

## ABSTRACT

JEONG, KA YEON. Establishment and Stabilization of pH in Container Root Substrate.  
(Under the direction of Dr. Paul V. Nelson.)

A rapid and accurate method to quantify the acidity of sphagnum peat moss was developed to establish initial pH of peat-base root substrate within a recommended optimal range. This acid-base titration method can predict the calcite requirements of acidic peat moss for target pH levels  $\leq 6.2$ . To monitor substrate pH during crop production, a non-destructive system for periodically sampling substrate solution is required. The Rhizon soil moisture sampler (RS) was tested in 1.8L volume pots containing lettuce plants. Significant pH gradients, 6.3 – 5.6 from bottom to top of pot were found. The pH level in a diagonally oriented RS extract was found to be equivalent to that in the lower 20% of the substrate column. The pH levels of substrate solution extracted by diagonally oriented RS, pour-through, and saturated media extract methods were found to be 6.2, 6.6, and 5.5, respectively.

The effects of plant species (13 floricultural crops), potential acidity/basicity rating of fertilizer, and concentration of water soluble fertilizer on substrate pH during crop production were assessed. Petunia, fibrous begonia, and osteospermum were very light acidifier species, pansy, impatiens, New Guinea impatiens, and geranium were light acidifier-species, and vinca, tomato, and Reiger begonia were medium acidifier-species. These three acidification groups of plants were fertilized at the rate of  $100 \text{ mg}\cdot\text{L}^{-1} \text{ N}$ . Pot mum, sunflower, and kalanchoe were heavy acidifier-species. This group was fertilized with  $200 \text{ mg}\cdot\text{L}^{-1} \text{ N}$  to comply with fertilization recommendations. Solutions of the basic, neutral, and acidic

fertilizers tested were acidic and caused a decline in substrate pH whenever plant uptake was low and substrate EC was increasing. Conversely, when a high proportion of the fertilizer was accumulated by plants, the acidity/basicity rating of the fertilizer was expressed in substrate pH shifts. To establish and stabilize substrate pH, mature dairy cow manure compost (DMC) was evaluated as a soilless substrate substitute for dolomitic limestone and peat moss. Although pH declined during plant production, the decline was similar for agricultural limestone and the 20 to 30% DMC treatments, indicating that the buffering capacity was equal for limestone and DMC. All limestone and a portion of peat moss were effectively replaced with DMC. Initial EC levels for DMC substrates were within the acceptable range for seedlings and bedding plants. End of crop tissue analysis indicated that DMC resulted in higher leaf concentrations of potassium, sulfur, copper, iron, and manganese, lower, but adequate, calcium and magnesium, and similar nitrogen, phosphorus, boron, and zinc concentrations. The impact of DMC on physical properties of a peat moss-perlite substrate with and without plant growth was evaluated at the beginning and end of a crop. DMC resulted in increased dry bulk density, but there was no change in dry bulk density between initial and final substrate for each treatment. The total porosity (TP) of all substrates was higher than 84%. Container capacity increased in all substrates over time and also with DMC addition at the beginning and end of the crop. A significant decrease in air space (AS) was observed with DMC addition in both initial and final substrates. The initial AS for all substrates decreased over 12 weeks. An increase in plant shoot dry weight resulted from DMC addition in substrates.

Establishment and Stabilization of pH in Container Root Substrate

by  
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A dissertation submitted to the Graduate Faculty of  
North Carolina State University  
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## DEDICATION

To my father and mother,

정해수, 손복순

I love you with all my heart.

## **BIOGRAPHY**

Although I grew up in Seoul, the biggest city in Korea, I spent my summers in the beautiful small mountainous town where my grandmother lived. The memories in her flower and vegetable gardens and in nature are still so vivid and beautiful and always make me smile. I believe the power of the beauty of nature and plants can help people have a better life.

When I was obtaining a Bachelor of Science degree in Biological Resources and Technology at Yonsei University in Korea, my eyes were opened by the course work related with crop science and soil science. During my junior year, I was selected for an exchange student program at Maryville College in Tennessee. This one year experience encouraged me to come to the U.S.A. for my graduate study in horticultural science. My education in horticulture started in Ohio where I received my M.S. degree at the Ohio State University under the direction of Drs. Claudio Pasian and David Tay. My research involved development of cultural recommendations for six begonia species conserved at the Ornamental Germplasm Center, Columbus, Ohio.

