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The Effect of Moisture Content in the Substrate on Rooting of Seedlings in Plug Trays

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Abstract.

Root growth, especially adventitious or basal, is important for both transplant quality and plug stability for mechanical transplanting. Two experiments were done on the effects of moisture level in the plug substrate on root morphology of six species grown in 288-plug trays. Two species per substrate moisture class [dry (verbena and zinnia), medium (coleus and petunia) and wet (begonia and impatiens)] were chosen based on the recommended moisture level. After radicle emergence, the moisture content of the substrate was maintained at three different moisture levels within each class (Experiment 1), or the plug substrate was allowed to dry out to one of three moisture levels before the trays were rewetted to the original moisture level for germination (Experiment 2). The number and the position of lateral and adventitious roots on the primary root, total root length and the length of the primary root were determined after the first leaf had fully expanded (3 weeks), while the number of lateral and adventitious roots and dry weights of the root and shoot were determined after 6 weeks. Two species showed a reduction in the number of adventitious roots (verbena, dry; and coleus, medium moisture), while total root length was reduced for coleus and the primary root length of impatiens (wet) decreased with increased moisture stress (Experiment 1). Total root length decreased for coleus (medium) and impatiens (wet) with increased moisture stress in Experiment 2. Root dry weight after 6 weeks was not affected by moisture stress in either experiment. An increased moisture deficit showed either no effect (zinnia, petunia and begonia) or resulted in a lower number and reduced total root length of both adventitious and lateral roots. Also the different moisture deficits did not affect the position of the lateral and adventitious roots on the primary root.

INTRODUCTION

In dicotyledonous seedlings, there are four types of roots: (i) the radicle (or primary root emerging from the germinating seed); (ii) adventitious roots (initiated from the hypocotyl); (iii) lateral roots (initiated from the primary taproot) and (iv) root hairs (Nicola, 1998). Some reports mention basal roots, which are special roots also initiated at the hypocotyl. These different types of roots do not develop all at the same time. First the radicle will emerge from the seed followed by lateral roots on the tap root and then the adventitious / basal roots from the hypocotyl. Moreover it has been shown that adventitious roots emerge acropetally (towards the shoot apex) while lateral roots develop basipetally (towards the radicle apex) (Weinhold, 1967). So depending on the time of observation, the number and length of lateral and adventitious roots differ.

The type, number and length of the different root types may have implications on the transplanting success of a seedling plug either into a cellpack or directly into a field soil. Water management plays an important role. Leskovar (1998) showed that overhead irrigation provided more and longer basal roots on bell pepper seedlings than an ebb and flow (subirrigation) system while the latter system had longer lateral roots. However, little work has been done on the effects of the amount and timing of irrigation on the architecture and root growth of bedding plants.

Plug production depends on high germination of the seeds, which usually requires a high degree of control of soil moisture and temperature. There are species that require high moisture content in the plug substrate (e.g., impatiens and begonias) while other

species (verbena and zinnia) require a low moisture content to germinate (Karlovič and Koranski, 1994). The initial germination (emergence of radicle, stage I) often takes place in a sweat chamber with high temperature and humidity requiring about 2-5 days. The next stage is when the radicle penetrates the substrate and the first true leaves starting to appear (stage II). The main purpose of stage II is to obtain true leaves and increase branching of the root system, so that high quality plugs are obtained which can be transplanted mechanically.

The main objective in this study was to study the effects of substrate moisture content on the root development of various bedding plant species differing in moisture requirements.

MATERIALS AND METHODS

Two experiments were done, both using two species that germinate under either a wet, medium or dry substrate (stage I) and then grown 'on' for about 6 weeks under three different moisture regimes. In Experiment 1, three different moisture levels were maintained within a very narrow range ($\pm 5\%$), while in Experiment 2, the substrate was allowed to "dry out" to a certain target level and then irrigated to the initial moisture content used for germination.

