The impact of substrate and irrigation interval on the post-transplant root growth of container-grown zinnia and tomato

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Abstract

Substrate type and irrigation interval were studied to determine their impact on the post-transplant root growth of ‘Thumbelina’ zinnia (Zinnia elegans Jacq.) and ‘Celebrity Hybrid’ tomato (Lycopersicon esculentum Mill.). Seeds of both species were planted in 80 cm\(^3\) (2.7 fl oz) plug cells containing either Metromix 360\textsuperscript{TM} (MM360) or Ball Professional Growing Mix\textsuperscript{TM} (BPGM) and, following germination, the seedlings were transplanted into 450 cm\(^3\) (27.5 in\(^3\)) plastic pots containing the same substrate. Evapotranspiration (ET\(_{0}\)) was measured gravimetrically each day and the water lost via ET\(_{0}\) added back to the substrate at intervals of 24, 48 or 96 hr. For zinnia, root growth was consistently better for seedlings grown in BPGM, a substrate with greater water holding capacity and air-filled porosity. For plants grown in BPGM and irrigated every 48 hr, root dry weight was significantly greater than it was for any of the remaining treatments. For tomato, root growth was greater for seedlings grown in BPGM and for transplants irrigated at 96 hr intervals; but, unlike zinnia, no significant interactions between substrate type and irrigation interval were observed. The results of this study show that root growth of plug-grown transplants can be improved by selecting a substrate with high porosity that allows for optimum oxygen and water exchange, and by extending the irrigation cycle to 48 hr (zinnia) or 96 hr (tomato).

Index words: irrigation scheduling, moisture stress, plant establishment, soilless growing media, transplant production.

Species used in this study: ‘Thumbelina’ zinnia (Zinnia elegans Jacq.); ‘Celebrity Hybrid’ (Lycopersicon esculentum Mill.).

Significance to the Horticultural Industry

The current trend in ornamental and vegetable transplant production is to grow the plants in plug-cell trays. In an effort to improve irrigation management practices for the production of container-grown plug-cell transplants, we evaluated the effect of substrate type and irrigation frequency on the root growth [root dry weight (RDW), specific root length (SRL), root tissue density (RTD) and root length density (RLD)] of zinnia and tomato. The data show that, for soilless cultivation of zinnia and tomato, root growth can be increased by seeding in a high porosity substrate that allows good air and moisture exchange. In our study, plug-cell transplants grown in BPGM and re-watered at intervals of two (zinnia) or four (tomato) days showed significantly more root growth than similar transplants watered daily. Since the goal of irrigation scheduling is to maintain adequate substrate moisture while maximizing plant-water use efficiency, the choice of an appropriate growing medium possessing good moisture release characteristics is one of the most important decisions a grower can make. With proper substrate selection, irrigation scheduling options become more manageable, resulting in better root growth and greater transplant success.

Introduction

While greenhouse, nursery and floriculture production contributes in excess of $20 B to the U.S. gross domestic product (Hodges et al. 2015), industry growth has slowed in recent years and, in some parts of the industry, has actually declined (Hall 2010). To reduce costs and increase efficiency, horticultural crop production has shifted toward the use of container-grown plants. It is estimated that in the U.S., about 90% of all horticultural plants grown in the nursery, greenhouse and floriculture industries is now produced in containers (U.S. Dept. Agric. 2007). Containerization allows for increased plant density per unit growing area, offering the potential for greater economic return. This is especially true in bedding plant production, where the trend has been to grow seedlings in small plug-cells where the root system remains intact during transplanting, thereby reducing transplant shock and shortening the time required for root establishment (Biembaum and Versluis 1998).