Since 2004, I have worked at the OFA Short Courses as a volunteer in the decoration department and as a monitor during the courses. I met people who are educators, researchers, growers, and are also working for industries serving horticulture. I experienced a range of aspects of horticulture. That was also the place where I met my advisor, Dr. Paul Nelson for the first time. Subsequently, I was accepted into a Doctor of Philosophy program with Dr.

Nelson at North Carolina State University. The research focused on the development of a system to stabilize substrate pH during crop production and introduction of compost material as a partial substitute for sphagnum peat moss in soilless substrate for stabilizing pH.

My career goal in horticulture is to connect horticulture and people with my following motto.

**“Greens, Colors, and Soils for Healthier and Happier Life”**

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## CHAPTER 1

### **Impact of Composted Dairy Manure on pH management and Physical Properties of Soilless Substrate**

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#### **Abstract**

Dairy cow manure compost (DMC) was evaluated as a soilless substrate substitute for dolomitic limestone and peat moss in two experiments. The objectives were 1) to quantify the impact of DMC on substrate pH establishment and stabilization throughout crop time and 2) to test the effect of DMC on physical properties of substrate peat moss plus DMC (at 5 to 30% by volume) was held constant at 75% volume and perlite at 25% without limestone. Two additional control treatments of 75% sphagnum peat moss and 25% perlite were formulated with and without agricultural dolomitic limestone. Pot chrysanthemum ‘Kory’ plants were transplanted into 16.5 cm diameter (1.4 L) plastic pots and fertilized at each

irrigation with 17N-2.2P-14.1K neutral fertilizer. Additions of 0 to 30% DMC resulted in initial substrate pH levels of 3.1 to 6.5. Although pH declined during plant production, the decline was similar in the agricultural limestone and the 20 to 30% DMC treatments that had similar initial pH levels. Thus, pH buffering capacity of DMC was similar to the limestone. The initial EC levels for all substrates were within the acceptable range for seedlings and bedding plants. Magnitude of shrinkage did not relate to addition of DMC and was of little commercial significance. Irrespective of time in the cropping cycle, DMC resulted in increased dry bulk density ( $D_b$ ), decreased total porosity (TP) and container capacity (CC), and little effect on air space (AS). AS levels were in a good range of 15% and above for the 7.6 cm tall test cylinders. End of crop tissue analysis indicated that DMC resulted in higher leaf concentrations of potassium, sulfur, copper, iron, and manganese, lower, but adequate, calcium and magnesium, and similar nitrogen, phosphorus, boron, and zinc concentrations. Maximum plant growth (dry weight) occurred with 15% DMC in Expt. 1 and with 10% DMC in Expt. 2. All limestone and a portion of peat moss were effectively replaced with DMC.

## **Introduction**

Use of dairy manure compost (DMC) has contributed to sustainable agricultural production through recycling of animal waste and improving chemical and physical properties of soil (Klausner et al. 1998; Eghball et al. 2004; Butler and Muir, 2006; Butler et al. 2008). Composting of manure benefits the handling of manure waste by reducing volume,

weight, and odor, and can kill weed seeds and pathogens (Rynk et al., 1992). Application of compost to soil significantly increased pH, organic matter content, and soil-water holding capacity (Murray, 1981; Butler and Muir, 2006; Butler et al., 2008; Butler et al., 2009). The effects of compost residuals lasted up to four years by guarding against soil acidification and nutrient depletion problems in corn production (Eghball et al., 2004). DMC and similar composted materials such as fiber from digested slurry or composted cattle slurry fiber were shown to serve as a substitute for peat moss in a growing mix for a number of crops (Bradley et al., 1996; Chen et al 1986; Prasad 2008). Information is lacking on the effects of compost on pH stabilization in container root substrates as well as on the impact compost could have on the physical properties of these substrates. The objectives of this study were 1) to quantify the impact of DMC on substrate pH establishment and stabilization throughout crop time and 2) to test the effect of DMC on physical properties of substrate.

