crown transplant (Netafim 2010). With sub-surface drip, a significant amount of soil surface moisture evaporation is avoided facilitating lower water use requirements. Buried trickle line has the additional potential benefit of facilitating fertilizer or pesticide application directly to the root zone where they can be most effective. Since drip systems can be broken into zones, water pressure and pump requirements are low, reducing costs. Trickle tubing can also be placed on the surface annually but tube malfunctions and total combined installation and removal costs are greater long-term compared to buried tubing.

Irrigation could prove valuable in Michigan production for several reasons. Asparagus planted with irrigation is at a lower risk of establishment failure, and yield may be increased significantly through higher per-picking yield and a more aggressive harvest schedule (Schaller and Paschold 2008; Bussell et al. 1984; Sterrett et al. 1990). In irrigated systems, it may be possible to increase planting densities, making harvest, fumigation, spraying and cultivation more efficient. The need for fumigation at planting could be reduced with alternate chemical applications through use of irrigation throughout the production cycle and increased nutrient use efficiency could provide value in addition to replacing transpired water (Netafim 2010).

## **Previous irrigation studies in asparagus**

Depending on the plant's age or variety, soil conditions, and average rainfall, irrigation trials have illustrated drastically increased yields in many parts of the world, including rain-fed climates similar to Michigan (Hartmann 1981; Rolbiecki and Rolbiecki 2007).

Several irrigation studies conducted in rain-fed asparagus production regions of Europe suggest that the potential benefits are large. For example, Rolbiecki and Rolbiecki (2007) found gains of over thirty percent with many cultivars in Polish variety trials. Hartmann (1981,1996) found number of spears increased 30% in sandy soils in Germany. He advocated use of trickle irrigation in his climate because of water savings and noticeable benefit with higher producing cultivars; European varieties showed particularly significant production increases with Grolim yields more than doubling with the addition of irrigation.

The method of irrigation can also have a dramatic effect on asparagus crop response, profitability, and environmental impact. (Sterrett et al., 1990) found that buried trickle irrigation greatly improved establishment survival rate in young asparagus, particularly with transplants. Lifespan and maintenance cost of buried tubing also were noted to be better. Netafim, one of the world's larger trickle manufacturers, actively promotes subsurface installation of its products in asparagus (2010). Overhead sprinkler systems had asparagus survival rates below un-irrigated control with noted wear to fragile ferns (Sterrett et al. 1990).

The effects of irrigation depend not only on climate and method, but also on the specific variety being evaluated. For example, Schaller and Paschold (2009a) showed significant differences in physiological drought response between Backlim and Grolim varieties. Likewise, Rolbiecki and Rolbiecki (2007) and Sinton and Wilson (2008) demonstrated dramatic yield differences between cultivars when subjected to drought stress. Several trials (Wilson and



Sinton 1996; Rolbiecki and Rolbiecki 2007) have found that the Jersey Giant variety—the most prevalent Michigan variety—is also one of the least impacted by supplemental irrigation. One of the core purposes of these trials is to evaluate the economic cost and benefit of overhead and sub-surface irrigation in Michigan asparagus. To our knowledge, no studies have compared the effects of irrigation on yield with varieties such as Guelph Millennium and Jersey Supreme, currently being planted in Michigan.

### **Goals and scope of this work**

With this background in mind, experimental trials were planned to collect information on the potential costs and benefits of irrigation applicable to Michigan's field conditions and climate. In the first study (described in detail in Chapter 2), the effects of two irrigation systems (subsurface drip versus overhead) on two asparagus varieties (Guelph Millennium and Jersey Supreme), representing the most commonly planted MI asparagus genetics were evaluated in a large field experiment. Yield and multiple aspects of plant health were evaluated over three successive seasons. The goal of this trial was to provide Michigan growers with applicable data that would assist them in deciding whether to invest in irrigation, and how to optimize irrigation systems to improve profits. We hypothesized that: 1) if typical historic drought stress periods occurred during the study, irrigation would increase fern growth, root carbohydrate storage and ultimately yield; 2) the yield benefits of irrigation would depend on asparagus variety, with the newer Millennium variety responding more favorably to irrigation than Jersey Supreme; 3) yield benefits would also depend on delivery systems, with sub-surface drip improving yields and reducing costs relative to overhead irrigation; and 4) sub-surface drip irrigation would result in

fewer weed and disease problems relative to overhead irrigation due to reduced leaf and soil-surface wetting.

This large-scale irrigation trial was supplemented with a greenhouse irrigation trial (described in detail in Chapter 3) aimed at better understanding the response of Millennium to baseline irrigation and drought treatments. We hypothesized that drought periods of two weeks or more in summer would have a negative impact on fern growth, carbohydrate storage, and yield the following spring. An important secondary objective of this trial was to better understand how root growth and carbohydrate storage responded to different forms of drought stress.

## **CHAPTER 2**

### **Effects of Sub-surface and Overhead Irrigation on Michigan Asparagus**

#### **Introduction**

As the weather patterns in the Great lakes region shift, intense summer heat and more sporadic rainfall have occurred with increased regularity. For example, since 1980 Michigan's average temperature has warmed by two degrees, and although precipitation has also increased in the last 50 years, this increase is concentrated during the cooler seasons (MSU Enviro-weather, 2013). During the critical period of fern growth and carbohydrate replenishment (July-August), rainfall is often well below evapo-transpirational demand (Figure 2-1), resulting in drought stress in un-irrigated fields. Evapotranspiration (ET) is the amount of soil moisture lost to the air from all surfaces in a cropping area. In water requirement forecasting, each crop is given a coefficient that represents its water requirement relative to a reference crop at various crop growth stages.

Relatively little is known about the impact of alternative irrigation systems on commonly grown asparagus varieties under Michigan weather and soil conditions. Drought stress may be an important factor contributing to the decline in asparagus fern health and yield. Although asparagus is deep rooted and relatively drought tolerant, soil water content during fern growth is an important determinant of crop yields (Drost and Wilcox-Lee 1997; Hartman 1981). Drought stress during fern growth can limit the capacity of plants to produce the soluble carbohydrates in roots necessary for high yields in subsequent seasons (Drost and Wilcox-Lee 1997). Stressed plants may also be more susceptible to fungal diseases that increasingly plague the asparagus industry, including *Fusarium* (Morrison et al. 2012) and *Phytophthora* (Saude et al. 2008). Warmer temperatures and more variable rainfall patterns observed in MI in recent years make irrigation an increasingly important tool for reducing risks of yield loss in asparagus production.

Irrigation may also create opportunities for valuable complementary practices including cover-cropping and fertigation. In irrigated systems, cover crops growing below the fern canopy may be established with reduced risk of competition for water with the asparagus crop. However, care must be taken to avoid competition for water with the crop. Living mulches in un-irrigated asparagus in Wisconsin suppressed weeds but in some cases reduced yields (Paine et al. 1995). In Michigan, winter rye sown as a living mulch immediately following asparagus harvest was found to be beneficial for suppressing certain weeds including *Conyza canadensis*, but also reduced volumetric water content to dangerously low levels during fern growth in the absence of irrigation (Brainard et al. 2012). Under irrigated systems, cover crops may be more widely adopted without adversely impacting asparagus.

Irrigation may also facilitate delivery of pesticides and fertilizers to improve the efficiency and efficacy of agrichemicals, and reduce their adverse environmental impacts. Fertigation and chemigation through drip tape is common in many vegetable crops, but has not been extensively explored in asparagus. By targeting agrichemicals directly to the root zone through sub-surface drip tubing, losses through leaching and volatilization are minimized. Asparagus growers adopting overhead irrigation report improvements in the efficacy of soil-applied herbicides requiring moisture for activation. This is particularly important following asparagus harvest when the optimal window for herbicide application is very short. Dry conditions during this period result in reduced herbicide efficacy. Some growers are forced to delay herbicide applications and extend harvest while waiting for rain to activate herbicides (Brainard, personal communication); this practice stresses asparagus by both limiting the period of fern growth for carbohydrate replenishment and drawing energy for more production.

With these potential benefits in mind, MI asparagus growers have begun to adopt irrigation into their systems, but little information is available to guide them in their choices regarding delivery system (overhead vs. drip). Almost all investment in asparagus irrigation so far has been in center-pivot systems. Theoretically, drip irrigation systems will save water and fuel costs associated with irrigation. Drip systems also minimize the risk of foliar disease by avoiding leaf-wetting. By avoiding moistening the soil surface, drip systems may also be less likely to stimulate weed and foliar fungal growth (e.g. rust). However, overhead irrigation systems are viewed by growers as less costly to maintain and provide several potential advantages including activation of soil-applied pesticides, spear cooling during harvest, and uniform establishment of cover crops. Before growers will consider less familiar alternatives like drip, they need data on its potential economic and biological advantages.

The impacts of irrigation on asparagus are likely to vary considerably with variety, but little information is available regarding response of typical Michigan varieties to irrigation. University of Guelph's Millennium has not been included in many previous international cultivar trials because of the relative recent entry into world markets. In Ontario and Michigan production of Millennium in soils nearing loam classification have been very good, suggesting having higher available water in sand would be beneficial. New Zealand irrigation trials found that Jersey Giant asparagus was not responsive to irrigation but those trials were in soils with high moisture retention ability and adequate natural rainfall; Jersey Supreme will probably share some of those responses because of shared paternity. In North Carolina variety trials, Millennium and Supreme both performed in the top quartile of available varieties over a five-year period (Cantaluppi 2011).

To address grower interests, and fill existing knowledge gaps, a large-scale field experiment was initiated in 2010 to evaluate the long-term impact of irrigation system (none, sub-surface drip, or overhead) on fern growth, yield and profitability of both Guelph Millennium and Jersey Supreme asparagus varieties. Secondary objectives included assessment of irrigation systems impact on the incidence of insect, disease, and weed pests. The long-term objective of the trial is to evaluate the development and subsequent decline of asparagus stand in each of the six treatments, to assess whether irrigation can delay the decline of productive asparagus fields. We hypothesized that 1) irrigation during fern growth would increase yield and quality of asparagus; 2) these benefits would be greater for Guelph Millennium compared to Jersey Supreme; and 3) sub-surface drip irrigation would provide equivalent yield improvements to overhead, while reducing delivery costs as well as disease and weed problems.

## **Materials and Methods**

### **Site characteristics and experimental design**

A field experiment was established in Oceana county, western Michigan in 2010. Six experimental treatments were examined consisting of three irrigation systems (none, overhead, and sub-surface drip) and two varieties (“Jersey Supreme” and “Guelph Millennium”). Four replicates of each treatment were arranged in a split plot design with irrigation as the main plot factor, and variety as the subplot factor. Each of the 24 subplots measured 6 x 18.2 m with four rows of asparagus spaced 1.5 m apart. Data were collected from the inside two rows of each plot to minimize edge effects.

Soils were representative of those found locally in asparagus production; a well-drained glacial moraine mixture of Spinks loamy fine sands and Perrinton loams, on slopes of up to six

degrees with southern exposure. Initial soil tests indicated a soil composition of 86% sand, 6% silt, and 8% clay averaged through the usable soil profiles, a surface pH slightly above neutral and slightly acidic subsoil. The experimental area had no reported history of previous asparagus production and had been either fallowed or cover-cropped for several years prior to planting. Soil chemical characteristics immediately prior to planting fell into the following range: (pH 6.3-7.0 with levels soil organic matter at 1.1 to 1.3%)

### **Trial establishment**

Asparagus was established from one-year-old crowns grown locally from seed on soil that had been fumigated to reduce risk of fungal pathogens. Two asparagus varieties representing the most common breeding programs were used for the trial: “Jersey Supreme” (JS) from Vilmorin Seed, and University of Guelph’s “Millennium” (GM). The growth physiology of these two varieties differs noticeably but both have been top performers in MI variety trials for spear yield and quality.

On 17 May 2010, crowns were planted at a density of 36,889 ha. in crested furrows with alternating placement on 150 cm row spacing. The weight of GM crowns was less than JS but both varieties appeared un-desiccated and disease free. Liquid 10-34-0 fertilizer was applied as directed by MSU soil test in furrow bottom of all plots. Netafim Uniram tubing with 45 cm emitter spacing and 1.59 liter per hour output was placed below the crowns in-furrow in sub-surface drip irrigation plots. Irrigation tubes were connected to buried 2.5 cm poly-pipe headers but irrigation was not initiated during the first season because of limited evapotranspiration with young plants and ample rainfall. Furrows were filled in slowly to hill over crowns with several cultivations during June and July as plants became established.

During the summer of 2010, asparagus spear emergence was relatively uniform between plots and treatments after planting. Fungicides, insecticides and herbicides were applied in accordance with standard grower practice for newly planted fields. In accordance with grower practice in first year after establishment, two harvests were taken in 2011 with combined yield of approximately 225 kg. Detailed yield data was not recorded since irrigation had not been initiated the season before and significant frosting had occurred in early May.

### **Moisture monitoring and irrigation**

Soil volumetric water content (VWC) was monitored with Sentek's Diviner 2000 system (Stepney, AU); one meter long specialized PVC tubes were hand-augured into each plot to record moisture at ten-cm intervals to a depth of 100 cm. Readings were conducted at least on a weekly basis throughout the fern growth period in both 2011 and 2012, with additional readings taken before and after each irrigation event. In addition, VWC at crown depth was data-logged hourly using two Decagon (Pullman, WA) EC-5 moisture sensors installed in each plot.

Irrigation water was supplied from a bored well through a steel reservoir tank with supplemental electric pressure pump. Turbine waterflow meters (GPI, Wichita, KS) were installed at the manifold to record water use in each irrigation zone. From the edge of each overhead plot, irrigation was applied through Nelson Orbitors (Model R-10, Walla Walla, WA) with road guard blinders with an overlapping, alternating 5.8 m radius semi-circle pattern spray on top of 1.5 meter PVC risers. Irrigation events occurred as dictated by Diviner readings and required from 24 to 30 hours to complete. Moisture levels were allowed to depreciate to approximately 50% available water in the rooting zone before irrigation events were initiated. Irrigation was applied to replenish water in the rooting zone to field capacity. In both 2011 and



2012, six and seven irrigation events occurred, with application rates ranging from 1.2 to 3 cm (Figure 2-2). Initial calculations for necessary irrigation time for the sprinklers did not adequately raise soil moisture levels so additional time was added in those treatments. Due to water pressure limitations, only a portion of the sprinkler plots could be irrigated simultaneously so a zone system was established.

