

DEVELOPMENT OF MANAGEMENT PRACTICES FOR ARTICHOKE
PRODUCTION IN SOUTHWEST TEXAS

A Thesis

by

TOGO SHINOHARA

Submitted to the Office of Graduate Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

December 2008

Major Subject: Horticulture

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Approved by:

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ABSTRACT

Development of Management Practices for Artichoke Production in Southwest Texas.

(December 2008)

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Co-Chairs of Advisory Committee: Dr. Daniel I. Leskovar
Dr. Bhimanagouda S. Patil

This research included studies for transplant and field crop management with the purpose of optimizing stand establishment, crop performance and nutritional quality of artichoke (*Cynara scolymus* L.) grown in southwest Texas.

Post-transplanting heat (35/20°C vs. 25/10°C, day/night temperatures) or drought [30% Water holding capacity (WHC) vs. 60% WHC] stress alone or in combination significantly reduced shoot or/and root growth of artichoke seedlings. Combined heat and drought stresses strongly affected shoot water status and root growth. Results from this study imply that it is desirable to improve stand establishment by either conditioning the seedlings to improve root growth or by preventing leaf dehydration by these stresses. Therefore, effects of plant growth regulators (PGR) on root growth and shoot water status were examined.

Ethylene regulators, including precursors or a releasing compound [DL-methionine (MET), 1-aminocyclopropane-1-carboxylic acid (ACC) and ethephone (ETH)], and inhibitors [amino-ethoxyvinylglycine (AVG) and 1-methylcyclopropene (1-MCP)] were applied to seedlings to evaluate their effect on root growth and

development. ACC and ETH ($1-100 \mu\text{M}\cdot\text{L}^{-1}$) enhanced root hair, root area and lateral roots (only with ETH at $30 \mu\text{M}\cdot\text{L}^{-1}$).

The effects of film-forming antitranspirants and abscisic acid (ABA, $500-2000 \text{ mg}\cdot\text{L}^{-1}$) foliar application on physiological responses, water status and hardness of artichoke transplants were examined under drought stress. ABA at $1000 \text{ mg}\cdot\text{L}^{-1}$ enhanced drought tolerance of transplants which was associated with the maintenance of shoot water status via stomatal closure. Film-forming antitranspirants were not effective to mitigate drought stress. These results suggest that ACC and ETH as root enhancers, and ABA as a plant water conditioner, could be useful PGR's to enhance stand establishment in artichoke seedlings.

Field artichoke performance in response to irrigation [50, 75 and 100% crop evapotranspiration (ETc)] and N ($0-180 \text{ kg}\cdot\text{ha}^{-1}$) rates were investigated during three seasons at Texas A&M AgriLife Research in Uvalde, TX. Irrigation was more effective than N rates to optimize artichoke yield. Yield reduction by 50% ETc was associated with a decrease in head number and weight. The highest yield was obtained with 100% ETc and $120 \text{ kg}\cdot\text{ha}^{-1}$ N. This study also showed that deficit irrigation significantly improved artichoke head quality, such as phenolic content, but with significant yield losses.

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My deepest appreciation goes to my mother and father for their encouragement.

NOMENCLATURE

ψ_w	water potential
1-MCP	1-methylcyclopropene
ABA	abscisic acid
ACC	1-aminocyclopropane-1-carboxylic acid
A_{CO_2}	net photosynthetic rate
ADF	acid detergent fiber
ANOVA	analysis of variance
AS	Antistress
AVG	amino-ethoxyvinylglycine
cv.	cultivar
DAT	days after transplanting or treatment
dpi	dot per inch
DW	dry weight
E	transpiration
EC	electric conductivity
EL	electrolyte leakage
ET	evapotranspiration
ET_0	reference evapotranspiration
ETc	crop evapotranspiration
ETH	ethephone

FAO	Food and Agriculture Organization
FW	fresh weight
g_s	stomatal conductance
GA ₃	gibberellic acid
g_s	stomatal conductance
HPLC	high performance liquid chromatography
Kc	crop coefficient
kPa	kilopascal
LSD	least significant difference
MDG	mean days of germination
Met (DL-MET)	methionine (DL-methionine)
MPa	megapascal
n	total sample size
NA	data not available
ND	data not detectable
NDF	neutral detergent fiber
NH ₄ -N	ammonium nitrogen
N.S.	not statistically significant
NO ₃ -N	nitrate nitrogen
NUE	nitrogen use efficiency
PAR	photosynthetically active radiation
PGR	plant growth regulator

RWC	relative water content
SAM	S-adenosyl methionine
SDI	sub-surface drip irrigation
TS	Transfilm
TW	turgid weight
UAN	urea ammonium nitrate
VG	Vapor Gard
WHC	water holding capacity
WUE	water use efficiency

Cultivar

GGI	Green Globe Improved
IS	Imperial Star

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CHAPTER I

INTRODUCTION

Artichoke (*Cynara scolymus* L.) is a native Mediterranean crop. The edible part of the immature floral buds (heads) are well known as an excellent source of nutritional compounds, such as dietary fiber and potassium, and also antioxidants (e.g., chlorogenic acid and cynarin). Currently, in the United States, almost 100% of the commercial fresh artichokes are grown in California and are supplied to the market throughout the year with a retail price ranging from \$1.5 up to \$4 per head. Our previous studies in the Wintergarden of Texas confirmed the feasibility of artichoke production with high yield and quality during spring (Leskovar et al., 2007). To effectively introduce this novel and valuable crop in southwest Texas, it is necessary to develop a crop management program, from seedling growth in the nursery to crop growth in the field.

Vegetable seedlings are often exposed to environmental stresses during the nursery growth, transport, and post-transplanting in the field. Artichoke seedlings appear to be very susceptible to water stress and high temperatures, which may result in poor stand establishment or significant plant losses. This is particularly important in the southwest Texas climate because of extreme high temperatures and drought episodes during late summer, but not when transplanting in the fall. Consequently, it is necessary to evaluate and develop strategies to improve abiotic stress tolerance of artichoke

seedlings. We expect that enhancing transplant root growth will improve field establishment, while reducing leaf transpiration will prevent excess water loss during high temperature and/or drought episodes.

Crop productivity can be strongly affected by management strategies, including irrigation and fertilization. This is even more important for artichokes which have a long growing season, up to 7 months. Environmental factors, such as temperature, rainfall and soil type, as well as their interaction with specific crop strategies can also have significant effects on crop growth and yield. Therefore the establishment of optimum field crop management needs to be specific to the growing environment. Irrigation and nitrogen fertilizer management also influences vegetable quality such as soluble solids and dry matter content. In addition, it is also known that levels of plant secondary metabolites are influenced by climatic conditions and growing location. In this context, it is possible that the nutritional quality of artichoke heads may also be improved by the development of site-specific field crop management practices.

The goal of this project is to develop a production management system applicable throughout the year for artichoke, a potential specialty crop in southwest Texas. Studies include transplant management in the nursery and crop management in the field to optimize stand establishment, crop yield, quality and nutritional components of artichoke heads. The establishment of management practices for artichoke production is a prerequisite to introduce this new crop into commercial production in southwest Texas. We expect that the information generated from this project will also contribute to

the creation of new market opportunities in the Texas agriculture and partially substitute shipments from California.

Specifically, the research project aimed at improving transplant and field crop management with emphasis on, 1) root development and seedling stress tolerance and 2) irrigation and nitrogen fertilizer management.

In the area of transplant management, the objectives were: to determine the effects of drought and heat stress on seedling growth, to improve seedling drought and heat stress tolerance with foliar application of abscisic acid (ABA) and/or commercial antitranspirants and to enhance seedling root growth by the application of ethylene regulators.

In the area of field crop management, the objective was to determine yield, sensory attributes and nutritional quality components of artichoke heads in response to differential irrigation and nitrogen rates.

CHAPTER II

REVIEW OF LITERATURE

2.1 Artichoke

2.1.1 Botany, Culture and Uses

Artichoke (*Cynara scolymus* L.) is a thistle-like perennial plant that belongs to the Compositae family. In a commercial production, plants can be grown as an annual or perennial with an average of 5 years and up to 10 years (Schrader and Mayberry, 1997; Ryder et al., 1983; Tesi et al., 2004). The main marketable part of artichoke is the immature flower bud, also known as head. The edible part of heads consists of tender lower internal bracts, the receptacle or basement of florets and a thickened stem. The artichoke plant has a deep taproot (120 cm) and the canopy can grow 60-120 cm tall, with a rosette of large leaves (shoots) coming from the crown base (Sims et al., 1977). Once the plant achieves an adequate shoot biomass and meets certain cumulative cold temperatures, the main stem bearing the head elongates from the crown. After the terminal or crown head develops from the main stem, axillary or secondary heads develop from lateral branches (Ryder et al., 1983; Sims et al., 1977).

Artichoke can be propagated either by seeds or vegetative organs. For vegetative propagation, several organs are used: rooted offshoots, base of unrooted stem or rhizome, rhizome with root and shoot section, also known as stamps (Mauromicale et al.,

2004; Ryder et al., 1983). Growers practically obtain those materials from mother plants in established field after harvesting the heads. Vegetative propagation is traditionally done in Europe, such as Italy, Spain and France (Macua, 2007). New techniques such as micropropagation through *in-vitro* culture using meristematic tissue provide virus-free explants (Repetto et al. 1996). Historically, the vegetative propagation systems have been a common practice that favors a fast establishment, enhance earliness, size and uniformity of heads (Sims et al., 1977). However, current trends are to establish the crop by seeds through transplants. Even though, this system may increase the annual cost of establishment, but it can significantly reduce operational costs for pest and weed management, which are typically higher in perennial compared to annual systems (D.I. Leskovar, personal communication). In the perennial production cycle, artichoke shoots are cut back to just above ground level after the harvest season, allowing re-growing the new offshoots for the next season (Ryder et al., 1983; Schrader and Mayberry, 1997).

Artichoke heads are marketed as fresh, frozen, or canned products. For fresh market, large heads are sold individually with a thickened stem of 5-10 cm long. Small heads, also known as 'baby artichokes', are sold in packages (e.g., 12 per pack) or in bulk. Frozen, canned, and bottled artichoke products are processed (boiled and/or marinated) using only the main heart parts (internal bracts and receptacle). If heads are not harvested, matured bracts open and become a large and attractive purple flower, which can be used for ornamental purposes. Furthermore, dry, fresh, juice or extracts of artichoke leaves can be prepared for pharmacological uses. Leaf products contains high amounts of phenolic compounds such as chlorogenic acid and cynarin which have

significant effect on choleresis and use for treatments against mild dyspepsia and indigestion caused by high fat diets (Bianco, 2007). The bitterness flavor of the famous Italian liquor named ‘Cynar’ coming from artichoke extracts is used as an after-dinner digestive (Gruppo Campari, 2007).

2.1.2 Production in Europe

Artichoke is an economically important and traditional crop in southern Europe, widely cultivated in the Mediterranean countries of Italy, Spain, France and Greece. Sonnante et al. (2007) recently reviewed the history of artichoke domestication based on both scientific work and classical artistic records and summarized that artichoke was domesticated in the Roman era in Sicily, an autonomous region of Italy, and then introduced to Arabs in early Middle Ages.

The top artichoke producing country in the world is Italy, with 468,964 tones in 2006 representing 37% of total world production (Fig. 1; FAOSTAT, 2006). The artichoke production areas in Italy are located mainly in southern (Puglia, Sicilia, and Sardegna) and central (Lazio, Campania, and Toscana) regions, with year-around or seasonal spring harvests, respectively (Cardarelli et al., 2005). In Europe, Spain and France were also ranked as the top 10 producers, while two South American countries, Argentina and Peru, were ranked third and fifth in the world (Fig. 1).

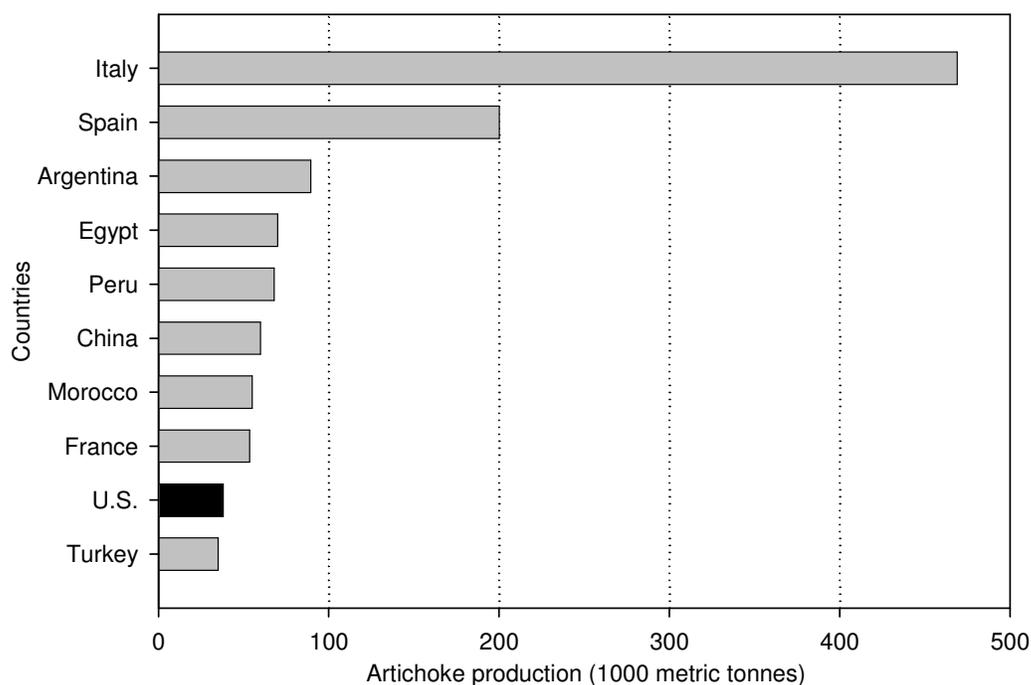


Figure 1. Top 10 artichoke producing countries in 2006 (FAOSTAT, <http://faostat.fao.org/>).

2.1.3 Production in U.S.

Artichoke was first introduced into the United States from southern Europeans in early 1800's. Commercial artichoke production occurred first in late 1800's in Louisiana by French immigrants, and in California by Italians and Spaniards (Boriss, 2005; Sims et al., 1977). The main artichoke industry was established in Castroville, California in 1920's (California Artichoke Advisory Board, 2008). In 2000's, artichokes are nearly 100 % commercially grown in California, with approximately 3,200 hectares, producing

38,000 tones of heads yearly (FAOSTAT, 2006; USDA, 2008). The United States placed ninth in total production in 2006 (Fig. 1). The major artichoke producers in California are located both in the central coastal counties and in southern California. In the central coastal counties of Monterey, Santa Cruz, and San Mateo artichokes are grown mostly as perennials and harvests occur year-round; while in southern California counties of Orange, Imperial, and San Diego counties the production cycle goes from fall to spring harvests. The largest artichoke producers are located in Castroville, Monterey, providing 70% of the total production in California (Schrader and Mayberry, 1997).

In the United States, artichoke is known as a specialty crop which is sold with a retailing price ranging from \$1.5 to 4.0 per head. The production value of artichoke in California is higher than other common vegetable crops grown in Texas (Table 1). Therefore, if commercial artichoke production is successfully introduced in Texas, this alternative vegetable would provide a great market opportunity for the Texas agriculture. Recent research efforts and trials confirmed that artichoke has the potential to grow in other U.S. states. In fact, researchers in the Northeast, such as Virginia, New York and Connecticut, have been trying to establish regional artichoke production (Bratsch, 2006; Hill, 2001; Rangarajan et al., 2000; Welbaum, 1994). Dr. Leskovar's group at Texas A&M University has been evaluating the technical feasibility of artichoke production since 2004, demonstrated that they can be grown well in southwest Texas (Leskovar et al., 2007).

Table 1. Production value of artichoke in California and common fresh vegetables in Texas during 2004-2006 (Agricultural Prices 2006 Summary, NASS; USDA, 2007).

Year	2004	2005	2006
Commodity	Production values (\$·ton ⁻¹)		
Artichoke in California	994	1001	937
<u>Common Vegetables in Texas</u>			
Cabbage	410	351	324
Cantaloupes	492	659	798
Carrots	573	639	439
Chile peppers	1709	957	827
Haneydew melons	575	313	683
Spring Onion	498	655	441
Spinach	849	340	586
Watermelon	220	282	190

2.2 Transplant Quality and Stand Establishment

Transplanting is a common horticultural establishment technique used especially for higher valued vegetables to improve stand establishment, growth performance and eventually final yields. However, right after field transplanting young seedlings are often exposed to environmental constrains, of various levels depending on the season, such as drought, flooding, heat and/or cold temperatures. The unbalanced plant water status caused by these environmental stresses may lead to sudden and severe water deficits (transplant shock) commonly resulting in temporal wilting, leaf yellowing, burning, abscission, and/or apical de-topping (Leskovar, 1998; Nitzsche et al., 1991). Therefore,

to achieve a high stand establishment, it is important to produce 'hardened' seedlings which have the capacity to withstand transplant shock.

Root growth and development of seedlings during the nursery stage is one of the important factors influencing vegetable stand establishment, and their subsequent growth (Leskovar and Stoffella, 1995), and they can be affected by environmental conditions (e.g., temperature and soil moisture) and cultural practices (e.g., irrigation, transplant age, pot size and nutrition).

Shoot growth of cucumber seedlings were highly inhibited by the combination of 37/37 °C daytime air/root-zone temperature regimes when compared with 26/26, 26/37 and 37/26 °C combinations, while root growth (dry weight) was significantly reduced at 37 °C of root-zone temperature regardless to air temperature (Wang and Tachibana, 1996). Root elongation of rice and pea seedlings were not affected by short term drought (48 hours), while in maize and cotton seedlings mild drought (-80 kPa ψ_w) enhanced root elongation, but severe drought (-900 kPa ψ_w) significantly reduced it by 20 % when compared with control (-10 kPa ψ_w) (Iijima and Kato, 2007).

In bell and jalapeno pepper seedlings, less basal and more lateral root development were observed under flotation irrigation compared with overhead irrigation system (Leskovar and Cantliffe, 1993; Leskovar and Heineman, 1994). Transplant age, container size and nutrition can also impact the overall performance of seedlings. For example, in onion, field survival rates decreased when using younger seedlings (6 weeks) compared with older seedlings (up to 12 weeks) (Leskovar and Vavrina, 1999). In cabbage, heavier heads were obtained when seedlings were grown in larger cell size

(80 cm³ vs. 7 cm³) (Marsh and Paul, 1988). In addition, optimum nutrient application such as 100 mg·L⁻¹N for pepper seedlings in the nursery also improve post-transplant performance (Aloni et al., 1991).

2.3 Ethylene Regulators on Roots

Plant root development is influenced by several phytohormones. Ethylene, a gaseous form of phytohormones, is biologically synthesized from Methionine (Met) via the following pathway according to Adams and Yang (1979):

Met → SAM (S-adenosyl methionine) → ACC (1-aminocyclopropane-1-carboxylic acid) → Ethylene (C₂H₄).

It is well known that increased level of endogenous ethylene inhibits primary root elongation, but promotes lateral and adventitious roots and root hair development, through interactions with other hormones (Aloni et al., 2006; Clark et al., 1999; De Smet et al., 2006; Kuroha and Satoh, 2007). The hormonal signals causing plant root growth responses are primarily triggered by environmental factors such as nutrients and water availability (Lopez-Bucio et al., 2003).

The role of ethylene in plant root development has often been studied by applying ethylene regulators, such as ethylene precursors or releasing substances (Met, ACC or Ethephon/Ethrel), and inhibitors of ethylene synthesis or action [AVG (amino-ethoxyvinylglycine) and Ag⁺, Co²⁺, and 1-MCP (1-methylcyclopropene)]. Soil amended with ethylene precursors, Met and ACC, increased the level of ethylene in soil and

induced ethylene-specific plant responses (triple response: short, thick and apical hook) in etiolated pea (Khalid et al., 2006). Both Met and ACC also increased the number and length of root hairs in Brassica species such as *Arabidopsis* and *Brassica rapa* (Hasegawa et al., 2003; Jason Pitts et al., 1998; Zhu et al., 2006). The application of the ethylene releasing-substance, Ethepon, increased tiller production of spring cereals without changing root/shoot ratio (Rajala and Peltonen-Sainio, 2001). Ethylene inhibitors, AVG and Co^{2+} , stimulated root elongation of barley seedlings (Locke et al., 2000) and AVG increased main root length but reduced lateral root density on common bean (Borch et al., 1999). In a study with canola, the application of ethylene action inhibitor, 1-MCP, increased root length without affecting shoot growth (Saleh-Lakha et al., 2005). To date, no information is available on how and what effects these ethylene regulators have on root development of artichoke seedlings.

2.4 Antitranspirants on Stress Tolerance

Antitranspirants are chemical compounds applied to plants to reduce transpiration and maintain high plant water status. These compounds can be categorized into three major groups based on their mode of action. Film-forming materials which provide a coating on leaf surface with wax, gel or plastics impeding excess water loss from leaves, and thus improving plant water status and increased growth under water stress condition (Moftah and Al-Humaid, 2005; Nitzsche et al., 1991). Reflecting materials, such as kaolin clay and chitosan, decrease leaf temperature by increasing leaf

reflectivity, resulting in lower transpiration and higher water use efficiency (Bittelli et al., 2001; Moftah and Al-Humaid, 2005). The third group of materials can be categorized as physiological antitranspirants, such as abscisic acid (ABA), which induces stomatal closure through metabolic processes in leaves.

ABA has been known as a stress induced phytohormone. Plant endogenous ABA levels increase in response to adverse environmental stimuli (Leung and Giraudat, 1998). ABA induces stomatal closing, thus reducing excess water loss from leaves and consequently water uptake from roots. Based on this stress alleviation function, ABA can be considered as a metabolic antitranspirant to protect plants under stress conditions (Mansfield, 1976). Exogenous ABA application has been examined and used on various vegetables including cucumber, pepper, and tomato and under different environmental stresses (Berkowitz and Rabin, 1988; Leskovar and Cantliffe, 1992; Li et al., 1996; Takahashi *et al.*, 1993). Tomato seedlings treated with $0.5 \text{ mg}\cdot\text{L}^{-1}$ ($2 \text{ }\mu\text{M}$) of ABA through hydroponic culture solution had a higher cold tolerance to $5 \text{ }^{\circ}\text{C}$ as compared to untreated controls, response that was attributed to reduced water loss from leaves (Takahashi et al., 1993). In cucumber leaves, ABA at 1.0 mM induced heat tolerance (under $45 \text{ }^{\circ}\text{C}$) and the duration of this effect depended on the concentration of ABA which was high up to 6 days (Li et al., 1996). Berkowitz and Rabin (1988) reported that 1.0 mM ABA significantly improved field survival of pepper seedlings due to increased leaf resistance and water potential, with a subsequent increase in yield. However, overall plant responses to ABA may vary according to concentration, application method, plant species, and stage of plant maturity. The effect of ABA application to enhance stress

tolerance of artichoke seedlings exposed to stress conditions has not been investigated. Besides, s-ABA (VBC-30074), a new ABA product from Valent Biosciences Corp (Libertyville, IL), has been accepted as a three year experimental use label by EPA (U.S. Environmental Protection Agency) on June, 2007. This opportunity allows us to expand our investigations with ABA under commercial nurseries conditions.

2.5 Irrigation and Nitrogen

2.5.1 Importance of Irrigation and N Inputs

Irrigation is an essential agricultural practice, especially in semiarid to arid regions. Even though the irrigated area in U.S. is less than 20% of cropland (USDA, 2002), irrigation impacts may significantly influence growth, yield and quality in fruits and vegetables, as seen in some of the most productive states such as California and Florida. In Texas, 28% of the harvested cropland was irrigated in 2002 (USDA, 2002). Irrigation makes agriculture possible in areas receiving less rainfall than the seasonal crop water use (evapotranspiration) and to ameliorate the negative effects of drought episodes during the growing season. Therefore, irrigation is used to compensate for crop evapotranspiration in these areas. Irrigation not only improves seasonal crop growth, yield and quality, but also enables intensive year-round production or double cropping.

One of the essential plant nutrients is nitrogen (N), which is a necessary for proteins, enzymes and metabolic processes involved in synthesis and transfer of energy.

Since plants have to acquire N from the soil, it is not surprising that N application frequently limits crop productivity in agricultural systems because of significant accumulation of N in crops are removed at harvest. Therefore to sustain optimum crop yields, addition of N to cropland is a prerequisite for most non legume cropping systems (Havlin et al., 1999). When the N supply is below the plant N sufficiency levels, plants usually become yellow and stunted, resulting in large yield reduction. For example, the N analysis of foliage biomass (Σ leaf + stem + heads) showed that $400 \text{ kg}\cdot\text{ha}^{-1}$ of soil N was absorbed by vegetative propagated artichoke, cv. Blanca de Tudela (Rincon et al., 2007). While higher N uptake ranging $388\text{-}625 \text{ kg}\cdot\text{ha}^{-1}$ was obtained by seed grown artichoke (Pomares et al., 2004).

2.5.2 Irrigation and N Impact on Yield and Yield Components

Since 100% of artichokes are commercially grown in California, very little information is currently available on irrigation and nitrogen fertilizer management in other regions of the U.S. Artichoke yield is highly influenced by irrigation. Based on studies conducted in Europe, artichoke appears to be a high water requirement crop compared to other vegetable crops, in part due to their large foliage biomass and longer production cycle, which can be up to 6-7 months when grown as an annual production cycle. Crop water use is directly related to evapotranspiration (ET), which can be determined by multiplying the reference evapotranspiration (ET_0) by specific crop coefficients (K_c). The ET_0 is calculated using climatological records of solar radiation,

air temperature, humidity and wind speed obtained from weather stations and applied to the Penman-Monteith equation (FAO, 1998). The K_c takes into account the crop characteristics and averaged effects of evaporation from the soil. The single crop coefficient (K_c) recommended by FAO in artichoke is 0.5, 1.0 and 0.95 as a value of K_c initial, K_c mid and K_c end stages of plant development, respectively (FAO, 1998). A lysimeter experiment in Italy found that the maximum ET of a seed propagated artichoke cv. 044 was more than 900 mm in one season (Boari et al., 2000). In another study, when five different irrigation rates were applied with a sprinkler system on artichoke cv. Blanca de Tudela, after two seasons, yield and the number of heads increased with increasing irrigation up to 630 mm (Macua et al., 2005). However, another study conducted under drip irrigation in Spain reported no yield differences between 100% and 125% of ET_c , which was equivalent to 547 mm and 726 mm of total water applied on cv. Imperial Star (Pomares et al., 2004). Comparing irrigation methods for artichoke in Tunisia, water use efficiency (WUE) was higher with drip ($24.2 \text{ kg}\cdot\text{mm}^{-1}$) than furrow irrigation ($18.6 \text{ kg}\cdot\text{mm}^{-1}$), which corresponded with a higher number of heads, 7 and 5 heads/plant, respectively (Mansour et al., 2005).

The detection of an optimum N application rate for artichoke appears to be even a more complex issue than irrigation rate. Commercial artichoke growers usually apply 112-224 N $\text{kg}\cdot\text{ha}^{-1}$ in California (Schrader and Mayberry, 1997) and 150-280 N $\text{kg}\cdot\text{ha}^{-1}$ in southeastern France (Ryder et al., 1983), but studies in Spain reported minor yield differences when N was applied between 0 and 300 N $\text{kg}\cdot\text{ha}^{-1}$ under furrow irrigation system, and 0 and 270 N $\text{kg}\cdot\text{ha}^{-1}$ under drip irrigation system for cvs. Imperial Star and

Num7144 (Pomares et al., 2004). Conversely, artichoke head weight and number were higher when N was applied at 200 kg·ha⁻¹ in Italy (Paradiso et al., 2007). Therefore, to establish artichoke production in a new region such as the Wintergarden of Texas, it is necessary to determine the optimum irrigation and N fertilizer rates.