## **Materials and Methods**

*Plant Culture* Two experiments were conducted in a glass greenhouse at 35 °N latitude in Raleigh, NC. The experiments were initiated in August 2007 (Expt. 1) and May 2008 (Expt. 2) and were conducted for 12 and 11 weeks, respectively. Five rooted cuttings of pot chrysanthemum 'Kory' (*Dendranthema x grandiflora* (Ramat.) Kitam.) were transplanted into green plastic pots containing 1.4 L of root substrate and measuring 16.5 cm in diameter at the top and 11.2 cm deep. Fertilizer formulated in deionized water was applied to the top of the substrate at each irrigation with approximately 20% leaching. Fertilizer consisted of

17N-2.2P-14.1K neutral water soluble fertilizer (Greencare 17N-5P<sub>2</sub>O<sub>5</sub>-17K<sub>2</sub>O, Kankakee, IL). It was applied at a concentration of 300 mg L<sup>-1</sup> N in Expt. 1 and 250 mg L<sup>-1</sup> N in Expt. 2. Frequency of irrigation ranged from twice a week at the beginning to daily at the end of the experiments. At 14 days after transplanting (DAT), all plants were pinched to leave an average of 9 leaves per plant. In Expt. 1, black cloth was applied from 7:00 p.m. to 7:00 a.m. beginning at 14 DAT and was continued for 9 weeks to induce flowers. In Expt. 2, incandescence light was applied at an intensity of 2 μmol m<sup>-2</sup> s<sup>-1</sup> for the first two weeks. After two weeks, plants were shaded with black cloth from 6:00 p.m. until 7:00 a.m. daily until color was well developed in buds. Plants were sprayed with daminozide plant growth regulator at a concentration of 2,500 mg L<sup>-1</sup> 21 DAT in Expt. 1 only.

Compost A stable mature compost of dairy cow manure plus spoiled-silage (DMC) adjusted initially to a C:N of 30, (Woods End Laboratories, Inc, Mt Vernon, Maine) was prepared by turned-pile method, using a tractor front-end loader to lift and mix a conical-shaped pile (dimensions 4.3m d x 1.8m H) 5 times in the course of 90-days. Temperature in the core of the pile rose within 7-days of mixing to 57-60°C and remained very warm (49 – 57°C) for 6-weeks. After cooling to less than 30°C piles were stored outdoors by covering with Compostex® compost fabric, a polypropylene spun fabric permeable to air but which sheds water. Prior to use, a cubic meter sample of DMC was sieved through a 13 mm screen and mixed in a Twister™ II Batch Mixer, (Bouldin and Lawson, McMinnville, TN). Compost samples were tested by the North Carolina Department of Agriculture and Consumer Services (NCDA&CS) waste analysis lab. Total dry weight concentrations of nutrients are

presented in Table 1.1. Other measurements include a cation exchange capacity of 36.7 meq  $100 \text{ cm}^{-3}$  determined by summation of cations; a base saturation of 100%; a pH level of 8.0 measured in a 2:1 deionized water filtrate; a saturated paste EC level of  $5.1 \text{ mS cm}^{-1}$ , a C:N ratio of 13.3; and calcium carbonate equivalence 1.67% (dry weight bases).

*Treatments* Root substrate treatments had fixed volumes of 25% perlite and 75% sphagnum peat moss (Sun Gro Horticulture, Bellevue, WA) plus DMC (at 5, 10, 15, 20, 25, or 30% by volume). No limestone was applied in these treatments. Additionally, two control treatment of 75% sphagnum peat moss and 25% perlite were formulated with and without agricultural dolomitic limestone ( $6 \text{ g L}^{-1}$ ). All treatments included wetting agent (AquaGro 2000 G, Aquatrols, Paulsboro, NJ) at the label rate of  $0.6 \text{ g L}^{-1}$ . Anhydrous calcium sulfate ( $\text{CaSO}_4$ ) at  $0.9 \text{ g L}^{-1}$  was added into all treatments in Expt 2.