Experiment 1. Constant Moisture Level

Plug trays (288 cells – TLC universal plug tray) with 1.9×1.9 cm cells and a depth of 3.2 cm, were filled using a germination substrate (Premier Pro-mix PGX, Premier Tech Ltd., Riviere-du-Loupe, PQ, Canada). Prior to filling, the substrate was wetted to a 200% moisture content (based on weight of moisture/ dry weight of substrate). The trays were filled to a heap (3-4 cm high) and then dropped from about 3-4 cm height onto a solid surface three times. The trays were then scraped off with a ruler until all cells were filled evenly. All filled trays weighed about 770-800 g (empty tray: 165 g).

Two blocks of 72 cells of the plug trays were then dibbled and singularly seeded on April 29, 2005. Following seeding of six species, the trays were then moistened with a fine mist until achieving the maximum moisture content (equivalent to 0% moisture loss) depending on the moisture category required for germination (Table 1). Two species were used for each moisture category, namely for the category Dry (verbena 'Obsession scarlet' and zinnia 'Star orange'), Medium (coleus 'Wizard pink' and petunia 'Celebrity blue') and Wet (begonia 'Victoria green leaf pink' and impatiens 'Super Elfin lavender').

The moistened trays were then covered with a thin layer of clear plastic (except for verbena: black plastic) and moved into a seed germinator maintained at constant 25°C with side fluorescent (cool-white) lighting at an irradiance of $5 \mu\text{mol} \cdot \text{m}^{-2} \cdot \text{sec}^{-1}$ for $16 \text{ h} \cdot \text{day}^{-1}$. Upon emergence of the radicle (1-3 mm), the trays were removed from the germination chambers (2-4 days for all species except begonia). Then, all moistened trays were cut in one-half so that each half tray contained 144 cells (12×12 cells) with 72 cells sown.

Within each moisture category (Table 1), there were three levels of moisture loss: 0, 10 and 20% for Dry; 0, 20 and 40% for Medium; and 0, 30 and 60% for the Wet category.

Once germination stage I was reached, the trays were moved to a glass greenhouse and placed randomly onto a bench with an open mesh support and allowed to dry out to either 0, 10, 20, 30, 40 or 60 % moisture loss depending on the moisture category (Table 1). The air temperature inside the greenhouse compartment was set at 23°C for day and night.

Trays were weighed three times per day (800, 1200 and 1600 HR). If weight of the tray was within 20 g from the minimum stated value, the tray was watered with deionized water using a backpack sprayer until the maximum weight was reached (Table 1). The plugs were fertilized two times with a complete nutrient solution ($\text{EC}=1.1 \text{ mS} \cdot \text{cm}^{-1}$).

When the first true leaves appeared (3 weeks after seeding), the germination percentage and the geometry of the seedling roots were determined. For this purpose, five

plugs were randomly chosen and removed from each plug tray followed by carefully rinsing the substrate from the root system. The roots were then stained with 0.01% Toluidine blue and placed in a Petri dish. The length of the main root (starting from the dicotyledons), as well as the location on the main root (distance from the dicotyledons to the implant) of all the lateral and adventitious roots and their individual length were measured by placing the Petri dish on mm-grid paper. Based on the location of the implant of the adventitious and lateral roots on the radicle, the number of lateral and adventitious roots on the main radicle were divided in three sections: Section I contained roots formed within 10 mm from the top surface of the plug; Section II contained those roots formed in the bottom one-half (between 10 and 20 mm from the soil surface) of the plug while Section III included those roots that were formed at the base of the plug (> 20 mm from the plug surface).

After about 6 weeks (June 6, 2005), only the length of the main root as well as fresh and dry weights of both root and shoot systems were determined.