Climate data was first gathered from MSU's Enviroweather station four miles to the west and later by a Decagon weather station (EM50) on site for the 2011 and 2012 growing seasons. Data gathered by the station included: ET, rainfall, PAR, and soil temperature. Additional data was gathered during the growing season from repeated visual evaluation of ferns, stems, and spears. These data parameters included: spear number, height, physiological stage, disease, and insect prevalence. Uniformity in data collection standards with concurrent greenhouse trials was attempted whenever possible. When visually evaluating fern canopy years of field scouting experience allowed us to recognize and quantify unexpected, observable trends including differences in foliar damage from herbicides, fern canopy closure rates, pest prevalence, and autumn fern senescence rates.

### **Light interception**

Canopy density was evaluated by measuring light interception with a portable PAR-sensing bar (Lightscout 6 quantum bar, Spectrum Technologies, Aurora, IL). Photosynthetically active radiation, PAR, is the portion of the light spectrum usable to plants. In each plot, one reading was taken above the canopy and five readings taken below the canopy at approximately 30 cm above ground level. Light interception (LI) was calculated according to the following equation:

$$(1) \text{ LI} = 100 - (\text{PARb}/\text{PARa}) * 100$$

Where PARb is PAR at 30 cm below the canopy and PARa is PAR above the canopy.

### **Root sampling**

Soil cores were taken with 7.6 cm diameter hand augers (Sentek 47 mm auger) throughout each season to monitor root development in multiple soil profiles both in the senescence period and post-harvest. Soils were kept in coolers and processed on campus within several weeks of collection. Roots were separated from soils with sieves, weighed, and analyzed for carbohydrate levels according to the method developed for the New Zealand Aspire root carbohydrate system (Wilson et al. 2002). Root samples were collected, washed, and placed into the freezer overnight to separate fructose from cell tissue. Just prior to analysis, root samples were thawed, dab dried with paper, cut into 0.5 cm sections and at least 2g were crushed in a garlic press to collect root sap. The refractive index (RI) of extracted sap was evaluated using an Atago PAL-1 digital handheld refractometer. This device reports RI readings in degrees Brix based on the relationship between RI and the sucrose concentration (% w/w) of a pure sucrose solution at 20 °C. Samples were taken at both the crest and trough of carbohydrate stages at senescence and early canopy establishment stages to evaluate usable carbohydrate levels for the crown. These samples could be referenced against each other and with existing databases of asparagus field root carbohydrate levels. Root fresh weights were taken initially but the auger method was deemed too inaccurate for usable biomass distribution projections using the soil core method (Drost and Wilson 2003).

## **Yield and quality assessment**

Yields were determined from the center two rows of each sub-plot on fifteen different harvest dates from 19 April to 29 May 2012. Harvest was done by hand, using the snapping method common in Michigan. Spears were harvested at 15-25 cm length. Those spears with diameters below 1 cm, as well as prematurely ferning or damaged spears were discarded. Cumulative fresh weight of marketable yield was determined and used for analysis. In addition, spear quality was assessed from all spears on two harvest dates (14 May and 24 May), classifying spears into the following size grades by diameter at the butt: Small (between 7.9 and 12.7 mm), Regular (12.7 to 17.5 mm) and Jumbo (greater than 17.5 mm). The total number and fresh weight of spears in each size grade was evaluated, and mean weight per spear calculated by dividing the total number of spears by their total weight for each size category.

## **Fern evaluation**

Fern samples were collected twice per season in 2011 and 2012. Once before cladophyll (photosynthetic fernlet) loss in early fall (4 Oct, 2011 and 28 Sept 2012) and again after winter dormancy had fully occurred after first frost in late November (15 Nov 2011 and 9 Nov 2012). At the first sampling date five ferns were randomly selected from each subplot, cut at the soil surface and weighed fresh. A subsample of five plants from each subplot was then dried, separated into cladophyll and stem tissue, and weighed. At the November sampling date, all fern from four 1 m subsamples per subplot were cut off at the soil surface and weighed in the field. Field weights were taken for each plot with an accuracy of 5 g. In addition a 25 fern subsample from each plot was weighed fresh, dried, and weighed again to obtain an estimate of fern moisture content at the main research greenhouse in Lansing, where it was air-dried for a week

in open paper grocery bags. At this point fern samples were separated into stem and cladophyll proportions with hand shaking in paper bags and with manual separation. Dried stems and branches were weighed as were the dried cladophylls taken from them. By compiling this data differences in the amount of leaf surface potential between treatments could be analyzed.

### **Pest evaluation**

Generally pest pressure was monitored and controlled using IPM standards set by MSUE and local vegetable cropping consultants to minimize interference with treatment effects. When scouting resulted in abnormal trends with entomological or pathological pests, relevant specialized MSU labs were consulted for appropriate reactions. Pest pressure and populations were evaluated on either per plant or per m of row basis from data rows depending on the situation.

### **Statistical analysis**

The fixed effects of irrigation (none, overhead or drip) and variety (Millennium or Jersey Supreme) on asparagus stem number, stem dry weight, yield, root soluble carbohydrate content (Brix), weed and insect density, disease ratings and volumetric water content were analyzed using PROC MIXED procedures in SAS (SAS Institute, 2009) with replicate (block) treated as a random effect. To improve assumptions of normality and homogeneity, stem number data was either log- or square root-transformed; cladophyll data was log transformed; and weed and insect density data square root transformed. All other responses did not require transformation. P-value 0.10 was used as the statistical benchmark of significant differences in these trials due to the variability inherent with asparagus field data; P-values below 0.10 are included in all tables to

allow the reader to see when greater confidence levels were observed in trends. Volumetric water content (VWC) data was aggregated by depth to analyze season long changes in water availability. To evaluate differences in water use by depth (10 cm increments to 1 m in depth) between varieties, the change in VWC during extended dry periods (7 July – 24 July, 2011 and 17 June – 14 July, 2012) was evaluated in un-irrigated treatments. For each dry period, daily water loss from each soil depth was calculated by dividing total water loss from that depth by the number of days. For this analysis, only the fixed effect of variety was evaluated, with replicate (block) treated as a random effect. Differences between treatments were evaluated using the pdiff LSMEANS option in the PROC MIXED procedure.

## **Results**

### **Rainfall, irrigation and soil moisture**

Rainfall was below long-term averages in both 2011 and 2012, with significant periods of drought occurring in both years (Figures 2-2 and 2-3). At our experimental site, dry periods occurred mostly during July and August in 2011 (Figure 2-2), and mostly in June during 2012 (Figure 2-3). Approximately 15 cm of overhead irrigation was applied in six (2011) and seven (2012) separate application events each year.

Volumetric water content (VWC) to 1 m depth revealed that in 2011, irrigated treatments had significantly greater VWC for an extended period ranging from late July until late August (Figure 2.4). In contrast, differences in VWC in 2012 were limited to the first two weeks in July, and the last week in August (Figure 2-5). These patterns were as expected given the timing of rainfall and irrigation in each year (Figures 2-2 and 2-3).

Not surprisingly, overhead irrigation resulted in higher soil moisture at the soil surface during dry periods in both years. For example, on 28 August 2011, VWC in the top 20 cm of soil was approximately twice as high in overhead compared to sub-surface drip treatments (Figure 2-6). In contrast, VWC between 20 and 60 cm was higher in sub-surface drip treatments.

Although effects of variety on soil VWC on specific dates were generally not detected (data not shown), changes in VWC during dry periods following rainfall events revealed significant differences in both total moisture removal by variety, as well as the depth of water removal (Figures 2-7 and 2-8). In both years, Jersey Supreme removed more total water during dry periods than Guelph Millennium, and also removed water from deeper in the soil profile. In 2011, total water loss from the top 1 m of soil between 11 July and 24 July was 1.46 mm/day in Jersey Supreme treatments, compared to 1.03 mm/day for Millennium (Figure 2-7). Similarly, in 2012, total water loss was 2.46 mm/day for Jersey Supreme compared to 1.83 mm/day for Millennium (Figure 2-8). In Millennium treatments, approximately 58% of this water loss came from the top 30 cm of soil, whereas for Jersey Supreme, only 45% came from the top 30 cm of soil.

### **Fern development**

In both 2011 and 2012, fern stem number in August was affected by variety, with Jersey Supreme having more total stems, and more mature stems in both years (Table 2-2). Irrigation had little effect on stem number in either year. In 2012, more new stems were detected in un-irrigated treatments, presumably due to delayed fern development prior to August counts.

Light interception from developing fern—an indirect measurement of fern leaf area—was significantly higher in both irrigated treatments compared to the un-irrigated control 2011, and

significantly higher for Jersey Supreme compared to Millennium in both years (Table 2-3).

During the 2012 season a significant impact of irrigation on light interception existed with the New Jersey variety at the 6 August sampling date, but with Millennium asparagus differences declined below significant levels.

During the 2011 season Jersey Supreme fern dry weight was more than 25% greater than that of Guelph Millennium at both early and late fall sampling dates (Table 2-4). Sub-surface drip irrigation increased cladophyll dry weight by 55% in 2011 over control plots.

### **Root brix**

Analysis of root carbohydrates followed the seasonal trends we generally expected as laid out in the Aspire carbohydrate system (Wilson et al. 2002). Root brix levels decreased slightly with harvest, troughed as the fern canopy developed, and moved up to peak levels by the time of fall senescence as carbohydrates were added due to fern photosynthesis (Table 2-5). The effects of irrigation on brix levels were not significant at most sampling dates. However, in fall 2011, drip irrigation increased brix levels in New Jersey Supreme but reduced brix levels in Millennium.

### **Pest evaluation**

Contrary to expectations, irrigation treatments had no detectable effect on purple spot severity or on Marestalk density in 2011 (Table 2-6). We had anticipated that overhead irrigation might increase purple spot severity by increasing leaf wetness relative to sub-surface drip and non-irrigated treatments. Rust and purple spot fungi were present in trials but remained under threshold level of control with foliar sprays as dictated by the IPM program. Rust levels

remained under control both seasons through preventative applications of fungicide and also were not particularly severe in nearby production areas.

Herbicide applications in the 2011 season were limited due to asparagus's susceptibility to chemical damage before it becomes fully established. Summer annuals were not a major problem in this trial due to good chemical control after 2010; hand weeding and selective herbicides in the initial season maintained field cleanliness. However, numerous Marestalk seedlings were evident in fall 2010, and large Marestalk weeds were present in all plots by the end of 2011. We had hypothesized that overhead irrigation would promote weed growth by increasing moisture availability at the soil surface. However, no irrigation effect on weeds was detected (Table 2-6) in this trial. Marestalk density was higher in Guelph Millennium treatments relative to Jersey Supreme treatments, presumably due to greater light penetration under the smaller Millennium fern during the fall of 2010. In 2012, greater residual herbicide use was possible because of increased tolerance with older asparagus so weed populations were diminished. In addition, canopy closure for both varieties was greater in 2012, leading to very few weeds.

Certain insect populations in the field were at times allowed to rise above IPM threshold levels due to close proximity with ongoing entomological trials. Invasive Japanese beetles populations in the region were unusually high in 2011 and 2012 and caused some topical foliar damage in this trial. Insect monitoring conducted in early July 2012 revealed that asparagus beetle densities were much greater in overhead irrigation plots (Table 2-7). This effect was particularly noticeable in New Jersey Supreme where beetle densities in overhead-irrigated plots were nearly thirty times more prevalent than in un-irrigated controls. By August of both years defoliation of ferns was noticeable in some plots as a result of beetle feeding. In contrast, no



effects of irrigation on either Japanese beetles, or asparagus miner damage were detected (Table 2-7). Common asparagus beetle and asparagus miner populations were in the normal population range during the years of the trial. Asparagus aphid, cutworm, yellow striped armyworm were problematic in nearby production fields during this trial but were not noticeable in our plots.

### **Yield and quality**

Asparagus yield in 2012 was influenced both by variety and by irrigation (Table 2-8). The average yield difference with both varieties together in the abbreviated harvest season was an additional 11 to 12%. Precipitation and degree-day accumulation in 2010 were not extreme, allowing for good initial plant set for all plots. But due to high temperatures and spotty precipitation in the 2011 and 2012 there were numerous stressful drought periods in Western Michigan. Yield increases in weight of marketable asparagus ranged from 6-9% with the New Jersey Supreme to 13-21% with Millennium. Local evaluation trials have established that baseline un-irrigated yields with both these cultivars exceed the existing local production average, so these production gains are substantial in real terms. Millennium spear weight increased 7-12% for irrigated plots; a production shift which lowers per unit harvest cost. Overhead irrigation also increased Millennium spear number in 2012 but did not increase spear number in drip plots. New Jersey yields showed an alternate trend with a 15% increase in spear number for drip irrigation but no significant increase in numbers with overhead irrigation treatment.

## **Soil chemical properties**

At this stage of the trial major changes in soil conditions between treatments are not yet evident. Although annual soil tests have indicated several gradual changes are occurring: In overhead irrigation plots both the pH and Mg levels increased, possibly as a result of deposition of minerals from hard-water from the well. Potassium levels appear to be low in soils of the New Jersey overhead irrigation plots so further available nutrient testing at crown level in future seasons may be justified. Inclusion of cover crops into the system would play a positive role in increasing soil organic matter and buffering nutrient levels but thus far the practice has been avoided to avoid confounding data from ongoing experimental treatments.

## **Summary and Discussion**

Overall, short-term results from this study confirmed most of our original hypotheses. Most notably, irrigation during in the 2011 growing season resulted in 2012 crop yield increases of 6 to 21% depending on variety and delivery system (Table 2-8). Given prices received by growers in 2012, these yield increases corresponded to an increase in gross revenue of approximately \$250 to \$1500 per hectare. These yield improvements are particularly impressive given that yields in 2012 were reduced substantially by a late frost. Given estimated cost of approximately \$2,500 per hectare to install irrigation, yield improvements similar to what we observed in 2011 need to occur 2 to 10 times during the anticipated 15-year life of the asparagus planting to justify installation of irrigation.