*Data* Root substrate pH and EC were measured in substrate solution extracted using the pour-through technique (Wright, 1986) in Expt. 1 and the Rhizon Soil Moisture Sampler in Expt. 2 (Soil Moisture Equipment Corp., Santa Barbara, CA, [www.soilmoisture.com](http://www.soilmoisture.com)). Both tests were designed to sample unaltered bulk solution. The Rhizon sampler consisted of a 10 cm long hollow, hydrophilic polymer PVC tube that was inserted diagonally into the pot from 0.5 cm below the substrate surface to the bottom of the pot. Substrate solution was drawn through the sampler and into a collection vial under a vacuum of -138 kPa.

Root substrate shrinkage during the crop production period in Expt. 1 was measured as the difference in depth of the substrate below the pot rim at day 1 and harvest date. Prior to

the first measurement, plants had been transplanted and watered to settle the substrate. Substrate depth was determined as the average of three measurements of the distance from the pot rim to the substrate surface.

In Expt. 1 the five plants in each pot were cut at the substrate surface, dried to a constant weight in a forced draft oven at 70°C, and the combined weight of the five plants was measured. In Expt. 2, leaves one third of the distance from the terminal end of lateral shoots were harvested. These leaves were washed in 0.2 N HCl for 1 min, rinsed in deionized water, dried in a forced draft oven at 70°C, and weighed. Total dry shoot weight was determined as the sum of the weight of sampled leaves plus the weight of the remainder of the combined five shoot in each pot. The dry leaf samples were ground in a Thomas-Wiley Intermediate Mill (Arthur H. Thomas Co., Swedesboro, NJ 08085) with a stainless steel cutting chamber to pass a 20 mesh sieve (1 mm particle size). A 0.15 g sample was digested in a microwave digester (MARS; CEM Corp, Matthews, NC) using a modified EPA method (EPA method 3051 with additional peroxide step). Nutrient concentration, except N, was determined with inductively coupled plasma optical emission spectroscopy (ICP-OES; Model IRIS Intrepid II, Thermo Corp., Waltham, MA). Total nitrogen was determined with a combustion analyzer (model 2400, Perkin Elmer, Waltham, MA).

Average substrate physical properties in a 7.6 cm tall column were measured in Expt. 2 at the beginning and the end of the experiment. Measurements included dry bulk density ( $D_b$ ,  $\text{g}\cdot\text{cm}^{-3}$ ), total porosity (TP, % substrate volume), container capacity (CC, % substrate

volume), and air space at CC (AS, % substrate volume) using the NCSU porometer method (Fonteno 1996). At the initial date, three 7.6 diameter by 7.6 cm tall cylinders were taped together end to end. This combination cylinder was filled with substrate and was compacted by dropping it a distance of 15 cm three times. The center cylinder with its substrate was used for physical properties testing. At the end of the experiment (77 DAT), substrate was removed from the center of pots by coring. Three 7.6 cm diameter cylinders were taped together end to end. The bottom cylinder was 3.8 cm tall and had a beveled cutting edge while the two cylinders above it were each 7.6 cm tall. This compound cylinder was hammered through the substrate in the pot to the pot bottom. The center cylinder with its substrate was used for physical properties testing.

Experimental Design & Analysis Both experiments were arranged in a randomized complete block design with five blocks. Each plot consisted of one complete row of three pots across the 122 cm wide bench. Initial substrate pH and EC values of each substrate were regressed using the PROC REG to determine the best-fit, linear or quadratic model. Terms of the model were evaluated for significance based on a comparison of  $F$  values at  $\alpha=0.05$ . Means of plant tissue nutrient concentrations were separated by T-test at  $P\leq 0.5$ .