2.6 Chemical Constituents of Artichoke

2.6.1 Nutritional Components

Artichoke is one of the oldest crops and traditionally used by ancient Greeks and Romans as a medicinal plant, and believed to provide beneficial effect for dyspeptic and hepatic disorders (Abad Alegria and Gonzalez Vivanco, 2004). Artichoke heads are known as a good source of minerals such as potassium (3.7 mg·g⁻¹FW) and magnesium (0.6 mg·g⁻¹FW) (Ocean Mist Farms, 2008; USDA, 2007). The edible part of artichoke heads (heart) also can provide high amounts of vitamin C if they are served as raw or cooked with light boiling (Abad Alegria and Gonzalez Vivanco, 2004; Gil-Izquierdo et al., 2001).

Artichoke is also known as an excellent source of dietary fibers, including non-starch polysaccharides such as cellulose, lignin and inulin, a unique carbohydrate, polymer of fructose, found in Compositae family. These components are especially useful for people with diabetics because it can help to maintain blood sugar levels stable after meals, although insoluble (e.g., cellulose and lignin) and soluble (e.g., inulin)

components of dietary fiber behave differently in metabolic system (Abad Alegria and Gonzalez Vivanco, 2004; Wolever, 1990). The levels of dietary fiber content (alcohol insoluble residues, AIRs) were reported to be higher in artichoke head (56–99 g·kg⁻¹ FW) when compared to cauliflower (45–93 g·kg⁻¹ FW) and chicory (5–45 g·kg⁻¹ FW) (Femenia et al., 1998). Another study found the high degree of polymerization of inulin in artichoke heads which may have valuable physiological properties (e.g., digestion in small intestine, and colonic fermentation) (Flamm et al., 2001; Schütz et al., 2006).

2.6.2 Phenolic Compounds

Recent studies revealed that artichoke contains high amount of phenolic compounds which have beneficial health effects due to their antioxidant abilities (Alamanni and Cossu, 2003; Curadi et al., 2005; Wang et al., 2003). In fact, a study conducted to investigate the antioxidant amount in the American diet revealed that the antioxidant content of artichoke ranked fourth out of more than 1000 food products and was the highest over several selected vegetable crops (Fig. 2; Halvorsen et al., 2006).

An antioxidant is known to have a property to prevent or alleviate the oxidative cell damage either by removing free radicals or by being oxidized itself. Cell damage is caused by free radicals via lipid peroxidation or oxidizing DNA and proteins, hence leading to aging, cancer, and cardiovascular diseases (Prior and Cao, 2000). In fact, various studies showed the significant antioxidative effect of artichoke leaf extracts or heads including on oxidation of low density lipoprotein (LDL) *in vitro* (Brown and Rice-

Evans, 1998), on production of intracellular reactive oxygen species which induced by oxidized LDL or inflammatory mediators in cultured endothelial cells and monocytes (Zapolska-Downar et al., 2002), on hydroperoxide-induced oxidative stress in cultured rat hepatocytes and human hepatoma cells (Gebhardt, 1997; Miccadei, 2008), and on oxidative stress of human leukocytes (Perez-Garcia et al., 2000).

Two main phenolic compounds in artichoke are chlorogenic acid (*5-O*-caffeoylquinic acid) and cynarin isomer (*1,5-di-O*-caffeoylquinic acid) (Table 2; Schütz et al., 2004). Although it is known that many phenolic compounds shows a strong antioxidative capacity when compared with vitamin C, E, and β -carotene (Vinson et al., 1995), the content of phenolics and antioxidant activity of artichoke may vary among plant parts and cultivars (Curadi et al., 2005; Romani et al., 2006; Wang et al., 2003) as well as head maturity, storage period and processing (Gil-Izquierdo et al., 2001; Halvorsen et al., 2006; Llorach et al., 2002; Wang et al., 2003). In addition, there is no information on how pre-harvest management practices such as irrigation and nitrogen fertilizer affect the phenolic content and antioxidant activity of artichoke.

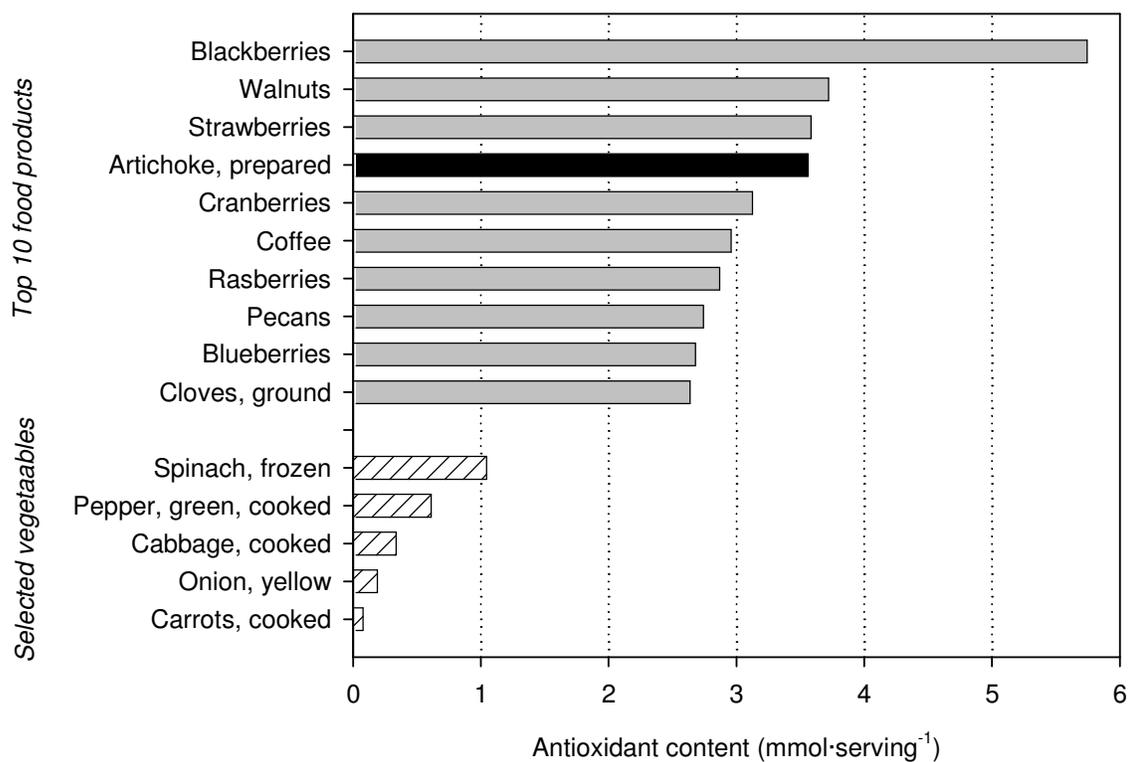


Figure 2. Antioxidant content of top 10 ranked foods and 5 selected vegetables (Halvorsen et al., 2006)

Table 2. Phenolic content of artichoke heads, cv. Green Globe (Schütz et al., 2004).

Phenolic Compounds	Artichoke Heads (mg·kg ⁻¹ DM)
1- <i>O</i> -caffeoylquinic acid	758.5
3- <i>O</i> -caffeoylquinic acid	150.9
5-<i>O</i>-caffeoylquinic acid (chlorogenic acid)	3143.0
4- <i>O</i> -caffeoylquinic acid	109.1
1,3-di- <i>O</i> -caffeoylquinic acid (cynarin)	95.0
3,4-di- <i>O</i> -caffeoylquinic acid	172.9
3,5-di- <i>O</i> -caffeoylquinic acid	762.0
1,5-di-<i>O</i>-caffeoylquinic acid (cynarin isomer)^x	3889.9
4,5-di- <i>O</i> -caffeoylquinic acid	119.5
dicafeoylquinic acid	239.0
luteolin 7- <i>O</i> -glucoside	188.4
luteolin 7- <i>O</i> -glucuronide	363.3
apigenin 7- <i>O</i> -glucoside	399.7
apigenin 7- <i>O</i> -glucuronide	1002.8
naringenin 7- <i>O</i> -glucoside	54.0
luteolin 7- <i>O</i> -rutinoside	130.5
apigenin 7- <i>O</i> -rutinoside	476.4
narirutin	56.8

^x In our study, the isomer 1,5-di-*O*-caffeoylquinic acid was measured as a cynarin.

CHAPTER III

TRANSPLANT MANAGEMENT

3.1 Heat and Drought Stress Effect on Artichoke Transplant

3.1.1 Materials and Methods

3.1.1.1 Experimental treatments

Artichoke, cv. Green Globe Improved (Condor Seed Production, Inc., Yuma, AZ) seedlings were grown in 128-cell polystyrene trays (cells with 3.5 cm × 3.5 cm; diameter × depth) in the greenhouse at the Texas A&M AgriLife Research & Extension Center at Uvalde, TX. Seeds were sown in a growth media (peat-lite mix, Speedling, Sun City, FL) and water saturated. After emergence, seedlings were irrigated daily and fertilized biweekly with blended fertilizer (24-8-16, Miracle-Gro® Garden Feeder) for 60 days. After seedlings reached 3-4 true leaves (ready for transplant in field), they were transplanted on plastic pots (9 cm × 9 cm; length × depth, square side), and filled with 5 mm sieved soil collected from the experimental field (sand: loam: clay = 22%: 40%: 38%). Prior to transplanting, soil was air-dried and then soil moisture was adjusted to 30 or 60% of maximum water holding capacity (WHC), which was considered as dry or normal soil moisture conditions, respectively. Water loss by evapotranspiration was compensated by daily watering (see peaks, Fig. 3). The maximum WHC of the soil was 53.4 ml H₂O·100 g⁻¹ soil and this value was obtained from the following calculation:

Maximum WHC ($\text{ml H}_2\text{O} \cdot 100 \text{ g}^{-1}$) = ($\text{ml H}_2\text{O}$ applied – $\text{ml H}_2\text{O}$ leached out – $\text{ml H}_2\text{O}$ in air-dried soil) / ($\text{g air-dried soil} - \text{g H}_2\text{O}$ in air-dried soil). Transplants were placed in growth chambers with temperature adjusted to 35/20 °C (considered heat stress) or 25/10 °C (normal) with 14/10 h day/night photoperiod (Fig. 4). During day time PAR was $400 \mu\text{molm}^{-2}\text{s}^{-1}$. Relative humidity (RH, %) inside the growth chamber was controlled to be above 50% RH by a portable humidifier (Fig. 4).

3.1.1.2 Shoot and root growth

Plant samples were taken at 0 (before transplanting on plastic pots), 3, 7, and 14 days after transplanting (DAT) and partitioned into shoot (leaves and stem) and root. Roots from each transplant were carefully washed with tap water and spread onto a clear plastic container (25 cm × 40 cm) containing a thin water layer. Roots were scanned with a resolution of 300 dpi and total root length and scanned area were analyzed by WINRHIZO LA-1600 (Regent instruments Inc., Quebec, Canada). After fresh weight measurements, shoot and root were oven-dried at 70 °C for 3 days.

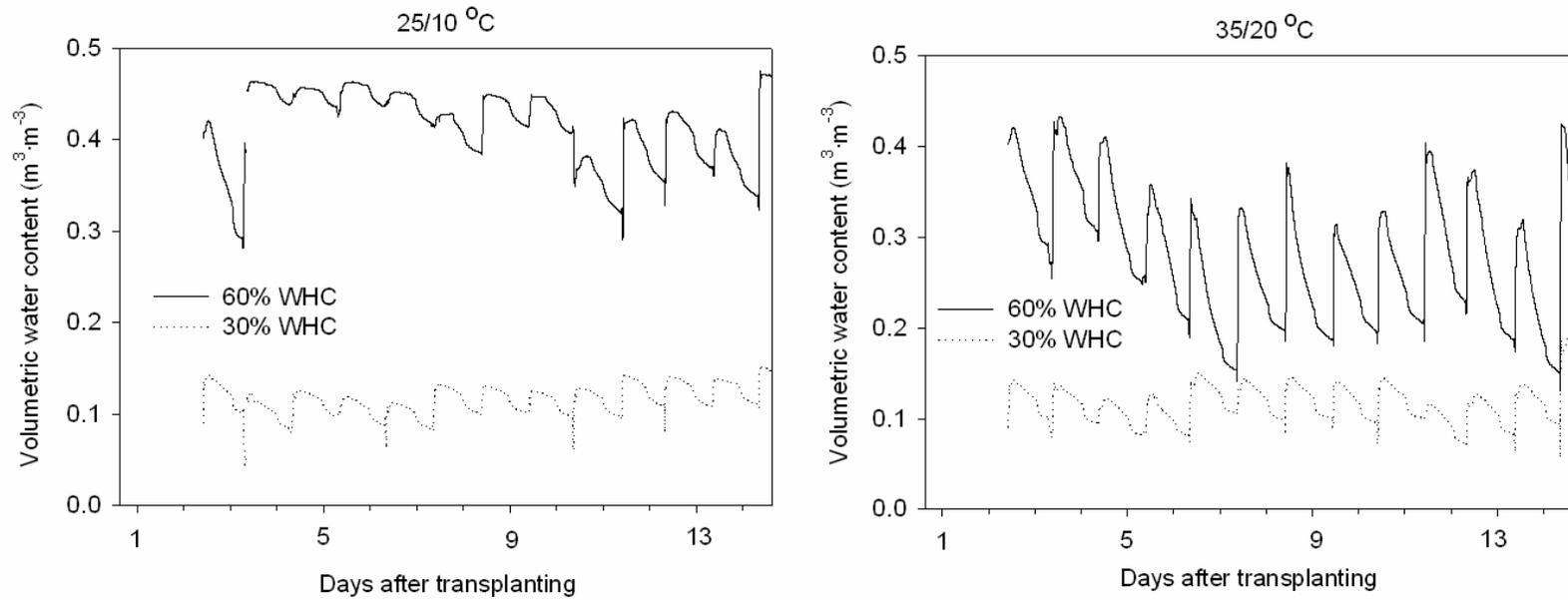


Figure 3. Volumetric water content dynamics over times for artichoke transplants in pots, subjected to normal (60% WHC, solid line) or dry (30% WHC, dot line) with daily watering and placed in controlled growth chambers at 25/10 °C (left) and 35/20 °C (right) with 14/10 h day/night photoperiod. Volumetric water content was continuously measured with ECH₂O® soil moisture probes (EC-5, Decagon Devices Inc., WA).

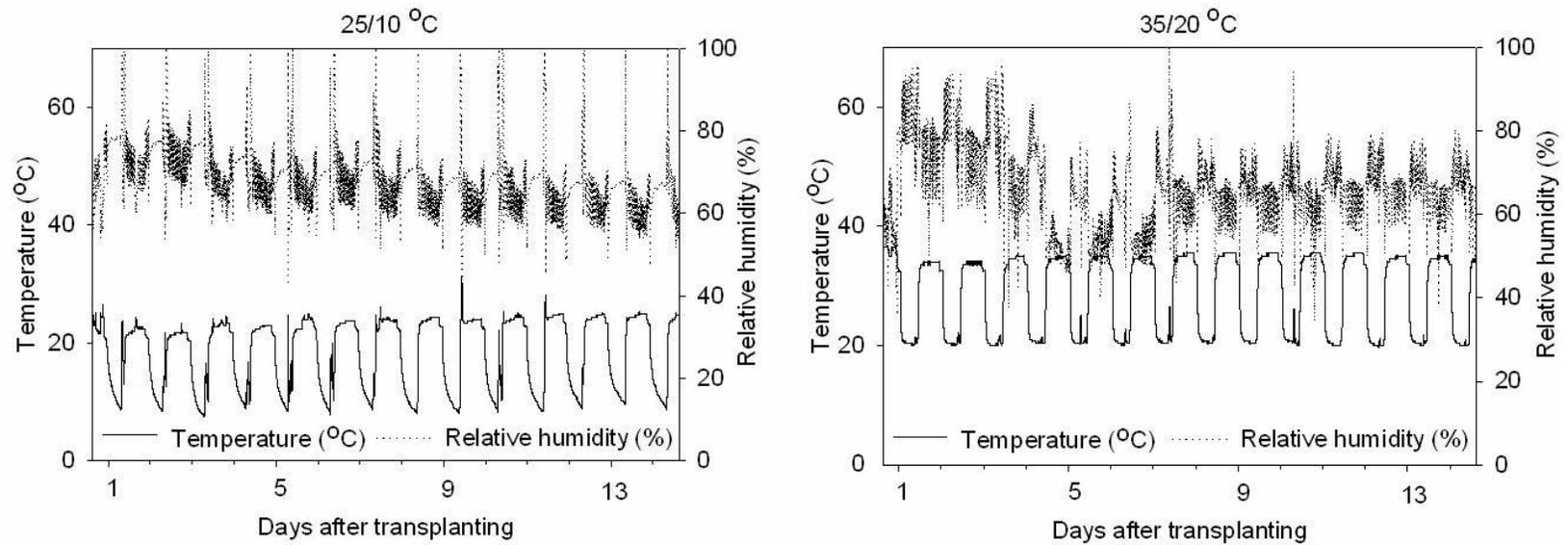


Figure 4. Temperature ($^{\circ}\text{C}$) and relative humidity (%) changes in controlled growth chambers at 25/10 $^{\circ}\text{C}$ (left) and 35/20 $^{\circ}\text{C}$ (right) with 14/10 h day/night photoperiod. Temperature and relative humidity were measured periodically during the experiment with an RH/TempLog Datalogger (Oakton[®], Verson Hills, IL).

3.1.1.3 *Plant physiological responses*

Net photosynthetic rate (A_{CO_2} , $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), transpiration (E , $\text{mmol H}_2\text{O}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and stomatal conductance (g_s , $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) were measured during daytime (measurements started 6 h after lights on), by a portable LI-6400 photosynthesis system (Li-Cor Inc, Lincoln, NE). Measurements were conducted at 0, 1, 3, 7, 10 and 13 DAT and the light strength, atmospheric CO_2 concentration, and stomatal ratio (the ratio of stomata on one side of the leaf to the other) were adjusted to $400 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ PAR, 400 ppm, and 0.5 (equivalent to 2.0) as a Li-Cor's default setting, respectively.

3.1.1.4 *Plant water status and cell membrane stability*

Plant water relations were determined by measurements of leaf water potential (ψ_w , MPa) and relative water content (RWC, %). Water potential was measured with a Scholander-type pressure bomb (Soil Moisture Equipment Corp., Santa Barbara, CA). Relative water content (RWC), an evaluation of absolute water amount in plants, was determined using leaf segments obtained from leaves of the same age as used for ψ_w . Leaf segments were collected using a hole-puncher (1 cm diameter) and weighed (fresh weight: FW). Leaf segments were then submerged in distilled water for 4 hours to obtain turgid weight (TW). The segments were dried in an oven over 24 hours till completely dry and measured dry weight (DW). The RWC was calculated using the equation described below (Turner, 1981):

$$\text{RWC (\%)} = 100 \times [(\text{FW}-\text{DW})/(\text{TW}-\text{DW})]$$

In addition, to estimate the cell membrane dysfunction by stress was measured. Three leaf segments electrolyte leakage (EL, %) were washed and placed in sealed culture tubes (25 mm × 150 mm) filled with 10 ml of distilled water and shaken in a water bath at room temperature (25 °C) for 24 h. Electrical conductivity (EC) of the solution was measured (EC₁), and then the solution with leaf segments was autoclaved at 120°C for 20 min. After cooling to 25 °C (>1 h), the second EC (EC₂) was measured (Lutts et al. 1996). The EL value was obtained by the following equation:

$$EL (\%) = 100 \times EC_1 / (EC_1 + EC_2)$$

All these destructive measurements were conducted right after the measurement shoot fresh measurement at 0, 3, 7 and 14 DAT.

3.1.1.5 Statistical analysis

The experiment was conducted using a completely randomized design. All destructive measurements including plant growth parameters and water status were taken in four subsamples (plants). Plant physiological measurements were taken in three subsamples. All data were statistically analyzed by two-way ANOVA using SPSS (version 14.0 for Windows; SPSS Inc., Chicago, IL). Differences among treatments were performed using Fisher's Least Significant Difference (LSD) at $p = 0.05$. If the Levene test for homogeneity of variance and/or the Shapiro-Welk test for normality of experimental error were significant, then nonparametric analysis using Friedman test with the comparison for main factors was performed.

3.1.2 Results

3.1.2.1 Shoot and root growth

Shoot growth on a fresh weight basis was significantly suppressed by both heat (35/20 °C) and drought (30% WHC) stresses compared with normal temperature (25/10 °C) or wet soil moisture condition (60% WHC) when measured 14 DAT, while shoot growth based on dry weight (dry matter accumulation) was significantly suppressed by heat stress only (Table 3). The partitioning of the significant temperature × soil moisture interaction of shoot fresh weight at 7 DAT indicated that the combination of heat and drought showed more stress on shoot growth (3.62 ± 0.61 g) compared to normal soil moisture condition with heat stress (6.76 ± 0.64 g) or than any soil moisture condition with normal temperature.

Root dry weight measured before transplanting was 0.29 g. Root growth (dry matter accumulation) was significantly reduced by drought stresses compared with normal soil moisture at 7 and 14 DAT, but was similar at both temperatures (Table 3). The significant temperature × soil moisture interaction of root dry weight at 3 DAT indicated that the combination of heat and drought showed more stress on root growth (0.28 ± 0.06 g) than normal soil moisture condition of heat stress (0.47 ± 0.04 g) or than any soil moisture condition of normal temperature.

Root length and root scanned area measured before transplanting was 886.6 cm and 122.8 cm^2 , respectively. The significant temperature × soil moisture interaction at 3 DAT indicated that the combination of heat and drought stresses negatively affected root

length (728.5 ± 78.6 cm) and area (249.7 ± 13.5 cm²), but was not affected by any other conditions. Overall, temperature and soil moisture conditions did not have a significant effect on these parameters at $p = 0.05$, but a negative trend was shown by both heat and drought stress when measured at 14 DAT (Table 4).

3.1.2.2 Plant physiological response

The net photosynthetic rate (A_{CO_2}) was significantly affected by temperature regime and soil moisture content. At 3 DAT A_{CO_2} was significantly higher at 35/20 °C than at normal temperatures (25/10 °C), but inversely A_{CO_2} was significantly higher at normal temperature than heat temperature when measured 14 DAT (Fig. 5). After 24 hours of transplanting, drought stress significantly decreased A_{CO_2} compared with normal soil moisture and remained lower throughout the experiment (Fig. 5).

Transpiration (E) was significantly higher at 35/20 °C than at normal temperatures when measured 3 and 5 DAT, but E was significantly suppressed by drought stress from 1 DAT to 14 DAT (Fig. 5).

Table 3. Shoot and root weight of artichoke transplants in response to temperature and soil moisture status.

Status	Shoot FW (g)			Shoot DW (g)			Root DW (g)		
	<i>Days after transplanting</i>								
	3	7	14	3	7	14	3	7	14
<u>Temperature (T)</u>									
25/10°C	7.73	6.33	8.21 a	1.12	1.12	1.48 a	0.35	0.46	0.56
35/20°C	6.50	5.19	6.04 b	1.09	0.95	1.24 b	0.38	0.38	0.55
<u>Soil Moisture (M)</u>									
60 % WHC	7.83	6.36	8.18 a	1.19	1.04	1.36	0.39	0.49 a	0.62 a
30% WHC	6.39	5.16	6.07 b	1.02	1.04	1.36	0.33	0.35 b	0.49 b
<u>Interaction</u>									
T × M	N.S ^x	*	N.S	N.S	N.S	N.S	*	N.S	N.S

Shoot fresh and dry weight (g), root dry weight (g) and shoot to root ratio at Day 0 were 5.70, 0.75, 0.29 and 0.39, respectively.

Means within columns followed by different letters are significantly different (LSD or Friedman test; p = 0.05).

^x N.S: Not significantly difference.

* Significant at p < 0.05.

Table 4. Root length and surface area of artichoke transplants in response to temperature and soil moisture status.

Status	<u>Root length (cm)</u>			<u>Root scanned area (cm²)</u>		
	<i>Days after transplanting</i>					
	3	7	14	3	7	14
<u>Temperature (T)</u>						
25/10°C	994.7	1103.7	1805.1	141.3	173.5	310.8
35/20°C	1034.8	1402.7	1662.0	151.5	184.1	255.1
<u>Soil Moisture (M)</u>						
60 %WHC	1098.4	1399.4	1800.4	161.1	195.4	304.9
30 % WHC	931.1	1107.1	1666.7	131.7	162.1	261.0
<u>Interaction</u>						
T × M	**	- ^x	N.S ^y	**	N.S	N.S

Root length (cm) and root surface area (cm²) at Day 0 were 886.6 and 122.8, respectively.

^xFriedman test with only main factors.

^y N.S : Not significantly difference

**Significant at $p < 0.01$.

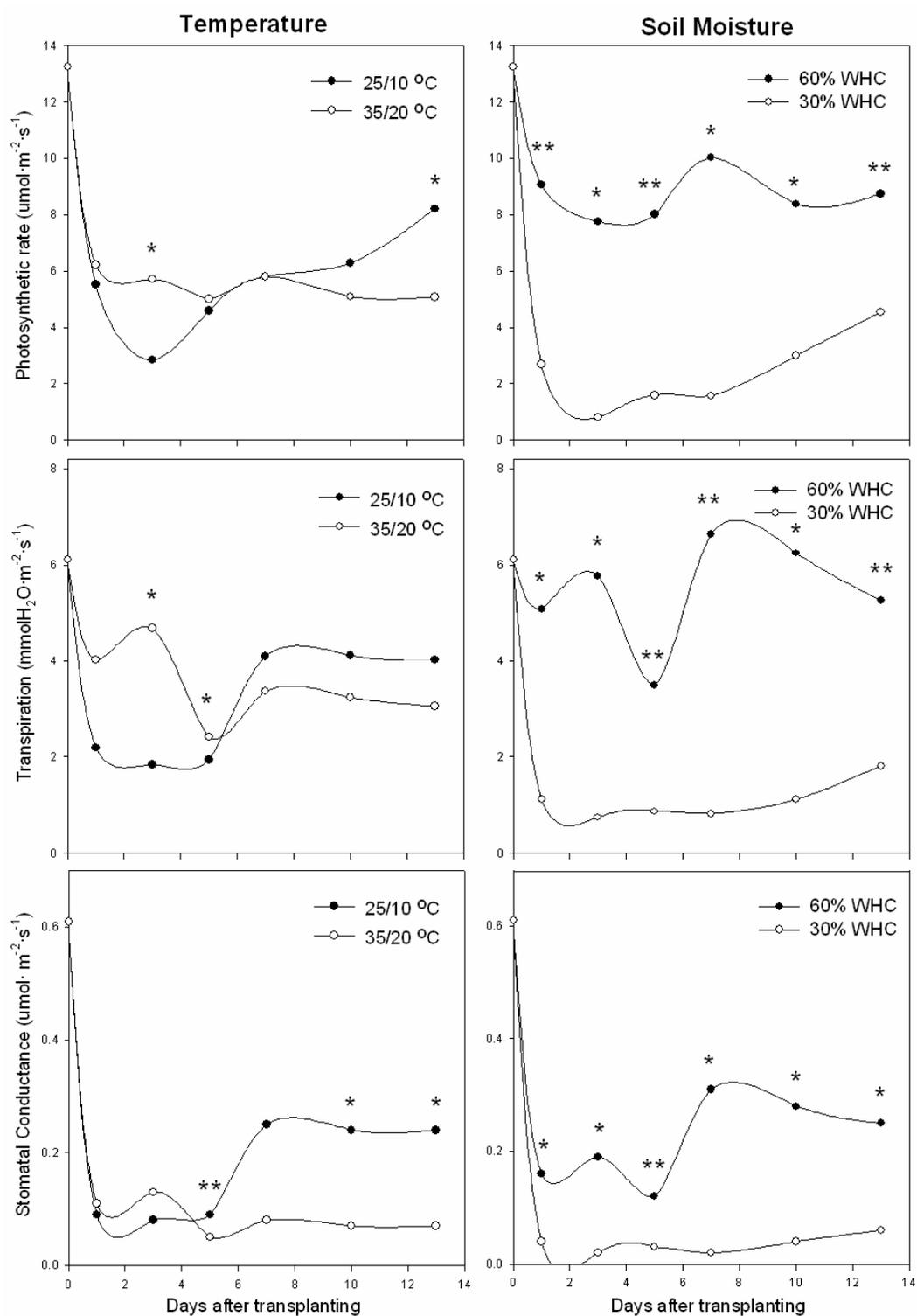


Figure 5. Physiological responses of artichoke transplants in response to temperature and soil moisture. The level of significance shown are * $p < 0.05$ or ** $p < 0.01$.