Plug-cell production, while increasing the efficiency of growing a crop, has complicated the issue of plant-water management. The decreased size of the root zone in plug-cells, ranging from 2 cm\(^3\) to 25 cm\(^3\) (0.12 to 1.53 in\(^3\)), requires the application of very precise amounts of

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irrigation water. After transplanting, and during the period of plant establishment, substrate moisture levels must be kept sufficiently high to encourage root growth while, at the same time, avoiding media saturation and low oxygen levels in the root zone. The balance between too much and not enough substrate moisture is often difficult to maintain since the water-absorbing capacity of transplanted root systems is often reduced (transplant shock) while, at the same time, the young transplants have insufficient new root growth to compensate for the amount of water lost via transpiration. Consequently, two important considerations in successful plug-cell transplant production have evolved: (1) selecting a growing medium with the physical characteristics necessary to provide adequate quantities of water and oxygen, and (2) determining the correct frequency, or interval, between successive watering events to ensure that substrate moisture is maintained within a range sufficient to support new root growth and development.

Published information on irrigation interval is available for a number of field-grown crops, including wheat [Triticum aestivum L. (Bstawi et al. 2011)], maize [Zea mays L. (El-Halim et al. 2014)], sugarcane [var. of Saccharum officinarum L. (Ethan et al. 2013)] and tomato (Capsicum annuum L.), Ismail and Ozawa (2009) reported growth of potted pepper (Capsicum annuum L.), Imsam and Ozawa (2009) reported increased stress tolerance by causing more roots to develop (those grown in containers. Using container-grown pepper (Capsicum annuum L.), Imsam and Ozawa (2009) reported that increasing the time interval between irrigations increased stress tolerance by causing more roots to develop at the base of the container where moisture levels were higher. Kim et al. (2015) found that both substrate type and irrigation frequency were important in determining the shoot growth of potted Leucospermum species. While there are an abundance of studies dealing with the performance of nursery crops grown in different media (Allaire et al. 1996), little has been done to study the effects of irrigation frequency on root growth and root system architecture (Blom et al. 2008). As a result, the current study was undertaken to investigate the impact of changes in both substrate composition and irrigation frequency on post-transplant root growth of two important horticultural species, zinnia and tomato.

Materials and Methods

Substrate physical properties. Prior to the start of the investigation, the physical properties of the two commercial soilless substrates used in this investigation [Metromix 360® (MM360; Sun Gro Horticulture, Agawan, MA), and Ball Professional Growing Mix® (BPGM; Ball Horticulture Co., W. Chicago, IL) were determined using methods described by Niedziela and Nelson (1992). While some of the major components of both substrates are similar (e.g. sphagnum peat moss, vermiculite, dolomitic limestone and gypsum), MM360 contains bark ash and coconut coir, while BPGM contains composted bark, perlite and shredded peanut hulls. The test results for both substrates are presented in Table 1, and will be referenced in the Results and Discussion section of this paper.

| Substrate   | Bulk density (g cm\(^{-3}\)) | Particle density (g cm\(^{-3}\)) | Air-filled porosity (%) | Container capacity (%) | Total porosity (%) |
|-------------|-------------------------------|----------------------------------|-------------------------|------------------------|-------------------|
| MM360       | 0.1\(^{b}\)                  | 0.3\(^{a}\)                      | 5.9\(^{b}\)             | 64.6\(^{b}\)           | 70.5\(^{b}\)      |
| BPGM        | 0.2\(^{a}\)                  | 0.7\(^{b}\)                      | 12.7\(^{a}\)            | 71.0\(^{a}\)           | 83.7\(^{a}\)      |

*Each value represents the mean of five replications. Each sample consisted of a water:substrate ratio of 2:1 (v/v). Values in the same column differ significantly when followed by a different letter, Tukey\(_{0.05}\).*  
*MM360®*  
*Ball Professional Growing Mix®*  

Table 1. The physical properties of two commercial soilless substrates used to study the impact of substrate type and irrigation interval on the post-transplant root dynamics of container-grown zinnia and tomato seedlings.