## **Results and Discussion**

### Substrate pH

Initial substrate pH in Expts. 1 and 2, measured after watering newly transplanted plants with deionized water, was 5.8 and 6.0 in the control treatment with limestone but no

DMC and 3.2 and 3.1 in the control treatment without both limestone and DMC, respectively (Fig. 1.1). Initial substrate pH increased with each increase in DMC in a quadratic fashion ( $P < 0.0001$ ). At 30% DMC initial substrate pH was 6.3 and 6.6 in experiments 1 and 2, respectively. The initial substrate pH declined over time in all treatments with the exception of the un-limed 0, 5, and 10% DMC treatments in Expt. 1 and 0% DMC in Expt. 2 (Fig. 1.2). Substrate pH in these latter treatments was always below 5.0 and rose slightly by the end of the experiments. Substrate pH in the limestone control treatment over time was between the 20% and 30% DMC treatments in Expt. 1 and between the 20% and 25% treatments in Expt. 2. The declines in substrate pH over time in the limestone control and 20% to 30% DMC treatments were fairly parallel, indicating that the pH buffering capacity of DMC was of a similar magnitude to the agricultural limestone. A downward shift in pH was anticipated because applied fertilizer was neutral, there was no alkalinity in the irrigation water, and plant root respiration was expected to have an acidifying effect. The residual component of limestone was inadequate to counteract acidification.

#### Substrate EC

The EC of DMC, as determined by the saturated media extraction procedure, was  $5.1 \text{ mS} \cdot \text{cm}^{-1}$ . The EC of substrate solutions in Expt. 1, obtained through the pour-through extraction procedure one hour after watering the newly transplanted plants with deionized water ranged from 2.0 to  $3.0 \text{ mS} \cdot \text{cm}^{-1}$  (Fig. 1.3). The EC levels in all substrates were within the safe range

for seedlings and bedding plants as set forth for the pour-through technique by Whipker, et al. (2000).

### Plant Growth

The 15% and 10% DMC treatment in Expt. 1 and Expt. 2, respectively, produced the largest plant dry weight which is significantly greater growth compared to both control treatments that did not contain DMC (Fig. 1.4). Similarly, the addition of composted separated fiber from cattle manure improved the growth of tomato seedling (Prasad 2008).

### Substrate Physical Properties

Because the procedures for sampling substrate were, out of necessity, different at the beginning and end of the experiment, comparison of physical properties between the two times would not be valid. Consequently, interpretation of physical properties is limited to the impact of DMC within each of the initial and final treatment series.

Initial and final substrate dry  $D_b$  increased from 0.10 to 0.23  $\text{g}\cdot\text{cm}^{-3}$  and from 0.11 to 0.19  $\text{g}\cdot\text{cm}^{-3}$ , respectively, with DMC increases from 0 to 30% (Fig. 1.5A). These are in the range found by Chen et al., (1986). Initial TP decreased from 87.5% to 79.6% with DMC increases from 0 to 30% (Fig. 1.5B). Final TP likewise decreased from 91.2% to 85.2% with increased DMC. Substrate CC followed a similar pattern to TP (Fig. 1.5C). Initial CC declined from 70.0% to 62.7% and final CC from 75.2% to 70.0% with increases of 0 to 30% DMC. Initial AS increased slightly up to 10 to 15% DMC and then declined moderately at

higher DMC levels (Fig. 1.5D). Final AS was random across DMC treatments without fitting any significant regression line. Irrespective of time in the cropping cycle, DMC resulted in increased  $D_b$ , decreased TP and CC, and little effect on AS. AS levels were in a good range of 15% and above for a 7.6 cm tall column of substrate.

### Nutrient Uptake

The control substrate with limestone and the substrate with 20% DMC were selected in Expt. 2 for a comparison of leaf nutrient concentrations because the substrate pH levels in these treatments were similar and in the desired range. Plants grown in the substrate with 20% DMC contained significantly higher concentrations of K, S, Cu, Fe, and Mn and lower concentration of Ca and Mg than plants in the limestone control treatment (Table 1.2). Higher Ca and Mg concentrations would be expected in the limestone control treatment plants due to the supply of these nutrients in dolomitic limestone. Higher concentrations of the other nutrients were reasonable given the supply of these in manure compost.

### **Conclusions**

This study demonstrated that DMC can be used in the place of limestone to set initial substrate target pH and buffer it as well as limestone over 77 days of production. Air space at container capacity did not differ and substrate volume shrinkage was insignificant in the DMC substrates. However, bulk density was higher and container capacity was lower in the DMC substrates. Substrate EC values increased with DMC addition but remained in acceptable levels.