Stomatal conductance (g_s) measured before transplanting was $0.61 \text{ mmolm}^{-2}\text{s}^{-1}$, and 24 h after of transplanting, a strong depression of g_s occurred with values in the range of 0.02 to $0.2 \text{ mmol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ for all transplants regardless of treatments (Fig. 5). After 5 DAT, g_s significantly increased at normal temperatures, but it stayed lower by the heat stress treatment. Drought stress significantly suppressed g_s compared to normal soil moisture and remained lower throughout the experimental period (Fig. 5).

3.1.2.3 Plant water status

Even though no significant effect of heat stress was observed during early establishment (3 and 7 DAT), heat stress significantly decreased relative water content (RWC) when measured 14 DAT (Table 5). At 3 DAT, RWC was significantly decreased by drought stress. Thereafter, soil moisture content did not affect RWC (Table 5).

Similarly, leaf water potential (ψ_w) was significantly reduced by heat stress compared with normal temperature at 14 DAT. Drought stress significantly decreased ψ_w at 3 and 7 DAT, but it recovered by 14 DAT (Table 4).

Cell membrane stability measured as electrolyte leakage (EL) before transplanting was 16.3%, and was not significantly affected by either temperature regime or soil moisture content (data not shown).

Table 5. Relative water content, leaf and stem water potential of artichoke transplants in response to differential temperature and soil moisture status.

Status	Relative water Content (%)			Water potential (MPa)		
	<i>Days after transplanting</i>					
	3	7	14	3	7	14
<u>Temperature (T)</u>						
25/10°C	79.1	78.3	82.5 a	-0.85	-0.76	-0.67 a
35/20°C	79.6	81.0	76.6 b	-0.87	-0.73	-0.94 b
<u>Soil Moisture (M)</u>						
60 % WHC	82.5 a	82.3	81.0	-0.50 a	-0.61 a	-0.77
30 % WHC	76.1 b	77.1	78.2	-1.22 b	-0.88 b	-0.84
<u>Interaction</u>						
T × M	N.S ^x	- ^y	N.S	N.S	N.S	N.S

Relative water content (%) and leaf and stem water potential at Day 0 were 84.0 and 0.87, respectively.

Means within columns followed by different letters are significantly different (LSD or Friedman test; $p = 0.05$).

^x N.S: Not significantly difference.

^y Friedman test with only main factors.

3.1.3 Discussion

Transplant performance is an important component for stand establishment of artichoke and their subsequent growth. In southwest Texas, seedlings are frequently subjected to high air temperatures and rapid soil drying at transplanting. In this study, the effects of individual or combined heat and drought stresses on growth, physiological responses and water status of artichoke transplants were examined.

Heat stress (35/20 °C, day/night) significantly inhibited shoot growth but not root growth. The reduction of shoot FW was 27% and DW was 16% compared to normal temperatures when measured 14 DAT (Table 3). During the 14 d of experimental period, plant physiological responses also differed in response to heat stress. At early stage of post-transplanting (1-3 DAT), transpiration (E) was significantly higher by heat stress exposure as a leaf-cooling process, and response was also associated with increases in g_s and A_{CO_2} (Fig. 5). However as heat stress continued there was a reduction of plant water status (RWC and ψ_w) and a decline of g_s , E and A_{CO_2} that maintained low when measured 13 DAT (Fig. 5 and Table 5). Unaffected root growth by heat condition is reasonable to assume that root growth is not primarily related to photosynthesis, as compared to leaf growth. Similarly, in pepper transplants, root growth was not affected by heat stress (35/25 °C), but shoot growth was severely inhibited 4 days after transplanting when compared with normal temperatures (26/18 °C) (Aloni et al., 1992). In addition, cucumber seedlings growing at 38 °C root temperature for 6 days showed higher proportion of carbon translocation into roots than shoots, even though a large

amount of assimilate in roots was consumed by respiration (Du and Tachibana, 1994). These findings support our results and indicate that shoot growth is more susceptible to heat stress than root growth.

Drought stress significantly inhibited root growth (19%) based on a dry weight basis when measured 14 DAT, while shoot growth was minimally affected (Tables 3 and 4). Drought stress sharply reduced stomatal conductance (within 24 hours) and leaf water status (shoot fresh weight, RWC and ψ_w) within 3 d after transplanting, causing a decline of A_{CO_2} (Fig. 5 and Table 5). Even though A_{CO_2} , E and g_s declined at transplanting, regardless to soil moisture, only plants under well-watered conditions showed full recovery of A_{CO_2} , E and g_s 7 DAT. Although mild drought stress is known to promote plant root growth (Aloni et al., 1992; Sharp and Davies, 1979), a significant inhibition of root growth was observed in this experiment. Our results partially agree with their findings, because we observed higher root weight (20%) at 3 DAT by drought stress at 25/10 °C, however this difference was diminished as drought stress was prolonged. Therefore, transient drought stress (up to 3 days) at optimum growth temperatures may induce root growth, but inhibition may occur with extending the period of drought stress. However, it is well known that root growth in response to drought stress depends on stress intensity as well as cultivar/specie (Iijima and Kato, 2007; Rahman et al., 1999).

The combination of both heat and drought stress caused significant reduction of plant growth. The depression of A_{CO_2} , E and g_s measured initially within 24 hours of exposure did not recover throughout the 14 days of experimental period (Fig. 5). The

negative effects of combined heat and drought stresses was shown on shoot water status and root growth as early as 3 DAT (Tables 3 and 5). These results indicate that heat and drought stresses have additive effects, which and induce more severe plant growth inhibition as compared to either stress factor alone.

In conclusion, heat or drought alone, or in combination significantly reduced shoot or/and root growth of artichoke seedlings by lowering A_{CO_2} and shoot water status. Under simultaneous conditions of both stresses, which will typically occur during summer and/or early fall in southwest Texas, a delay or failure in transplant establishment is expected. The detrimental effects of the combined stresses severely affected root growth and shoot water status. This suggests that to prevent transplant shock by heat and drought stresses it is desirable to either condition seedlings to improve early root growth or to prevent leaf dehydration. Therefore, effects of plant growth regulators (PGRs) treatment on root growth and shoot water status were examined in the following study.

3.2 Ethylene Regulator Effect on Artichoke Roots

3.2.1 Materials and Methods

3.2.1.1 Experimental design

Study 1: Effect of ethylene on artichoke seed germination and root growth

Artichoke, cv. Green Globe Improved (Condor Seed Production, Inc., Yuma, AZ), seeds were sterilized in 1.0 % sodium hypochlorite bleach (Clorox® a.i. 6.0%) for 15 min, washed with running tap water for 5 min and rinsed with distilled water 3 times. Cracked and/or floated seeds were discarded after soaking. A total of 16 rooting treatments: five ethylene regulators, at three concentrations each, and a control were evaluated. Those were: 1, 10 and 100 $\mu\text{M}\cdot\text{L}^{-1}$ of 1-MCP [active ingredient (a.i.) 3.8% 1-methylcyclopropene, Valent BioSciences Co., Libertyville, IL,], and 1, 30, and 100 $\mu\text{M}\cdot\text{L}^{-1}$ of ACC (a.i. 98.0% 1-aminocyclopropane-1-carboxylic acid, CALBIOCHEM®, San Diego, CA), and 1, 10, and 100 $\mu\text{M}\cdot\text{L}^{-1}$ of AVG (ReTain®, a.i. 15.0% amino-ethoxyvinylglycine; Abbot Laboratories, Abbott Park, IL), and 1, 30, and 100 $\mu\text{M}\cdot\text{L}^{-1}$ of Ethephon (FLOREL®, a.i. 3.9% 2-chloroethylphosphonic acid; Lawn and Garden Products, INC., Fresno, CA) and 1, 30, and 100 $\mu\text{M}\cdot\text{L}^{-1}$ of DL-Met (a.i. 99.7%, Phytochrome Inc., Tokyo, Japan) and control (distilled water). Prior to seeding, 5 ml of each solution were added to petri-dishes containing a blue blotter paper. Five seeds were placed on each blotter paper and germinated in a thermo-gradient table controlled by thermocouples (SD 10, Shimaden Co Ltd, Tokyo, Japan) and temperature was adjusted

to constant 23.0 ± 0.2 °C by thermostat cooling system (VWR 1160S, VWR®, West Chester, PA) in dark. This experiment was repeated using the same treatments plus a higher concentration, $1000 \mu\text{M}\cdot\text{L}^{-1}$, of each ethylene regulator (total 21 treatments).

Study 2: Effect of ethylene on early root development

Artichoke cv. Green Globe Improved seeds were sterilized as previously described. Artichoke seeds were seeded on water soaked paper towels (25 cm × 40 cm) covered by oil-paper (25 cm × 40 cm) to maintain adequate moisture for germination and placed in plastic containers which were placed in a thermo-gradient table in dark at 23.0 ± 0.2 °C for 8 d. After 8 d, time when most seeds germinated, five uniform artichoke seedlings were selected and transferred to 9 cm petri-dishes containing a blue blotter paper with each concentration of ethylene regulators, as used in *Study 1*. Concentrations were 1, 10 and $100 \mu\text{M}\cdot\text{L}^{-1}$ of 1-MCP, 1, 30, and $100 \mu\text{M}\cdot\text{L}^{-1}$ of ACC, 1, 10, and $100 \mu\text{M}\cdot\text{L}^{-1}$ of AVG, 1, 30, and $100 \mu\text{M}\cdot\text{L}^{-1}$ of Ethephon, 1, 30, and $100 \mu\text{M}\cdot\text{L}^{-1}$ of DL-Met and control (distilled water) and 5 ml of each solutions were added to blue blotter paper. Petri-dishes were placed in the thermo-gradient table in dark at 23.0 ± 0.2 °C for an additional 3 days (11 days after seeding).

3.2.1.2 Seed germination and root evaluations

In the first study, germination was classified into normal (with complete morphological parts); abnormal (with broken cotyledons, less than one cotyledon, missing primary root, poorly developed, or absence of hypocotyls), and dead. At 8 days

after seeding, each seedling was scanned with a resolution of 600 dpi and root length (cm) as well as root scanned area (cm²) were analyzed with the software WINRHIZO LA-1600 (Regent instruments Inc., Quebec, Canada).

In the second study, after treatments were imposed, rooted seedlings were evaluated between 8-11 days after seeding (0-3 days after treatment). Each seedling was scanned with a resolution of 600 dpi and the primary root length and hypocotyls length were analyzed by WINRHIZO LA-1600. Lateral roots coming from the primary root were also counted.

3.2.1.3 Statistical analysis

These experiments were conducted using a completely randomized design with three to six replications. All measurements were taken in 3-5 subsamples of 9-21 seedlings per treatment, unless otherwise indicated. All data were statistically analyzed by ANOVA using SPSS (version 14.0 for Windows; SPSS Inc., Chicago, IL). Differences among treatments were performed using LSD at $p=0.05$. If the Levene test for homogeneity of variance and/or the Shapiro-Welk test or Kolmogorov-Smirnov test for normality of experimental error were significant, Kruskal-Wallis test was performed instead of ANOVA and multiple comparisons were performed using Mann-Whitney test.

3.2.2 Results

3.2.2.1 Study 1

Ethylene regulators (range 1 to 1000 $\mu\text{M}\cdot\text{L}^{-1}$) did not affect on germination rate, mean days of germination (MDG) and seedling performance. Overall the average germination rate was 94.2% and 95.1% and MDG was 5.0 days and 4.4 days in the first (data not shown) and the second assay (Table 6), respectively.

Root length for the control was 1.36 cm, and significantly increased by AVG application, being the longest (2.79 cm) with 10 $\mu\text{M}\cdot\text{L}^{-1}$ of AVG (Fig. 6). No significant difference of root length was observed with other ethylene precursors, including DL-MET, ACC and ETH or with the ethylene inhibitor 1-MCP (Fig. 6). Root scanned area for the control was 0.89 cm^2 , and significantly increased with ETH application, being the largest (1.64 cm^2) with 30 $\mu\text{M}\cdot\text{L}^{-1}$ (Fig. 6). There was also an increasing trend of root surface area with ACC 30 $\mu\text{M}\cdot\text{L}^{-1}$ (1.69 cm^2). AVG did not affect root scanned area, rather it tended to decrease at high concentration (100 $\mu\text{M}\cdot\text{L}^{-1}$) when compared with control (Fig. 6). AVG (10 $\mu\text{M}\cdot\text{L}^{-1}$) clearly induced root elongation but decreased root hair formation. However, root elongation was highly inhibited by AVG at high concentration (100 $\mu\text{M}\cdot\text{L}^{-1}$) (Fig. 7). In contrast to AVG treatment, ethylene precursors (ACC and ETH) induced a dense and long root hair formation (Fig. 7).

In the second assay, root length for the control was 2.43 cm, and was significantly increased (3.72 cm) with AVG at 10 $\mu\text{M}\cdot\text{L}^{-1}$ (Fig. 8). An inhibitory effect of ethylene regulators on root elongation was observed at the highest concentration (1000

$\mu\text{M}\cdot\text{L}^{-1}$) of ACC, ETH and AVG (1.15, 1.36 and 1.13 cm, respectively) (Fig. 8). Both DL-MET and 1-MCP did not affect root elongation even at the highest concentration of 1000 $\mu\text{M}\cdot\text{L}^{-1}$. Root scanned area for the control was 1.23 cm^2 , and significantly increased with ethylene precursors including DL-MET at 1 or 30 $\mu\text{M}\cdot\text{L}^{-1}$, ACC 1 $\mu\text{M}\cdot\text{L}^{-1}$ and ETH at 30 or 100 $\mu\text{M}\cdot\text{L}^{-1}$ (Fig. 8). AVG at 10 $\mu\text{M}\cdot\text{L}^{-1}$ showed larger root scanned area (1.56 cm^2), but AVG 100 and 1000 $\mu\text{M}\cdot\text{L}^{-1}$ severely reduced the root area (Fig. 8).

Table 6. Germination and seedling growth of artichoke in response to ethylene regulators (second assay, Study 1). Test was performed in petri-dishes at 23 °C in dark for 8 d.

Treatment	Conc. ($\mu\text{M}\cdot\text{L}^{-1}$)	Germination (%)	MDG (day)	Seedlings		
				Normal (%)	Abnormal (%)	Dead (%)
Control	0	100	4.2	100	0	0
DL-MET	1	94.3	4.2	94.3	0	5.7
	30	97.1	4.0	91.4	5.7	2.9
	100	94.3	4.0	94.3	0	5.7
	1000	96.4	4.5	93.6	2.9	3.6
ACC	1	91.4	4.4	88.6	2.9	8.6
	30	94.3	4.6	91.4	2.9	5.7
	100	93.6	4.6	87.9	5.7	6.4
	1000	97.1	4.2	94.3	2.9	2.9
ETH	1	97.1	4.6	97.1	0	2.9
	30	97.1	4.5	91.4	5.7	2.9
	100	97.1	4.1	94.3	2.9	2.9
	1000	100	4.3	93.6	6.4	0
AVG	1	85.7	4.5	77.1	8.6	14.3
	10	91.4	4.7	88.6	2.9	8.6
	100	94.3	4.5	94.3	0	5.7
	1000	100	4.3	94.3	5.7	0
1-MCP	1	91.4	4.6	91.4	0	8.6
	10	94.3	4.6	94.3	0	5.7
	100	97.1	4.6	94.3	2.9	2.9
	1000	94.3	5.0	94.3	0	5.7
Overall Mean		95.1	4.4	92.4	2.8	4.8

MDG: Mean days of germination.
Number of samples (n=7).

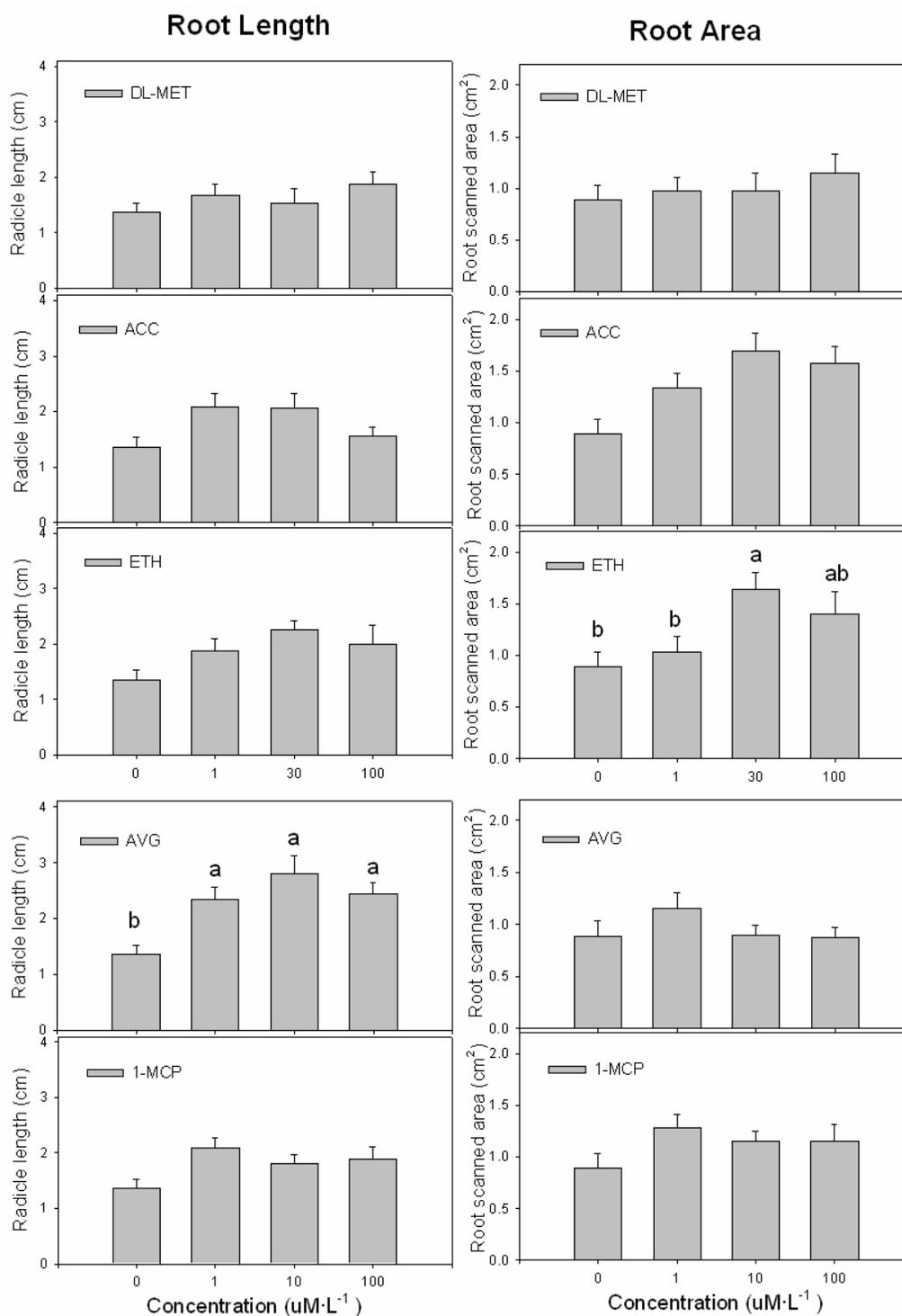


Figure 6. Root length and root scanned area of artichoke seedlings in response to ethylene regulators after 8 d of incubation (first assay, Study 1). Vertical bars indicate mean \pm SE ($n = 10-14$). Means within columns followed by different letters are significantly different (LSD or Mann-Whitney test, $p = 0.05$).

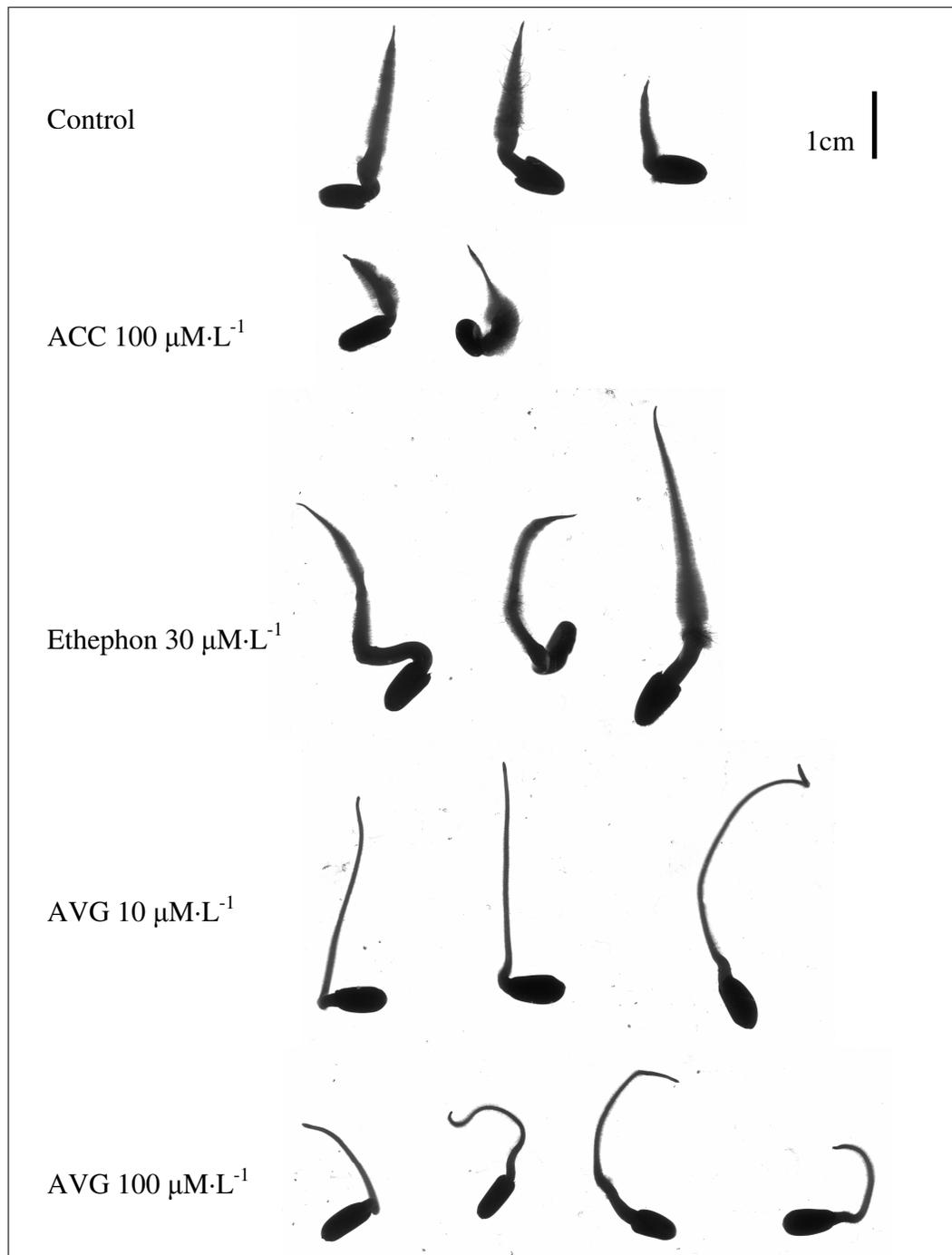


Figure 7. Root morphology in response to ethylene regulators, 8 days after seeding (first assay, Study 1).

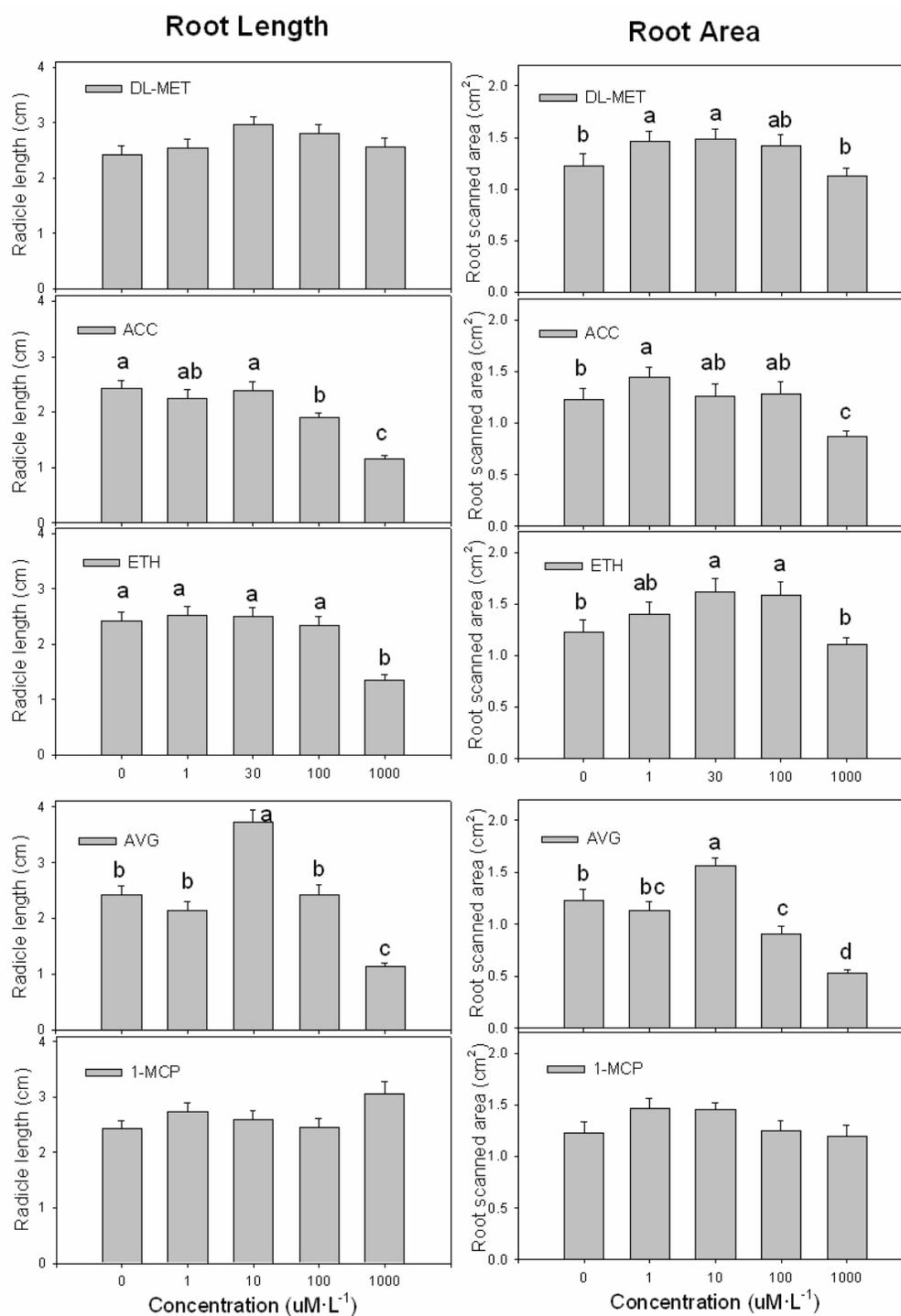


Figure 8. Root length and root scanned area of artichoke seedlings in response to ethylene regulators for 8 d (second assay, Study 1). Vertical bars indicate mean \pm SE ($n = 20-21$). Means within columns followed by different letters are significantly different (LSD or Mann-Whitney test, $p = 0.05$).

3.2.2.2 Study 2

Primary root length was very variable between plants as well as among treatments. Overall all ethylene regulators (DL-MET, ACC, ETH, AVG and 1-MCP) tended to inhibit root elongation as concentration increased from 1 to 100 $\mu\text{M}\cdot\text{L}^{-1}$, and the largest inhibition was caused by 1-MCP (Fig. 9).

Lateral root initiation was significantly induced by 30 $\mu\text{M}\cdot\text{L}^{-1}$ of ETH with significantly higher number of lateral roots as compared to control or other ethylene regulators when measured 3 days after treatment (DAT). Other ethylene precursor such as DL-MET and ACC did not affect the initiation of lateral roots. Ethylene inhibitors did not promote lateral root initiation when seeds were incubated at 10 and 100 $\mu\text{M}\cdot\text{L}^{-1}$ of AVG or 100 $\mu\text{M}\cdot\text{L}^{-1}$ of 1-MCP (Fig. 10).