Experiment 2. Fluctuating Moisture Level

A similar procedure was followed as described above except for the difference in moisture management. In this experiment, the substrate was allowed to dry out to a given moisture content before the trays were re-irrigated (with a dilute fertilizer solution, EC=0.6 mS /cm) until the initial moisture content at seeding (0% moisture loss) was reached. Again within each moisture category, there were still three levels of moisture losses based on the respective moisture category (Table 1), except that the target weight of the 0% moisture loss within the 'wet' category was adjusted from 1800 to 1700 g. The seeding took place on June 17, 2005 and the moisture content of the substrate at filling was 182% (w/w).

After 4 weeks, the roots were measured in a slightly different manner than described in the previous experiment. All adventitious roots that originated at the primary root within 5 mm from the soil surface were counted and their individual length determined. Each root was abscised from the seedling and transferred to mm-grid paper for determining its length.

After 6 weeks, the lateral and adventitious roots were counted and fresh and dry weights of both root and shoot systems were determined.

The experimental design for both experiments was a completely randomized design with moisture loss level (3) per species and the replicates (2) as the factors. Five samples were taken per experimental plot at each date. The statistical analysis was done on a per species basis using SAS Proc GLM (SAS Institute Inc., Cary, NC; version 8).

RESULTS

Experiment 1. Constant Moisture Level

1. Germination. The total germination percentages were slightly different depending on species, namely begonia (84%), coleus (96%), impatiens (96%), petunia (91%), verbena (86%) and zinnia (93%), but there was no effect from the watering practices on the percent germination at 3 weeks after seeding (data not shown).

2. Week 3 (at First True Leaf). The number of lateral and adventitious roots was reduced in two species, namely verbena and coleus, while total root length was reduced for coleus with increased moisture stress compared to the control (0%) (Table 2). The length of the primary root was reduced with increased moisture stress for impatiens. For the other three species (begonia, petunia and zinnia), there were no statistical (or visual) differences in the number of roots, total root length or the length of the primary root due to the moisture levels. Begonia had about twice the number of lateral roots compared to the other five species.

Although the number of lateral / adventitious roots positioned between 0 and 10 mm (distance from the dicotyledon node) section were greater than the number in the 10-20 or >20 mm section, there was no difference due to the moisture levels (data not

shown).

3. Week 6. The root dry weight of petunia for the 30 and 60% moisture stress was twice to the control, but there were no statistical differences in the total dry root weight for the other five species due to the moisture treatment (data not shown).

Experiment 2. Fluctuating Moisture Level

1. Germination. The germination percentages were for begonia (72%), coleus (98%), impatiens (91%), petunia (90%), verbena (71%) and zinnia (87%). No effects from the watering practices were observed on the percent germination.

2. Week 4 (after First Leaf Expansion). The number of adventitious roots did not depend on the moisture regime of the plugs on any of the species. The overall root length of all adventitious roots decreased for coleus and impatiens with increased stress before irrigation (Table 3). The mean root length of the adventitious roots decreased with increased moisture stress for coleus. Verbena, begonia and petunia did not show any effects on the measured parameters as a result of the different moisture deficits.

3. Week 6. The number of adventitious and lateral roots decreased for impatiens with an increased stress, while the root dry weight of petunia was lower with increased moisture stress (Table 4). No effects were observed on verbena, begonia and zinnia.

DISCUSSION

The hypothesis that root growth and/or architecture would be affected by either a constant water stress or a fluctuating water stress could not be universally supported by the results among the species studied. The hypothesis that a 'dry' growing substrate would improve branching from the main taproot was also not supported in either experiment. No species showed an increase in the number or the total root length of the lateral and adventitious roots with increased moisture deficit. In some cases, the root number (Experiment 1) and total root length (Experiment 1 + 2) actually decreased with increased moisture deficit. Also the results of the root dry weight after 6 weeks for any of the species did not support that increased moisture stress creates a carbohydrate sink in the root system. It is of course possible that the stress applied did not provide sufficient stress in order to find significant results in our experiments.