Due to the relatively short time frame of these experiments (28 days for zinnia, 16 days for tomato), we did not reanalyze the physical properties of either substrate at the end of the study. It should be noted, however, that substrate physical properties may change over time because of settling, shrinkage and/or organic matter decomposition (Mohammadi-Ghehsareh 2015). Some greenhouse crops have a production cycle of up to four months (Owen & Lopez 2015) and, over this period of time, substrate parameters (e.g. water, air and nutrient fluxes as well as transport gradients) may change (Fields et al. 2015).

Zinnia growth experiment. A single seed of ‘Thumbe-lina’ zinnia (Livingston Seed Co., Columbus, OH) was sown into each cell [80 cm\(^3\) (2.7 oz)] of two 50-cell plug trays, one tray containing MM360, and the other BPGM. After seeding, the trays were covered with plastic wrap and sub-irrigated with tap water (pH 8.8; EC 1.59 dS m\(^{-1}\)) to keep the substrate moist. Both trays were placed beneath fluorescent lights (95 \(\mu\)mol m\(^{-2}\) s\(^{-1}\) photosynthetic photon flux; 12 hr photoperiod) in a laboratory where temperature was maintained at 68 to 74 F and RH at 45 to 70%. After radicle emergence (week 1), the plastic wrap was removed from each tray and the germinated seedlings allowed to grow an additional three weeks under fluorescent lights. During this time the plants were sub-irrigated twice weekly with a water-soluble fertilizer [20N-2.6P-18.3K (JR Peters, Inc., Allentown, PA)] at a N rate of 200 mg L\(^{-1}\) (0.01 oz-1.10 qt\(^{-1}\)). By the beginning of week 4, the plants had developed 3 to 4 true leaves (plug stage 3) and were ready for transplanting.

At the time of transplanting, 24 uniformly-sized seedlings growing in MM360 were removed from the plug tray and replanted into each of twenty-four 9-cm\(^2\) (3.5-in\(^2\)) plastic pots filled with 425 cm\(^3\) (25.9 in\(^3\)) of the same substrate. The identical procedure was followed when transplanting 24 uniformly sized seedlings growing in BPGM. Groups of eight seedlings in each of the two substrates were randomly assigned time interval irrigation events of 24, 48 or 96 hr. The amount of water added back to the substrate at each irrigation interval was equal to the amount of moisture lost via evapotranspiration (ET\(_{0}\)) over...
the same period of time. A plastic saucer was placed beneath each container to collect any leachate which, if any, was subsequently reabsorbed. Gravimetric measurements of ET₀, similar to those described by Grant et al. (2012) were made daily at 0700hr, just prior to the beginning of each 12-hr photoperiod.

Irrigation treatments were repeated for 28 days, after which the experiment was terminated and the root ball of each plant separated from the shoot at the root collar. After removal from its container, each root ball was placed in a No. 18 sieve [1 mm (0.04 in)] and repeatedly washed with a fine spray of tap water to remove substrate debris. After this initial washing, each root system was transferred to a 14-cm (5.5-in) diameter plastic petri dish containing water, and the roots further cleaned using a dissecting needle and forceps. Cleaned root systems were then placed in 45 mL (1.5 oz) capped centrifuge tubes containing 50% ethanol and stored at 8 C (46 F) until they could be optically scanned and digitized (WinRHIZO Pro, Reagents Instruments, Victoria, BC, Canada) to determine the following root measurements: length, surface area and volume.

Finally, root dry weight (RDW) was recorded after oven drying each root system at 80 C (176 F) for 48 hr. From the RDW and WinRHIZO data, the following root growth parameters were calculated: specific root length, SRL (root length/RDW); root tissue density, RTD (RDW/root volume); root length density, RLD (root length/substrate volume); and the roots further cleaned using a dissecting needle and forceps. Cleaned root systems were then placed in 45 mL (1.5 oz) capped centrifuge tubes containing 50% ethanol and stored at 8 C (46 F) until they could be optically scanned and digitized (WinRHIZO Pro, Reagents Instruments, Victoria, BC, Canada) to determine the following root measurements: length, surface area and volume.