Hypocotyl growth was significantly inhibited by ACC at 3 DAT, ETH at 2 and 3 DAT and AVG at 3 DAT. This effect was concentration dependent, with a severe inhibition at 100 $\mu\text{M}\cdot\text{L}^{-1}$ of ETH and AVG (Fig. 11). No clear effect on hypocotyl growth was observed with both DL-MET and 1-MCP.

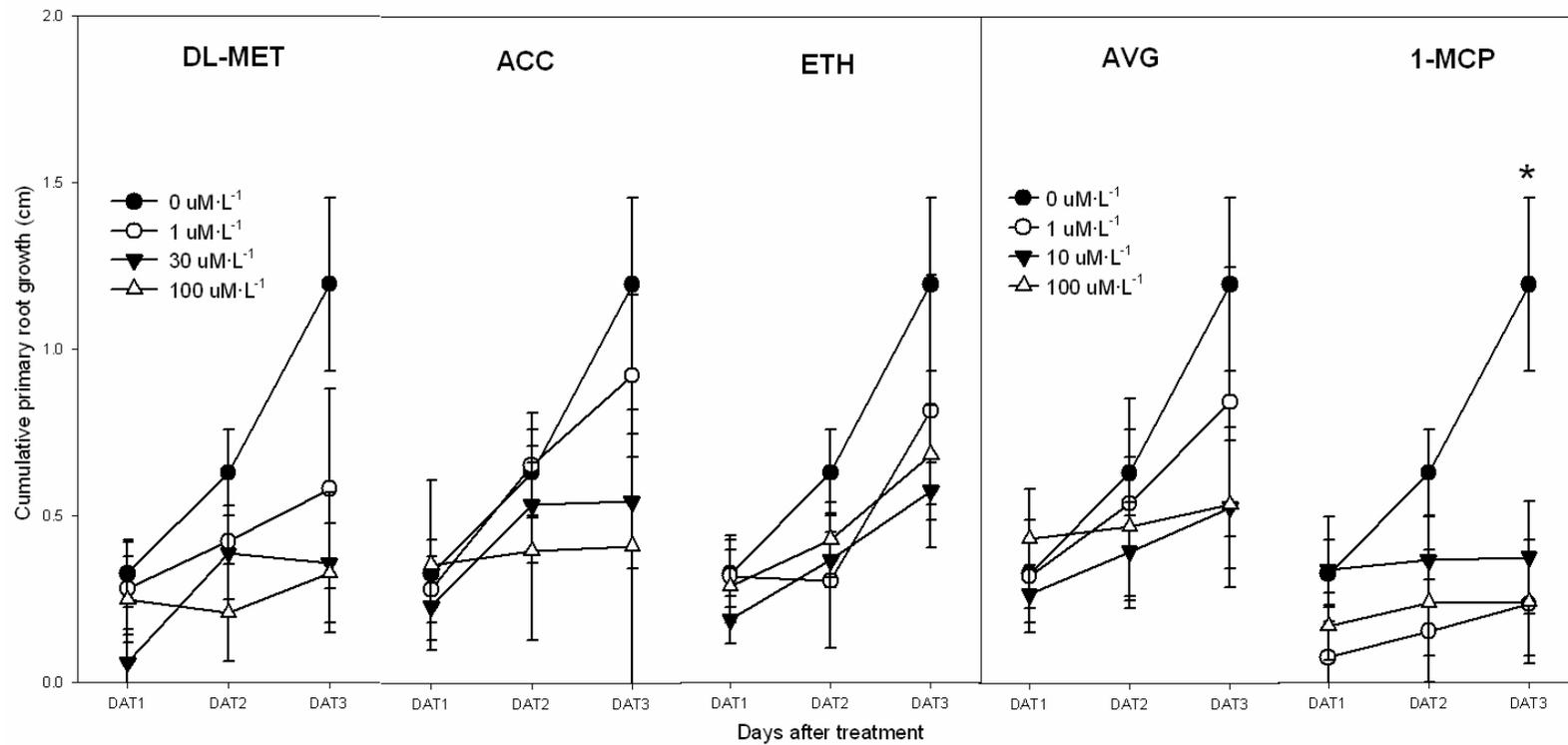


Figure 9. Primary root growth of artichoke seedlings in response to ethylene regulators. Treatments were imposed from 8 (0 days after treatment; DAT) to 11 (DAT 3) days after seeding (Study 2). Vertical bars indicate mean \pm SE (n = 20). *p < 0.05.

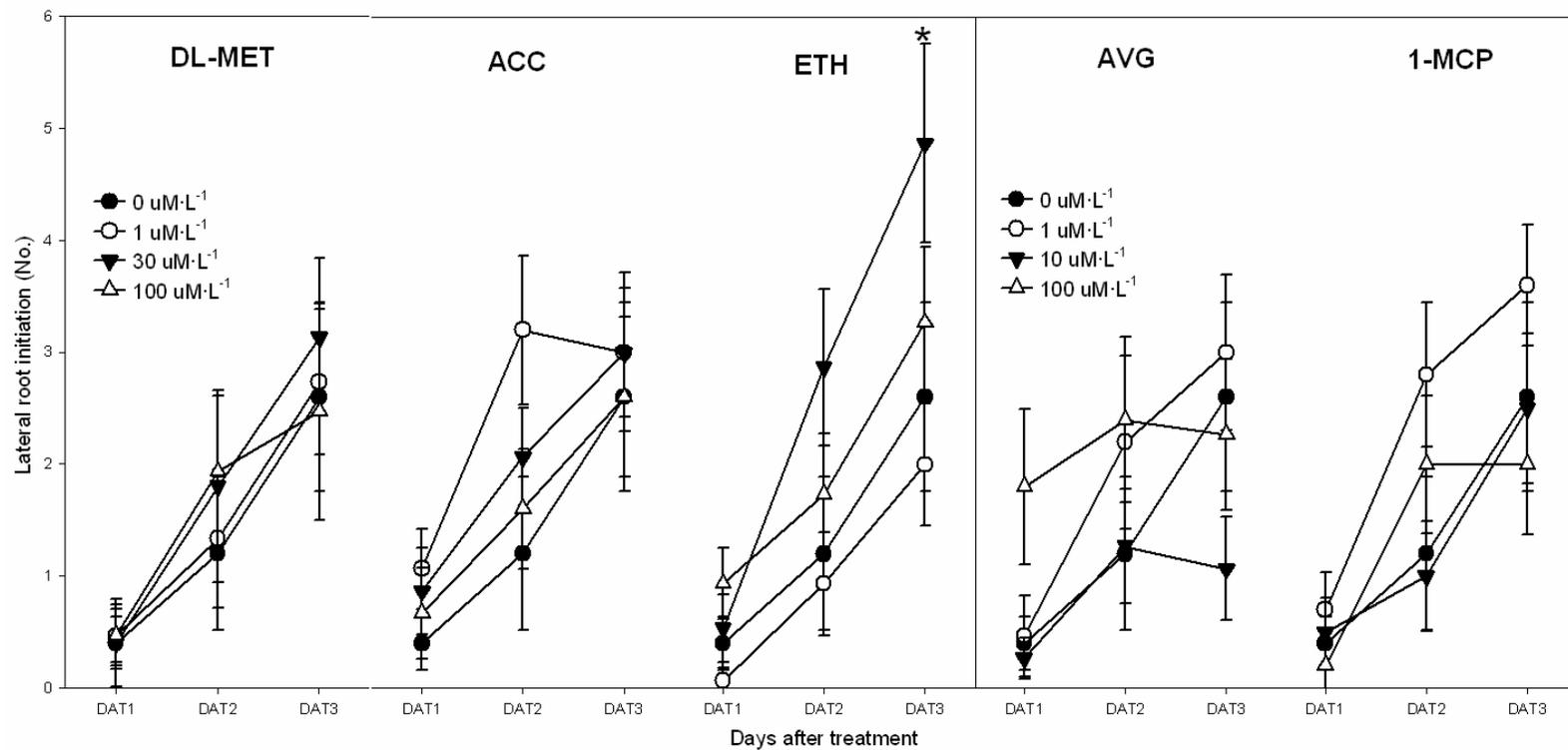


Figure 10. Lateral root initiation of artichoke seedlings in response to ethylene regulators. Treatments were imposed from 8 (0 DAT) to 11 (DAT 3) days after seeding (Study 2). Vertical bars indicate mean \pm SE (n = 20). *p < 0.05.

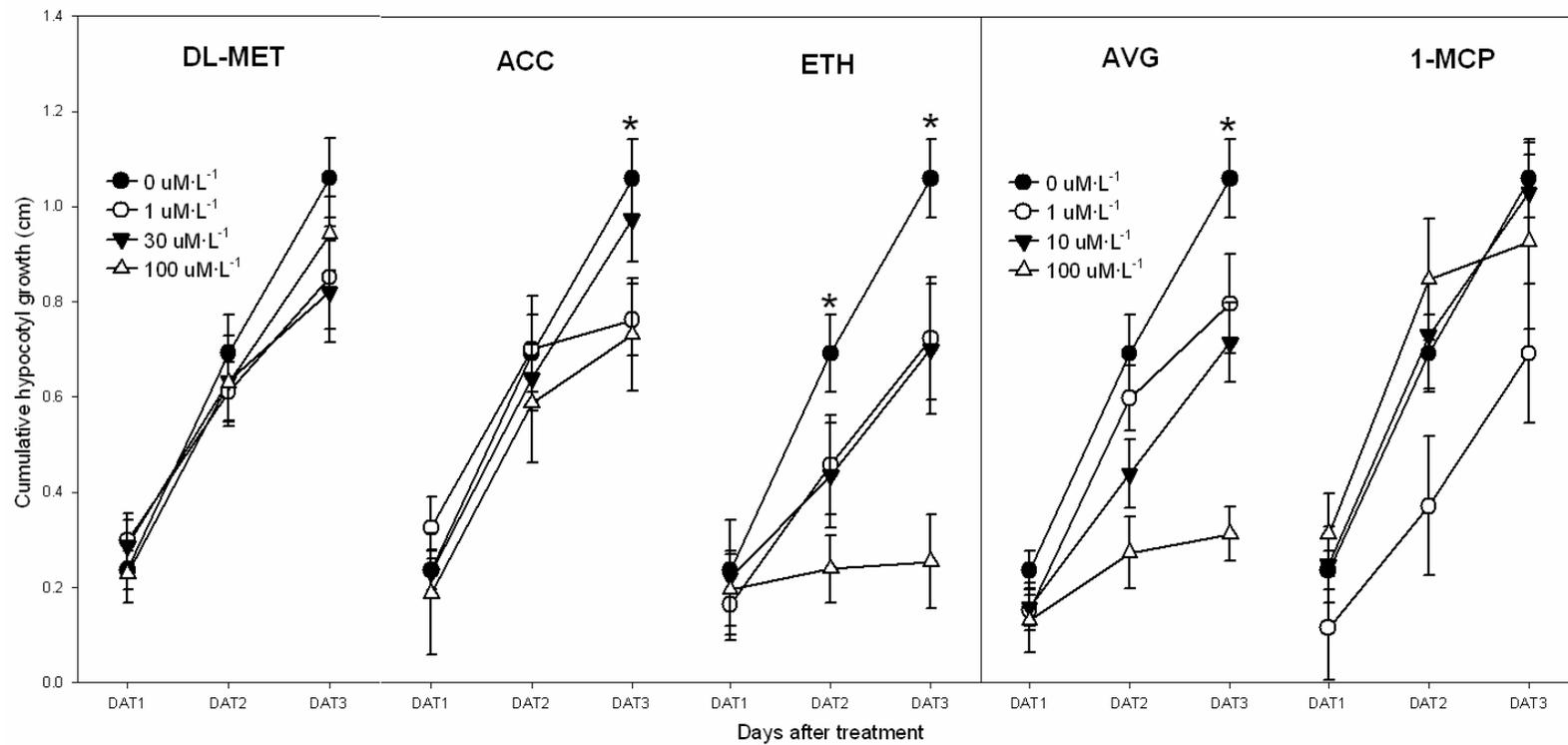


Figure 11. Hypocotyl growth of artichoke seedlings in response to ethylene regulators. Treatments were imposed from 8 (DAT 0) to 11 (DAT 3) days after seeding (Study 2). Vertical bars indicate mean \pm SE (n = 20). *p < 0.05.

3.2.3 Discussion

3.2.3.1 Study 1

Ethylene regulators including ethylene precursors (DL-MET and ACC), an ethylene releasing compound (ETH), an ethylene biosynthetic inhibitor (AVG) and an ethylene action inhibitor (1-MCP) were applied to germination media of artichoke seeds for 8 days in darkness to evaluate this effect on germination and radicle growth of seedlings.

Overall, both the percentage and the speed of artichoke seed germination were not affected by any ethylene regulators (Table 6). It is reported that ethylene stimulates seed germination in many plant species, but this response dominantly occurs only when endogenous ethylene level is low, which is particularly in dormant seeds or when seeds are exposed to adverse environmental conditions such as drought, salt or high temperature, or inhibitors such as abscisic acid or methyl jasmonate (Kepczynski and Kepczynska, 1997). In this study, no stress was imposed on artichoke seeds, except for the application of ethylene regulators. Since we did not observe any adverse effects on germination by these ethylene regulators, it seems reasonable to assume that ethylene action may not have affect germination process in artichoke, as shown in other vegetable crops such as tomato, onion and cucumber (Lalonde and Saini, 1992).

Contrary to germination, root elongation and root scanned area were significantly affected by ethylene regulators. ACC and ETH ($1-30 \mu\text{M}\cdot\text{L}^{-1}$) induced dense and long root hair formation, but did not stimulate root elongation; in fact, it was inhibited at

higher concentrations (Figs. 6, 7 and 8). Consistent with root hair responses, root scanned area tended to increase with ACC and ETH compared to control seedlings (Figs. 6 and 7). AVG, an ethylene inhibitor, significantly induced root elongation at $10 \mu\text{M}\cdot\text{L}^{-1}$ but it was inhibited at higher concentrations. Furthermore, root hair formation was significantly inhibited at all concentrations of AVG (Figs. 6, 7 and 8). Therefore, the increase in root elongation by AVG did not contribute to an increase in root area due to less of root hair mass (Figs. 6, 7 and 8). Our results strongly suggest that ethylene is a positive regulator of root hair formation as shown in other plant species (Jason Pitts et al., 1998; Tanimoto et al., 1995) as well as a negative regulator of root elongation (Locke et al., 2000; Smith and Robertson, 1971). In this study, 1-MCP did not show any clear effects on root elongation or root scanned area (Figs. 6 and 7). Since 1-MCP acts as gaseous form, perhaps it was partially diffused from un-tightly closed petri-dishes then reducing the potential concentration effect on seeds. Therefore, this lack of response by 1-MCP could be partly due to the incubation methodology.

3.2.3.2 Study 2

In this study, the same ethylene regulators as in Study 1 were applied to 8 day-old artichoke seedlings for 3 days in darkness to evaluate their effect on early root development of seedlings.

In general, primary root growth (length) was very variable among seedlings and treatments. Overall, all ethylene regulators seem to inhibit the primary root growth as concentration increased up to $100 \mu\text{M}\cdot\text{L}^{-1}$ (Fig. 9). Lateral root initiation was

significantly induced by ETH ($30 \mu\text{M}\cdot\text{L}^{-1}$), but inhibited by AVG (10 and $100 \mu\text{M}\cdot\text{L}^{-1}$) and 1-MCP ($100 \mu\text{M}\cdot\text{L}^{-1}$) (Fig. 10). This response support the general hypothesis that ethylene interrupts polar auxin transport in the vascular bundles and pericycle, which induce lateral root formation (Aloni et al., 2006). Plants treated with higher concentration of ACC or ETH showed inhibited hypocotyl growth and apical hooking, which are well known as seen in our assay (Fig. 11) and as reported by Ecker (1995).

In conclusion, the ethylene precursor ACC and the ethylene releasing compound ETH enhanced root hair, lateral roots formation, and root area. These results open the opportunity to further evaluate the potential use of exogenous ethylene regulators as root enhancers. Improving root growth and/or function for nursery seedlings is very important practice to improve post-transplant performance especially under heat and drought conditions, which typically occur during summer and early fall in southwest Texas region. Root hair and lateral root (vs. taproot) developments may both play an important role in unfavorable field conditions and/or under environmental stresses (Drew and Saker, 1975; Jungk, 2001). Enhancing root growth by ACC or ETH may improve the seedling ability for higher water and nutrient uptake under those stresses. However, the physiological basis of ethylene-induction on roots required further investigations.

3.3 Antitranspirant Effect on Artichoke Stress Tolerance

3.3.1 Materials and Methods

3.3.1.1 Experimental design

Study 1: Effect of antitranspirants on stress tolerance

This experiment was conducted in a greenhouse at the Texas A&M AgriLife Research & Extension Center at Uvalde during March 2006. Sixty days-old artichoke seedlings cv. Green Globe Improved (Condor Seed Production, Inc., Yuma, AZ), were grown in 128-round polystyrene cell trays (3.5 cm × 3.5 cm, diameter × depth). Seeds were sown in a growth media (peat-lite mix, Speedling, Sun City, FL) and water saturated. After emergence, seedlings were irrigated daily and fertilized periodically with blended fertilizer (24-8-16, Miracle-Gro® Garden Feeder). Seedlings were transferred into 6-square cell trays (4 cm × 5.5 cm, square side length × depth), with additional growth media and water saturated. Four antitranspirants were evaluated in this study. Three film forming commercial antitranspirants were Antistress (AS; Enviroshield Products Co., Houston, TX), Transfilm (TF; PBI/Gordon Corp., Kansas City, MO), and Vapor Gard (VG; Miller Chemical and Fertilizer Corp., Hanover, PA). The metabolic antitranspirant used was abscisic acid (ABA, Valent BioSciences Co., Libertyville, IL). The concentration of each antitranspirant spray solution was prepared based on manufacturer recommendations, 2.2% for AS and VG, and 3.8% for TF, and 2000 mg·L⁻¹ for ABA. Foliar applications were done with a hand sprayer on the upper

side of leaves until runoff. Untreated control plants were sprayed with tap water. Drought stress as water withholding was imposed right after spray applications and maintained for 4 d. In the greenhouse, average daily minimum and maximum temperatures during experiment were 15 °C and 38 °C, respectively, with a relative humidity ranging from 32% to 93%. Light source was provided by both natural solar radiation and artificial light, and the midday photosynthetic active light intensity was maintained between 600 and 1200 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ PAR.

Study 2: Effect of ABA on stress tolerance

This experiment was conducted in a greenhouse at Texas A&M AgriLife Research at Uvalde during spring 2006. Artichoke seedlings cv. Green Globe Improved (Condor Seed Production, Inc., Yuma, AZ) were grown as described in study 1. Four concentrations of ABA (0, 500, 1000, and 2000 $\text{mg}\cdot\text{L}^{-1}$) were prepared. Foliar applications were done by a hand sprayer on the upper side of leaves until runoff. Untreated control plants were sprayed with tap water. Drought stress (water withholding) was imposed right after spray applications and maintained for 5 d. After the desiccation period, transplants were re-watered daily for 6 consecutive days until 11 days after transplanting. In the greenhouse, average daily minimum and maximum temperatures during the experiment were 7 °C and 31 °C, respectively. Relative humidity ranged from 33% to 90%. Light was provided by both natural solar radiation and artificial light and the midday photosynthetic active light intensity in the greenhouse was maintained between 600 and 1200 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ PAR.

3.2.1.2 Plant physiological responses and water status

Plant physiological responses were determined by daytime measurements of net photosynthetic rate (A_{CO_2} , $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and stomatal conductance (g_s , $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). Measurements were done using a portable LI-6200 photosynthesis system (Li-Cor Inc, Lincoln, NE). Measurements were conducted at 0, 1, 3 and 4 DAT (drought stress period, Study 1) and at 0, 3, 5 (drought stress period) and 11 DAT (6 days after re-watering, Study 2).

Plant water status and cell membrane stability under temporal drought and foliar application of antitranspirants were determined. Water potential (ψ_w , MPa), relative water content (RWC, %) and electrolyte leakage (EL, %) were measured as previously described in section 3.1.1.4. All destructive measurements were conducted at 0, 3 and 4 DAT (Study 1) and at 0, 3, 5 and 11 DAT (6 days after re-watering) (Study 2).

3.2.1.3 Statistical analysis

These experiments were conducted using a randomized complete block design with four replications. All measurements were taken in two subsamples. Data were statistically analyzed by ANOVA using SPSS (version 14.0 for Windows; SPSS Inc., Chicago, IL). Differences among treatments were performed using LSD at $p = 0.05$. If the Levene test for homogeneity of variance and/or the Shapiro-Welk test or Kolmogorov-Smirnov test for normality of experimental error were significant, then a nonparametric analysis using Kruskal-Wallis test was conducted instead of ANOVA. Multiple comparisons were performed using Mann-Whitney test.

3.3.2 Results

3.3.2.1 Plant physiological responses

Study 1: Effect of antitranspirants on stress tolerance

Net photosynthetic rate (A_{CO_2}) measured before treatment applications (Day 0) was $13.0 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$. After 24 h of drought stress (1 DAT) no significant differences for A_{CO_2} was detected among film-forming antitranspirants (AS, TF and VG) and control, but a significant depression of A_{CO_2} ($0.7 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) occurred with ABA. Conversely, after 3 d of drought stress (3 DAT) A_{CO_2} sharply decreased for the control and the three film-forming treatments ($1.6 - 2.3 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), while A_{CO_2} at ABA-treated plants was reversed ($5.4 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and maintained significantly higher compared with the control and film-forming antitranspirants at 3 DAT. These differences in favor of ABA were maintained at 4 DAT, while A_{CO_2} was significantly reduced at all other treatments (undetectable at AS and VG) (Table 7).

Stomatal conductance (g_s) measured before treatment application was $2.30 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, and after 24 hours of drought stress (1 DAT) g_s slightly decreased both for the control ($1.88 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and film-forming antitranspirants (ranging from 1.68 to $1.96 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), and with no significant difference among these treatments. Conversely a sharp drop of g_s ($0.08 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) was measured for the ABA treatment and this lower level of g_s for the ABA treatment was maintained throughout the experiment. As drought stress progressed, g_s was significantly reduced for the control and film-forming antitranspirants (undetectable for AS- and VG-treated plants at 4 DAT). Although, ABA-

treated plants kept relatively low g_s values at 3 and 4 DAT, they were significantly higher than control and film-forming antitranspirants (Table 7).

Study 2: Effect of ABA on stress tolerance

Net photosynthetic rate (A_{CO_2}) before treatment application was $16.9 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, and after 24 h of drought stress it declined as concentration increased, with the lowest value of $4.0 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ at $2000 \text{ mg}\cdot\text{L}^{-1}$ of ABA. After 3 d of drought stress, A_{CO_2} decreased for the control ($1.1 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and $500 \text{ mg}\cdot\text{L}^{-1}$ of ABA ($5.80 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), while A_{CO_2} increased in ABA-treated plants at higher concentrations. The A_{CO_2} was completely reversed after rewatering for plants treated with the higher concentration of ABA ($1000 \text{ mg}\cdot\text{L}^{-1}$ and $2000 \text{ mg}\cdot\text{L}^{-1}$) compared to control and $500 \text{ mg}\cdot\text{L}^{-1}$ ABA-treated plants. Eventually, plants completely wilted for the control and $500 \text{ mg}\cdot\text{L}^{-1}$ of ABA and A_{CO_2} for these plants were not detectable 11 DAT or 6 d after rewatering (Table 8).

Stomatal conductance (g_s) measured before drought stress was $1.06 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$, and after 24 h of drought stress sharply dropped in a concentration dependent manner, being the lowest level for the $2000 \text{ mg}\cdot\text{L}^{-1}$ ABA ($0.10 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and the highest for control ($0.39 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). After 3 d of drought stress, g_s was significantly lowered for the control ($0.09 \text{ mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), but it was maintained for ABA-treated plants. After rewatering (8 and 11 DAT), higher g_s was measured for the two highest concentrations of ABA and the lowest at $500 \text{ mg}\cdot\text{L}^{-1}$ (8 DAT). At 11 DAT g_s was not detectable both for the control and $500 \text{ mg}\cdot\text{L}^{-1}$ of ABA-treated plants, but was maintained lower rate at the two highest concentrations of ABA (Table 8).

Table 7. Leaf photosynthesis and stomatal conductance of artichoke transplants in response to antitranspirant foliar application with 4 d of desiccation (Study 1).

Antitranspirants	Photosynthetic rate ($\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)			Stomatal conductance ($\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$)		
	Day 1	Day 3	Day 4	Day 1	Day 3	Day 4
Control	14.7 a	1.6 b	0.7 b	1.88 a	0.11 c	0.01 b
ABA	0.7 b	5.4 a	3.0 a	0.08 b	0.20 a	0.05 a
Antistress	11.8 a	1.4 b	ND ¹	1.68 a	0.13 bc	ND
Transfilm	12.5 a	1.5 b	0.79 b	1.96 a	0.14 bc	0.02 b
Vapor Gard	12.1 a	2.3 b	ND	1.79 a	0.13 b	ND

Photosynthetic rate and stomatal conductance at Day 0 were 13.0 and 2.30, respectively.

Means within columns followed by different letters are significantly different. (LSD or Mann-Whitney test, $p = 0.05$)

¹ ND: data not detectable.

Table 8. Leaf photosynthetic rate and stomatal conductance of artichoke transplants in response to ABA foliar application with 5 d of desiccation and 6 d of rewatering (Study 2).

ABA (mg·L ⁻¹)	Photosynthetic Rate (μmol·m ⁻² ·s ⁻¹)				Stomatal Conductance (mol·m ⁻² ·s ⁻¹)			
	Day 1	Day 3	Day 8 (3) ¹	Day 11 (6)	Day 1	Day 3	Day 8 (3)	Day 11 (6)
0	15.4 a	1.1 c	ND ²	ND	0.39 a	0.09 b	ND	ND
500	8.7 b	5.8 b	2.0 c	ND	0.15 b	0.14 a	0.05 c	ND
1000	5.7 c	10.0 a	5.6 b	8.4 b	0.12 b	0.16 a	0.09 b	0.18 b
2000	4.0 c	7.3 b	7.6 a	10.5 a	0.10 b	0.13 a	0.12 a	0.24 a

Photosynthetic rate and stomatal conductance at Day 0 were 16.9 and 1.06, respectively.

Means within columns followed by different letters are significantly different. (LSD or Mann-Whitney test , p = 0.05).

¹ Number in parentheses shows the days from start daily watering.

² NA: data not detectable.

3.3.2.2 *Plant water status and cell membrane stability*

Study 1: Effect of antitranspirants on stress tolerance

Relative water content (RWC) was 80.7% when measured before drought stress, and after 3 d of drought stress it was significantly decreased for both control (58.0%) and film-forming transpirants (60.7 – 62.5%), while the highest level of RWC was maintained in ABA-treated plants (88.8%). The reduction of RWC was more pronounced for the control (41.3%) and film-forming antitranspirants (36.8 – 41.5%) after 4 d of drought stress, but not for ABA-treated plants (83.9%) (Table 9).

Electrolyte leakage (EL) was 8.6% when measured at 0 DAT, and after 3 days of drought stress ABA-treated plants had lower EL (10.4%) compared with control (12.9%) and film-forming antitranspirants (13.6 – 18.4%). Higher EL (>30%) was pronounced for all treatments except ABA-treated plants at 4 DAT (Table 9).

Leaf water potential (ψ_w) was -0.48 MPa when measured at 0 DAT, and significantly decreased for all treatments except ABA-treated plants (-0.37 MPa) after 3 d of drought stress. The decline of ψ_w was pronounced for both control and film-forming antitranspirants at 4 DAT, but was significantly higher ψ_w (-0.75 MPa) for ABA-treated plants (Table 9).

After 4 d of drought stress, control or plants treated with film-forming antitranspirants were severely wilted and stunted, while ABA-treated plants maintained turgidity (Figure 12).