## **ACKNOWLEDGMENTS**

Mention of a trademark, proprietary product, or vendor does not constitute a guarantee or warranty of the product by the U.S. Dept. of Agriculture and does not imply its approval to the exclusion of other products or vendors that also may be suitable.

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Table 1.1. Total nutrient concentrations in dairy manure compost.

	N	P	K	Ca	Mg	S	Fe	Mn	Zn	Cu	B
	(% )						(mg·kg <sup>-1</sup> )				
Mean	1.77	0.42	0.76	1.8	0.56	0.23	3486.33	351.67	134.67	758	24.63
S.E. <sup>1</sup>	0.11	0.01	0.01	0.03	0.01	0	147.74	6.69	1.86	9.45	0.37

<sup>1</sup> Standard error (n=3)

Table 1.2. Nutrient Concentrations with standard errors (n=5) in chrysanthemum leaves grown in 0% DMC with lime (Control) and in the substrate with 20% DMC without lime (20% DMC) at the end of Expt. 2 (77days after transplant).

Macro Nutrients (% of Dry Weight)						
Treatment	N	P	K	Ca	Mg	S
Control	6.40±0.17	0.78±0.11	5.21±0.16	2.15±0.12	0.56±0.02	0.19±0.02
20% DMC	5.81±0.11	0.59±0.01	5.98±0.27	1.77±0.04	0.44±0.01	0.27±0.00
Significance <sup>1</sup>	NS	NS	*	*	**	**
Micro Nutrients (mg kg <sup>-1</sup> of Dry Weight)						
Treatment	B	Cu	Fe	Mn	Zn	Na
Control	65.3±10.5	9.1±1.5	155.1±18.3	308.8±23.6	38.4±1.7	883.9±21.0
20% DMC	53.4±2.8	15.8±1.3	269.6±42.2	462.4±28.2	58.8±9.9	412.8±51.0
Significance	NS	*	*	**	NS	***

<sup>1</sup>T-test significance \*, \*\* and \*\*\* at P≤0.05, 0.001, and 0.0005, respectively.

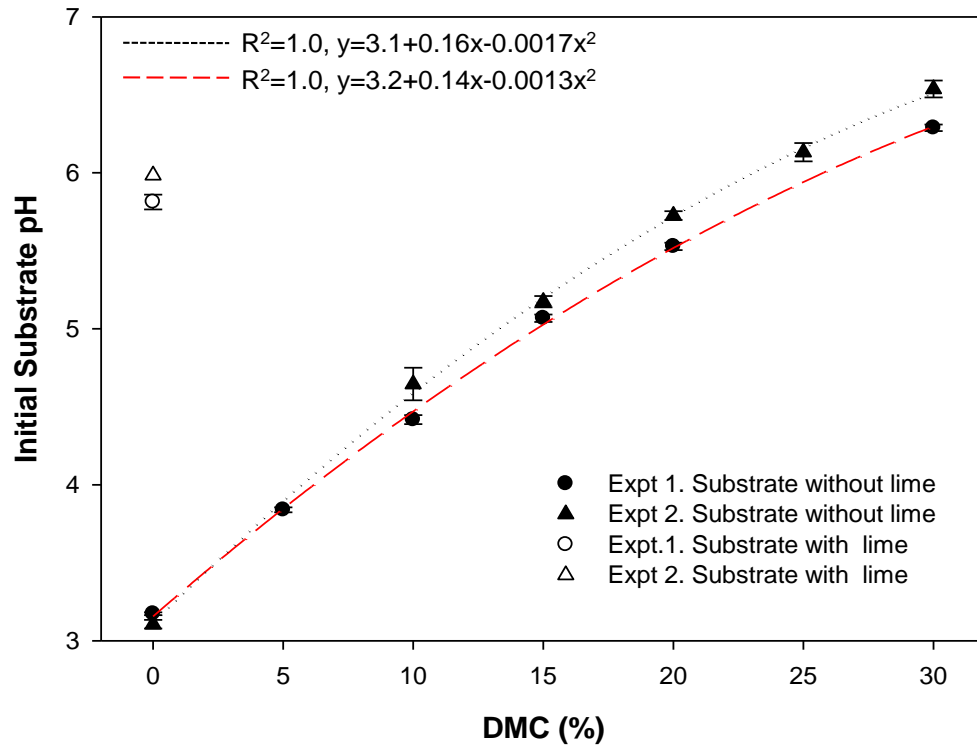


Figure 1.1. Response of initial substrate pH to volumetric quantity of dairy manure compost (DMC) in Expts. 1 and 2.