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### Tomato growth experiment

Using the same methods and procedures described for zinnia, a single seed of ‘Celebrity Hybrid’ tomato (Livingston Seed Co., Columbus, OH) was sown directly into each of two 50-cell plug trays, one tray containing MM360, the other BPGM. Two weeks after sowing, when the plants had developed fully-expanded cotyledons, 24 uniformly-sized seedlings were removed from the plug trays and replanted into each of twenty-four 9-cm² (3.5-in²) plastic pots as described earlier. Groups of eight seedlings in each of the two growing media were randomly assigned irrigation treatments of 24, 48 or 96hr. Irrigation treatments were repeated for 16 days, after which, because of seedling size, the experiment was terminated, the plants harvested, and measurements of root growth made using the same methods described for zinnia.

The experimental design for both studies was a balanced 2 by 3 factorial with two substrates, each with twenty-four replications, arranged randomly in two blocks, each block containing an equal number of plants irrigated at either 24, 48 or 96hr. The data were transformed to natural logarithms prior to analyses in order to obtain residuals that were normally distributed. Models for both designs were fit and analyzed using statistical software (Statistix 10, Analytical Software, Tallahassee, FL), and differences in treatment means compared using Tukey’s pairwise comparison test, P ≤ 0.05.

### Results and Discussion

**Zinnia.** In the current investigation, root growth, as measured by changes in RDW, was 30% greater for zinnias grown in BPGM than for those grown in MM360 (Table 2). Irrigation interval, though, did not impact RDW at the frequency levels used in these experiments. Most importantly, the data showed a significant interaction between substrate type and irrigation interval in some cases, thereby preventing the discussion of main effects. For plants grown in BPGM and irrigated every 48 hr, RDW was 11% to 56% greater than for any of the remaining treatments (Table 2). Since successful transplant production seeks to minimize plant stress, a favorable balance between substrate moisture content and aeration is important. For the substrates used in this study, air-filled porosity (AFP) and container capacity (CC), and the ratio between them, were consistently higher in BPGM than in MM60 (Table 1), suggesting that the physical characteristics of BPGM, especially when irrigated at 48 hr intervals, provided better conditions for zinnia root growth and development.

Specific root length is a frequently calculated growth parameter and is often used to characterize the economic aspects of root growth [i.e. for each unit of growth (root length), there is a corresponding unit of resource investment (root dry weight)] (Ostonen et al. 2007). Determinations of SRL made in the current study showed that zinnias grown in BPGM exhibited higher SRLs than those grown in MM360, regardless of the irrigation interval.

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**Table 2. The effect of substrate and irrigation interval on root growth, total evapotranspiration and average daily water loss of 4-week-old Zinnia elegans seedlings 28 days after transplanting from plug trays into marketable containers.**

| Substrate | Irrigation interval (hr) | RDW (mg) | SRL (cm·mg⁻¹) | RTD (mg·cm⁻³) | RLD (cm·cm⁻³) | ET₀ (g) | DWL (mL) |
|-----------|--------------------------|----------|----------------|---------------|--------------|--------|---------|
| MM360     | 24                       | 279.0    | 5.3            | 30.2          | 4.2          | 862    | 30.7    |
|           | 48                       | 230.6    | 4.9            | 27.0          | 4.5          | 804    | 28.7    |
|           | 96                       | 219.3    | 4.3            | 27.4          | 3.9          | 867    | 31.0    |
| BPGM      | 24                       | 300.4    | 5.7            | 31.2          | 4.2          | 988    | 35.3    |
|           | 48                       | 341.5    | 8.1            | 32.5          | 4.7          | 1111   | 39.7    |
|           | 96                       | 308.5    | 8.1            | 31.0          | 4.7          | 893    | 31.9    |