The results of the implants of the adventitious and lateral roots indicated that after 3 weeks of maintaining different moisture regimes, there was no effect of the moisture regime on either the number of roots or in total root length in any of the three sections for any of the species in either of the experiments.

Our results (Experiment 1 + 2) showed that an increased moisture stress (both constant and fluctuating levels) showed either no effect or resulted in a lower number and reduced total root length of both adventitious and lateral roots depending on the species, and is consistent with previous literature (Leskovar, 1998; Nicola, 1998). Begonia, petunia and zinnia did not show any effects. Also the different moisture deficits did not affect the position of the implant of the lateral and adventitious roots on the primary root.

Literature Cited

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Tables

Table 1. Weight of plug tray (tray + substrate + water + seedling) within individual moisture loss and category. For Experiment 1 (Constant moisture level), weight tolerance is indicated within parentheses. Trays were irrigated when minimum weight was reached and terminated upon reaching the maximum value. For Experiment 2 (Fluctuating moisture level), the trays were irrigated when the weight of the trays dropped to the value belonging to the percent moisture loss and irrigated until reaching the weight at 0% moisture loss.

Moisture category	Moisture loss (%) *					
	0	10	20	30	40	60
Dry	1200 g (1140-1260)	1120 g (1065-1175)	1040 g (990-1090)			
Medium	1500 g (1425-1575)		1280 g (1215-1345)		1060 g (1005-1115)	
Wet	1800 g (1710-1890)			1380 g (1310-1450)		960 g (910-1010)

* Moisture loss was based on the amount of moisture in the tray at seeding. Weight of empty tray (165 g) + air-dry substrate (200 g) was 365 g.

Table 2. The number and total length of lateral and adventitious roots and length of primary root for six plant species maintained at three moisture categories (dry, medium and wet) and three constant moisture losses (Experiment 1). Measurements were taken after first true leaf was developed. Data are the mean of 10 seedlings.

Moisture category and loss ^z (%)	Lateral + adventitious roots (no.) ^y	Total root length (mm)	Length primary root (mm)	Lateral + adventitious roots (no.)	Total root length (mm)	Length primary root (mm)
Dry		<i>Verbena</i>			<i>Zinnia</i>	
0	14.4 ± 1.0	302 ± 30	55 ± 6.2	15.8 ± 1.9	303 ± 39	41 ± 2.4
10	16.4 ± 1.8	338 ± 37	74 ± 10	16.4 ± 1.6	322 ± 34	53 ± 7.7
20	11.8 ± 0.7	248 ± 20	56 ± 6.3	16.0 ± 1.5	313 ± 34	53 ± 6.3
Significance ^y	*	ns	ns	ns	ns	ns
Medium		<i>Coleus</i>			<i>Petunia</i>	
0	19.6 ± 0.8	371 ± 15	73 ± 7.4	13.8 ± 0.8	299 ± 14	41 ± 3.0
20	17.7 ± 0.7	339 ± 16	73 ± 5.3	15.5 ± 0.5	342 ± 15	48 ± 4.7
40	12.8 ± 0.7	268 ± 8	58 ± 7.1	14.3 ± 0.9	279 ± 34	44 ± 4.1
Significance ^y	*	*	ns	ns	ns	ns
Wet		<i>Begonia</i>			<i>Impatiens</i>	
0	28.2 ± 2.6	293 ± 32	41 ± 2.9	13.2 ± 0.7	361 ± 16	117 ± 12
30	28.8 ± 2.2	340 ± 27	45 ± 2.3	13.9 ± 0.9	404 ± 24	92 ± 7.1
60	31.9 ± 2.0	375 ± 33	40 ± 2.1	15.0 ± 0.5	404 ± 19	82 ± 8.1
Significance ^y	ns	ns	ns	ns	ns	*

^z Plug trays weighed 1200, 1500 and 1800 g for 0% loss for 'Dry', 'Medium' and 'Wet', respectively. The percent loss is the moisture reduction relative to the 0%.