Although there was no statistical difference between yield responses of Jersey Supreme and Millennium to irrigation in 2012, our study revealed some interesting differences in varietal response. First, fern growth of Jersey Supreme was consistently more responsive to irrigation

than that of Millennium. This was reflected both in fern dry weight at the end of the season (Table 2-4), and in light interception rates during fern growth (Table 2-3). Second, Millennium yield improvements were equivalent or greater than those of Jersey; these results suggest that fern growth is a poor indicator of irrigation response in Millennium. Trends in both fern dry weight and light penetration suggest that Jersey Supreme fern growth may be more responsive to sub-surface drip irrigation compared to overhead irrigation, and vice-versa for Guelph Millennium. Third, in 2012 the effect of irrigation on cladophyll dry weight differed significantly by variety; overhead irrigation significantly increased cladophyll dry weight for Jersey Supreme, but had not detectable effect on cladophylls of Millennium. Fourth, the root carbohydrate concentrations of Jersey and Millennium varieties also responded very differently to irrigation (Table 2-5). As expected, Jersey Supreme root soluble solids concentration (brix) increased with irrigation. In contrast, soluble solids in Millennium were either unaffected or declined with irrigation. These results suggest that previous attempts among researchers in New Zealand (Wilson et al. 2002) to monitor the status of total root carbohydrate storage by examining brix from root samples are likely highly misleading and counterproductive for making management decisions for the variety Millennium. As noted by the original proponents of this system, brix sampling must be accompanied by sampling of total storage root biomass to get a more accurate measurement of total carbohydrate storage (Drost and Wilson 2002). Unfortunately, sampling of total root biomass is a very labor-intensive process given storage roots heterogeneous distribution in the soil. Potential inaccuracies due to the inclusion of inactive storage roots in older plants could further complicate carbohydrate testing's validity (Read 2009).

Our results suggest that had intensive root sampling been conducted in this trial, Millennium may have been shown to respond to irrigation by increasing total storage root biomass rather than simply increasing carbohydrate concentration. Future research elucidating the irrigation response mechanisms which increased Millennium yields would be helpful for developing and adapting applied analysis methods, which in turn would lead to improved field management tools. The greenhouse trial described in Chapter 3 is a first step towards achieving that goal.

## **CHAPTER 3**

### **Effects of long- and short-duration drought stress on asparagus**

#### **Introduction**

There has been little grower interest in irrigating asparagus in Michigan until recently since it was relatively drought tolerant and lacked high profit margins during the past several decades. In recent years the cost of producing asparagus has risen sharply, along with the farm gate price, so risk is more intrinsic in production of this uninsurable crop. Irrigation is increasingly seen as an insurance policy to protect grower's substantial investments from direct drought loss as well as a means to maximize yield potential. There were several years in the 2000's when Western Michigan experienced notable drought conditions. Yields and asparagus stand health were noted to be generally lower in the years following stressful drought conditions. It was uncertain what percentage of the yield decrease could be attributed to drought induced stunting and how much was the result of other related factors, including increased incidence of root disease from abiotic stress. Local growers became more interested in getting information about the drought tolerance of their asparagus and potential plant responses to abiotic stress so they could make better production decisions.

Although field irrigation experiments provide the most relevant information to growers, greenhouse experiments have the distinct advantage of providing controlled conditions to better understand the response of asparagus to specific drought conditions. Greenhouse experiments in confined containers also allow more thorough sampling of root tissue than is feasible under field conditions.

Numerous previous greenhouse studies of asparagus drought have been conducted since the 1990's (Schaller and Paschold 2009; Liddycoat and Wolyn 2009; Hebner et al. 2006;

Bhowmik et al. 2001; Drost and Wilcox-Lee 1989; Drost and Wilcox-Lee 1997; Nicola and Basoccu 2000; Elmer 1995), but few have direct relevance to Michigan soils, varieties and current growing practices. Most greenhouse irrigation research has been too short in duration to gain perspective on the cumulative impact of drought stress on asparagus development (Drost and Wilcox-Lee 1989; Elmer 1995). Likewise, most greenhouse experiments have been too limited in physical scale—using small greenhouse pots—to adequately assess impacts of drought stress without root restriction. In some cases, potting soil was used instead of the droughty sands found in most asparagus production areas. Additionally, in many trials asparagus was started from seedlings whose drought tolerance differs dramatically from the crowns typically utilized in Michigan. Finally, in many studies, archaic non-hybrids or foreign white spear cultivars were used that don't share enough traits with the varieties currently being planted in Michigan for meaningful extrapolation of results.

Nonetheless, several greenhouse trials provide insight into asparagus response to drought stress that may be relevant for Michigan. Drost and Wilcox-Lee (1989, 1997), using relatively small pots and high organic potting soil demonstrated that yield the following season was impacted by drought pressure at fern stage; growth of rhizome buds was negatively impacted by limited water supply and asparagus showed a linear decrease in crown mass accumulation when subjected to high water deficits. Paschold and Schaller (2003, 2009) have done extensive work evaluating effects of drought stress on asparagus using sandy soils in large-volume containers over several seasons. Although this work focused primarily on varieties relevant in Europe, it demonstrated the value of large container evaluations, and that stomatal activity and hydraulic xylem activity can already be inhibited before asparagus is visibly drought stressed.

For Michigan asparagus growers, several important information gaps remain. Historic climate data from Michigan's primary asparagus growing region (Hart, MI), demonstrate that there are often extensive periods during asparagus fern growth in which evapotranspiration exceeds rainfall (Figure 2-1 and data not shown). For example, during the years 2005-7 three to four weeks of drought stress occurred. Over the past 15 years, drought periods of 3-4 weeks in July and August occurred 7 times. The extent to which commercially grown asparagus varieties in Michigan (e.g. Guelph Millennium) may be able to tolerate such acute drought conditions is unknown. Nor is it well understood how longer-duration, lower level drought stress may influence asparagus growth. Better understanding of the conditions contributing to yield loss from drought stress should help growers make more informed decisions about whether to invest in irrigation, and how to best to utilize irrigation systems already in place.

The project goal was to evaluate the effects of both long- and short-duration drought stress under controlled conditions on: asparagus fern establishment, yield, as well as root biomass and carbohydrate accumulation. This trial's relevance to local production was ensured by using sandy soils in a large soil volume container with a popular newer variety: Guelph Millennium. Millennium was selected because of its widespread commercial availability, and its noted absence in many previous irrigation trials. Additionally, surprisingly high yield gains with many European varieties that share Millennium parentage (Panka and Rolbiecki 2009; Rolbiecki and Rolbiecki 2008; Lamparski and Rolbiecki 2006) indicate that Millennium might be more sensitive to drought than New Jersey Supreme, which shares lineage with the relatively drought tolerant New Jersey Giant cultivar. Our hypothesis was that drought stress would lead to reduced fern and root growth rates and reduced spear yields. We further anticipated that the nature of

drought stress (prolonged low level stress vs. short-duration acute stress) would differentially impact fern growth, root growth, carbohydrate concentration and yields.

## **Materials and Methods**

### **Site characteristics and experimental design**

This trial was established in the spring of 2011 in the Michigan State University Horticulture Farm head house. This is a dual layer plastic, year-round structure that is six meters tall with thermostatically controlled forced air heaters and limited cooling capability from a venting roof. With greenhouse trials water inputs could be finely controlled, allowing for testing the cumulative impact over several seasons of drought conditions typical of Oceana County. Four treatments were examined in a complete factorial design with four replications. Factors included baseline water level (high or low) and drought stress (none or 2-3 weeks of water withholding) imposed over two growing seasons, with an imposed dormancy period separating the two seasons. The entire experiment was repeated in time, with the first cycle occurring over two asparagus seasons during 2011-12, and the second cycle occurring over two asparagus seasons during 2012-13. The two cycles were conducted in MACX (Decade Products, Grand Rapids, MI). The same soils were used during both cycles since there was no evidence of soil pathogens and since root tissue had been separated so it would not have deleterious effect on new crowns. The timing of key activities for this trial is summarized in Table 3-1.

### **Trial establishment**

Eight MACX produce bins were divided in half with impermeable 3 cm thick foam board partitions to create sixteen equal 115 x 50 x 78 cm plots. Plot soil volume was 391 L with a 10



cm lip above soil level to ensure plot separation. This substantially larger soil volume compared with most previous trials allowed for reduced edge effects in general, interplant root interaction, and longer periods of growth before root restriction became severe. MACX boxes are also useful, because they are designed for easy lifting and transport using a forklift and since transport to cold environments was necessary to induce dormancy. Boxes were placed with approximately 20 cm separating each box to facilitate airflow and access for data collection.

Glacial sand of similar texture to West Michigan's was collected near an existing asparagus field trial at the "Sand Hill" farm in East Lansing. Soil was collected from the A, B, O, and C profiles with a backhoe from two nearby excavation holes at the same site; soil composition was 83% sand, 12% silt, 5% clay. Half of the A-B soil was sterilized with steam for 12 hours to kill weed seeds and put aside to be used as a topsoil layer over the asparagus crowns. The remaining soil was hand shoveled into 10 cm soil layers from two trailers to simulate real soil profiles and to equalize pH imbalances between the soils from the two holes, which ranged from moderately acidic to alkaline. Between each profile a mixture zone was worked up with a spade to avoid potential root limiting layers.

Year old Millennium asparagus crowns were donated by Michigan commercial growers (Ron Richter and Oomen Farms). Disease-free crowns with several viable growth buds and storage roots were weighed and selected for uniformity; 16 sets of five crowns were selected with combined weights for each plot within 5 g of each other.

Fertilizer was added based on soil tests and MSU recommendations for asparagus. Fertilizers were applied following typical grower practice, with an initial application of mono-ammonium phosphate fertilizer (12-52-0) applied below the crowns at planting. Additional urea

and 17% N-P-K fertilizer applications were applied to the soil surface and raked in after each successive harvest period as dictated by soil tests.

Supplemental lighting was provided with timer-controlled 400W metal halide lights suspended 3 m directly above each MACX box. Light levels were logged hourly with a Hobo U-12 data logger (Onset, Bourne, MA), along with temperature and relative humidity to calculate growing degree-day (GDD) accumulation.

Growing season length in the greenhouse was calibrated to roughly match the five-year GDD base 40°F average of 2500 in Hart, MI in the center of the asparagus-producing region. After achieving 2500 GDD each growing season, plants were removed via forklift from the greenhouse and placed either outside if temperatures were near freezing point or in a walk-in cooler kept dark at 3°C to simulate winter dormancy (Table 3-1).

### **Moisture monitoring and irrigation**

Baseline irrigation consisted of either a “high” level of irrigation slightly in excess evapotranspiration value of asparagus. “Low” irrigation treatments received one half the amount of water as high treatments, and were established to assess the impact of a prolonged period of continuous sub-optimal soil moisture levels. In acute drought stressed treatments irrigation was withheld for 2-3 weeks at the end of the fern canopy development stage (Table 3-1).

Irrigation was achieved primarily through buried Netafim Uniram tape with pressure compensating emitters on 30 cm spacing; water input was regulated within ten percent of 0.19 L per minute. Each plot had five emitters; four were buried at crown depth and one was slightly above soil level. During the initial watering of each growing season a watering wand was used to bring soil in all plots to field capacity.

Moisture was monitored with the same equipment as in the larger field trial described in Chapter 2. A Sentek Diviner tube was cut to 75 cm and used to track moisture throughout the soil at depths from 10-60 cm in each plot. The top 10 cm values were discarded because of lack of good soil contact. Tubes were installed 20 cm from both the front and sidewalls to ensure accurate readings without grossly infringing on the asparagus crown's growth area. Soil moisture levels were taken with the Diviner on at least a weekly basis throughout the growing season with additional readings taken prior to or following irrigation events. One Decagon moisture sensor (EM10) per plot was also used to data-log (EM5) soil moisture hourly at crown level, and to serve as a backup data gathering system in the event that Diviner-2000 measurements failed. Due to the reliability of the Diviner probe Decagon sensor use was discontinued after the first two growing seasons.

### **Root sampling**

Root sampling protocols followed Drost (2002) and the Aspire soluble carbohydrate monitoring system developed in New Zealand (Wilson et al. 2002). Fleshy root samples were taken at time of planting from surplus crowns to establish a baseline soluble root carbohydrate level. Additional fleshy root samples were taken at the end of the first growing season, post-drought the second season, and at the end of experimental cycle when the plants were exhumed after two growing seasons. Several 5-10 centimeter fleshy roots were dug out from at least two crowns per partition with a hand trowel after use of soil cores to obtain root samples was judged to be overly destructive to the plants.

Root samples were collected, washed, and placed into the freezer overnight to separate fructose from cell tissue (Wilson et al. 2002). Just prior to analysis, root samples were thawed,

dab dried with paper, cut into 0.5 cm sections and at least 2g were crushed in a garlic press to collect root sap. The refractive index (RI) of extracted sap was evaluated using an Atago PAL-1 digital handheld refractometer. This device reports RI readings in degrees Brix based on the relationship between RI and the sucrose concentration (% w/w) of a pure sucrose solution at 20°C. Care was taken to avoid sample dilution from thawed surface moisture and each sample reading was taken three times and averaged.

### **Fern evaluation**

Fern evaluation occurred on a regular basis throughout the growing season at roughly two-week intervals. Stems were counted in each plot and either all stems were measured for height or a subsampling was recorded to document the development stage of the fern. At several sampling dates, stem counts were conducted separately for stems that were visually assessed to be fully mature (older tissue based on leaflet color), newly mature (fully expanded cladophylls but light green tissue), immature (small light green shoots with minimal cladophyll development), and dead (aborted by the plant).

Fern growth was also evaluated at several critical stages for new shoot growth, by measuring the length of new shoot tissue at 5 locations on the developing fern. New shoot growth appears as a distinctly lighter shade of green. Randomly selected branchlets were evaluated for amount of fresh growth at two points on both the north and south side of the fern, and one at the pinnacle. This method was used to determine fern growth without destructive sampling or reliance on a light sensor.

After the asparagus had reached 2500 GDD, boxes were either brought outdoors for frost exposure or placed in a walk-in cooler to induce dormancy. After fern senescence dead fern was

collected by cutting stems at ground level with hand pruners. From each plot stems were combined, dried and weighed after air-drying in the greenhouse. It should be noted that during the first cycle, stem and cladophyll tissue was largely intact at the time of fern biomass sampling. However, during the second cycle most cladophyll tissue and some stem tissue had senesced at the time of sampling, resulting in underestimates of fern dry weight and possible sample bias.

### **Yield and quality assessment**

Following the dormancy period, boxes were returned to the greenhouse and re-watered to field capacity (to mimic typical spring soil moisture conditions in Michigan) to initiate the second season of growth (see Table 3-1 for timings). Fresh asparagus spears were harvested to evaluate the effects of first season irrigation and drought stress treatments on yield, and to mimic harvest pressure typical of second year asparagus. Spears were snapped at 20 cm height directly above the soil level in the manner asparagus harvest occurs in Michigan. Spear number per plot was documented along with weight of harvested spears. Spears were initially categorized as either: marketable, small-diameter, or unmarketable appearance. A subsample of spears were weighed fresh, dried and weighed again to determine the fresh to dry weight ratio.