Study 2: Effect of ABA on stress tolerance

Relative water content (RWC) was 83.1% when measured before drought stress, but decreased as ABA concentration decreased (56.3% at 500 mg·L⁻¹ and 71.2% at 2000 mg·L⁻¹ of ABA) after 3 d of drought stress. As drought stress progressed (5 DAT), RWC decreased for all treatments, but completely recovered after rewatering for only the higher concentrations of ABA (1000 and 2000 mg·L⁻¹) but not for control or 500 mg·L⁻¹ of ABA (Table 10).

Electrolyte leakage (EL) was 10.7% when measured at 0 DAT. After 3 d of drought stress all ABA-treated plants were able to maintain lower levels of EL (ranging from 11.2 to 12.3%) than control (31.0%). As drought stress progressed (5 DAT), EL was significantly higher for the control, while lower levels were measured for ABA at 1000 mg·L⁻¹. EL was significantly reduced for higher concentrations of ABA (1000 and 2000 mg·L⁻¹) compared to ABA 500 mg·L⁻¹ (Table 10).

Figure 13 shown complete wilting for control plants, medium wilting for 500 mg·L⁻¹, less or none wilting for 1000 and 2000 mg·L⁻¹ of ABA-treated plants at 4 d after drought stress.

Table 9. Relative water content (RWC), electrolyte leakage (EL), and leaf water potential (ψ_w) of artichoke transplants in response to antitranspirant foliar application with 4 d of desiccation (Study 1).

Antitranspirants	RWC (%)		EL (%)		ψ_w (MPa)	
	Day 3	Day 4	Day 3	Day 4	Day 3	Day 4
Control	58.0 b	41.3 b	12.9 ab	31.0 ab	-1.68 b	-3.00 b
ABA	88.8 a	83.9 a	10.4 b	11.1 b	-0.37 a	-0.75 a
Antistress	60.7 b	41.3 b	13.6 ab	44.1 a	-1.54 b	-2.96 b
Transfilm	62.3 b	41.5 b	18.4 ab	31.1 ab	-1.61 b	-2.64 b
Vapor Gard	62.5 b	36.8 b	15.7 a	42.8 ab	-1.60 b	-3.38 b

RWC, EL, and ψ_w at Day 0 were 80.7, 8.6 and 0.48, respectively.

Means within columns followed by different letters are significantly different. (LSD or Mann-Whitney test , $p = 0.05$).

Table 10. Relative water content (RWC) and electrolyte leakage (EL) of artichoke transplants in response to ABA foliar application with 5 d of desiccation and 6 d of rewatering (Study 2).

ABA (mg·L ⁻¹)	Relative water content (%)			Electrolyte leakage (%)		
	Day 3	Day 5	Day 11 (6) ¹	Day 3	Day 5	Day 11 (6)
0	45.8 c	20.1 c	12.2 b	31.0	90.3 a	NA ²
500	56.3 bc	29.7 b	24.8 b	12.3	71.6 ab	71.2 a
1000	63.4 ab	39.7 a	74.5 a	11.2	53.4 b	15.0 b
2000	71.2 a	34.1 b	79.5 a	11.3	71.0 ab	12.9 b

RWC and EL at Day 0 were 83.1 and 10.7, respectively.

Means within columns followed by different letters are significantly different. (LSD or Mann-Whitney test , p = 0.05).

¹ Number in parentheses shows the days from start daily watering.

² NA: data not available.

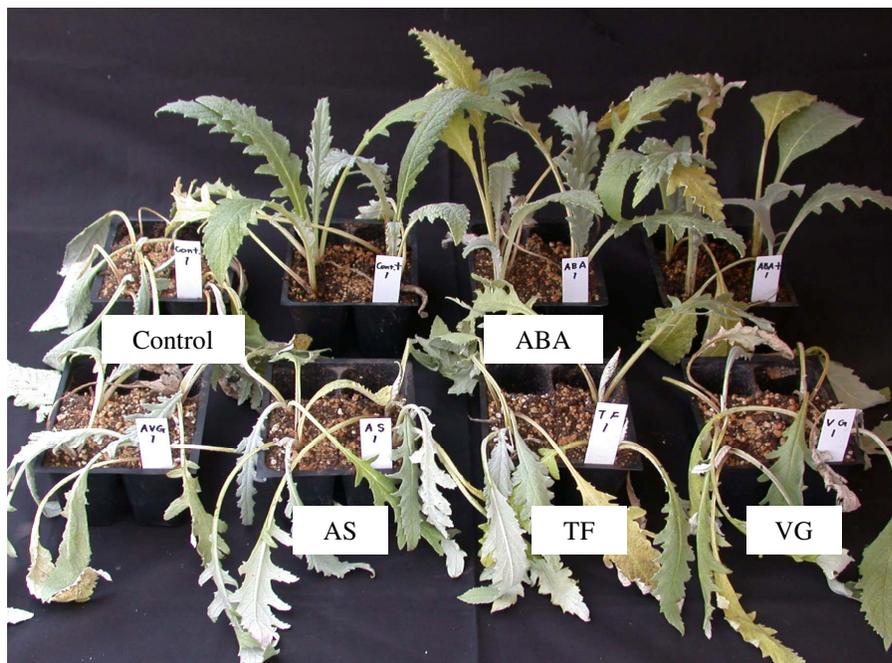


Figure 12. Artichoke seedlings treated with antitranspirants (ABA; abscisic acid, AS; Antistress, TF; Transfilm, VG; Vapor Gard and water as Control) 4 d after desiccation (Study 1).

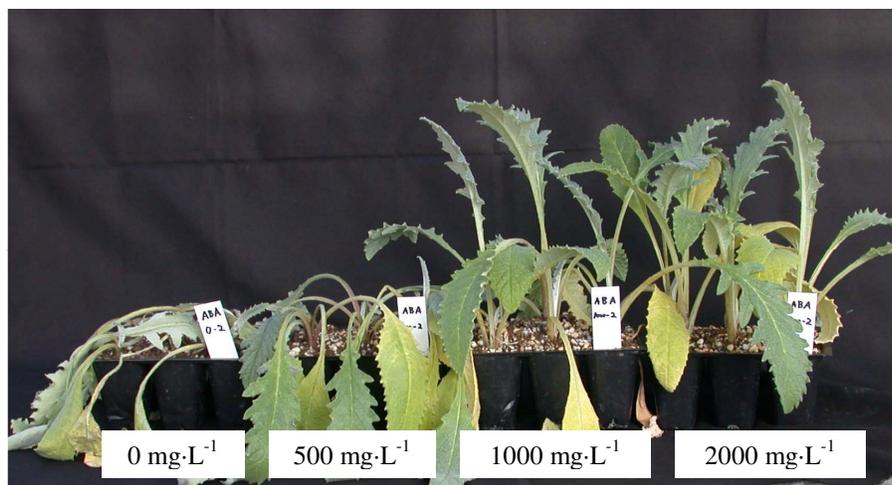


Figure 13. Artichoke seedlings treated with ABA (0 as Control, 500, 1000 and 2000 $\text{mg}\cdot\text{L}^{-1}$) 4 d after desiccation (Study 2).

3.3.3 Discussion

3.3.3.1 Study 1

In this study, the effects of film-forming antitranspirants and ABA foliar application on physiological responses (A_{CO_2} and g_s), water status and cell stability (RWC, EL and ψ_w) of artichoke transplants under drought stress were examined.

Overall, film-forming antitranspirants AS, TF and VG did not show any positive effect on either physiological response or water status of artichoke transplants when compared with control plants. As the period of drought stress was prolonged, A_{CO_2} and g_s of plants treated with all film-forming antitranspirants gradually decreased with time (Table 7). The physiological responses were well related with their water status (RWC and ψ_w) (Table 9). The reduction of RWC and ψ_w in leaves eventually caused severe wilting and cell membrane injury (Table 9 and Fig. 12). In contrast to film-forming antitranspirants, ABA positively affected physiological responses and water status of artichoke transplants. Stomatal conductance (g_s) for ABA-treated plants was reduced to 95% compared to control at 1 DAT, and depression of A_{CO_2} was associated with the stomatal closure (Table 7). However, g_s partially recovered over time as compared to control (Table 7). The rapid plant response of stomata helped to maintain a high seedling water status and thus cell hardiness under drought stress (Table 9 and Fig. 12).

Film-forming antitranspirants are widely used commercially for a variety of horticultural crops, orchard trees and turfs, and former studies indicate significant improvements of plant performance (EnviroShield Products Co., 2005; Irmak et al.,

2003; PBI/Gordon Co., 2007). However our results in artichoke seedlings do not support these claims. Presumably, the film-forming antitranspirants applied foliarly act as a physical barrier on leaf surfaces, primarily in the adaxial surface. This effect is mainly specie dependent, due to different leaf morphological characteristics (shape, wax deposition, trichomes, stomata number and size). In fact, artichoke young leaves are covered by dense glandular trichomes and relatively higher number of stomata are located in the abaxial vs. adaxial surface (Brutti et al., 2002). Our results agree with those of Goreta et al. (2007) and Leskovar et al. (2008) who also reported that application of film-forming antitranspirants were not as effective as ABA for drought stress mitigation in pepper and tomato seedlings.

3.3.3.2 Study 2

In this study, the effects of ABA foliar application with 4 different concentrations (0 as control, 500, 1000 and 2000 mg·L⁻¹) on physiological responses (A_{CO_2} and g_s), water status and cell membrane stability (RWC and EL) of artichoke transplants were examined during a 5 d period of drought stress and following after 6 d of re-watering.

After 24 hours of application and dehydration, both A_{CO_2} and g_s were readily decreased by ABA, except for control plants (Table 8). Those effects were highly concentration dependent, with stronger responses with higher ABA concentrations (Table 8). These responses were transient, particularly for the photosynthetic rates which partially recovered when measured 3 DAT, while it tended to be prolonged on stomatal conductance, which remained low even after re-watering (Table 8). Consistent with

stomatal closure, leaf RWC was higher compared with control when measured 3 DAT, but as drought stress progressed it gradually decreased (Table 10). Higher RWC contributed to maintain plant hardiness (EL), but EL also increased as dehydration got severe (Table 10). Relative water content (RWC) and EL were fully recovered after 6 d of re-watering with 1000 and 2000 mg·L⁻¹ ABA, but while not for plants treated with 500 mg·L⁻¹ of ABA or for control plants.

The results from these studies indicate that ABA treated transplants had an improved drought stress tolerance which we attribute to a higher maintenance of seedling water status during the drought stress period and a rapid recovery to normal water status after rewatering. In addition to the involvement of ABA in regulating stomatal openings, its transient effect on photosynthesis reduction and higher water retention in plants, other protective plant responses have been reported in different plants under environmental stresses. Those ABA effects include increased adventitious roots such as shown in tomato transplants (Takahashi et al., 1993), inhibition of ethylene synthesis in jack pine seedlings under drought stress (Rajasekaran and Blake, 1999), and amelioration of photosynthetic enzyme activity in wheat under drought (Nayyar and Kaushal, 2002). Whether the beneficial effects of ABA on artichoke transplants under drought stress could also be in part related to those growth and enzymatic response is unknown.

A partial leaf chlorosis was observed at 2000 mg·L⁻¹ ABA application during this experiment. ABA application could affect leaf temperature. This effect may be because of heat injury caused by inhibition of transpiration, thus leaf cooling effect, which is

associated with highly decreased stomatal conductance. Similar symptoms were also observed in cucumber leaves treated with 1.0 mM (250 mg·L⁻¹) of ABA application at high temperature (45 °C) conditions (Oda et al., 1994).

In conclusion, ABA (1000 mg·L⁻¹) foliar application effectively enhanced drought tolerance of artichoke transplants associated with the maintenance of leaf water status via stomatal closure. These results suggest that exogenous ABA could be a useful transplant conditioner, thus improving hardiness of artichoke transplants allowing them to withstand temporal drought stress. We expect ABA could reduce transplant shock and enhancing stand establishment. Improved stand establishment is a pre-requisite for a rapid resumption of growth and to optimize crop performance and yield of artichoke grown in semi-arid conditions.

CHAPTER IV

FIELD CROP MANAGEMENT

4.1 Effects of Irrigation and Nitrogen on Artichoke Growth, Yield and Quality

4.1.1 Materials and Methods

4.1.1.1 Experimental design

Annual - biennial production system (2005-07)

A two-year field experiment was conducted at the Texas A&M AgriLife Research and Extension Center at Uvalde, TX to determine the impact of irrigation regimes and nitrogen rates on yield and nutritional quality of artichoke heads cvs. Imperial Star and Green Globe Improved (Condor Seed Production, Inc., Yuma, AZ). The soil was a Uvalde clay loam (pH 7.9, organic matter 3.3%, with a textural composition of 22% sand, 40% silt, and 38% clay). Pre-plant soil physical and chemical properties (0-15 cm depth) are summarized in Table 11. Two month old artichoke seedlings were transplanted on beds (2.03 m between rows, 0.76 m within row, giving a plant population of 6,465 plants·ha⁻¹) on 16 Dec 2005. After the first season harvest ended (2005-06), plants were cut back to the ground level on 19 July 2006 for 'Imperial Star' and 25 July 2006 for 'Green Globe Improved' and new off-shoots were allowed to re-growth for the second season (2006-07) production (Fig. 14).

Main plots were allocated to three irrigation regimes: 50, 75 and 100% crop evapotranspiration rates (ET_c). The values obtained for ET_c were based on climatic parameters that are incorporated in the calculation of the reference evapotranspiration (ET_o), which was updated daily in the lysimeter facility located at the experimental site. The ET_o values were adjusted by these phenological crop coefficients (K_c), which were referred as FAO's values (K_c ini = 0.5, K_c mid = 1.0, K_c end = 0.95) (FAO, 1998) based on crop canopy characteristics with slight modification. Irrigation water was supplied using a subsurface drip system (SDI) with drip tape placed at 15 cm below the soil surface and emitters spaced every 30 cm, and a flow rate of 340 L·h⁻¹·100m at 0.55 bar (T-Tape® 500 U.S. Model, T-systems International, San Diego, CA). Differential irrigation started after stand establishment on 31 Jan 2006 (first season, 2005-06) or after plants reached uniform growth on 17 Oct 2006 (second season, 2006-07). Total water inputs (irrigation + rainfall) in each irrigation rates during the experiment were described in Table 12. Subplots within main plots were allocated to four N fertilizer rates: 0, 60, 120, and 180 kg·ha⁻¹ applied as a granular ammonium nitrate (NH₄NO₃) by side dressing in two equal split doses, the first at stand establishment (20 Jan 2006 for the first and 9 Oct 2006 for the second season) and the second at bud initiation (24 Mar 2006 for first and 1 Feb for second season) (Fig. 14).

Table 11. Physical and chemical properties of soils, 2005.
Uvalde, TX

Properties	Values
Textural	Clay Loam
Sand	22 %
Silt	40 %
Clay	38 %
pH	7.9
EC	435 μ mhos/cm
Organic Carbon	1.9 %
Organic Matter	3.3 %
Total Nitrogen	1396.7 ppm
NH ₄ -N	3.7 ppm
NO ₃ -N	77.4 ppm
P	64.6 ppm
K	717.0 ppm
Ca	14320.7 ppm
Mg	243.7 ppm
S	40.4 ppm
Na	143.9 ppm
Fe	2.7 ppm
Zn	1.1 ppm
Mn	9.7 ppm
Cu	0.5 ppm

Soil samples (0-13 cm depth) were collected prior to planting. Samples were air-dried, sieved, and analyzed at the Texas A&M Soil, Water and Forage Testing Laboratory, College Station.

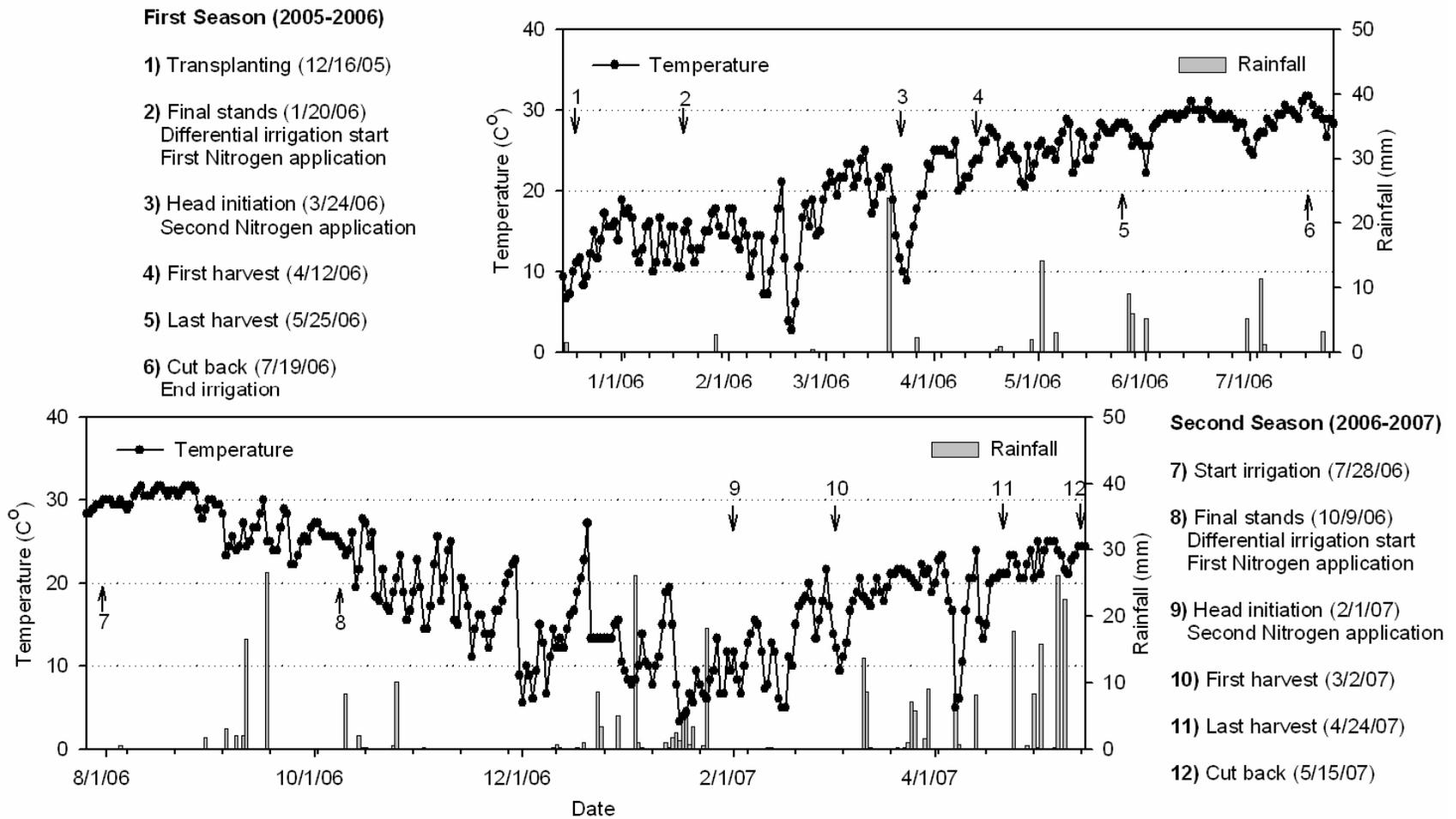


Figure 14. Time-course of average temperatures (solid lines), rainfall (bars) and specific agronomic practices (arrows) during the first and second season, 2005-07. Uvalde, TX

Table 12. Frequency (No.) and amount (mm) of irrigation applied or rainfall received during the artichoke production cycle, 2005-07. Uvalde, TX.

2005-06	Frequency and amount of irrigation or rainfall						Total applied			
	Dec.	Jan.	Feb.	Mar.	Apr.	May				
	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>			
<u>Irrigation</u>										
50% ETc	6-136 ^x	6-54	4-27	5-37	10-78	13-127	44-404			
75% ETc	6-136 ^x	6-57	5-41	6-56	10-119	13-179	45-530			
100% ETc	6-136 ^x	6-60	5-53	6-72	10-161	13-234	45-656			
Rainfall	1-1.5	1-2.8	2-0.8	3-26	3-3.6	4-33	14-68			
2006-07	Jun.–	Sep	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	Total applied
	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>
50% ETc	22-204 ^y	10-75	6-50	6-41	3-21	0-0	6-45	5-36	7-54	65-536
75% ETc	22-204 ^y	10-75	6-59	6-61	3-32	0-0	6-68	5-62	7-81	65-642
100% ETc	22-204 ^y	10-75	6-68	6-81	3-42	0-0	6-90	5-87	7-109	65-757
Rainfall	10-39	5-50	6-22	1-0.3	8-20	14-68	2-0.5	11-48	4-35	61-282

^xValues include pre-planting irrigation, 4 times with 112 mm.

^yValues include pre-awaking irrigation, 7 times with 69 mm.

Annual production system (2007-08)

Based on the results of the first two years, a third year field experiment was conducted in the same location and conditions as in the biennial system, except for some modifications (e.g., foliar fertilizations and plasticulture) aimed at optimizing growth and productivity. Two month-old artichoke seedlings were transplanted on beds (2.03 m between rows, 0.91 m within row, giving a plant population of 5,390 plants·ha⁻¹), and covered by black plastic mulch (0.038 mm thickness) on 24 Oct 2007.

Main plots were allocated to the same three differential irrigations as previously described (50, 75 and 100% evapotranspiration rates; ET_c). Since black plastic mulch was applied, evaporation from bare soil ($K_c \text{ bare} = 0.2$) was deducted from the FAO's K_c and calculated the water application in the mulch throughout the season. Therefore the adjusted K_c values were $K_c \text{ ini} = 0.3$, $K_c \text{ mid} = 0.8$ and $K_c \text{ end} = 0.95$, with modifications based on canopy size. Irrigation water was supplied using a drip tape placed at 15 cm below surface with emitters spaced every 30 cm and a flow rate of 170 L·hour⁻¹ at 0.55 bar (T-Tape® 500 U.S. Model, T-systems International, San Diego, CA). Differential irrigation started after stand establishment on 28 Nov 2007. Water inputs (irrigation + rainfall) during the experiment are described in Table 13. Subplots within main plots were allocated to four N rates: 10, 60, 120, and 180 kg·ha⁻¹ as liquid UAN applied by fertigation, Fertijet™ (NETAFIM™, Fresno, CA). A 10 kg·ha⁻¹ of N (UAN, 32%) was applied to all plots as a starter fertilizer on 14 Nov 2007 in order to enhance stand establishment. The rest of N (0, 50, 110 and 170 kg·ha⁻¹) was applied in four equal split doses on 28 Nov, 17 Dec in 2007, 8 Jan and 5 Feb in 2008 (Fig. 15).

Third Season (2007-2008)

1) Transplanting (10/24/07)

2) Final stands (11/28/07)
Differential irrigation start
First Nitrogen application

3) Head initiation (2/21/08)
Second Nitrogen application

4) First harvest (3/20/08)

5) Last harvest (4/28/08)

6) Cut back (5/9/08)
End irrigation

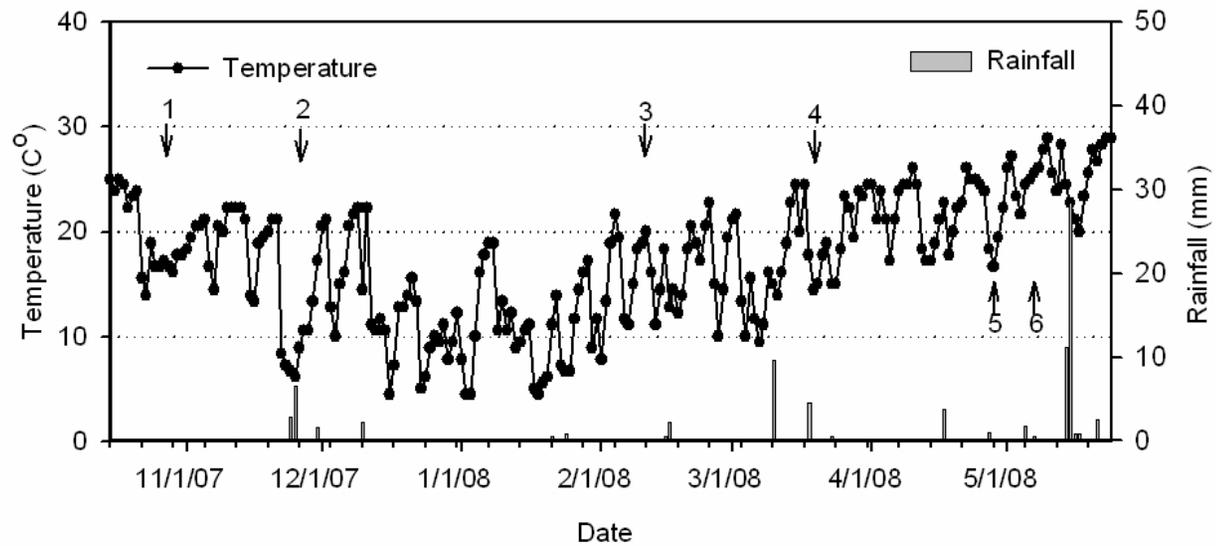


Figure 15. Time-course of temperature (solid lines), rainfall (bars) and specific agronomic practices (arrows) during the annual artichoke production cycle 2007-08. Uvalde, TX

Table 13. Frequency (No.) and amount (mm) of irrigation applied or rainfall received during the artichoke production cycle on black plastic mulch, 2007-08. Uvalde, TX

2007-08	Frequency and amount of irrigation or rainfall							Total applied
	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	
	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>	<u>No.-mm</u>
<u>Irrigation</u>								
50% ETc	6-22 ^x	6-18	3-9	4-12	7-25	7-39	7-54	40-179
75% ETc	6-22 ^x	6-18	4-14	5-18	9-38	12-58	14-80	56-248
100% ETc	6-22 ^x	6-18	4-19	5-24	8-50	12-78	14-105	55-317
Rainfall	0-0	3-11	1-2	4-2	2-3	5-15	2-5	17-38

^xValues include pre-planting irrigation, 2 times with 12 mm.

4.1.1.2 Field management

In addition to N applications, 56 kg·ha⁻¹ of P and 60 kg·ha⁻¹ of K were applied each season as a water soluble granule nutrient mixture (Gainer[®] 0-50-30 with 0.02% B, 0.05% Cu, 0.1% Fe, 0.05% Mn, 0.0005% Mo and 0.05% Zn, RSA MicroTech, LLC, Marysville, WA) through a computer controlled fertigation system (Fertijet[™], NETAFIM[™], Fresno, CA). These nutrients were applied in four split doses with a 40%: 20%: 20%: 20% on 19 Dec 2005, 31 Jan, 8 Feb and 6 Apr 2006 for the first season (2005-06) and on 9 Aug, 9 Oct, 21 Dec 2006 and 21 Feb 2007 for the second season (2006-07), and on 31 Oct, 28 Nov, 18 Dec 2007 and 9 Jan 2008 for the third season (2007-08). Supplemental Ca and Zn (Ca-Zn, 5% Ca and 2.5% Zn, TRACITE[®]) were biweekly applied foliarly by a CO₂ backpack sprayer during harvesting period (5 L·ha⁻¹ and 14 L·ha⁻¹) in the 2005-06 and 2006-07 season, respectively and during the whole growth period (26 L·ha⁻¹) in 2007-08 season. Weeds were controlled by a pre-plant application of 1.1 kg·ha⁻¹ of Diuron 80DF (Diuron, 80% a.i.) and 5.8 L·ha⁻¹ of Goal[®] 2XL (Oxyflurofen, 23% a.i.) and by hand weeding throughout the cropping seasons, if necessary. Cucumber beetle control during the vegetative stage was performed with a single foliar spray of 2.3 L·ha⁻¹ of Endosulfan 3EC (Endosulfan, 34% a.i.) (second season, Study 1) or by periodic application of 70 ml·ha⁻¹ of Asana[®] XL (Esfenvalerate, 8.4% a.i.) (Study 2). For aphids, spider mites and corn earworm control during the growing season, 580 ml·ha⁻¹ of Provado[®] 1.6 (Imidacloprid, 17.4% a.i.) and/or 420 g·ha⁻¹ of Capture[®] 2EC (Bifenthrin, 25.1% a.i.) were used when needed in all seasons. During late spring to late summer in 2006 (Study 1), some artichoke younger offshoots were

suddenly wilted, leading to dead caused by cotton root rot (*Phymatotrichum omnivorum*). To our knowledge this is the first evidence of cotton root rot disease on artichokes, as identified in Texas by Dr. M.C. Black, Texas A&M AgriLife Extension in Uvalde. This disease finally resulted in extensive plant losses (approximately 40%).