F-test probabilities:
- Substrate (S) *** *** ** * *** ***
- Irrigation interval (I) NS NS NS * NS NS
- S x I * ** NS NS **

*Each value represents the mean of 8 replications. NS, *, **, ***; nonsignificant, or significant at P < 0.05, 0.01, 0.001, respectively (Tukey). Abbreviations: RDW, root dry weight; SRL, specific root length (root length/RDW); RTD, root tissue density (RDW/root volume); RLD, root length density (root length/substrate volume); ET₀, total evapotranspiration; DWL, daily water loss; MM360, Metromix 360™; BPGM, Ball Professional Growing Mix™.
Table 3. The effect of substrate type and irrigation interval on root growth, total evapotranspiration and average daily water loss of 2-week-old Lycopersicon esculentum seedlings 16 days after transplanting from plug trays into marketable containers.

| Treatment | Root growth | Substrate | Irrigation interval (hr) | RDW (mg) | SRL (cm$^{-1}$) | RTD (mg cm$^{-3}$) | RLD (cm$^{-3}$) | ET$_O$ (g) | DWL (mL) |
|-----------|-------------|-----------|--------------------------|----------|----------------|--------------------|----------------|-----------|------------|
| MM360     | 24          | 28.0      | 29.9                     | 5.9      | 2.2            | 352                | 21.9          | 22.9      |
|           | 48          | 28.9      | 25.6                     | 6.4      | 1.9            | 360                | 22.5          | 22.8      |
|           | 96          | 35.3      | 26.4                     | 7.8      | 2.4            | 352                | 22.0          | 22.0      |
| BPGM      | 24          | 32.3      | 27.0                     | 6.7      | 2.2            | 364                | 22.8          | 22.2      |
|           | 48          | 33.4      | 30.0                     | 6.8      | 2.5            | 352                | 22.0          | 22.0      |
|           | 96          | 42.6      | 27.2                     | 8.8      | 3.0            | 355                | 22.2          | 22.2      |

F-test probabilities:

| Substrate (S) | Irrigation interval (I) | NS | ** | NS | NS | NS | ** | NS | NS |
|---------------|-------------------------|----|----|----|----|----|----|----|----|

Each value represents the mean of 8 replications. NS, *, **, ***, nonsignificant, or significant at P $\leq$ 0.05, 0.01, 0.001, respectively (Tukey). Abbreviations: RDW, root dry weight; SRL, specific root length (root length/RDW); RTD, root tissue density (RDW/root volume); RLD, root length density (root length/substrate volume); ET$_O$, total evapotranspiration; DWL, daily water loss; MM360, Metromix 360™; BPGM, Ball Professional Growing Mix™.

(The Table 2). The combination of BPGM irrigated every 48 hr resulted in SRL values that were 42% greater than they were for zinnias grown in BPGM and irrigated every 24 hr (Table 2). One of the factors controlling SRL, root length, often declines as substrate moisture decreases (Manes et al. 2006). In the current study, this was true for zinnias grown in MM360, but not for those grown in BPGM, where total root length actually increased as the frequency of irrigation decreased from 24hr to 48hr (data not shown).

Root traits such as RTD and RLD are thought to be functionally linked with leaf and stem traits to maximize the efficiency by which plants acquire and use resources in the soil (Kramer-Walter et al. 2016; Comas et al. 2013). In the present study, RTD was not affected by irrigation interval, but RLD was 10% greater for zinnias irrigated every 48hr compared to those irrigated more frequently (Table 2). While it seems counterintuitive that plants irrigated less frequently (48hr) would have a higher RLD than plants irrigated more often (24hr), Franco et al. (2008) and Fernandez et al. (2006), among others, have reported increased root growth in plants exposed to mild or moderate water stress conditions, a situation which may have existed in the present study for zinnias grown in small containers (450 cm$^3$) and watered only once every other day. In the current investigation, RTD was 1% greater for zinnia transplants grown in BPGM, a high-porosity growing medium (Table 2). RTD has been shown to play a role in determining the rate of biomass turnover in plants (Garnier and Laurent 1994; Schlaper and Ryser 1996), and Eisenstat (1991) has reported that roots with low RTDs often have proportionately higher SRLs. This was the case in the present study, and these data support the hypothesis that plants with higher RTDs often exhibit slower tissue turnover rates and consequently, over time, are able to accumulate greater biomass (Ryser 1996).