### **Second year fern growth and root sampling**

Following harvest in the second year, fern was allowed to develop and irrigation and drought stress treatments were re-initiated, with the same plants receiving the same treatments for a second year (see Table 3-1 for timing). Evaluations of fern development were conducted during the second season of both cycles using the same protocols described above for the first season with one exception: stem height during the second growing season was taken for each

individual stem in each box, and the sum of all stem heights per box was calculated. This total stem height was considered a reasonable non-destructive estimate of total fern growth, although clearly it is not a perfect estimate, since stem diameter, branching, and cladophyll development are not captured by this method.

After completing a second fern growth season, MACX boxes were disconnected from the irrigation supply lines and exposed to frost one final time to return soluble carbohydrates to the crowns from the above ground biomass. Following senescence, dead fern was removed, dried and weighed as described above for season one. Boxes were then placed on a large sheet of diamond metal grate and were flipped over with a forklift to separate soil and fleshy root tissue. Asparagus crowns and fleshy root tissue remained above the grating as the soil was sieved out, while fine roots and soil passed through the metal grate. Care was taken to make certain that root and crown tissue from the two partitions did not mix on top of an open bin.

As each of the five crowns from each partition was freed of the majority of the soil they were cleaned by hand brushing to remove sand. This process was adequate in most cases to remove nearly all the soil particles. If necessary roots were rinsed off with a hose and dabbed dry with towels and air-dried for 30 minutes. Fresh fleshy-root and crown tissue was subsequently weighed. Approximately 5g samples of fleshy root were taken from each crown to assess carbohydrate levels following the protocol described above. The remaining crown and root material was dried and weighed.

## **Statistical analysis**

The fixed effects of baseline irrigation (high vs. low) and drought (none vs. 2-3 wks.) on asparagus stem number, stem height, stem dry weight, yield, root weight, and root soluble

carbohydrate content (Brix) were analyzed using PROC MIXED procedures in SAS (SAS Institute, 2009) with replicate (block) treated as a random effect. To improve assumptions of normality and homogeneity, spear number data was log-transformed. All other responses did not require transformation. One outlier for asparagus yield and stem number in this trial was detected and removed from the data-set prior to analysis; justification for removal was based both on its deviation from the mean, and the fact that outlier data came from the MACX box closest to a heat source in the greenhouse. This appeared to promote earlier and more vigorous spear emergence (data not shown) and subsequent fern growth. In most cases, significant interactions of fixed effects with year were detected, so analyses were conducted separately by year. However, for responses for which no significant year interactions were detected—including yield—combined data for both years are presented. Given the high degree of variability in many of the responses, mean separation was conducted whenever main or interactive effects had p-values of less than 0.10. These p-values are noted at the bottom of each table of results. Otherwise, significance is reported as “NS”. Differences between treatments were evaluated using the pdiff LSMEANS option in the PROC MIXED procedure.

## **Results**

This greenhouse experiment had a number of interesting results that helped explain and expound on the data taken in related asparagus field trials. Our key findings were based on stem number and weight data, yield, and root data taken over the three and a half growing seasons completed so far in this trial.

### **Stem number during first growing season**

During the first growing season, neither baseline irrigation nor drought had any detectable effect on stem number (Table 3-2). In the second repetition of the trial 79 days after planting (DAP) crowns we had significantly higher stem numbers in the high water supply plots over the low water supply. High irrigation increased stem number by over 20% in this second trial repetition. This significant trend was still evident a month later at 111 DAP. By that point in the growing season drought treatment had also significantly negatively impacted development of asparagus fern. Drought stress reduced the total stem number by 14% by August 20, 2012. In the two drought treatments there was a reduction of old (mature) stem numbers by 23% in the drought treatments compared to the two non-drought stressed plots (Table 3-2). This implies a photosynthetic surface disadvantage for the drought treatments during asparagus's prime photosynthetic period to recharge root storage carbohydrates. In a field situation, inability to develop a strong fern canopy shortly after harvest can lead to detrimental late season growth as the plants struggle to make up for a carbohydrate deficit before the winter season.

### **Fern dry weight**

In both repetitions of the experiment, fern dry weight at the end of the first growing season was not affected by baseline irrigation, drought stress, or their interaction (Table 3-3). Similar to the stem number results in the fall of 2011 there was no significant difference between treatments, although data indicated a trend likely was emerging between the drought treatments with greater weight fern in no-drought plots. By April 2012 after high/low water treatments had been imposed twice, there were significantly greater fern weights in the high water plots, 423g per plot versus 285g. When compiled between both growing cycles this represented a 22%



difference in cumulative fern weight between the high and low water treatments. In 2012 during the second run of the trial fern weight was highly variable after the first season so no significant differences were detected. This variability was probably related to how late in the season ferns were collected: many cladophylls had already fallen off because of exposure to frost and wind and all soluble carbohydrates had already been trans-located back to the crowns. This late collection date was purposeful since we did not want to extract usable carbohydrates from the more stressed treatments that had entered senescence more slowly than the high water no-drought plots.

### **Yield, spear number and spear size after one season**

Asparagus yield following the first season was variable, resulting in low power to detect significant differences. Nonetheless, when data was combined across both years, a 15% reduction in yield was detected due to the short-duration drought stress imposed in the first season (Table 3-4; P-value = 0.074). This yield reduction in acute drought stress treatments was attributable largely to a decreased number of harvested spears rather than a reduction in spear size; spear number was 14% lower in short-duration drought stress treatments (P-value = 0.064). In contrast, neither spear number, spear size, nor yield were affected by chronic low water stress treatments (Table 3-4).

### **Stem number and height in second growing season**

Following the first dormant and harvest periods multiple counts of stems occurred to document the development of the fern canopy. These plants were larger than they had been the previous growing season and their growth habits altered somewhat. In the first repetition of the

trial (2011-12), stem number at both 30 and 69 days after harvest (DAH) was significantly lower in low irrigation compared to high irrigation treatments (Table 3-5). In the second run of this greenhouse trial this effect was not detected, with stem number at 7 and 28 DAH unaffected by either baseline irrigation or drought stress (Table 3-5). Stem number information later in the season is not yet available.

Total stem height (sum of the height of all individual stems—see methods) during the second growing season was unaffected by treatment in both years at the first two sampling dates from 7 to 25 DAH (Table 3-6). In the first experimental repetition, total stem height at 35 DAH was 22% lower in low irrigation compared to high irrigation treatments, but unaffected by drought stress treatments. In contrast, total stem height at 36 DAH in the second experimental repetition was unaffected by baseline irrigation level, but reduced 13% by short-duration drought stress. Total stem height at 35-36 DAH in the two experimental repetitions was remarkably similar, ranging from 2 to 2.8 m per box in both years (Table 3-6).

### **Root weight and soluble solids**

At the time of destructive harvest of the plots following two growing seasons, low irrigation baseline treatments resulted in a 25.5% decrease in total storage root biomass (P-value = 0.034), while having no effect on the concentration of soluble sugars (brix) in root tissue (Table 3-7). In contrast, short-duration drought stress had no effect on root biomass, but resulted in a 11.9% reduction in brix levels (P-value = 0.058).

## Summary and Discussion

Overall, this trial demonstrated several interesting differences between drought stress treatments of different durations on the commonly grown variety, Guelph Millennium. In particular, our results demonstrated that prolonged low-level drought stress reduced fern growth (Table 3-3) and root weight (Table 3-7), while short-duration intense drought stress, typical in Michigan during July and August (Figure 3-1), had greater impact on root carbohydrate concentration (Table 3-7) and short-term yield (Table 3-4). The trial confirmed our suspicion that monitoring root carbohydrate levels is an inadequate method for guiding management decisions in the absence of more intensive sampling to evaluate changes in storage root biomass. The study also reinforced the conclusion from our field study, that targeted irrigation to avoid short-duration drought stress can improve asparagus yields. In particular, we observed a 15% improvement in yields one year after establishment when drought stress was avoided through irrigation. Moreover, although long-term yield impacts could not be evaluated, increases in root biomass and carbohydrate content due to irrigation, suggest that yield boosts in subsequent years would also be likely as a result of irrigation.

Although these results are interesting and shed light on the effects of different types of drought stress, they should be interpreted cautiously due to several problems encountered along the way. The basis of our greenhouse production methodology was from field practices used on my farm; professional greenhouse growers may have made different choices more adapted to indoor production. Among the major challenges encountered in this trial were: adapting field data collection methods and devising establishment and production protocols for a crop that's rarely produced indoors. The most complex issues were uncertainty about how to: initiate

senescence in the asparagus plant, adapt to atypical fern growth habits, and find effective non-destructive evaluation tools for determining net plant productivity.

## **Dormancy**

Typically asparagus fern in the field browns off in the middle of autumn. Krug (1999) reported that either freezing temperatures or severe drought stress could initiate “ripening” in the asparagus plant. Drought is the primary method of initiating senescence in desert climates in South America and California’s Imperial Valley. Due to the large cumulative water holding capacity of the soil within the MAXC boxes involved in this trial completely depleting available moisture would have altered the seasonal growth pattern too far beyond what could reasonably occur in Michigan’s climate. Drought conditions that severe could easily alter the crown development to an extent that would have rendered trial data un-transferable into local field conditions.

Walk in coolers were available for use but larger freezers could not be located on campus to freeze plastic containers. It was assumed that removal from daylight and acclimatization to near freezing temperatures would bring about dormancy. Ku and Woolley had found that asparagus had a chilling requirement of three weeks at 5°C to break dormancy (2008). Once the initial 2500 GDD had been achieved in the greenhouse plants were brought to the coolers and left in unlit 3°C coolers for over a month with no strong sign of senescence beginning. Addition of fifteen liters of ice to the soil surface per plot did not activate dormancy cycle by bringing temperatures down to freezing.

After the initiation of this greenhouse trial Landry and Wolyn (2011) published a report breaking down the senescence pattern of Millennium asparagus into three stages. Cues for each

stage were broken down into a mixture of: amount of crown nitrogen, moisture percentage in fleshy root, short day length signaling, and level of various proteins and carbohydrates in the crown. We concluded the simplest way to satisfy enough of these factors to induce dormancy was to bring the plants into contact with natural frosts after a full growing season as occurs in the field. After waiting another month in the unlit cooler ambient outdoor temperatures dropped and all boxes were placed outdoors and following one hard overnight frost event fern dormancy quickly occurred.

This natural frost method was not always possible, since the trial's time frames of dormancy induction did not always correspond to periods of cold outdoor temperatures (Table 3-1). If the growing season ended between October and April this was not a problem in Michigan but during the summer in the greenhouse 2500 GDD accumulated very quickly resulting in at least one problematic dormancy shift per calendar year. Potentially lower cumulative light absorption occurred in these growing seasons due to abbreviated growth time frames and asparagus's maximum photosynthetic capacity of around 1000  $\mu$  mole (Faville et al.1999).

### **Temperature regulation**

The headhouse was selected to accommodate the very large containers used in the experiment and forklift, but ventilation systems and temperature control were suboptimal, resulting in variable temperatures ranging from 10°C to 37°C between the seasons. Greenhouse temperatures were documented at levels well above plant photosynthesis shutdown threshold during the day in the summer because side venting or air-conditioning of the building was not possible.

Growth time had to be supplemented to some growing seasons to compensate for the hours over 35°C that rarely occur in the 2500 GDD Michigan growing season. In the summer of 2012 an additional 500 GDD was allowed to compensate for this shutdown time. Also Takatori (1971) noted finding that root growth dramatically slows when temperature exceed 30°C so the heat levels may have confounded differences in root development between the various treatments because humidity levels could have altered the soil temperature at root level. In a Michigan field situation this never would have occurred because of the consistently cool soils.

Another challenge conducting this trial was related to uneven heat dispersion due to placement of heaters at the ends of building and the lack of cooling capability in the greenhouse. This temperature induced growth disparity was particularly evident when outdoor temperatures were below freezing causing a greater temperature gradient in different areas of the building. Asparagus spear elongation rates are directly correlated with temperature (Ku 2008). Asparagus in plots in the first repetition (particularly the south-most box) often left dormancy more quickly than the rest of the trial because of heating disparities resulting from the forced air system's location. If this trial was repeated, the inclusion of a guard row on the first plot would be warranted, as would re-randomization of the plots before every growing season to improve data quality.

Radiative heat loss through the plastic layer was theorized to be the reason for frost damage shortly after harvest periods during the winter as quickly growing stems are very frost susceptible once no longer protected by proximity to the ground. Once fern was formed fully and hardened off it was resilient to minor radiative frost damage in the greenhouse.

Similar cosmetic frost damage to winter harvested spears led to the consolidation of the marketable and unmarketable spear categories. Combining all larger sized spears into one

production amount was more reflective of actual plant yield potential. The small diameter spear category was retained to represent similar small spears that emerge from minor growth buds in the field. Likewise we did not engage in trimming of over length spears and discount that weight as other trials have, since in a production field that mass could have been harvested between 20 and 30 cm with a daily harvest crew.

### **Unusually tall ferns in second season**

By the second season in the greenhouse trials fern heights were significantly taller than field-grown asparagus of similar age. The height of some individual ferns measured well over three meters while most field fern heights are closer to two meters. Ferns were very leggy, with weak stems resulting in pronounced canopy drooping by the end of the growing season. This may have been the result of either insufficient total light levels or altered light-spectrum composition from the supplemental metal halide lighting with natural light filtered through plastic. The lack of wind, which is known to discourage vertical growth, may have also played a role, as could have soil pH.

### **Future work**

Asparagus is a difficult crop to trial scientifically under unpredictable field conditions, so greenhouse work has value to complement field data. Further repetitions of similar large-scale greenhouse studies, where all tissue root can be captured could be used to better understand how asparagus responds to environmental pressures. Additional information on the response of soluble solid concentration and root growth in combination with measurements of stomatal conductance would be a logical extension of this work, and would help inform asparagus decision-tools like Aspire.

Rhizocam tubes had been installed in this trial at its inception to monitor feeder root development but the process proved cumbersome and beyond the resources of this trial. Use and analysis of digital rhizocams with root tracking software during both drought events and dormancy periods would be very useful to the body of knowledge. In addition, inclusion of a root pathogen factor, like *Fusarium* or *Phytophthora* could help explain the commonly referenced but little researched relationship between drought stress and asparagus soil pathogens.