^y ns,* non-significant or significant at $P < 0.05$, respectively.

Table 3. The effect of different watering regimes (Experiment 2: Fluctuating moisture loss regime) on the number as well as the total root length of adventitious roots (those originating within 5 mm from the soil-line) of six species when the first leaf was fully expanded (4 weeks after seeding). The species were divided into a 'dry', 'medium dry' and 'wet' group based on germination requirements. Data represent the mean of 10 seedlings.

Moisture group and % moisture loss ^z	Root number	Total root length (mm)	Mean root length (mm)	Root number	Total root length (mm)	Mean root length (mm)
Dry		<i>Verbena</i>		<i>Zinnia</i>		
0	6.9 ±0.7	271 ±30.2	39 ±2.8	14.5 ±1.3	592 ±60.9	41 ±1.7
10	9.5 ±1.3	365 ±35.7	38 ±2.3	13.3 ±1.8	507 ±53.5	38 ±1.8
20	8.3 ±0.9	327 ±37.3	39 ±1.8	11.3 ±0.7	430 ±34.1	38 ±1.8
Significance ^y	ns	ns	ns	ns	ns	ns
Medium		<i>Coleus</i>		<i>Petunia</i>		
0	11.7 ±0.6	564 ±26.6	48 ±3.0	12.0 ±1.2	629 ±74.3	52 ±2.5
20	10.4 ±0.7	467 ±39.6	45 ±3.3	11.1 ±1.0	488 ±57.4	44 ±2.5
40	10.4 ±0.6	423 ±27.0	40 ±3.0	10.8 ±1.2	553 ±79.0	51 ±2.6
Significance	ns	*	*	ns	ns	ns
Wet		<i>Begonia</i>		<i>Impatiens</i>		
0	11.1 ±1.2	150 ±22.4	14 ±1.1	9.8 ±0.2	735 ±35.8	75 ±3.9
30	11.2 ±1.0	214 ±27.0	19 ±1.3	9.8 ±0.4	702 ±39.2	72 ±4.0
60	10.6 ±0.7	214 ±17.3	20 ±1.3	8.1 ±0.3	552 ±36.8	68 ±3.7
Significance	ns	ns	ns	ns	*	ns

^z Moisture losses are relative to the amount of moisture at seeding (Table 1); at irrigation the moisture was brought back up to the initial moisture level at seeding (0%).

^y ns, * non-significant or significant at $P < 0.05$.

Table 4. The effect of different watering regimes (Experiment 2: Fluctuating moisture deficit) on the number of adventitious roots as well as the dry weight of total roots and shoots for six species after 6 weeks from seeding (n=10).

Moisture loss ^z	Adventitious + lateral roots	Root dry weight (mg)	Shoot dry weight (mg)	Adventitious + lateral roots	Root dry weight (mg)	Shoot dry weight (mg)
Group 'Dry'		<i>Verbena</i>		<i>Zinnia</i>		
0	12.8	29	59	29.4	27	58
10	10.2	26	51	32.8	29	76
20	12.5	25	47	28.1	26	41
Significance ^y	ns	ns	ns	ns	ns	ns
Group 'Medium'		<i>Coleus</i>		<i>Petunia</i>		
0	17.5	13	57	11.6	50	68
20	14.8	13	56	10.2	41	65
40	13.2	11	44	9.60	40	56
Significance ^y	ns	ns	ns	ns	*	ns
Group 'Wet'		<i>Begonia</i>		<i>Impatiens</i>		
0	15.7	6	23	12.9	24	64
30	21.6	12	28	12.8	19	62
60	14.8	8	13	10.6	16	54
Significance ^y	ns	ns	ns	*	ns	ns

^z Moisture losses are based relative to the amount of moisture at seeding (Table 1).

^y ns, *, non-significant or significant at $P < 0.05$.