Container-grown plants are normally irrigated to resupply the amount of water lost by total evapotranspiration, ET$_O$ (Million and Yeager 2012). In the current study, many of the environmental and plant-related factors that influence ET$_O$ (e.g. temperature, relative humidity, light intensity, plant size and container size) either were held constant or were similar between replications of the same treatment, making differences in ET$_O$ more likely attributable to changes in substrate composition or in irrigation frequency. In the present investigation, evapotranspiration was always greater for zinnias grown in BPGM, while irrigation interval had no significant impact on ET$_O$ (Table 2). Likewise, daily water loss (DWL), expressed here as the average volume of water added back to the substrate each day, responded similarly (Table 2). For both ET$_O$ and DWL there was a significant interaction (P = 0.013 and 0.014, respectively) between substrate type and irrigation interval when zinnia transplants were grown in BPGM and irrigated every 48hr (Table 2). The relationship between substrate type and water loss noted here appears to reflect differences in the physical properties of the two growing media used in these experiments. BPGM, a substrate with greater total porosity (Table 1), would allow for more surface moisture evaporation, freer water movement and better drainage, all factors likely to increase ET$_O$ and DWL.

**Tomato.** For tomato seedlings, root growth (RDW) averaged 18% higher for plants grown in BPGM than for those grown in MM360 (Table 3). RDW of transplants irrigated every 96 hr was 25% to 29% greater than for similar plants irrigated more frequently (24 hr or 48 hr). These results support earlier research by Franco et al. (2002), who reported that Lotus creticus L. seedlings irrigated two days per week exhibited greater root growth than similar plants irrigated more frequently (six days per week). The roots of plants exposed to periods of moderate soil moisture stress often exhibit greater root growth than similar plants irrigated more frequently. Substrate moisture availability is largely determined by the physical properties of the substrate. Mupondi et al. (2014) reported finding an increase in RDW of 4-week-old tomato plants grown in compost-amended substrate having a higher particle density (PD) and a greater container capacity (CC) than for tomatoes grown in media with lower PD and CC values. In the present investigation, PD and CC values for BPGM were significantly greater than they were for MM360 (Table 1).

Neither substrate composition nor irrigation frequency by themselves had a significant impact on SRL in tomato (Table 3). However, both RTD and RLD were significantly greater (32% and 23%, respectively) for tomato transplants
irrigated every 96 hr versus those irrigated every 24 hr. Since RTD and RLD both represent root traits that are important in determining the ability of plants to obtain water and nutrients from the substrate, data from the present study suggest that a regimen of less frequent irrigation may actually benefit root growth in some instances. These results further support the idea that plants subjected to slight or moderate levels of moisture stress may develop root tissues that are denser and that occupy a greater unit volume of substrate than similar plants irrigated more frequently. Although no significant differences in RTD were noted between tomato transplants grown in MM360 or in BPGM, root length per unit volume of substrate (RLD) was numerically 18% greater for transplants grown in the higher porosity medium (BPGM).

With tomato, there were no significant differences in ET₀ or DWL between transplants grown in either MM360 or BPGM, nor were there any differences in ET₀ and DWL between seedlings irrigated at 24, 48 or 96hr intervals (Table 3). The absence of any ET₀ or DWL differences in the tomato study (differences which did exist in the zinnia experiment) may have resulted from a greater variation in leaf area per plant for tomato than for zinnia (data not shown), or from the shorter experimental time period in the tomato study (16 days versus 28 days).

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