## **CHAPTER 4**

### **Discussion**

#### **Field and greenhouse trial**

Differences in fern number, cladophyll weight, and total dry fern weight were apparent during both field seasons from irrigation treatments, although the drought conditions in Oceana County weren't as severe as they were in much of the nation. The yield increases of 6 to 21% with irrigation treatments during the 2012 field season were an indication to me that there are effects in asparagus that cascade from one season to the next. I'd hypothesize that long-term this positive trend, resulting from irrigation, will continue benefitting both plant survival rates and yields; making irrigated plantings more lucrative enough to justify additional expenses. Cultivar reactions to environmental stress and irrigation treatments showed this may not be a "one size fits all" situation. Our Diviner 2000 data illustrated different water requirements from various soil depths for the two cultivars: UG Millennium was more responsive to surface watering, making it a good match with fields with center pivots installed. Planting Millennium seedlings or small crowns in sand under a center pivot would likely be more beneficial than planting large Millennium crowns in heavier soil (Sinton and Wilson 2008).

Our results suggest that some alterations in spray program may be warranted in irrigated fields, since irrigation resulted in large amounts of fresh fern growth that beetles appeared to prefer for shelter and feeding (Lamparski et al. 2010). The same holds true with purple spot and other foliar diseases related to leaf wetness hours, although we saw less of this than we expected in our trial. Inclusion of chemical stickers with fungicide sprays may also be warranted under center pivot irrigation.

## **Future work**

Typically trials of this variety end at the point when production is really coming into maturity, when growers sunk costs are covered and profits are made. Continuation of this field trial for several more years will yield a great deal of usable information; even if data collection rates are downsized the long-term trends observed in those plots will be much more applicable to the industry.

Both increased usage of cover crops and altered herbicide programs are options with overhead irrigation that would be logical extensions of future research. Soil health in our existing production system would probably benefit from including a cover crop besides the winter rye commonly employed, on alternating years with normal residual herbicide use to keep down perennial weed populations. Identifying systemic insecticides, fungicides, and nutrient packages that could be chemigated into trickle irrigated asparagus is another practical topic for future research.

Attempting higher density plantings with several modern varieties in irrigated Michigan fields would also be interesting; other irrigated production areas have already moved in this direction. Although Ku (2006) and Schaller and Paschold (2009) did some recent work on asparagus's photosynthetic rate, further investigation of modern North American cultivars in field conditions would be helpful to optimizing water supply scheduling.

It appears that the Aspire soluble carbohydrate model won't be commonly utilized by Michigan growers. One alternative approach, which may have practical application and warrant future research would be development of a temperature and accumulated light growth model to

better predict carbohydrate accumulation. Already growers depend on weather stations to monitor fungal development and hopefully will have insect pest modeling available soon.

Use of high tunnels both for drought research and altered harvest schedule production is another area for prospective future work. Given that asparagus does not readily exhibit drought stress symptoms, another topic that might benefit growers would be identification of easily visibly wilting “indicator-plants” that could be planted into the edges of irrigated asparagus fields in the place of electronic sensors for irrigation scheduling.

### **Industry implications**

The USDA NASS reported in 2010 that 1843 of Michigan’s 4310 asparagus producing hectares were planted prior to the year 2000; most of these were replant situations with the New Jersey Giant cultivar not known for its longevity in those situations. From 2005 to 2009 only 1112 hectares were planted in Michigan because of the depressed markets (USDA NASS 2010). The combination of stand age (+12 years), water stress during the 2011 and 2012 growing seasons, increased harvest pressure due to high prices, and continued attrition from soil diseases will quickly remove many of these older fields from production. Additionally, growers intended to plant only 1355 hectares from 2010 to 2015, and since 2009 increased seed costs and limited availability have further restricted new plantings (Bakker 2010-2012). A new challenge has developed for Michigan growers: supply of both spears and young plants during the past several years have been tight and there is no indication that this trend will reverse soon.

There is room for market growth as Michigan only supplies 2% of the nation’s asparagus; providing the Great Lakes region with its May and June fresh asparagus supply is a feasible goal (Martin 2010). However, maximizing production per unit of land is essential to make asparagus

profitable with the hand harvest system. Michigan's asparagus industry has to focus on consistent high quality production and improving yield per acre to meet this existing demand or other production areas will step in and fill the void. Midwestern asparagus production levels have the potential to rebound in the next several decades but significant adaptation of production methods may be required in order for a full resurgence to occur.

Michigan is not an arid environment, but even with a drought-hardy crop like asparagus, supplemental irrigation can be of substantial value for ensuring consistent, optimum production levels. Results from this study show that under weather conditions similar to the past several years, yield increases from irrigation, in addition to increased overall plant health are likely to justify the additional cost of irrigation for Michigan asparagus growers. Irrigation systems will not necessarily provide substantial benefit every year in Michigan, but the increased yield in dry years in conjunction with high plant survival rates over time due to greater plant health should keep fields in higher production ranges longer.

## APPENDIX

Table 2-1 Schedule of major events, 2010-2012

Hart, MI field trial

	2010	2011	2012
Dec.- March	NA	Senescence	Senescence
April	NA	Field mowed/ Spear emergence/ Fertilized/Harvest on 28th	Field mowed/ Spear emergence/ Multiple frosts
May	Field cultivated/Crowns planted	Harvests on 9&12th/ Layby herbicides applied	Harvest period of several weeks
June	Canopy Development/ Weed control	Canopy development/ First generation Miners/ Fungicides applied/ Irrigation started	Fern canopy development/ Multiple irrigation events
July	Full immature fern 1 Meter apx.	Full mature fern/ Drought period: multiple irrigation events/ Root samples	Full mature fern/irrigation
August	Full immature fern	Full fern/Second generation canopy ferns develop/Irrigation	Secondary canopy development/ Drought: irrigation events
September	Full immature fern	Final irrigation/Senescence initiated in Millennium	Full fern/Miner evaluation/Irrigation pulled
October	Senescence initiated	Semi-dormant/First fern sampling	Senescence initiated/ First Fern sampling
November	Fern dormant/ Root Samples	Full senescence/ Second fern sampling	Semi-dormant/Second fern sampling

Table 2-2 Stem number by category, 8 August 2011 and 2 August 2012.

Hart, MI

Treatment	2011				2012			
	Total	New	Mature	Dead	Total	New	Mature	Dead
	-----#/m-row-----							
Irrigation Main Effect								
None	28.6	1.6	25.4	1.6	28.3	<b>6.3 b</b>	21.0	2.4
Overhead	28.5	2.6	24.7	1.3	29.1	<b>4.4 a</b>	23.8	2.1
Drip	28.9	2.0	26.0	0.9	31.1	<b>3.6 a</b>	26.6	2.1
Variety Main Effect								
Jersey	<b>35.1 a</b>	2.5	<b>30.8 a</b>	1.7	<b>33.2 a</b>	4.8	<b>27.5 a</b>	2.5
Millennium	<b>22.3 b</b>	1.5	<b>19.9 b</b>	0.8	<b>25.8 b</b>	4.7	<b>20.1 b</b>	1.9
Interactive Effects								
Jersey								
None	<b>35.3 b</b>	2.0	<b>31.3 ab</b>	2.0	30.5	6.9	22.3	3.4
Overhead	<b>32.0 ab</b>	3.1	<b>27.0 b</b>	1.8	33.0	4.0	28.5	2.1
Drip	<b>38.0 a</b>	2.5	<b>34.3 a</b>	1.3	36.1	3.3	31.6	2.0
Millennium								
None	<b>21.9 c</b>	1.1	<b>19.6 c</b>	1.1	26.0	5.8	19.8	1.5
Overhead	<b>25.0 c</b>	2.0	<b>22.4 c</b>	0.6	25.3	4.8	19.1	2.1
Drip	<b>19.9 c</b>	1.5	<b>17.8 c</b>	0.6	25.9	3.9	21.5	2.1
ANOVA	-----P-value-----							
Variety	<b>&lt;0.0001</b>	0.236	<b>&lt;0.0001</b>	0.126	<b>0.012</b>	0.720	<b>0.010</b>	0.300
Irrigation	0.974	0.561	0.723	0.862	0.705	<b>0.044</b>	0.201	0.964
Variety x Irrigation	<b>0.066</b>	0.887	<b>0.008</b>	0.931	0.671	0.540	0.549	0.208
Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).								

Table 2-3. Light interception by asparagus fern, 2011 and 2012.

Hart, MI

Treatment	2011			2012		
	19-Aug	6-Sep	4-Nov	9-Jul	6-Aug	14-Sep
	-----% light intercepted-----					
Irrigation Main Effect						
None	<b>77.2</b> <b>b</b>	<b>74.7</b> <b>b</b>	<b>69.7</b> <b>b</b>	68.3	65.3	83.3
Overhead	<b>82.1</b> <b>ab</b>	<b>85.0</b> <b>a</b>	<b>78.7</b> <b>ab</b>	66.8	71.7	81.1
Drip	<b>82.7</b> <b>a</b>	<b>84.1</b> <b>a</b>	<b>80.1</b> <b>a</b>	68.3	72.6	84.2
Variety Main Effect						
Jersey	<b>86.1</b> <b>a</b>	<b>85.4</b> <b>a</b>	<b>79.5</b> <b>a</b>	<b>71.9</b> <b>a</b>	<b>75.3</b> <b>a</b>	<b>87.3</b> <b>a</b>
Millennium	<b>75.2</b> <b>b</b>	<b>77.1</b> <b>b</b>	<b>72.8</b> <b>b</b>	<b>63.7</b> <b>b</b>	<b>64.4</b> <b>b</b>	<b>78.4</b> <b>b</b>
Interactive Effects						
Jersey						
None	82.0	78.0	72.2	74.1	<b>65.5</b> <b>b</b>	88.8
Overhead	86.1	89.0	80.9	68.9	<b>78.2</b> <b>a</b>	87.9
Drip	90.2	89.2	84.9	72.5	<b>82.1</b> <b>a</b>	85.2
Millennium						
None	72.4	71.4	66.6	62.5	<b>65.0</b> <b>b</b>	77.8
Overhead	78.2	81.1	76.4	64.7	<b>65.2</b> <b>b</b>	80.6
Drip	75.2	78.9	75.2	64.0	<b>63.1</b> <b>b</b>	76.9
ANOVA						
	-----P-value-----					
Variety	<b>&lt;0.0001</b>	<b>0.001</b>	<b>0.009</b>	<b>0.001</b>	<b>0.001</b>	<b>0.002</b>
Irrigation	<b>0.078</b>	<b>0.019</b>	<b>0.053</b>	0.792	0.102	0.583
Variety x Irrigation	0.346	0.702	0.583	0.359	<b>0.027</b>	0.823

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).



Table 2-4. Fern stem and cladophyll dry weight, 2011 and 2012.

Treatment	Hart, MI		Early Fall				Late Fall	
	Cladophyll	Stem	Total	Cladophyll	Stem	Total	Total	Total
-----grams biomass/m2-----								
Irrigation Main Effect								
None	<b>136 b</b>	883	1018	116	999	1115	862	595
Overhead	<b>183 ab</b>	986	1210	141	953	1093	945	652
Drip	<b>211 a</b>	1024	1235	150	1003	1153	962	670
Variety Main Effect								
Jersey	195	1065	<b>1283 a</b>	134	<b>1106 a</b>	1240	<b>1028 a</b>	678
Millennium	157	852	<b>1010 b</b>	138	<b>864 b</b>	1001	<b>818 b</b>	600
Interactive Effects								
Jersey								
None	132	<b>971 ab</b>	<b>1122 ab</b>	<b>89 b</b>	935	1024	966	618
Overhead	202	<b>940 ab</b>	<b>1187 ab</b>	<b>168 a</b>	1110	1278	1011	710
Drip	250	<b>1282 a</b>	<b>1539 a</b>	<b>144 ab</b>	1273	1418	1107	708
Millennium								
None	141	<b>795 b</b>	<b>915 b</b>	<b>143 ab</b>	1062	1205	758	572
Overhead	159	<b>1046 ab</b>	<b>1241 ab</b>	<b>114 ab</b>	795	909	880	595
Drip	172	<b>764 b</b>	<b>931 b</b>	<b>156 ab</b>	733	889	817	633
ANOVA -----P-value-----								
Variety	NS	NS	<b>0.0592</b>	NS	<b>0.088</b>	NS	<b>0.0207</b>	0.3043
Irrigation	<b>0.0896</b>	NS	NS	NS	NS	NS	NS	0.6031
Variety x Irrigation	NS	<b>0.0898</b>	0.084	<b>0.099</b>	NS	NS	NS	0.9644

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Table 2-5. Root soluble solid concentration, 2011-2012.

Hart, MI

Treatment	2011		2012	
	Summer	Fall	Summer	Fall
-----Percent BRIX-----				
Irrigation Main Effect				
None	N/A	16.4	10.2	12.6
Overhead	9.6	16.5	9.3	12.5
Dri	7.5	16.0	9.2	12.1
p				
Variety Main Effect				
Jersey	<b>7.3 b</b>	16.2	8.9	12.6
Millennium	<b>10.4 a</b>	16.4	10.2	12.2
Interactive Effects				
Jersey				
None	N/A	<b>14.4 c</b>	9.4	12.5
Overhead	9.7	<b>16.2 bc</b>	8.6	13.6
Drip	N/A	<b>18.0 ab</b>	8.9	11.8
Millennium				
None	10.4	<b>18.3 ab</b>	11.1	12.7
Overhead	9.5	<b>16.8 bc</b>	10.1	11.5
Drip	11.6	<b>14.1 c</b>	9.4	12.3
ANOVA -----P-value-----				
Variety	<b>0.085</b>	0.798	0.632	0.537
Irrigation	0.800	0.914	0.969	0.826
Variety x Irrigation	0.102	<b>0.006</b>	0.775	0.362

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Table 2-6. Weed and disease severity, 2011.  
Hart, MI

Treatment	Marestail Density # weeds per m sq.	Purple spot severity visual rating
Irrigation Main Effect		
None	0.32	4.9
Overhead	0.19	4.5
Drip	0.25	4.3
Variety Main Effect		
Jersey	<b>0.15 b</b>	4.5
Millennium	<b>0.36 a</b>	4.6
Interactive Effects		
Jersey		
None	0.13	4.9
Overhead	0.14	4.3
Drip	0.17	4.4
Millennium		
None	0.51	4.8
Overhead	0.24	4.8
Drip	0.34	4.3
ANOVA		
		-----P-value-----
Variety	<b>0.0399</b>	NS
Irrigation	NS	NS
Variety x Irrigation	NS	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Visual rating (0=none; 10=severe)

Table 2-7. Density and fern damage from key insect pests, 2012.