4.1.1.3 Earliness and head emergence (first season)

Head emergence rate (%) of both cultivars 'Green Globe Improved' and 'Imperial Star' was monitored weekly on 4 Apr (109 days after transplant: DAT), 10 Apr (115 DAT) and 18 Apr (123 DAT) and 23 May (158 DAT, end of the first season) in 2006. Since bolting requires vernalization, cumulative temperatures [$\Sigma T(\text{mean day } ^\circ\text{C})$] and chilling hours [$\Sigma h(^{\circ}\text{C} \leq 10)$] were calculated using temperature records from a weather station at the Texas A&M AgriLife, Uvalde.

4.1.1.4 Plant growth and physiological response (third season)

During the third season (2007-08), leaf number on main stem was recorded throughout the crop season on 13 Nov (20 DAT), 26 Nov (33 DAT), 20 Dec (57 DAT), 10 Jan (78 DAT), 23 Jan (91 DAT), 7 Feb (106 DAT), 21 Feb (120 DAT), 5 Mar (133 DAT) and 19 Mar (147 DAT). Plant biomass accumulation was estimated by measurements of plant width and height, and recorded on 5, 18 and 27 Mar 2008. Plant physiological responses were determined by measurements of net photosynthetic rate (A_{CO_2} , $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), transpiration (E , $\text{mmolH}_2\text{O}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) and stomatal conductance (g_s , $\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). All indices were measured during midday by a portable photosynthesis

system (LI-6400, Li-Cor Inc, Lincoln, NE). Measurements were conducted on 8 Feb, 21 Mar, 10 Apr and 2 May 2008 and the light strength, atmospheric CO₂ concentration, and stomatal ratio were adjusted to 1500 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ PAR, 400 ppm, and 0.8 as cited in Brutti *et al.* (2002), respectively.

4.1.1.5 Yield and yield components

Harvests occurred periodically between 12 Apr and 25 May 2006 for the first season (2005-06) and between 2 Mar and 24 Apr 2007 for the second season (2006-07) and between 20 Mar and 28 Apr 2008 for the third season (2007-08). Marketable yield ($\text{kg}\cdot\text{ha}^{-1}$), head weight (g) and head number per plant ($\text{no}\cdot\text{plant}^{-1}$) were recorded. Water use efficiency (WUE) expressed in kg of heads per water input ($\text{kg}\cdot\text{ha}^{-1}\cdot\text{mm}^{-1}$) and nitrogen use efficiency (NUE) expressed in kg of heads per nitrogen rates ($\text{kg}\cdot\text{ha}^{-1}\cdot\text{kg}^{-1}$) were calculated.

4.1.1.6 Head size distribution

Artichoke head size under differential irrigation and N rates was determined at different harvest times. Harvest periods were separated as early and late. Early harvests were done on 13, 20, 26 Apr and 3 May 2006 for the first season (2005-06), 2, 12, 19 and 23, 28 Mar 2007 for the second season (2006-07), and 20, 24, 28 Mar, 1, 4 and 7 Apr 2008 for the third season (2007-08). Late harvests were done on 8, 12, 18 and 25 May for the first season (2005-06), 3, 10, 16 and 24 Apr 2007 for the second season (2006-07) and 11, 14, 18, 21, 25 and 28 Apr 2008 for the third season (2007-08). Head

size was classified into marketable heads based on four sizes; <7 cm, 7-9 cm, 9-11 cm or >11 cm and unmarketable, those with tip burn and open bracts.

4.1.1.7 Soil nitrogen content (first and second season)

Soil residual N was measured after the harvesting season to investigate soil N availability and also to estimate plant N acquisition during a crop cycle. Soil sampling (0-15 cm depth) was completed on 27 July 2006 for the first season (2005-06) and on 25 June 2007 for the second season (2006-07). Nitrate-nitrogen ($\text{NO}_3\text{-N}$, ppm) was extracted from soils using a 1N KCl solution. Nitrate was determined by reduction of nitrate ($\text{NO}_3\text{-N}$) to nitrite using a cadmium column followed by spectrophotometric measurement. Ammonium-nitrogen ($\text{NH}_4\text{-N}$) was extracted from soils using a 1N KCl solution. Ammonium-N was determined spectrophotometrically at 420 nm wavelength. Soil N analysis was conducted at the Texas A&M Soil, Water and Forage Testing Laboratory in College Station, TX.

4.1.1.8 Chemical constituents in heads (first and second season)

Chemical constituents of artichoke heads were determined at three times during harvests; 20 Apr, 3 and 25 May 2006 for the first season (2005-06) and 23 Mar, 3 and 24 Apr 2007 for the second season (2006-07). Before analysis, outer bracts were removed keeping the stems (5 cm length), chopped, dried in oven at 70 °C until constant weight, and ground to make powder samples, which were kept in sealed plastic bags until analysis. Total nitrogen and fiber content were analyzed at the Texas A&M AgriLife

Research at Uvalde, TX and sugar, total phenolics, chlorogenic acid and cynarin contents (methods are described below) were analyzed at the Vegetable and Fruit Improvement Center (VFIC) at College Station, TX under the direction of Dr. K. Yoo.

Nitrogen content: Total N content ($\text{g}\cdot 100\text{g}^{-1}$ DW) was analyzed by the Kjeldahl method. About 0.2 g of artichoke dry sample was digested by concentrated H_2SO_4 with Kjel-tabs (Catalyst tablets, VWR®, West Chester, PA). The distillation was conducted using a Kjeltec System 1028 Distilling Unit (Inc., Hoganas, Sweden) with 0.1 N NaOH. Distillates were collected into boric acid solution with indicators (bromocresol green and methyl red) and titrated with 0.1 M HCl. Total N content was calculated by the following equation:

$$\text{N (g}\cdot 100\text{g}^{-1} \text{ DW)} = [(\text{Ts}-\text{Tb}) \times \text{N} \times 1.4] / \text{sample dry weight}$$

where: Ts is ml to titrate sample, Tb is ml to titrate blank and N is normality of HCl (N = 0.0993).

Fiber content: Since fiber content of artichoke heads is an important constituent for their health promoting properties (e.g., prevent constipation) as well as their sensory quality (e.g., taste and texture), both neutral detergent fiber (NDF, $\text{g}\cdot 100\text{g}^{-1}$ DW, hemicellulose, cellulose and lignin) and acid detergent fiber (ADF, $\text{g}\cdot 100\text{g}^{-1}$ DW, cellulose and lignin) were measured (Van Soest, 1963). About 0.75 g artichoke dry sample was taken and NDF and ADF were analyzed using a FiberTec System I (Tector Inc., Hoganas, Sweden) with NDF and ADF solutions.

NDF solution: Four liters of distilled H_2O was placed in 6 L flasks, adding 450 g sodium lauryl sulfate and 150 ml 2-ethoxyethanol mixed until dissolved which were then

transferred to the tank. Distilled H₂O of 1.5 L was placed in a 2 L flasks, adding 279.2 g EDTA, disodium salt and 102.2 g sodium borate and mixed while heating, until dissolved, and then transferred to the 25 L tank. Distilled H₂O of 1.5 L was placed in 2 L flasks, add 68.4 g sodium phosphate, dibasic, and mixed, while heating, until dissolved which were then transferred to the tank, then distilled H₂O of 7.85 L was added and mixed.

ADF solution: Four liters of distilled H₂O in 6 L flasks and adding 300 g hexadecyltrimethyl-ammonium bromide. Sulfuric acid of 420 ml was added and mixed until dissolved which was then transferred to the 25 L tank, then distilled H₂O of 10.58 L was added and mixed.

Sugar composition: About 1 g artichoke dry sample was taken from plastic bag and placed into 15 ml centrifuge tubes, mixed with 30 ml 80% ethanol, and shaken for 24 hours. Extracts of 500 µl were collected and kept in freezer (<0 °C) until sugar analysis. The ethanol extracts were injected into a high performance liquid chromatograph (HPLC) system. The HPLC system included a binary pump, an autosampler, a refractive index detector, a sugar analysis column (Alltech 700 CH) with a guard cartridge and heated at 90°C. The solvent was deionized water at a flow rate of 0.5 ml·min⁻¹ and 20 µl sample was injected. Sugar concentrations of sucrose, glucose and fructose were calculated using external standards.

Phenolics content: Total phenolics content was measured using the Folin-Ciocalteu assay (Singleton and Rossi, 1965). The ethanol extract used in the sugar analysis was used for total phenolic measurements. Extracts of 50 µl were mixed well

with 9 ml of nano pure water in a 15 ml test tube. A 0.5 ml Folin-Chiocalteu reagent (F-C solution) was added, mixed well and waited for 5 min. A 1.5 ml of NO_2CO_3 solution ($20 \text{ g} \cdot 100 \text{ ml}^{-1}$) was added, mixed well again and kept for 2 hours at room temperature. The total phenolics content was determined by spectrophotometer (Spectronic 601, Milton Roy USA, Ivyland, PA) with absorbance at 760 nm. The content was standardized against chlorogenic acid and the linearity was ranging from $15.6 \text{ mg} \cdot \text{L}^{-1}$ to $2030 \text{ mg} \cdot \text{L}^{-1}$. The final value was converted to milligrams per gram dry weight ($\text{mg} \cdot \text{g}^{-1}$ DW).

Chrologenic acid and cynarin content: The ethanol extract used in the sugar analysis was used for chrologenic acid and cynarin analysis using an HPLC system. Standard compounds- A commercial standard cynarin (identified as 1,3-di-*O*-caffeoylquinic acid, Roth, Germany) and chlorogenic acid (Sigma, St. Louis, MO) were used. A major cynarin isomer, 1,5-di-*O*-caffeoylquinic acid, was purified from the concentrated ethanol extract by using a preparative column (Econosil C-18, 10 μm , Alltech, Deerfield, IL) with $4 \text{ ml} \cdot \text{min}^{-1}$ methanol gradient from 20% to 90% in 30 min. A 150 μl concentrated extract was injected and the matching fraction of the peak was collected. The concentration of the fraction was calculated by measuring absorbance at 326 nm ($E 1\%$, 1 $\text{cm}=616$, Merck Index Eleventh Edition). The same fraction was injected into the HPLC and the peak area was obtained and used as external standard of 5-*O*-caffeoylquinic acid, which was confirmed as chlorogenic acid by a matching spectrum max at 326 nm using a diode array detector. The commercial cynarin and chlorogenic acid were also run by the HPLC and calibrated as external standards. The

purity of the purified standard was similar or better to the commercial ones. The HPLC column used was a Water Spherisorb ODS-2, 5 μm (4.6 \times 250 mm). The solvent was programmed with 10% to 50% acetonitrile containing 0.5% phosphoric acid in 20 min. The flow was 1 $\text{ml}\cdot\text{min}^{-1}$, and after a 5 μl sample was injected, the detection was made at 326 nm. Cynarin content was determined by the isomer 1,5-di-O-caffeoylquinic acid, which is a major cynarin isomer in artichoke heads (Schütz et al., 2004).

4.1.1.9 Statistical analysis

First and second season (2005-07): The experiment was conducted using a split plot design with four replications (thirteen plants per each individual plot sized 2 m \times 9.9 m). Irrigation was assigned to the main plot, nitrogen levels to the subplots. Since the poor head emergence occurred in cv. Green Globe Improved in the first season due to lack of vernalization, yield data from cv. Green Globe Improved was not included.

Data (soil analysis, yield, yield components, head size and chemical constituents) were all analyzed by ANOVA using SPSS (version 14.0 for Windows; SPSS Inc., Chicago, IL). Differences among treatments were performed using LSD at $p = 0.05$. For the head emergence rate, both cvs. Green Globe Improved and Imperial Star, was evaluated by the analysis of data as a split-split plot design with cultivar as main plot and irrigation as sub, and N as sub-sub plots.

Third season (2007-08): The experiment was conducted using a randomized complete block design with four replications (seven plants per each individual plot sized 2 m \times 6.4 m). All plant (cv. Imperial Star) growth measurements including leaf number, plant width, and plant height were taken in two subsamples (total eight plants). Photosynthesis and other plant physiological measurements were taken in one subsample with replicated measurements (twice) (total eight plants). All data were statistically analyzed by ANOVA using SPSS (version 14.0 for Windows; SPSS Inc., Chicago, IL). Differences among treatments were performed using LSD at $p = 0.05$.

4.1.2 Results

4.1.2.1 Climate and water condition

In general, winter (Nov to Feb) temperatures for 2005-06, 2006-07 and 2007-08 seasons were similar, with average daily temperatures in the range of 13.4 to 14.3 °C. The main difference among seasons was that in 2005-06, the average temperature on Jan was significantly higher (14.3 °C) than the other two seasons (9.5 to 11.8 °C).

Rainfall trends of the first (2005-06) and third (2007-08) seasons were considered dry, while the second (2006-07) season was wet. In fact, total rainfall during the crop cycle was 68 mm in 2005-06, 282 mm in 2006-07 and 38 mm in 2007-08, respectively. Large amounts of rainfall in 2007-08 was because of a longer crop cycle and also higher precipitation recorded on Jan, Mar and Apr, 2006 as compared with the other two seasons (Tables 12 and 13).

Total irrigation applied during the crop cycles for the 100% ET_c was 656 mm in 2005-06, 757 mm in 2006-07, and 317 mm in 2007-08. Frequent irrigations occurred during harvest seasons in 2005-06 and 2007-08, but not in 2006-07 due to a large number of precipitation events. Overall, plastic mulch application saved approximately 52 to 58% of irrigation amounts in the third season (2007-08) compared with 2005-06 and 2006-07, respectively (Tables 12 and 13).

4.1.2.2 Earliness and head emergence (first season)

The first head was observed on 22 Mar 2006 (96 DAT) for Imperial Star (IS) which was significantly earlier than Green Globe Improved (GGI). Head emergence for IS was completed by 18 Apr 2006 (123 DAT). On the contrary, only 16% of GGI plants produced heads by 18 Apr 2006 (123 DAT) and eventually 58% of GGI plants did not produce heads by the end of a crop cycle in 2005-06 as measured at 158 DAT (Table 14). Higher irrigation rates significantly induced earliness regardless of cultivar, while N fertilizer rate did not show any effect on earliness (Table 14).

4.1.2.3 Plant growth and physiological response (third season)

In the third season 2007-08, leaf number was significantly affected by irrigation and N rate. Leaf number was significantly higher with increasing irrigation rate throughout the vegetative growth stage until 5 Mar (133 DAT), while plants under deficit irrigation had less leaf growth until the reproductive stage started (147 DAT) (Table 15). Increasing N rates to 180 kg·ha⁻¹ had only a slight improvement in leaf development when measured 88 DAT as compared to 10 kg·ha⁻¹ N, but in general N did not affect leaf number (Table 15). The partitioning of the significant irrigation × nitrogen interaction when measured 120 and 133 DAT indicates that the combination of 100% ET_c with any N rates showed similar leaf number, whereas deficit irrigation (50 and 75% ET_c) with differential N rates showed variable leaf number.

Plant size was also affected by irrigation and N rates (Table 16). Both plant width and height significantly increased with higher irrigation rates compared to 50% and 75%

ETc and this difference continued till reproductive stage (155 DAT) except for width (Table 16). Lower N rate of $10 \text{ kg}\cdot\text{ha}^{-1}$ significantly reduced plant size when measured 133 DAT, while $180 \text{ kg}\cdot\text{ha}^{-1}$ N slightly improved plant size. However, no significant difference was found among N rates at 155 DAT (Table 16).

Plant physiological responses to irrigation and N rates were evaluated with measurements of net photosynthetic rate (A_{CO_2}), transpiration (E) and stomatal conductance (g_s). Net photosynthetic rate (A_{CO_2}) was significantly reduced by deficit irrigation (50% ETc) compared to 75 and 100% ETc throughout the season. This difference was more evident as season progressed from 21 Mar to 2 May, 2008 (149 to 191 DAT). Nitrogen rates did not affect A_{CO_2} except 191 DAT when plants with time $180 \text{ kg}\cdot\text{ha}^{-1}$ N showed higher A_{CO_2} than other rates (Fig. 16).

Deficit irrigation (50% ETc) significantly reduced E compared to higher irrigation rates, and the difference was more pronounced as season progressed. However, transpiration was similar among N rates, except 191 DAT, time at which E was highest with $180 \text{ kg}\cdot\text{ha}^{-1}$ N (Fig. 16). Stomatal conductance (g_s) was clearly lower with deficit irrigation (50% ETc) compared to 75 and 100% ETc throughout the growing season. The difference was also pronounced as season progressed. Nitrogen fertilizer rates did not affect on stomatal conductance except 191 DAT (Fig. 16).

Table 14. Head emergence rate for artichoke in response to cultivar, irrigation and N rates, 2005-06.

Treatment	Head emergence rate (% of bolting plants)			
	Date (DAT ^x)			
	4 Apr (109)	10 Apr (115)	18 Apr (123)	23 May (158)
<u>Cultivar (C)</u>				
Green Globe Improved	4.0 b	10.7 b	16.1 b	42.0 b
Imperial Star	88.6 a	91.8 a	100 a	100 a
<u>Irrigation (I)</u>				
50% ETc	41.3 b	46.4 b	54.6	63.3 b
75% ETc	45.9 b	50.2 b	58.9	73.8 a
100% ETc	51.7 a	57.1 a	60.7	75.8 a
<u>Nitrogen (N)</u>				
	N.S	N.S	N.S	N.S
<u>Interaction</u>				
C × I	N.S	N.S	N.S	**
C × N	N.S	N.S	N.S	N.S
I × N	N.S	N.S	N.S	N.S
C × I × N	N.S	N.S	N.S	N.S

Cumulative temperatures [$\Sigma T(\text{day}^\circ\text{C})$] were 1716, 1853, 2053 and 2937 °C when measured 109, 115, 123 and 158 DAT, respectively. Total chilling hour [$\Sigma h(^\circ\text{C} \leq 10)$] on 109 DAT was 659 h, with the last chilling temperature ($^\circ\text{C} \leq 10$) observed on 26 Mar in 2006.

^xDAT: Days after transplanting.

Means within columns followed by different letters are significantly different (LSD, $p = 0.05$).

N.S: Not significant.

** $p < 0.01$.

Table 15. Leaf number of artichoke cv. Imperial Star in response to irrigation and N rates, 2007-08.

Treatment	Leaf number					
	<i>Date (DAT^x)</i>					
	10 Jan (78)	20 Jan (88)	7 Feb (106)	21 Feb (120)	5 Mar (133)	19 Mar (147)
<u>Irrigation (I)</u>						
50% ETc	4.2 c	5.5 b	10.0 c	13.0 c	17.1 b	19.9
75% ETc	4.6 b	5.7 a	10.6 b	13.7 b	17.9 a	19.9
100% ETc	5.1 a	6.4 a	11.1 a	14.4 a	18.3 a	19.9
<u>Nitrogen (N)</u>						
10 kg·ha ⁻¹	4.8	5.5 b	10.6 ab	13.3	17.4	20.1
60 kg·ha ⁻¹	4.7	6.0 a	10.5 ab	13.9	18.1	19.7
120 kg·ha ⁻¹	4.4	5.6 ab	10.2 b	13.4	17.4	20.1
180 kg·ha ⁻¹	4.6	6.3 a	11.0 a	14.2	18.3	19.8
<u>Interaction</u>						
I × N	N.S	N.S	N.S	**	**	N.S

^xDAT: Days after transplanting.

Means within columns followed by different letters are significantly different. (LSD, $p = 0.05$).

N.S: Not significant.

** $p < 0.01$.

Table 16. Plant width and height of artichoke cv. Imperial Star in response to irrigation and N rates, 2007-08.

Treatment	<i>Date (DAT^x)</i>					
	5 Mar (133)		18 Mar (146)		27 Mar (155)	
	Width	Height	Width	Height	Width	Height
<u>Irrigation (I)</u>						
50	160.5 c	37.9 c	177.0 c	47.1 c	172.2 b	66.3 c
75	177.6 b	46.1 b	193.7 b	67.1 b	192.6 a	89.5 b
100	184.4 a	50.0 a	207.9 a	74.2 a	196.5 a	95.1 a
<u>Nitrogen (N)</u>						
10 kg·ha ⁻¹	166.6 b	43.8	189.7 b	61.9	188.3	82.2
60 kg·ha ⁻¹	175.9 a	46.9	189.9 b	63.6	188.1	84.7
120 kg·ha ⁻¹	174.3 a	42.8	192.2 b	62.3	183.4	82.1
180 kg·ha ⁻¹	179.9 a	45.2	199.8 a	63.4	188.6	85.4
<u>Interaction</u>						
I × N	N.S	N.S	N.S	N.S	N.S	N.S

^xDAT: Days after transplanting.

Means within columns followed by different letters are significantly different. (LSD, $p = 0.05$).

N.S: Not statistically significant.

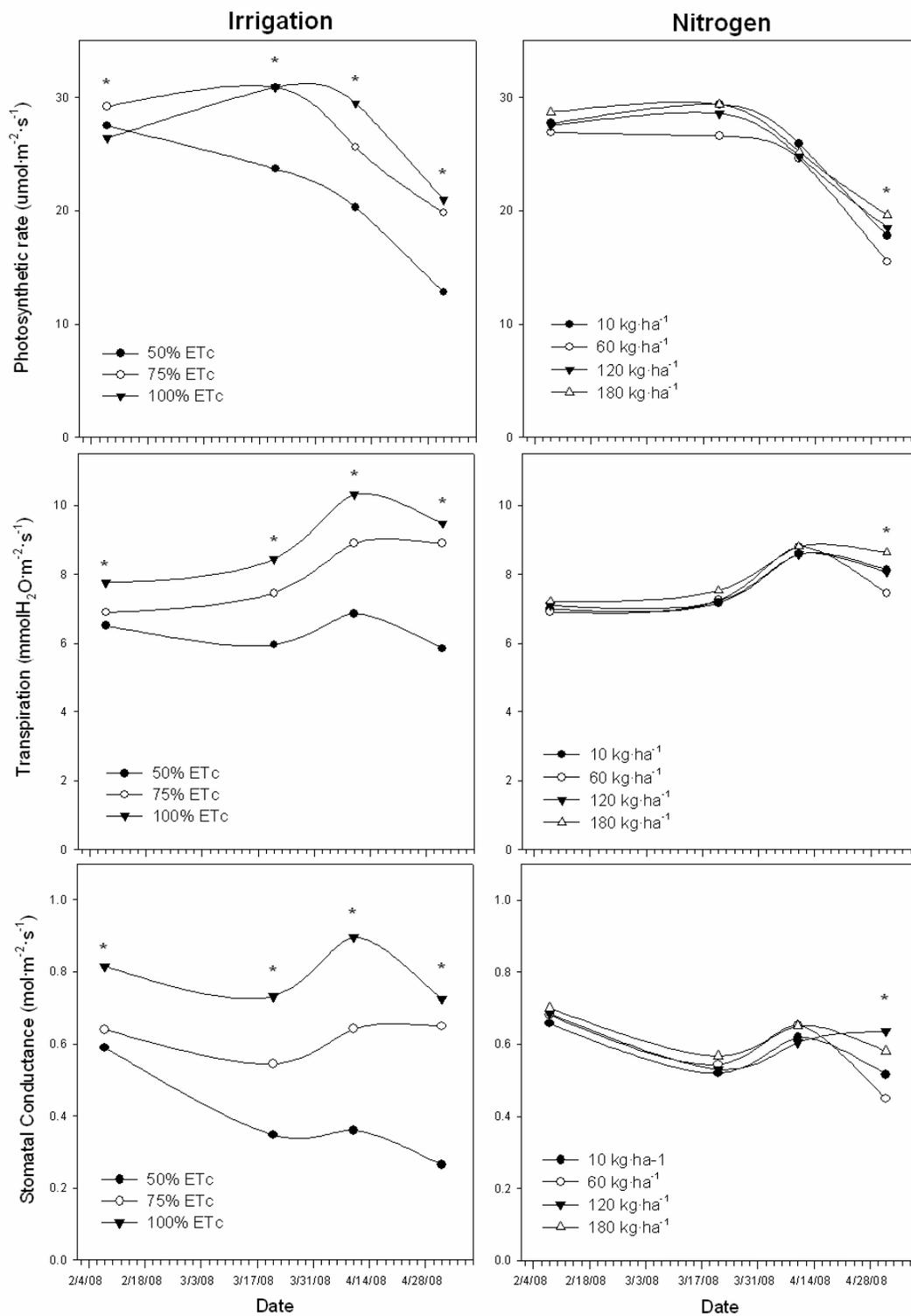


Figure 16. Plant physiological responses of artichoke cv. Imperial Star in response to irrigation (left) and N (right) rates, 2007-08. * $p < 0.01$.

4.1.2.4 Yield and yield components

In the 2005-07 seasons, marketable yields were significantly increased by 100% ETc (13.2 and 15.7 t/ha), whereas a 35 and 20% reduction in yield occurred under deficit irrigation (50% ETc) in the first (2005-06) and the second (2006-07) season, respectively. Nitrogen fertilizer did not significantly affect yield in both seasons (Table 17). The highest yields were achieved with the combination of 100% ETc and 60 kg·ha⁻¹ N (13.9 t·ha⁻¹) in the first season and 100% ETc and 120kg/ha N (17.6 t·ha⁻¹) in the second season. Higher irrigation rates significantly increased head number in both seasons, and only weight in the first season (Table 17).

Water use efficiency (WUE) was not affected by either irrigation or N rates. The higher values of WUE in the first compared to the second season were because of less water inputs due to a shorter crop cycle, 6 months in the first and 9 months in the second season) (Fig. 14 and Table 17). Nitrogen use efficiency (NUE) was significantly increased by 75% and 100% ETc in the first season, but not in the second season (Table 17). The significant irrigation × nitrogen interaction for NUE in the first season indicated that the combination of 100% ETc and 60 kg·ha⁻¹ N was more efficient for head

production than with 120 or 180 kg·ha⁻¹ N, or any other irrigation rates (Fig. 17).

In the third season (2007-08), marketable yield was significantly increased by 100% ETc (17.0 t·ha⁻¹), whereas a 30% yield reduction occurred under deficit irrigation (50% ETc) in 2007-08 (Table 18). Similarly, both head number and weight were significantly increased with higher irrigation rates as shown in the first season. However, yields were not affected by N rates (Table 18). The highest yield was achieved with the combination of 100% ETc and 60 kg·ha⁻¹ N (18.0 t·ha⁻¹).

Water use efficiency (WUE) was affected by irrigation rates, being significantly higher at 50 and 75% ETc than at 100% ETc, while N fertilizer did not affect WUE (Table 18). Nitrogen use efficiency (NUE) was significantly increased with higher irrigation rates (75% and 100% ETc). The significant irrigation × nitrogen interaction for NUE indicated that the combination of 100% ETc and 10 or 60 kg·ha⁻¹ N had higher NUE than 120 and 180 kg·ha⁻¹ N with 100% ETc, or 10 and 60 kg·ha⁻¹ N with any other irrigation rates (Table 18 and Fig. 14).

Table 17. Marketable yield, yield components, water use efficiency (WUE) and nitrogen use efficiency (NUE) of artichoke cv. Imperial Star in response to irrigation and N rates, 2005-07.