Hart, MI

Treatment	Insect density (7/9)				Asparagus Miner damage	
	Asp. beetle		Japanese beetle		7/9	9/8
	# insects per m of row				% of stems damaged	
Irrigation Main Effect						
None	0.3	b	0.5		10.1	26.4
Overhead	1.9	a	0.6		11.8	23.3
Drip	0.6	b	0.8		11.2	19.8
Variety Main Effect						
Jersey	0.9		0.7		13.4	18.7 b
Millennium	0.8		0.6		8.2	27.6 a
Interactive Effects						
Jersey						
None	0.1	b	0.8	bc	12.9	23.4
Overhead	2.8	a	0.0	c	14.5	16.1
Drip	0.3	b	1.1	ab	13.0	16.5
Millennium						
None	0.5	b	0.0	c	4.5	29.3
Overhead	1.0	ab	1.2	ab	9.2	30.5
Drip	0.9	ab	0.5	bc	9.4	23.1
ANOVA						
	-----P-value-----					
Variety	0.933		0.740		0.289	0.026
Irrigation	0.025		0.562		0.753	0.452
Variety x Irrigation	0.067		0.079		0.992	0.5428

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Visual rating of percentage of stems with significant miner damage

Table 2-8. Asparagus yield and quality, 2012.  
Hart, MI

Hart, MI			Quality assessment					
			Yield		Number		Spear wt	
Small	Large	Jumbo						
			kg/ha	#/ spear plot	g/spear	% of harvested spears		
Irrigation Main Effect								
None			2,916 b	153	14.6	25.8	73.2	1.0
Overhead			3,279 a	160	15.6	22.3	76.9	0.8
Dri			3,236 a	164	15.4	24.6	74.0	1.4
p								
Variety Main Effect								
Jersey			3,297 a	187 a	14.1 b	25.0	74.1	0.9
Millennium			2,991 b	131 b	16.3 a	23.5	75.3	1.2
Interactive Effects								
Jersey								
None			3,144	178 b	13.8 c	26.0	72.9	1.1
Overhead			3,317	180 a	14.9 bc	23.9	75.3	0.8
				b				
Drip			3,429	204 a	13.6 bc	25.0	74.1	0.9
Millennium								
None			2,688	129 c	15.3 a	25.7	73.4	1.0
					b			
Overhead			3,242	140 C	16.3 a	20.7	78.5	0.8
Drip			3,043	125 c	17.1 a	24.2	73.9	1.9
ANOVA			-----P-value-----					
Variety			0.0028	<0.0001	0.0001	0.1681	0.2294	0.3927
Irrigation			0.0141	0.4419	0.5143	0.5255	0.4433	0.4461
Variety x Irrigation			0.1427	0.082	0.0607	0.4639	0.3008	0.4524

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

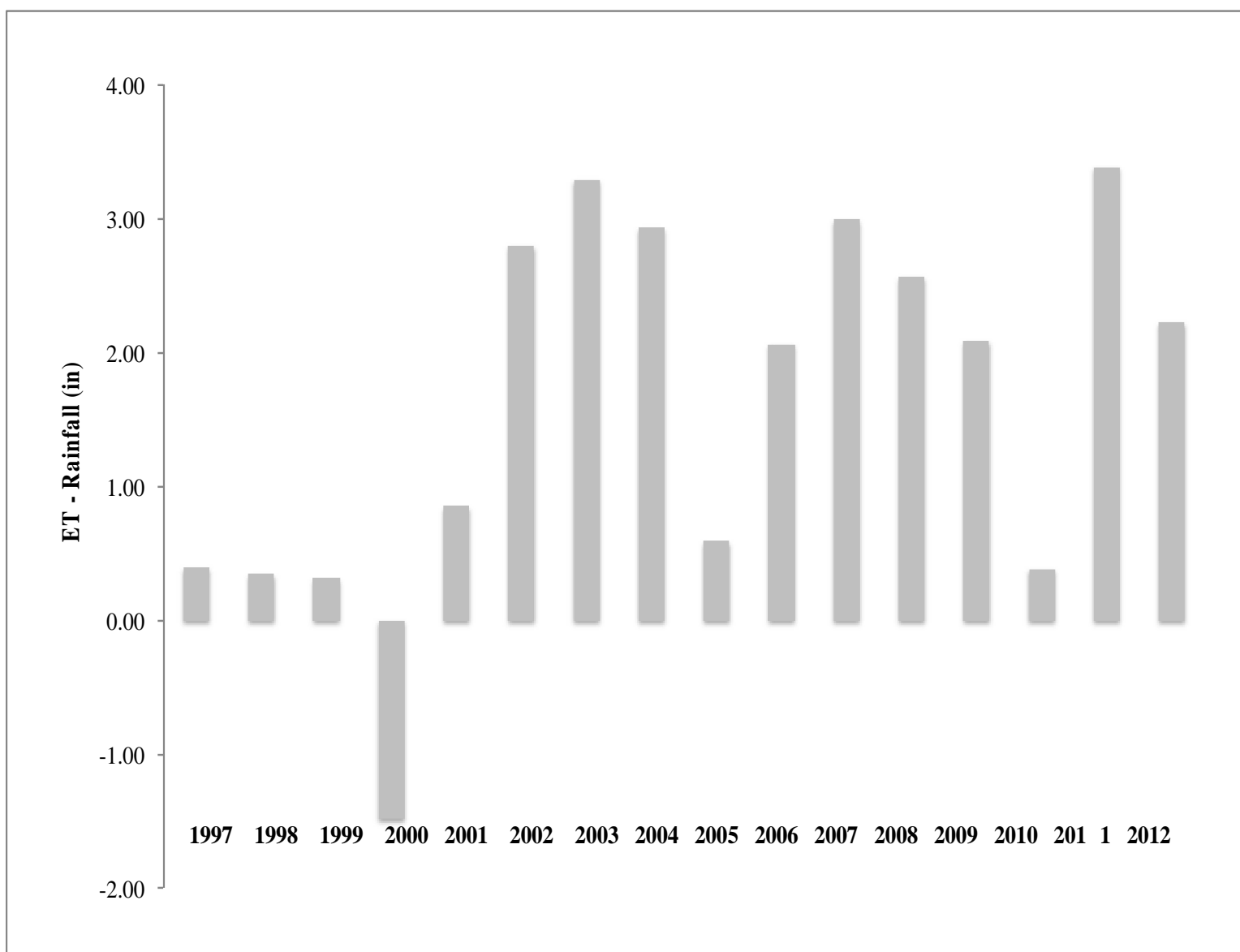
Quality measures were taken from subsample at three harvest dates only.

Table 2-9. Soil chemical properties, 2012.

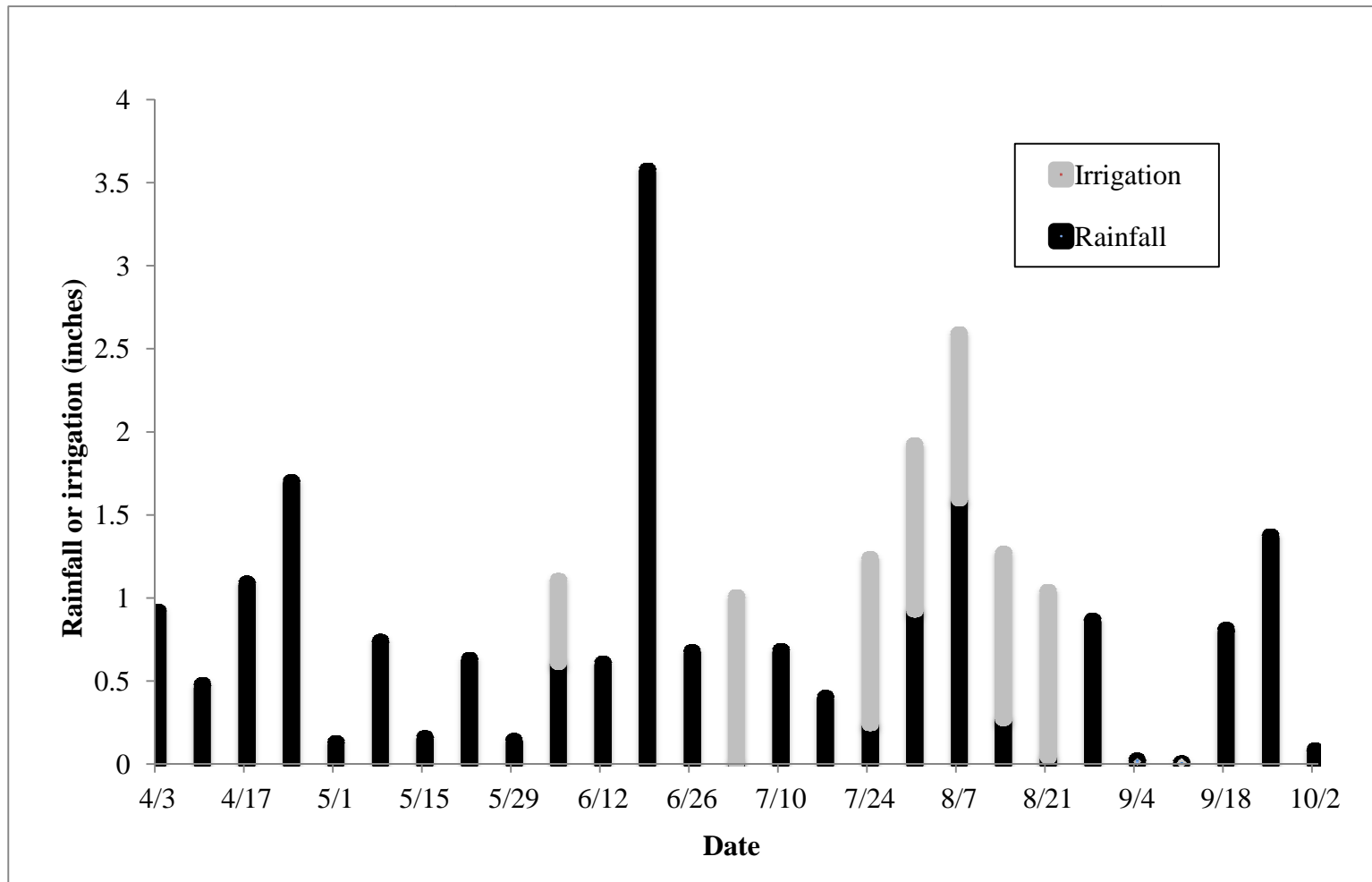
Hart, MI

Treatment	pH	K	P	Mg	Ca
		parts per million			
Irrigation Main Effect					
None	<b>6.6</b> b	111.4	98.1	<b>77.6</b> b	447.6
Overhead	<b>7.0</b> a	91.5	115.8	<b>97.4</b> a	488.5
Drip	<b>6.6</b> ab	106.4	103.4	<b>73.3</b> b	448.5
Variety Main Effect					
Jersey	6.7	103.8	105.9	83.5	473.3
Millennium	6.7	102.4	105.6	82.0	449.5
Interactive Effects					
Jersey					
None	6.6	<b>124.3</b> a	91.0	78.5	467.3
Overhead	7.0	<b>85.5</b> c	119.5	93.3	478.3
Drip	6.7	<b>101.5</b> bc	107.3	78.8	474.5
Millennium					
None	6.6	<b>98.5</b> bc	105.3	76.8	428.0
Overhead	7.0	<b>97.5</b> bc	112.0	101.5	498.0
Drip	6.6	<b>111.3</b> b	99.5	67.8	422.5
ANOVA	-----P-value-----				
Variety	NS	NS	NS	NS	<b>0.080</b>
Irrigation	<b>0.082</b>	NS	NS	<b>0.015</b>	NS
Variety x Irrigation	NS	<b>0.016</b>	NS	NS	<b>0.080</b>

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

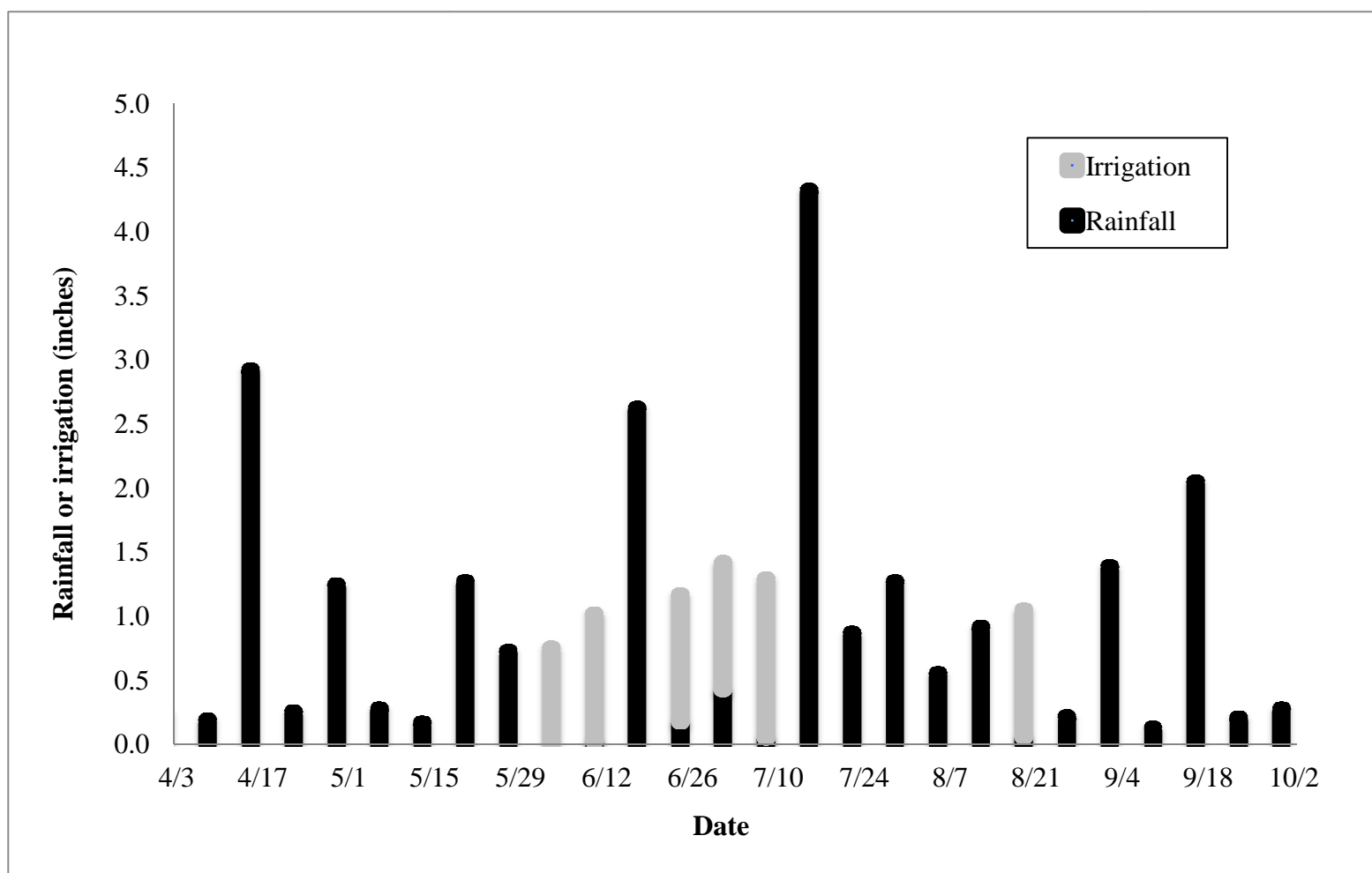


**Figure 2-1.** Estimated evapotranspiration (ET) minus rainfall between July and August, Hart, MI, 1997-2012. ET estimates were calculated as product of PET and crop coefficient for asparagus fern.

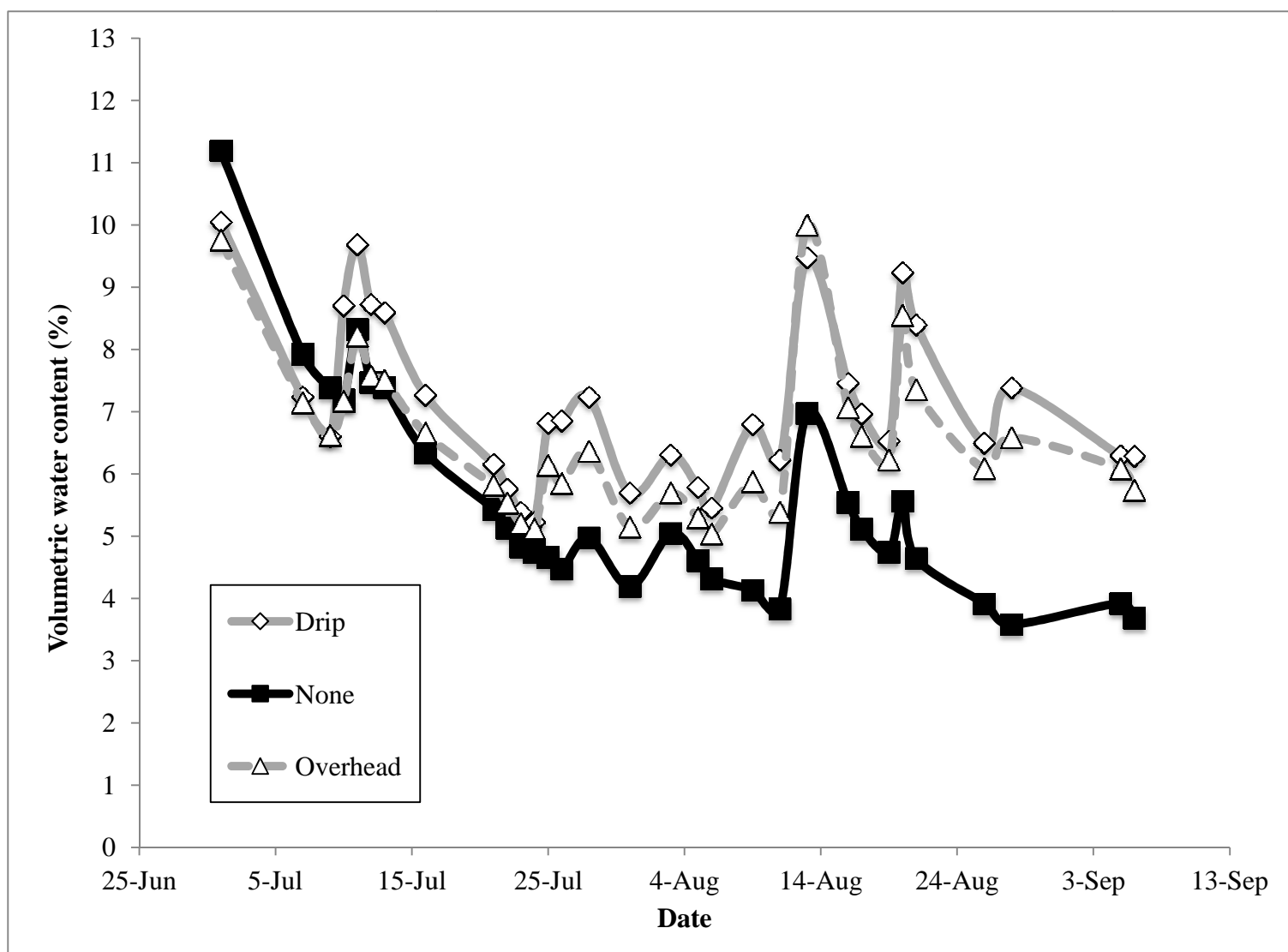


**Figure 2-2.** Rainfall (black bars) and irrigation (gray bars) events during 2011 growing season, Hart, MI.

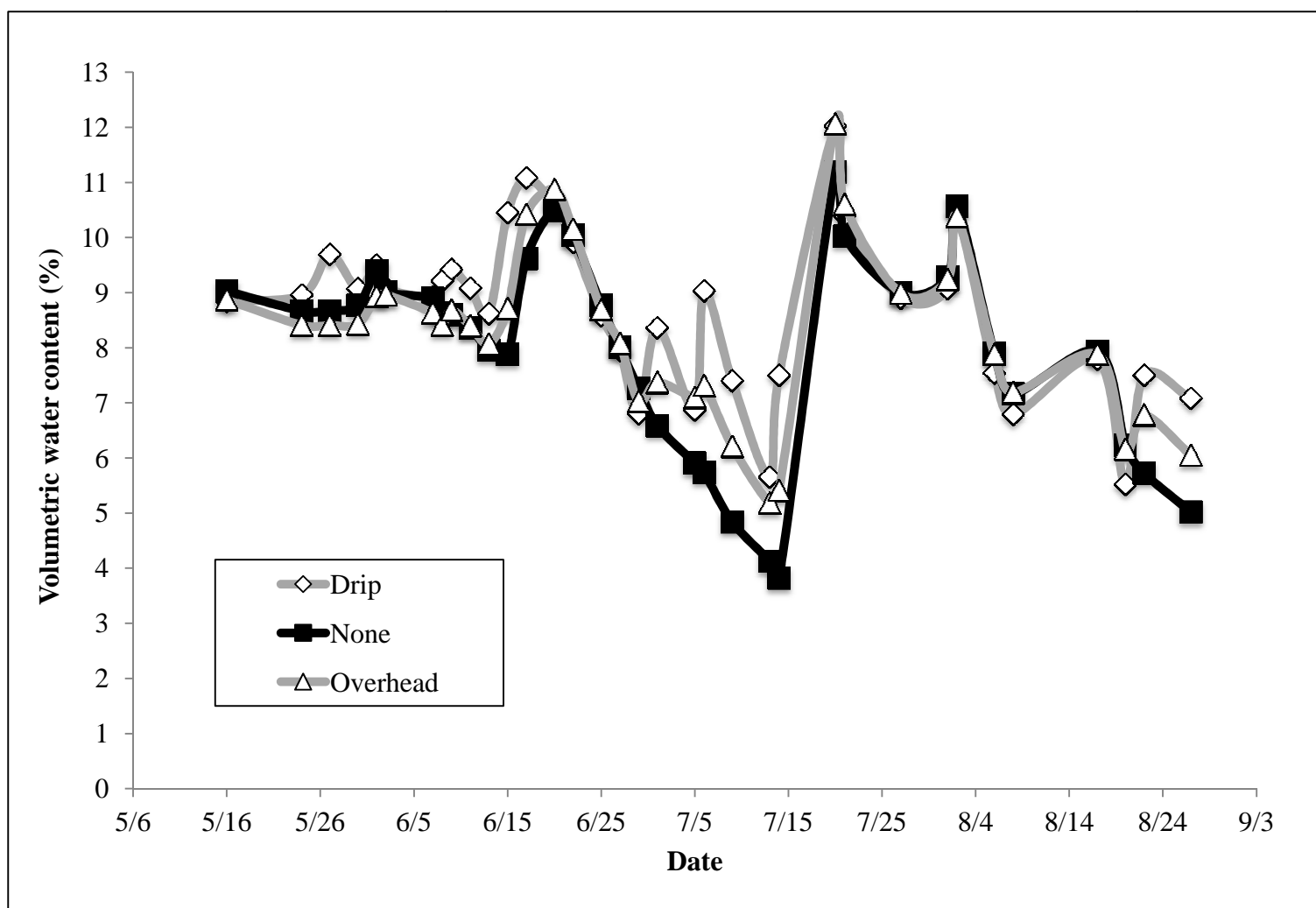




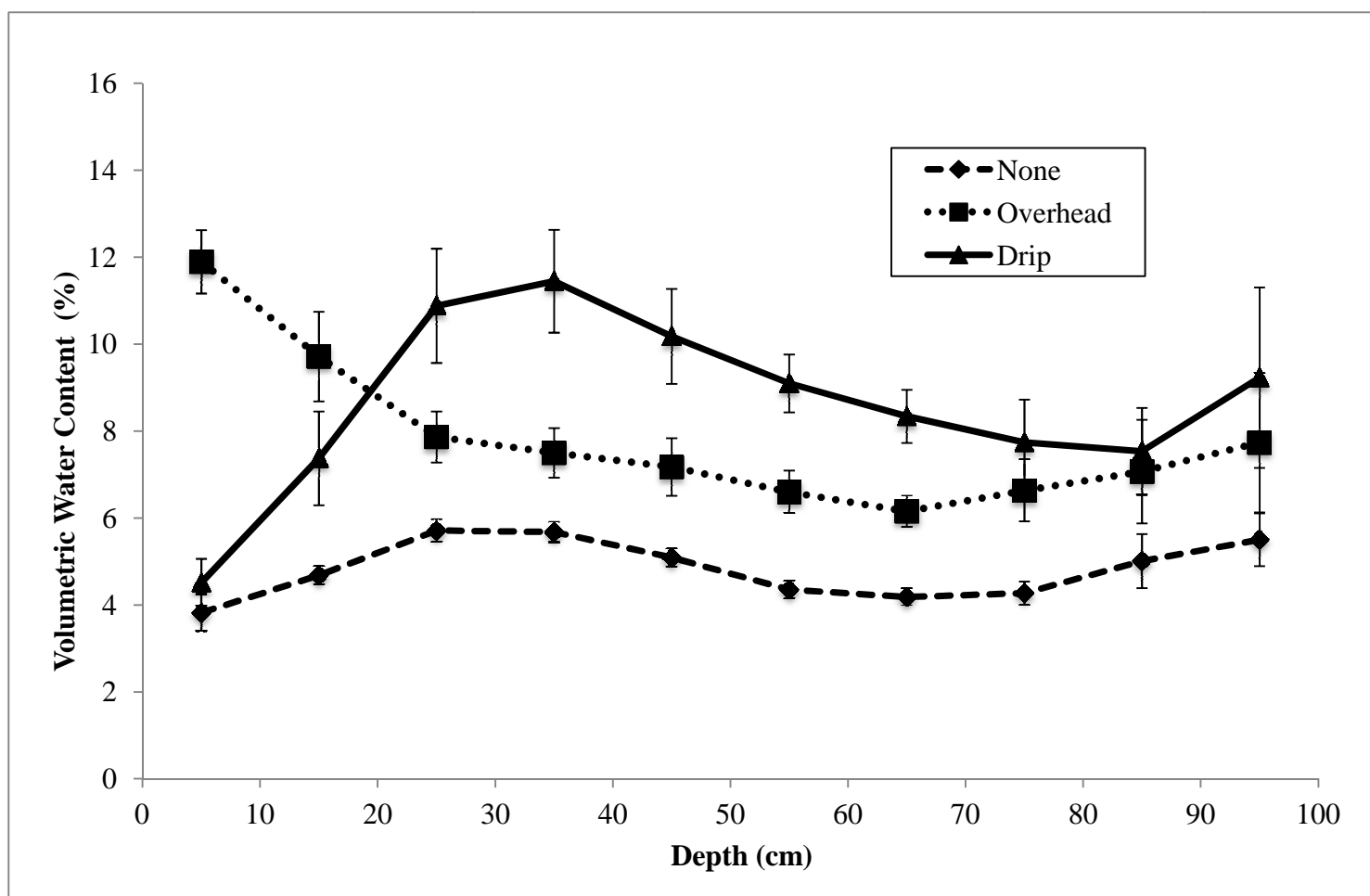
**Figure 2-3.** Rainfall (black bars) and irrigation (gray bars) events during 2012 growing season, Hart, MI.



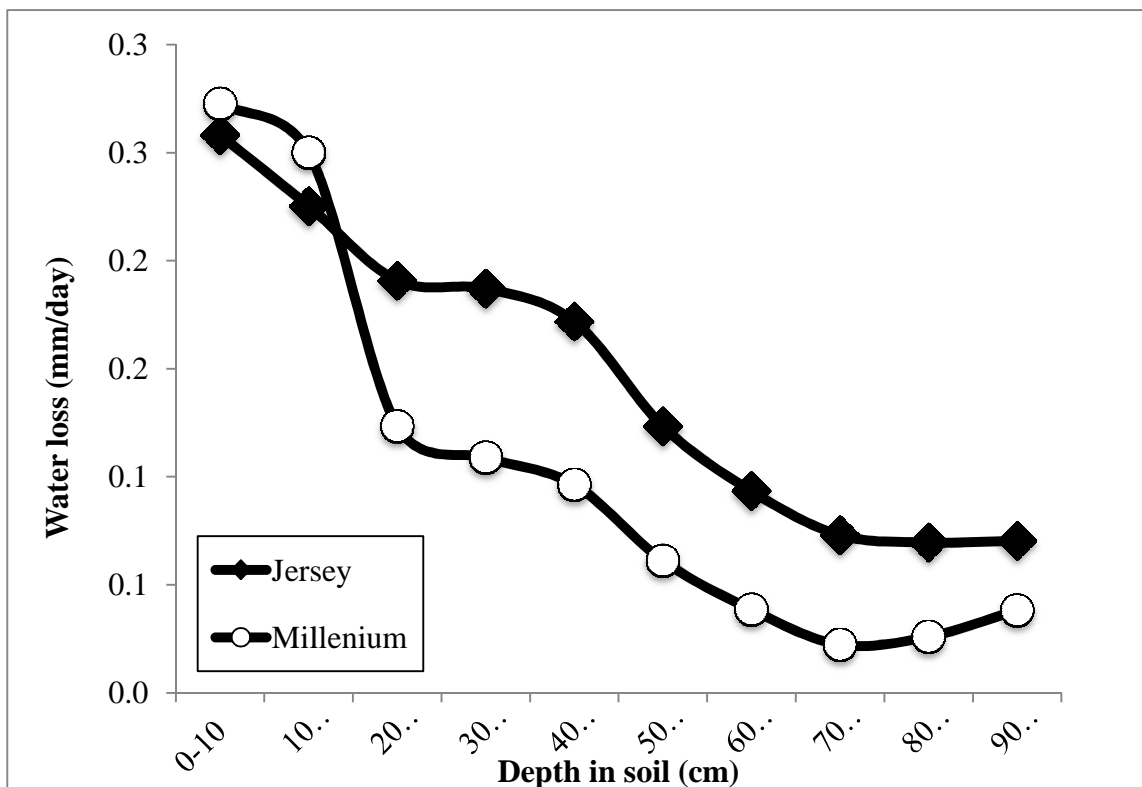
**Figure 2-4.** Mean volumetric water content by irrigation treatment, 2011.