Treatment	2005-06					2006-07				
	Yield (kg·ha ⁻¹)	Head No.	Wt. (g)	WUE (kg·ha ⁻¹ ·mm ⁻¹)	NUE (kg·ha ⁻¹ ·kg ⁻¹)	Yield (kg·ha ⁻¹)	Head No.	Wt. (g)	WUE (kg·ha ⁻¹ ·mm ⁻¹)	NUE (kg·ha ⁻¹ ·kg ⁻¹)
Irrigation (I)										
50% ETc	8473 c	8.3 c	170.9 b	18.0	85.6 c	12572 b	10.2 b	236.9	15.6	132.7
75% ETc	11571 b	11.1 b	187.2 a	19.3	114.1 b	14311 b	11.6 b	240.2	15.5	144.5
100% ETc	13153 a	12.3 a	188.2 a	18.2	137.3 a	15696 a	12.9 a	246.4	15.1	158.1
Nitrogen (N)										
0 kg·ha ⁻¹	11305	10.8	181.1	19.0		12669	10.6	227.7	13.7	
60 kg·ha ⁻¹	11063	10.5	184.9	18.4	184.4 a	13655	10.9	246.8	14.9	225.1 a
120 kg·ha ⁻¹	11164	10.6	181.8	18.6	93.0 b	15722	12.8	244.0	17.1	131.0 b
180 kg·ha ⁻¹	10730	10.3	180.6	18.0	59.6 c	14728	12.0	246.0	16.0	79.2 c
Interaction										
I × N	NS	N.S	N.S	N.S	**	N.S	N.S	N.S	N.S	N.S

Means within columns followed by different letters are significantly different. (LSD, p = 0.05).

N.S: Not significant.

**p < 0.01

Table 18. Marketable yield, yield components, water use efficiency (WUE) and nitrogen use efficiency (NUE) of artichoke cv. Imperial Star in response to irrigation (I) and N rates, 2007-08.

	Yield (kg·ha ⁻¹)	Head		WUE (kg·ha ⁻¹ ·mm ⁻¹)	NUE (kg·ha ⁻¹ ·kg ⁻¹)
		No.	Wt. (g)		
<u>Irrigation (I)</u>					
50% ETc	11696 c	8.9 b	280.8 c	54.1 a	378.8 b
75% ETc	15704 b	11.7 a	301.3 b	54.9 a	488.9 a
100% ETc	16962 a	12.3 a	312.4 a	47.8 b	544.4 a
<u>Nitrogen (N)</u>					
10 kg·ha ⁻¹	14258	10.7	293.1	50.5	1425.8 a
60 kg·ha ⁻¹	15096	11.1	302.2	53.0	251.6 b
120 kg·ha ⁻¹	14331	10.7	294.0	50.9	119.4 c
180 kg·ha ⁻¹	15464	11.3	303.5	54.7	85.9 c
<u>Interaction</u>					
I × N	N.S	N.S	N.S	N.S	*

Means within columns followed by different letters are significantly different. (LSD, p = 0.05).

N.S: Not significant.

*p < 0.05

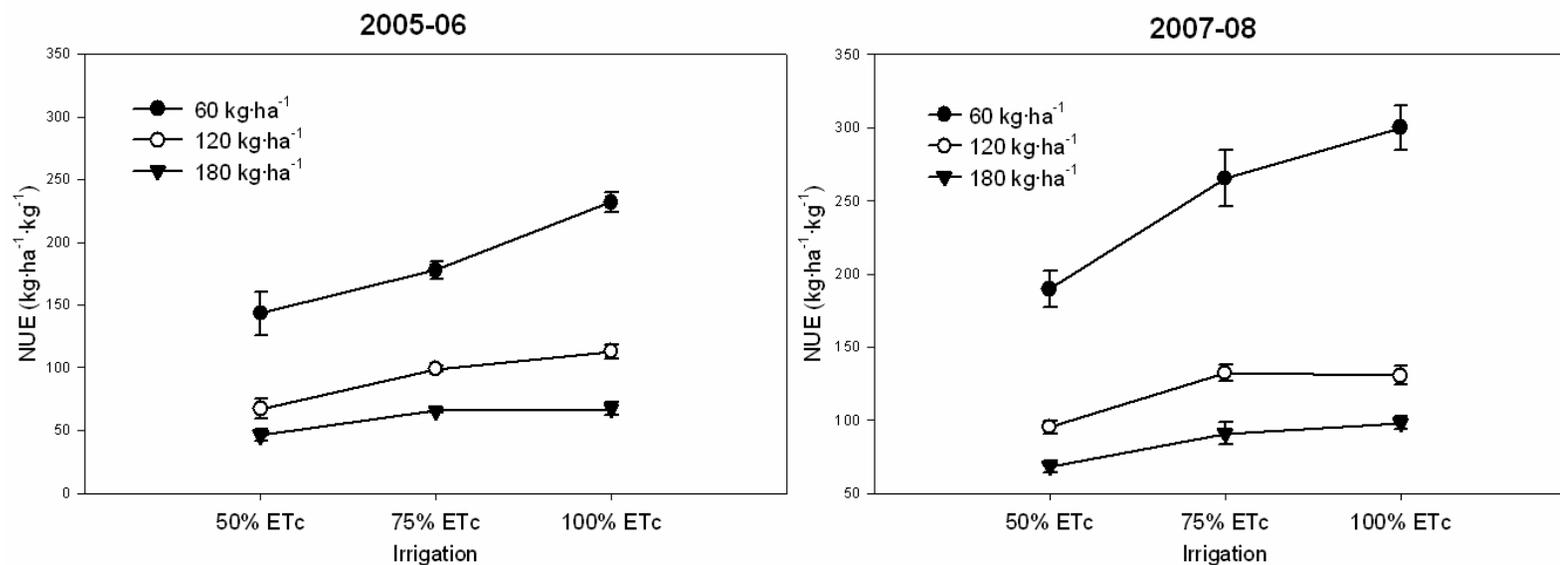


Figure 17. Nitrogen use efficiency (NUE, $\text{kg}\cdot\text{ha}^{-1}\cdot\text{kg}^{-1}$) in response to irrigation (ET_c) and N rates in 2005-06 (left) and 2007-08 (right). Vertical bars indicate mean \pm SE ($n = 4$)

4.1.2.5 Head size distribution

Artichoke heads for marketable yield were classified into size as small (<7 cm), medium (7-9 cm), large (9-11 cm) and jumbo (>11 cm) and those for unmarketable or cull heads with tip burn and open bracts. In the first season (2005-06), the distribution of head sizes were not affected by both irrigation and N rates at the early harvest; while a higher proportion of medium heads was obtained by 75% ETc compared with 50% ETc at late harvest. The proportion of open bracts on unmarketable heads was significantly lower at 75% ETc compared with 100% ETc at late harvest (Fig. 18). Tip burn was significantly increased by the combination of 50% ETc and higher N rates (120 and 180 kg-ha⁻¹ N) at late harvest (Fig. 18). In the second season (2006-07), the variability of head size was smaller than the first season and the majority of heads were of medium size ranging from 67 to 74% of total heads. Overall, irrigation and N rates did not affect the proportion for marketable heads, but a higher proportion of tip burn was obtained from 50 and 75% ETc at late harvest (Fig. 18).

In the second season (2007-08), the proportion for marketable heads was more clearly affected by irrigation rates. In the early harvest, a larger proportion of jumbo heads (27%) was obtained from 100% ETc, which resulted in a smaller proportion of large heads (73%) compared with 50% ETc (85%) (Fig. 19). A similar trend of head

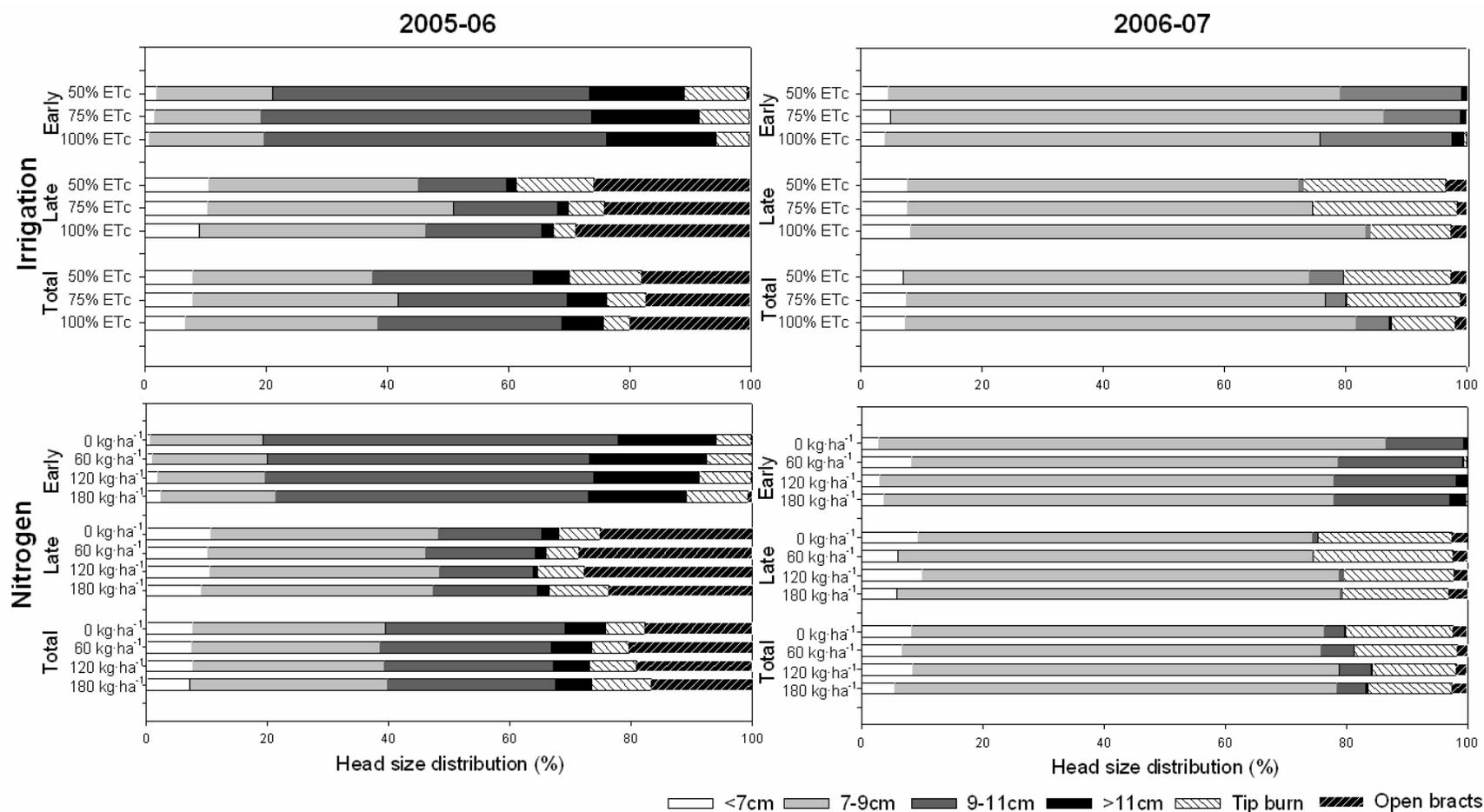


Figure 18. Head size distribution of artichoke cv. Imperial Star in response to irrigation (top) and nitrogen (bottom) rates in the first season (2005-06, left) and the second season (2006-07, right).

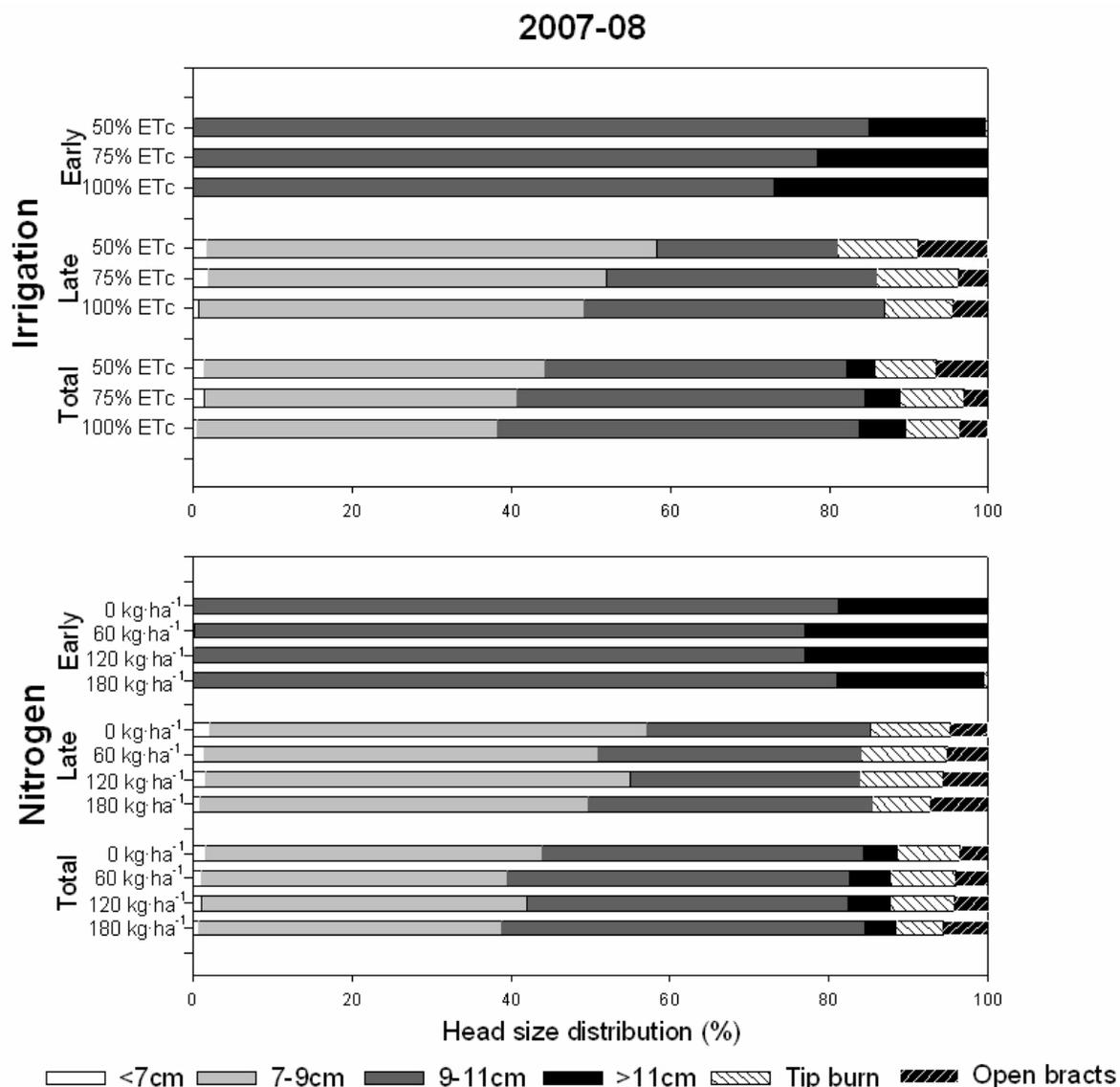


Figure 19. Head size distribution of artichoke cv. Imperial Star in response to irrigation (top) and nitrogen (bottom) rates in season, 2007-08.

distribution was measured in the late season, which had a higher proportion of large heads (38%), and corresponding smaller proportion of medium heads (48%) for 100% ETc compared with deficit irrigation (57%) (Fig. 19). Overall, N did not significantly affect head proportion for either marketable or unmarketable heads.

4.1.2.6 Soil nitrogen contents

Soil N content was evaluated each in 2006 and 2007 after harvest (end of a crop cycle). Preplant soil $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ were 3.7 and 77.4 ppm, respectively (Table 11). Irrigation did not significantly affect soil $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ when measured after the first season harvest, while both $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ were significantly affected by N fertilizer rates with the highest at $180 \text{ kg}\cdot\text{ha}^{-1}$ N (Table 19). The significant irrigation \times nitrogen interaction for $\text{NO}_3\text{-N}$ in the first season indicated that the combination of $180 \text{ kg}\cdot\text{ha}^{-1}$ N and higher irrigation rates (75% and 100% ETc) showed that a high concentration of $\text{NO}_3\text{-N}$ was present in the soil (0-13 cm depth) compared with other N rates in 75% and 100% ETc or any other N rates in 50% ETc (Fig. 20). In the second season, there were not significant differences among treatments, but there was an increasing trend for $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ content in response to N rates (Table 19).

Table 19. Soil nitrogen content after harvest for artichoke cv. Imperial Star in response to irrigation and N rates, 2005-07.

	2005-06		2006-07	
	NH ₄ -N (ppm)	NO ₃ -N (ppm)	NH ₄ -N (ppm)	NO ₃ -N (ppm)
<u>Irrigation (I)</u>				
50% ETc	3.5	171.1	5.6	34.7
75% ETc	9.3	335.4	9.2	35.2
100% ETc	7.3	304.4	6.0	40.7
<u>Nitrogen (N)</u>				
0 kg·ha ⁻¹	2.6 b	68.9 c	4.8	36.7
60 kg·ha ⁻¹	3.4 b	182.3 bc	6.3	37.7
120 kg·ha ⁻¹	4.4 b	280.1 b	6.9	38.6
180 kg·ha ⁻¹	16.4 a	549.9 a	9.8	34.6
<u>Interaction</u>				
I × N	N.S	*	N.S	N.S

Soil samples (0-13 cm depth) were collected on 27 Jul 2006 for the first season (2005-06) and 25 Jun 2007 for the second season (2006-07).

Preplant soil NH₄-N and NO₃-N were 3.7 and 77.4 ppm, respectively (2005).

Means within columns followed by different letters are significantly different. (LSD, p = 0.05).

N.S: Not significant.

*p < 0.05

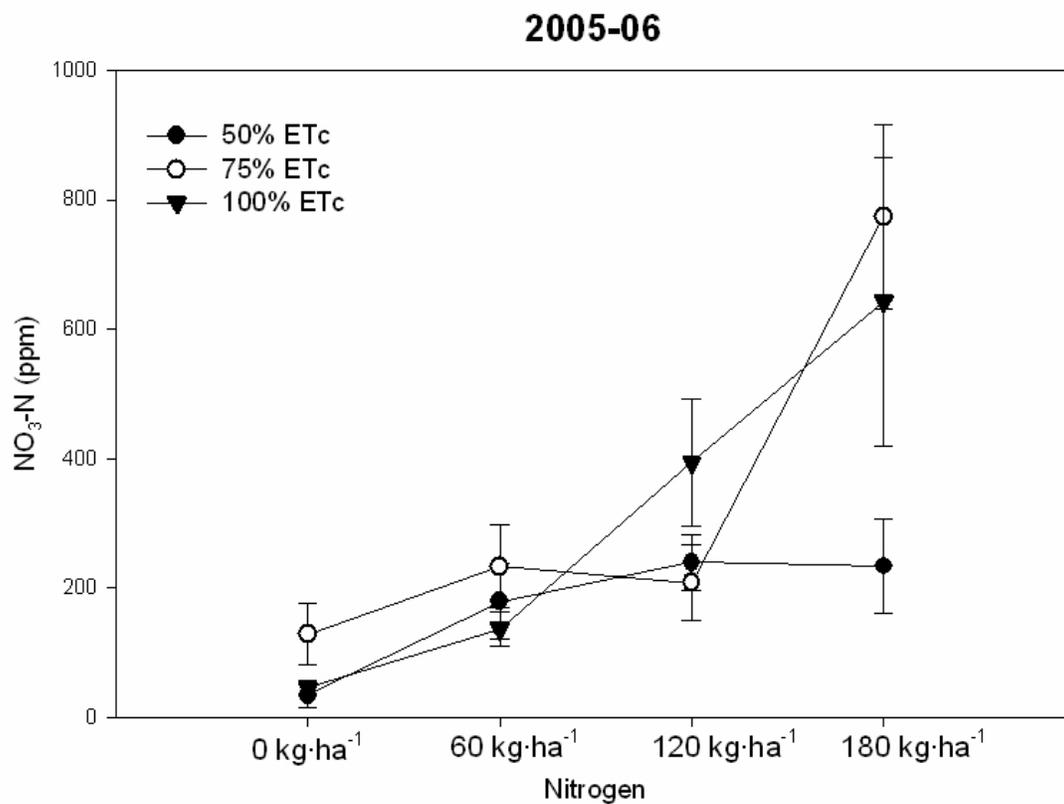


Figure 20. Soil NO₃-N content in response to irrigation (ETc) and nitrogen rates in the first season, 2005-06. Vertical bars indicate mean \pm SE (n = 4).

4.1.2.7 Chemical constituents in head

In general, total N content in heads in the first season (2005-06) was not affected by either irrigation or N rates, but it significantly decreased as harvest season progressed (Fig. 21). The lowest N content ($1.57 \text{ g} \cdot 100\text{g}^{-1} \text{ DW}$) was obtained from the combination of 100% ETc and $120 \text{ kg} \cdot \text{ha}^{-1} \text{ N}$ at late harvests. Also irrigation and N did not affect fiber content (NDF and ADF), but these were significantly decreased at late harvests (Fig. 22). The significant irrigation \times harvest season interaction for NDF indicated that 100% ETc at late harvest had lower NDF than other irrigation rates or than any other irrigation rates at early and mid harvests. Furthermore, total sugar content in heads was not affected by either irrigation or N rates and remained unchanged during harvesting season. The ratio of sucrose to monosaccharides in heads at each harvest was not affected by irrigation or N, but significantly increased as harvests progressed (Fig. 23). The largest ratio was obtained from the treatment combination of 50% ETc and $0 \text{ kg} \cdot \text{ha}^{-1} \text{ N}$ at late harvests.

Total phenolic content in heads was significantly increased up to 15% by deficit irrigation (50% ETc) compared with other irrigation rates, while it was not affected by N fertilizer when measured in the first season (2005-06) (Fig. 24). The significant irrigation \times harvest season interaction for phenolic content in the first season indicated that heads under deficit irrigation (50% ETc) harvested in late harvest contained higher phenolic content than under 75 and 100% ETc in the same late harvest or than any other harvest seasons (either early or middle). Phenolic content in late harvest, 2005-06 season, relatively decreased with higher N rate of 120 and $180 \text{ kg} \cdot \text{ha}^{-1} \text{ N}$ (Fig. 24). The

highest phenolic content ($30.0 \text{ mg}\cdot\text{g}^{-1} \text{ DW}$) was obtained from the combination of 50% ETc and $60 \text{ kg}\cdot\text{ha}^{-1} \text{ N}$ at late harvest in first season (2005-06). Similarly, in the second season (2006-07), no significant difference in phenolic content was obtained due to either irrigation or N rates in any of the three harvests, but it was significantly higher in late compared to early harvest (Fig. 24). Phenolic content in heads harvested in late harvest was approximately 30% higher than in early harvest.

Chlorogenic acid and cynarin are the main phenolic compounds present in artichoke heads. Chlorogenic acid in artichoke heads was significantly increased by deficit irrigation, but overall it was not affected by N rates (Fig. 25). Harvest season also significantly affected the chlorogenic acid content and increased as harvest season progressed (Fig. 25). The significant irrigation \times harvest season interaction indicated that this phenolic compound was maximized ($13 \text{ mg}\cdot\text{g}^{-1} \text{ DW}$) with deficit irrigation (50% ETc) at late harvest as compared with 75 or 100% ETc at late harvest. The trend for cynarin content in artichoke heads was relatively similar to that of chlorogenic acid content, though no significant difference existed among different irrigation, N and harvest season (Fig. 25).

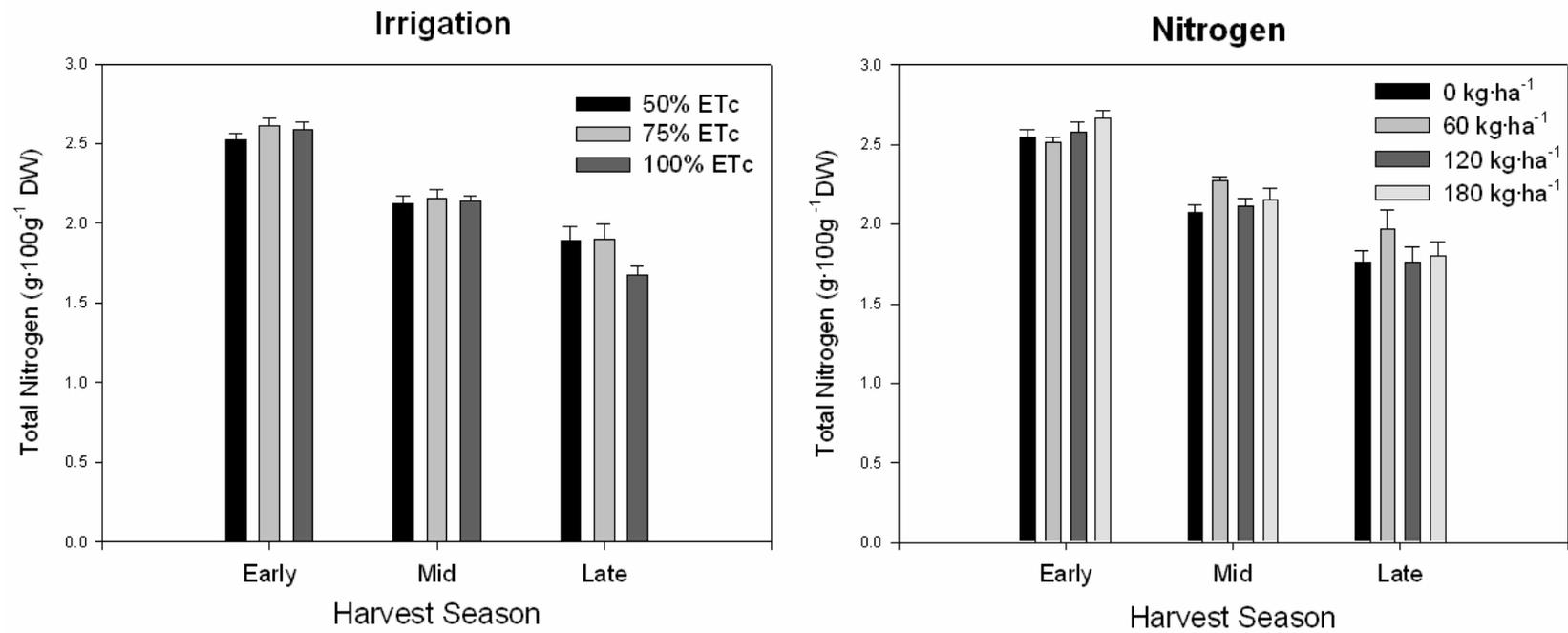


Figure 21. Total nitrogen content in artichoke head cv. Imperial Star in response to irrigation (left) and N (right) rates, 2005-06. Vertical bars indicate mean \pm SE (n = 11-16).

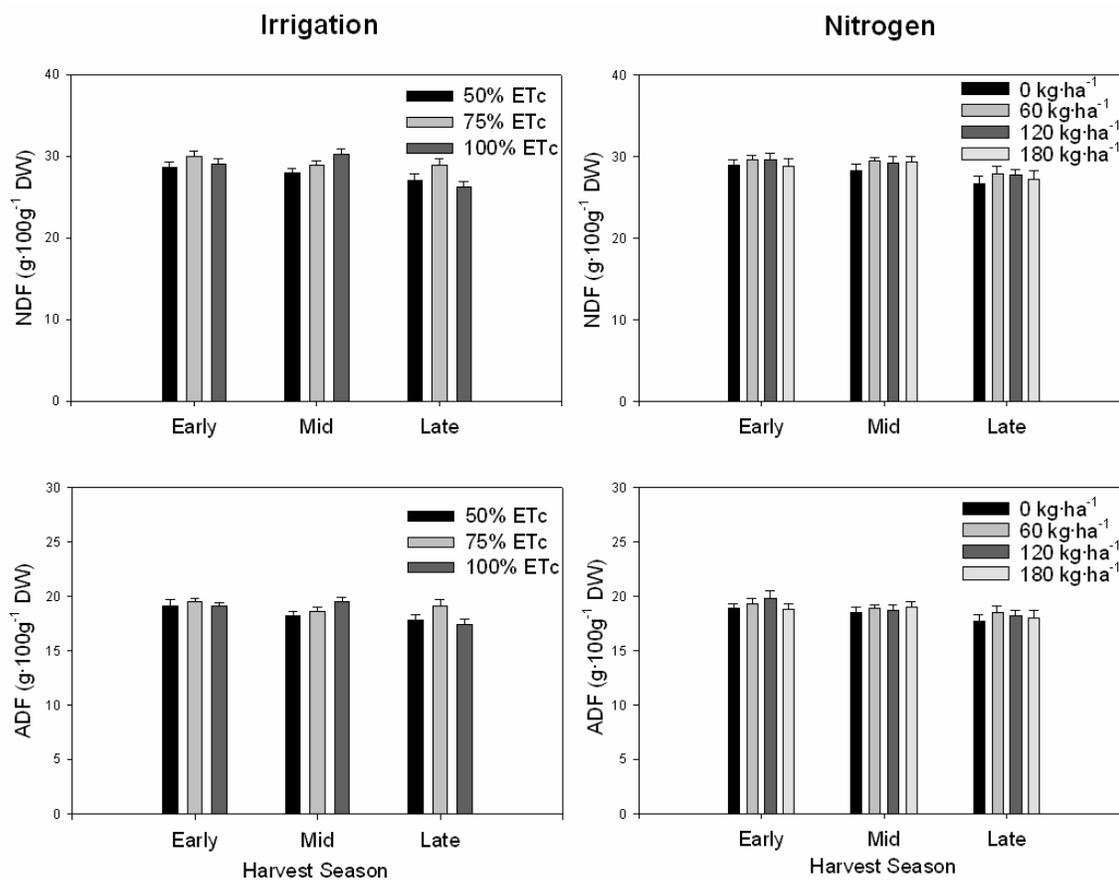


Figure 22. Neutral detergent fiber (NDF, top) and acid detergent fiber (ADF, bottom) in artichoke head cv. Imperial Star in response to irrigation (left) and N (right) rates, 2005-06. Vertical bars indicate mean \pm SE (n = 12-16).