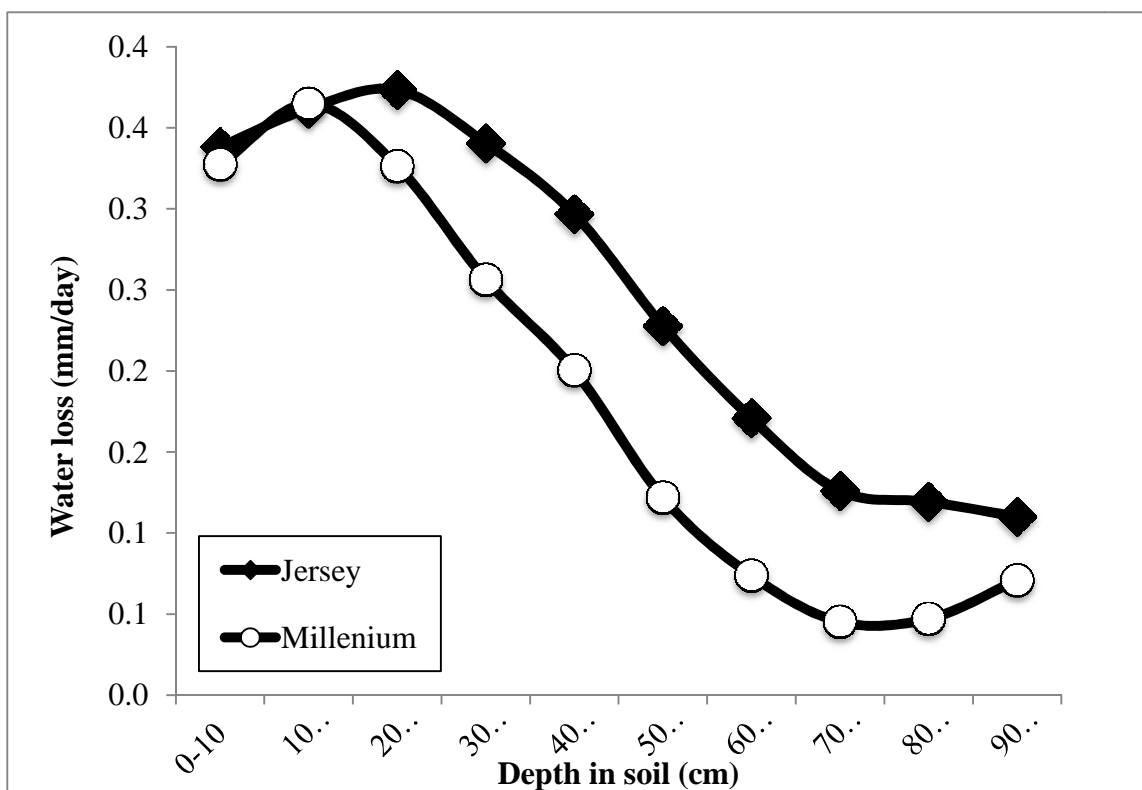
**Figure 2-5.** Mean volumetric water content by irrigation treatment, 2012



**Figure 2-6.** Mean volumetric water content by depth on August 28, 2011. This pattern of moisture distribution by depth was typical of conditions prevailing during most of the month of August, 2011 as well as early July 2012.



**Figure 2-7.** Mean daily water loss by depth between July 11 and July 24, 2011 from un-irrigated treatments containing either Jersey Supreme or Millennium asparagus varieties.



**Figure 2-8.** Mean daily water loss by depth between June 14 and July 17, 2012 from un-irrigated treatments containing either Jersey Supreme or Millennium asparagus varieties.

Table 3-1. Calendar of Activities.			
Activity		2011-12	2012-13
Season 1			
	Crowns planted	3/15/11	5/1/12
	First stem emergence	4/15/11	6/1/12
	Baseline irrigation differences initiated	6/1/11	6/27/12
	Drought stress initiated (trts xy)	6/13/11	6/28/12
	Drought stress terminated	6/28/11	7/16/12
	Root Brix sample	NA	7/23/12
	Cold dormancy initiated	8/2/11	8/20/12
	Fern dry weight samples	11/3/11	11/14/12
Season 2			
	Moved back to greenhouse and watered	11/3/11	1/9/13
	Spears up	11/7/11	1/21/13
	Harvest initiated	11/8/11	1/21/13
	Harvest terminated	12/5/11	2/13/13
	Fern stem counts and heights taken	12/16/11-2/10/12	2/20/13-3/19/13
	Baseline irrigation differences initiated	1/20/12	2/6/13
	Drought stress initiated (trts xy)	1/27/12	3/7/13
	Drought stress terminated	2/17/12	NA
	Cold dormancy initiated	3/21/12	NA
	Fern dry weight samples	4/6/12	NA
	Roots excavated	4/10/12	NA
	Root Brix sample	4/10/12	NA

Table 3-2. Total stem number during first season.

Treatment	2011-12			2012-13						
	5/23/11	6/23/11	7/28/11	6/18/12	7/19/12	8/20/12				
	69 DAP	100 DAP	135 DAP	48 DAP	79 DAP	111 DAP				
	Total	Total	Total	Total	Total	Total	Old	Fern	New	Dead
-----#/ box-----										
Irrigation Main Effect										
High	13.8	32.6	45.4	11.1	<b>20.3 a</b>	33.6	<b>20.4 a</b>	9.8	1.5	2.0
Low	12.4	30.8	44.1	12.8	<b>16.8 b</b>	29.0	<b>15.0 b</b>	9.8	2.1	1.4
Drought Stress Main Effect										
No drought	13.3	32.6	46.8	12.3	19.5	<b>33.8 a</b>	<b>20.0 A</b>	10.1	2.3	1.4
Drought	12.9	30.8	42.8	12.6	17.5	<b>28.9 b</b>	<b>15.4 B</b>	9.4	1.4	2.0
Interactive Effects										
High Irrigation										
No drought	14.8	33.5	47.5	11.3	21.5	36.8	<b>23.5</b>	9.8	2.0	1.5
Drought	12.8	31.8	43.3	13.0	19.0	30.5	<b>17.3</b>	9.8	1.0	2.5
Low Irrigation										
No drought	11.8	31.8	46.0	13.3	17.5	30.8	<b>16.5</b>	10.5	2.5	1.3
Drought	13.0	29.8	42.3	12.3	16.0	27.3	<b>13.5</b>	9.0	1.8	1.5
ANOVA	-----P -value-----									
Irrigation	NS	NS	NS	NS	<b>0.045</b>	0.123	<b>0.019</b>	NS	NS	NS
Drought	NS	NS	NS	NS	0.226	<b>0.038</b>	<b>0.001</b>	NS	NS	NS
Irrig. X Drought	NS	NS	NS	NS	0.755	0.498	<b>0.071</b>	NS	NS	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

DAP = Days after planting; Old = Fully mature; Fern = Newly mature; New =Not fully developed.



Table 3-3. Fern dry weight

Treatment	2011-12			2012-13
	10/27/11	4/11/12	Total	11/6/12
	-----g/box-----			
Irrigation Main Effect				
High	309	<b>423 a</b>	<b>732 a</b>	165
Low	288	<b>285 b</b>	<b>572 b</b>	151
Drought Stress Main Effect				
No drought	314	366	680	159
Drought	283	341	624	156
Interactive Effects				
High Irrigaiton				
No drought	326	423	749	174
Drought	293	423	715	155
Low Irrigation				
No drought	301	310	611	144
Drought	274	260	533	158
ANOVA	-----P -value-----			
Irrigation	0.2356	<b>0.0002</b>	<b>0.0007</b>	NS
Drought	0.1089	0.2900	0.1156	NS
Irrig. X Drought	0.8599	0.2900	0.5107	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

fern had dropped cladophylls at time of 2012-13 sampling

Table 3-4. Spear weight and number

Treatment	Spear Yield			Spear Number			Weight per spear		
	2011-12	2012-13	Combined	2011-12	2012-13	Combined	2011-12	2012-13	Combined
	g/box			#/box			g/spear		
Irrigation Main Effect									
High	152.0	151.8	151.9		19.6	16.6	11.3	7.4	9.6
Low	147.9	143.2	145.7		19.1	15.5	11.7	7.7	10.0
Drought Stress Main Effect									
No drought	160.2	161.5	<b>160.9 a</b>		20.9	<b>17.3 a</b>	11.6	7.7	9.9
Drought	139.6	132.1	<b>136.1 b</b>		17.7	<b>14.8 b</b>	11.4	7.5	9.7
Interactive Effects									
High Irrigation									
No drought	159.0	165.5	162.3	<b>ab</b>	18.5	17.1	11.7	7.5	9.9
Drought	144.9	138.0	141.5	<b>a</b>	20.8	16.1	10.8	7.3	9.3
Low Irrigation									
No drought	161.4	157.6	159.5	<b>a</b>	21.0	17.5	11.5	7.8	9.9
Drought	134.4	124.1	130.0	<b>b</b>	16.7	13.3	12.0	7.7	10.1
ANOVA									
Irrigation	NS	NS	NS	NS	NS	NS	NS	NS	NS
Drought	NS	NS	<b>0.074</b>	NS	NS	<b>0.064</b>	NS	NS	NS
Irrig. X Drought	NS	NS	NS	<b>0.099</b>	NS	NS	NS	NS	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Table 3-5 Total stem number following harvest. DAH = Days after harvest

Treatment	2011-12			2012-13	
	12-Dec	11-Jan	22-Feb	20-Feb	13-Mar
	7 DAH	30 DAH	69 DAH	7 DAH	28 DAH
	-----#/box-----				
Irrigation Main Effect					
High	8.3	<b>13.1 a</b>	<b>13.6 a</b>	9.3	10.8
Low	6.8	<b>10.1 b</b>	<b>10.3 b</b>	9.3	11.6
Drought Stress Main Effect					
No drought	7.8	12.0	12.4	9.5	11.5
Drought	7.3	11.3	11.5	9.0	10.9
Interactive Effects					
High Irrigaiton					
No drought	7.3	13.3	13.8	9.8	11.5
Drought	9.3	13.0	13.5	8.8	10.0
Low Irrigation					
No drought	7.3	9.0	11.0	9.3	11.5
Drought	6.3	8.5	9.5	9.3	11.8
ANOVA	-----P -value-----				
Irrigation	0.108	<b>0.004</b>	<b>0.001</b>	NS	NS
Drought	0.567	0.365	0.271	NS	NS
Irrig. X Drought	0.108	0.540	0.424	NS	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Table 3-6. Total stem height after harvest DAH = Days after harvest

Treatment	Sum of spear heights						Maximum height			
	2011-12			2012-13			2012-13			
	16-Dec	30-Dec	10-Feb	20-Feb	27-Feb	20-Mar	20-Feb	27-Feb	13-Mar	20-Mar
	11 DAH	25 DAH	36 DAH	7 DAH	14 DAH	35 DAH	7 DAH	14 DAH	28 DAH	35 DAH
cm/plot						cm				
Irrigation Main Effect										
High	394	682	<b>2,802 a</b>	435	1,255	2,282	114.0	208.0	<b>252.0 a</b>	264.0
Low	360	544	<b>2,174 b</b>	395	1,099	2,205	99.0	191.0	<b>229.0 b</b>	254.0
Drought Stress Main Effect										
No drought	337	607	2,580	412	1,231	<b>2,395 a</b>	102.0	202.9	244.2	265.0
Drought	417	620	2,395	418	1,123	<b>2,091 b</b>	110.7	196.5	236.5	251.0
Interactive Effects										
High Irrigaiton										
No drought	294	633	2,825	384	1,283	2,478	106.0	210.0	259.0	274.0
Drought	477	731	2,779	486	1,225	2,086	122.0	206.0	244.0	253.0
Low Irrigation										
No drought	378	580	2,337	440	1,179	2,312	98.0	196.0	229.0	257.0
Drought	333	509	2,013	350	1,020	2,097	100.0	187.0	229.0	251.0
ANOVA	P-value									
Irrigation	NS	NS	<b>0.003</b>	NS	NS	NS	NS	NS	<b>0.048</b>	NS
Drought	NS	NS	NS	NS	NS	<b>0.029</b>	NS	NS	NS	NS
Irrig. X Drought	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

Note drought stress in second year was applied at 58 DAH in 2012, and 22 DAH in 2013

Table 3-7. Final root weight and soluble solids, 2011-12  
2011-12

Treatment	2011-12	
	Root weight g/box	Sol. Solids % Brix
Irrigation Main Effect		
High	<b>1698 a</b>	16.2
Low	<b>1352 b</b>	15.2
Drought Stress Main Effect		
No drought	1549	<b>16.7 a</b>
Drought	1501	<b>14.7 b</b>
Interactive Effects		
High Irrigaiton		
No drought	1755	16.3
Drought	1640	14.1
Low Irrigation		
No drought	1342	17.1
Drought	1361	15.3
ANOVA	-----P -value-----	
Irrigation	<b>0.034</b>	NS
Drought	NS	<b>0.058</b>
Irrig. X Drought	NS	NS

Different letters within a column and effect category indicate significant differences according to LSMeans ( $\alpha=0.10$ ).

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## LITERATURE CITED

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