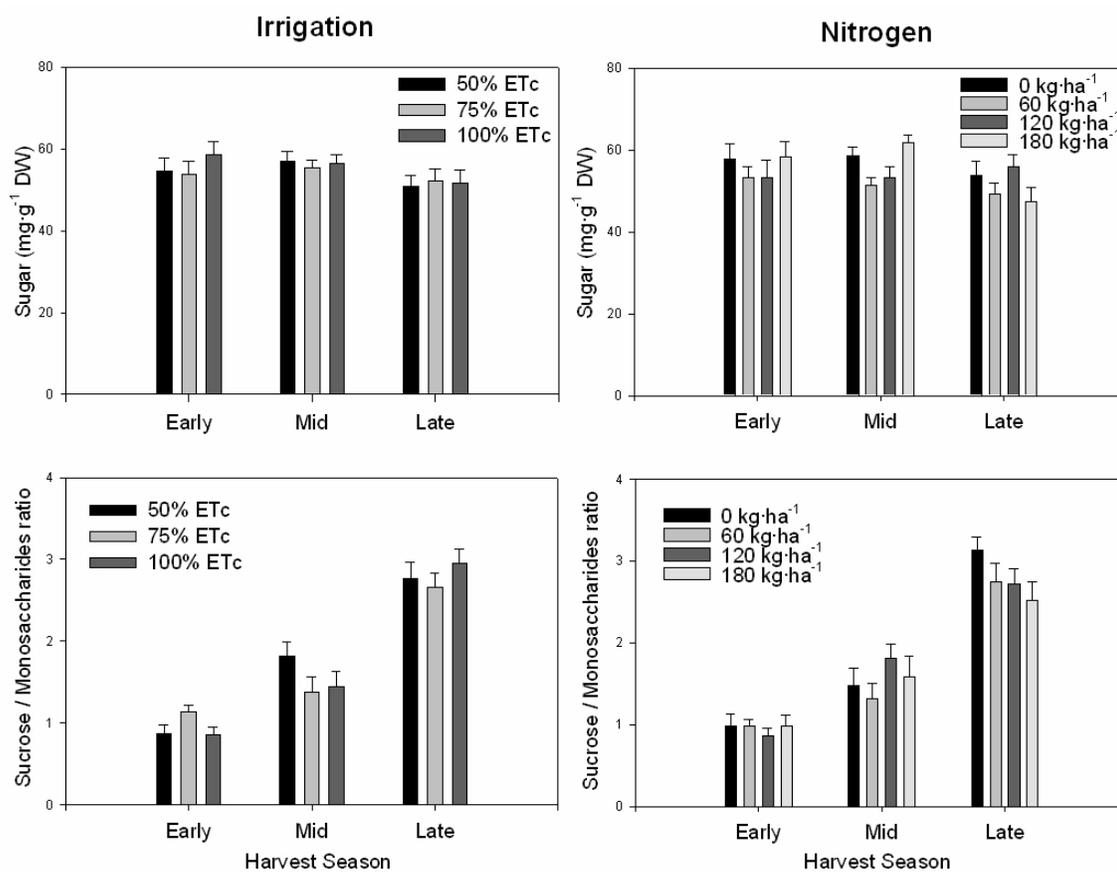


Figure 23. Sugar (glucose, fructose and sucrose) content (top) and the ratio of sucrose to monosaccharides (bottom) in artichoke head cv. Imperial Star in response to irrigation (left) and N (right) rates, 2005-06. Vertical bars indicate mean \pm SE (n = 10-16).

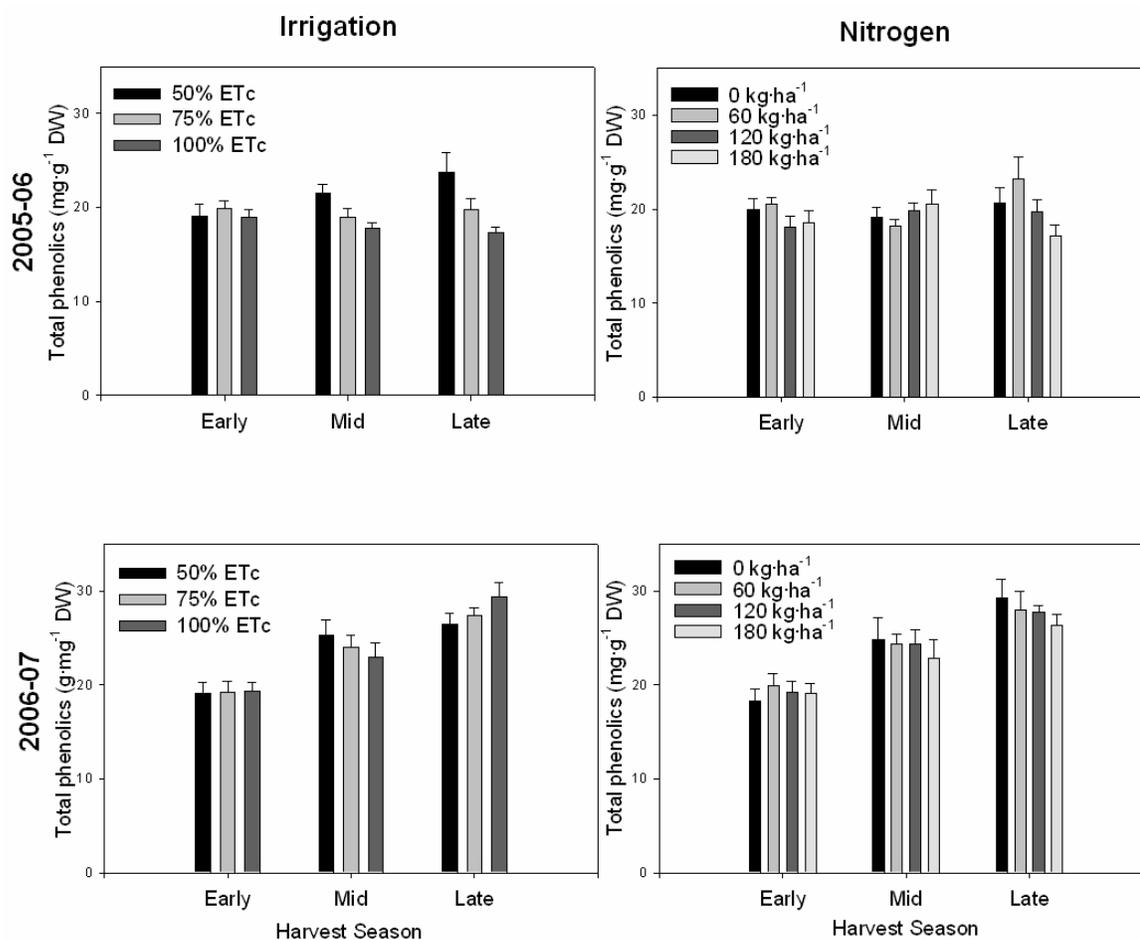


Figure 24. Total phenolic content in artichoke head cv. Imperial Star in response to irrigation (left) and N (right) rates in the first season (2005-06, top) and the second season (2006-07, bottom). Vertical bars indicate mean \pm SE ($n = 11-16$ for first season and $n = 6-14$ for second season).

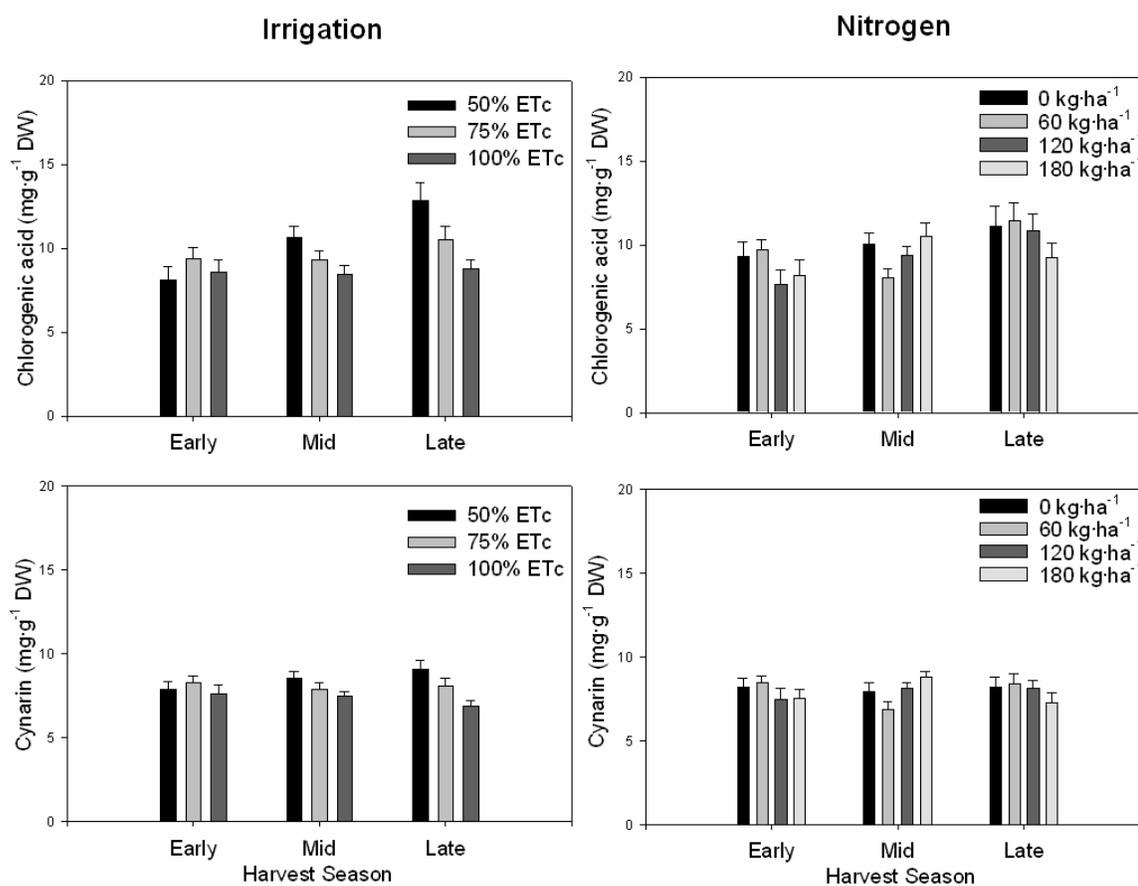


Figure 25. Chlorogenic acid (top) and cynarin content (bottom) in artichoke head cv. Imperial Star in response to irrigation (left) and N (right) rates, 2005-06. Vertical bars indicate mean \pm SE (n = 10-16).

4.1.3 Discussion

Previous studies confirmed the feasibility of artichoke production with high yield and quality during spring in the southwest Texas (Leskovar et al., 2007). To introduce artichoke production into commercial production in this region, it is important to develop field management strategies. In this field study, yield, yield components and the nutritional quality of artichoke in response to three irrigation and four N fertilizer rates were examined over two seasons (2005-07).

The average winter temperatures were 14.0 °C for the first season (2005-06) and 13.4 °C for second season (2006-07), which were within the range for optimum temperatures (7-29 °C) for artichoke growth (Schrader and Mayberry, 1997). In the first season, head emergence (bolting) successfully occurred for cv. Imperial Star (IS), but not for Green Globe Improved (GGI). Head emergence of IS was completed by 123 days after transplanting (DAT) with 659 hours of chilling (≤ 10 °C), while only 42% of GGI plants produced heads (Table 14). This result confirmed that delaying planting of late maturing cultivars more likely will reduce head emergence, leading to a serious yield reduction (Leskovar et al., 2007). This suggests that for late plantings it could be necessary to vernalize plants artificially by the application of gibberellic acid to insure the complete bolting (Schrader and Mayberry, 1997) when cultivars like GGI are used in southwest Texas region. The higher head emergence rate with 100% ETc rate observed in the first season may be partially related to improved vegetative growth (biomass accumulation) and advanced plant maturity as shown in third season (Tables 14 and 15).

Marketable yields were significantly increased by 100% ETc, while a 35% to 20% of yield reduction occurred at 50% ETc in the first and the second season, respectively (Table 17). High irrigation rate (100% ETc) increased head number in both seasons with greater weight of heads in the first season (Table 17). Consistent with the positive effect on the number and weight of heads, total yield increased with higher irrigation rates. Yield reduction, 12% in the first and 20% in the second season, were associated with lower head number and weight as also shown with 75% ETc. Our results agree with those of Saleh (2003) and Garnica et al. (2004), who also reported deficit irrigation decreased both head number and weight, while others reported only head number was affected (Macua et al., 2005; Pomares et al., 2004). Conversely, N fertilizer did not show any yield improvement in both seasons. Elia and Conversa (2007) recently reviewed the impact of N on artichoke earliness, yield and yield components. They highlighted that these responses did not follow a linear trend. Other studies showed significant impacts on earliness with N up to 200 kg·ha⁻¹ (Foti et al., 2005), yield with 200-300 kg·ha⁻¹ N (Ierna et al., 2006; Pomares et al., 1993), or head number and weight with N up to 200 kg·ha⁻¹ (Paradiso et al., 2007). However, no effect of N on yield was found in the range of 0-270 kg·ha⁻¹ N (Pomeras et al., 2004), 0-400 kg·ha⁻¹ N (Foti et al., 2005) and 100-215 kg·ha⁻¹ N (Saccardo et al., 2005). Our results indicate that acceptable head yields can be obtained with 120 kg·ha⁻¹ N; however, higher NUE was achieved with the combination of 60 kg·ha⁻¹ N and 100% ETc for both years (Tables 17 and Fig. 17).

Overall, size distribution of marketable heads was similar among treatments, and in general head size decreased as harvesting season progressed (Fig. 18). In the second season, there was less variability in head sizes than in the first season, with the majority of heads classified as medium size (Fig. 18). This difference in head size distribution between two seasons can be explained by the establishment methods used, transplants in the first season vs. offshoots in the second season. For transplants, a large main or crown head was developed from the main stem, following by 7-11 middle to small heads produced from lateral branches, whereas for offshoots (3-5 offshoots coming from a crown) only 2-4 medium and small axillary heads were produced after the completion of the large main head development.

Unmarketable heads or culls (open bracts and tip burn) increased as harvest season progressed in both season (Fig. 18). Significantly higher proportion of tip burn (or atrophy) was observed for deficit irrigation rates or by the combination of deficit irrigation with higher N rates (120-180 kg·ha⁻¹) (Fig. 18). It was reported that several environmental conditions can influence this physiological head disorder. These include lack of soil P, a high K/P ratio, high salinity, Ca deficiency, and/or high temperatures (Elia and Conversa, 2007; Francois et al., 1991). In addition to those environmental conditions, our results suggest that plants under either drought stress or drought combined with high N conditions may also induce head tip burn especially in the late harvesting season. Indeed, Dr. M. Miller (Weslaco AgriLife Extension) identified gray mold (*Botrytis cinerea*) on blackish parts of artichoke heads collected from our field. Since this fungus can develop via an infection from weaken or wound tissues, we may

also need to consider plant senescence during late season and/or physical stresses through handling at harvest or transport to cooling storage.

The large values of WUE for the first vs. the second season can be attributed to the longer crop cycle and thus higher irrigation frequency in the second season (28 July 2006 – 24 Apr 2007) compared with the shorter first season (15 Dec 2005 – 25 May 2006) (Fig. 14 and Table 12). From a viewpoint of water savings, the annual cycle would be a more cost profitable system than the biennial system, though relatively higher yield can be expected from the second crop season if transplanting is done late in the fall (e.g. Dec). In addition, weed and pest control issues will increase in the second year, and therefore growers may incur additional management costs (Mauromicale and Ierna, 1995). In fact, we lost a significant number of plants during the summer, between the first and the second season, by infection of cotton root rot (*Phymatotrichum omnivorum*).

Considering soil N, a relatively high concentration of available N ($\text{NO}_3\text{-N}$) at the soil surface (0-13 cm) was present in the field (77.4 ppm) before planting in 2005 (Table 11). After the first season, the post-harvest residual soil $\text{NO}_3\text{-N}$ was significantly higher at higher N rates under higher irrigation rates (75-100% ETc) (Table 19 and Fig. 20), but no difference was observed after the second season (Table 19). The lack of difference in the second season may be due to the long crop cycle (6 months vs. 9 months) and/or increased in the N leaching that may have occurred due to large amount of precipitations during Mar to Apr (29.6 mm vs. 83.0 mm). The significant irrigation (75 and 100% ETc) \times nitrogen ($180 \text{ kg}\cdot\text{ha}^{-1} \text{ N}$) interaction in the first season demonstrated that higher rates

of side-dressed granule N was dispersed well in the soil surface with higher irrigation rates under the subsurface drip system (Fig. 20). Although this may be beneficial for certain shallow rooted vegetables such as onion (Halvorson et al., 2008), our results indicated that artichoke did not respond well when high amounts of available N ($\text{NO}_3\text{-N}$) were presented in soil surface. Presumably, active nutrient uptake may be greater at deeper soil layers, where the majority of large roots (taproot and lateral roots) may be present. In fact, the rationale of very low fertilizer N use of field grown tomato in California was explained by the poor root system in N-rich soil surfaces (Jackson and Bloom, 1990). Further investigations on root growth dynamics is required to reveal the nutrient uptake characteristics of artichoke.

The nutritional quality of artichoke heads was clearly affected by harvest season, while both irrigation and N were less effective. In general, total N and fiber (NDF and ADF) content in heads were decreased as harvest season progressed (Figs. 21 and 22). According to Rincon et al. (2007), the rate of shoot N uptake and the artichoke head N concentration were smaller during the late than the early season. Therefore, it seems reasonable to assume that the decrease of N in heads is primarily related to plant aging processes. Since it has been shown that the reduction of fiber content occurs in many fruits during maturity (Larrauri et al. 1997; Eaks and Sinclair, 1980), fiber content in artichoke heads may also be degraded throughout the harvest season as plants mature and approach senescence. Total sugar content in heads was similar over the season, while their composition (the ratio of sucrose to monosaccharide) increased as harvest season progressed (Fig. 23). The compositional changes could be accompanied by a

decrease of invertase activity and an increase of sucrose metabolizing enzymes over the plant maturation process as shown in muskmelon fruit (Lingle and Dunlap, 1987).

In the first season, total phenolic and chlorogenic acid content in heads significantly increased by deficit irrigation (50% ETc), while it was not affected by N rates (Figs. 24 and 25). The significant effect of deficit irrigation was pronounced as harvesting season progressed (Figs. 24 and 25). In the second season, total phenolic content increased with harvest seasons, but irrigation and N rates were not as effective as harvest seasons (Fig. 24). Wang et al. (2003) also reported that higher amount of phenolics in artichoke mature heads when harvested on Oct than when harvested on Sept. We assume that phenolic content in artichoke heads may increase with plant maturity, as also shown in other vegetables and herbs (Ahamad et al., 2005; Deepa et al., 2007; Pandjaitan et al., 2005). Indeed, it has been proposed that plant environmental stresses such as cold, heat, water-deficit, and/or flooding exert a considerable influence on the levels of secondary metabolites (e.g., polyphenolics and phenylpropanoids) in plants (Dixon and Paiva, 1995, Kirakosyan et al., 2004). Therefore the increase in phenolic content of artichoke heads by deficit irrigation may be partly ascribed to plant defense mechanism against drought stress as shown in other vegetable (English-Loeb et al., 1997). The lack of drought stress effect on total phenolics in the second season might be due to high precipitation during harvest, which may have mitigated drought stress. Even though there are findings indicating that higher N application inversely affected phenolic content in artichoke leaves (Eich et al., 2005), our results showed no clear effect of N application on artichoke heads. It is also known that phenolic content in

artichoke heads can also change with cultivar, growing season, head maturity, storage, and processing (Curadi et al., 2005; Gil-Izquierdo et al., 2001; Llorach et al., 2002; Di Venere et al., 2004; Wang et al., 2003).

A follow up field study was conducted in the third season (2007-08) to confirm the impacts of irrigation and N fertilizer on artichoke as observed in the previous seasons. Plant growth and physiological responses were also examined to find the relationship of vegetative growth and reproductive growth (yield) in response to irrigation and N rates. Some management practices were adjusted based on previous results (planting date, N application method, planting distance, plastic mulch application and Ca-Zn micronutrient foliar amendment). All those modifications improved the overall plant performance and head quality.

Plant vegetative growth, leaf number and plant size (width and length), was significantly improved by higher irrigation rates, while only a slight improvement was obtained from 180 kg-ha⁻¹ N rate (Tables 15 and 16). These results confirmed that the higher irrigation rates might enhance earliness of artichoke, which was observed in first season (Table 14), mainly by improving plant size as well as maturity.

Plant physiological responses (A_{CO_2} , E , and g_s) were significantly different among irrigation rates, but all indices were well correlated, though the seasonal plant response was quite similar in general (Fig. 16). The higher irrigation rates significantly increased A_{CO_2} , E and g_s throughout a season, but the difference between 50% ETc and other rates (75% and 100% ETc) was more obvious as season progressed (Fig. 16). When plants are exposed to drought stress, stomata close progressively, decreasing g_s ,

with a parallel reduction in both E and A_{CO_2} (Tezara et al., 1999). The physiological responses to differential irrigation rates in our study indicate that artichoke plants subjected to deficit irrigation (50% ETc) exhibited a significant drought stress throughout the season, thus reducing vegetative growth and lowering yields as evident in the 50% ETc regime. Our results showed no significant effect of N fertilizer rates on these physiological responses (Figure 16).

Marketable yields were significantly increased by 100% ETc, while a 7% and 30% of yield reduction occurred at 75 % and 50% ETc, respectively (Table 18). As we observed in the first season, total yield at 100% ETc was consistent with the increase on both number and weight of heads. The proportion for marketable heads was similar to the first season, but showed higher proportion of larger heads at 100% ETc. Although our intention was not to compare seasons, certainly plastic mulch application can significantly increase WUE compared to bare soil as measured in the first and the second season (Tables 17 and 18). The total water input was 355 mm, including 38 mm of rainfall, for the 100% ETc rate (Table 13). Overall, plastic mulch application in the third season effectively saved 52% and 58% of irrigation water when simply compared 100% ETc rates with the first and the second season, respectively. Therefore, plasticulture system appears a promising management tool to improve crop performance and water saving in artichoke.

Averaged winter temperature (14.3 °C) in the third season was similar with other two seasons (Fig. 15). Although freezing temperatures (more than 5 h of continuous ≤ 0 °C) on 16, 23, 24, and 25 Dec 2007, and 3, and 8 Jan 2008 caused leaf injury, drying

mostly older leaves but without affecting younger leaves, all plants recovered soon after freezes were over. It may be possible that periodical foliar amendments of Ca and Zn to artichoke during the growing season might have improved plant tolerance to freezing by enhancing cold hardiness. The protective effect of this amendment was also recognized in orchard trees like apple and pear (Raese et al., 1996).

In conclusion, our results indicate that irrigation was more effective than nitrogen rates to optimize artichoke yield. Yield reduction was associated primarily with a decrease in head number and weight. Considering the three seasons of the study, threshold level of ≥ 700 mm of water inputs and $120 \text{ kg}\cdot\text{ha}^{-1}$ of N appears sufficiently enough to obtain high marketable yields for artichoke under our environmental conditions. However, growers need to check soil available N before planting so that rates can be adjusted. This study clearly shows that there are significant irrigation, season, and irrigation \times season effects on artichoke quality. Evidently, it is possible to enhance phenolic contents in artichoke heads by deficit irrigation, but at the cost of significant yield losses (average of 29% or 9% by 50% or 75% ETc, respectively). Based on our results and to achieve optimum yield and obtain valuable quality of artichoke heads, we recommend the annual production system with transplants established in Oct and total water inputs of ≥ 700 mm for the southwest Texas region.

CHAPTER V

SUMMARY

5.1 Transplant Management

In southwest Texas, simultaneous high air temperatures and drought episodes are very common during late summer and fall, time when artichoke seedlings are to be transplanted in the field. Therefore, high transplant quality is an important factor for stand establishment and subsequent growth of artichokes.

Heat (35/20°C, day/night temperature regimes) or drought stresses (30 % WHC) alone or in combination significantly reduced shoot or/and root growth of artichoke seedlings by lowering photosynthetic rate (A_{CO_2}) and shoot water status. The detrimental effects of those stresses combined severely affected seedling performance, primarily root growth and shoot water status. The results of this study imply that in order to prevent transplant shock, it would be necessary to enhance early root growth of seedlings while to preventing leaf dehydration.

The ethylene precursor ACC and the releasing compound ETH (1-100 $\mu\text{M/L}$) significantly enhanced root hair, lateral root formation (only 30 $\mu\text{M/L}$ of ETH), and total scanned root area. These results indicate the potential use of exogenous ethylene regulators as root enhancers. The increase in early root growth by ACC or ETH may improve the seedling ability of higher water and nutrient uptake. However, further investigations addressing the physiological activity of ethylene-induced roots will be required before any practical use can be conducted. Other reports indicate that root hair

and lateral root (vs. tap root) development play an important role for acquisition of soil nutrients especially in unfavorable field conditions (Drew and Saker, 1975; Jungk, 2001).

A foliar application of ABA (1000 mg/L) effectively enhanced drought tolerance of artichoke transplants maintaining a high leaf water status via a rapid stomatal closure response. These results suggest that exogenous ABA could be a useful plant growth regulator to improve hardiness of artichoke transplants and to condition transplants to withstand temporal drought stress conditions, thus reducing transplant shock. Improvement of stand establishment is a pre-requisite for a rapid resumption of growth and to optimize final yield potential of artichoke grown in semi-arid conditions, such as those encountered in southwest Texas.

5.2 Field Management

To introduce artichoke production into the commercial production in southwest Texas, it is important to develop field management strategies including planting season, irrigation, N fertilization rates, as well as selecting optimum establishment systems (e.g., transplants, plasticulture).

Vegetative growth, yield, yield components and nutritional quality of artichoke in response to different irrigation (50, 75 and 100 % ETc) and N fertilizer (0-180 kg/ha) rates under subsurface drip irrigation system (SDI) were examined in experimental fields in three seasons (2005-06, 2006-07 and 2007-08). Our results indicate that irrigation

rate management was more effective than nitrogen fertilizer rate to optimize artichoke yield. Yield reduction was associated primarily with a decrease in head number and weight. The highest yield was obtained from 100 % ETc with 60-120 kg·ha⁻¹ of N fertilizer. This study also shows that there are significant irrigation, season, and irrigation × season effects on artichoke nutritional quality. Evidently, it is possible to enhance phenolic contents in artichoke heads by deficit irrigation, though deficit irrigation may significantly reduce the total yield by a maximum of 35%. Based on our results from three seasons, we would recommend an annual production system by early transplanting (Oct) with sufficient irrigation (≥ 700 mm) and plastic mulch application to achieve optimum yield and quality of artichoke heads.

In addition to the information of irrigation and N rate management developed from this study, other techniques applied (e.g., plasticulture, macro-micro nutrients, pest control) might be useful information for establishing artichoke production systems with future growers in southwest Texas.

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