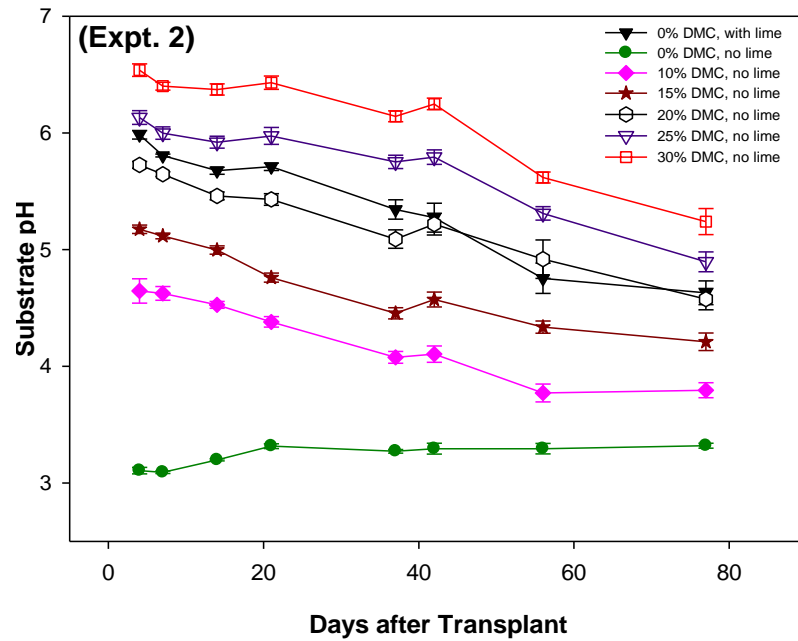
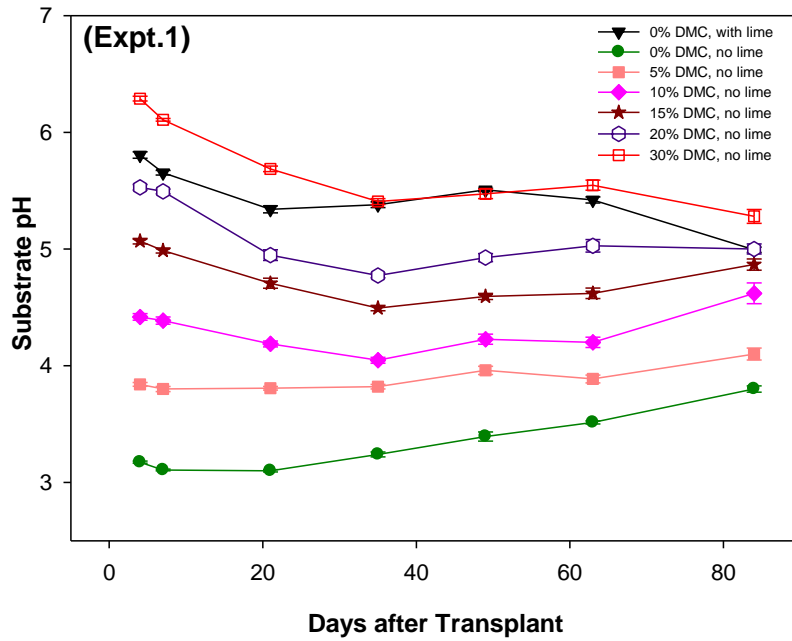


Figure 1.2. Substrate pH levels over time in treatments with and without limestone or dairy cow manure compost (DMC) in Expts. 1 and 2.

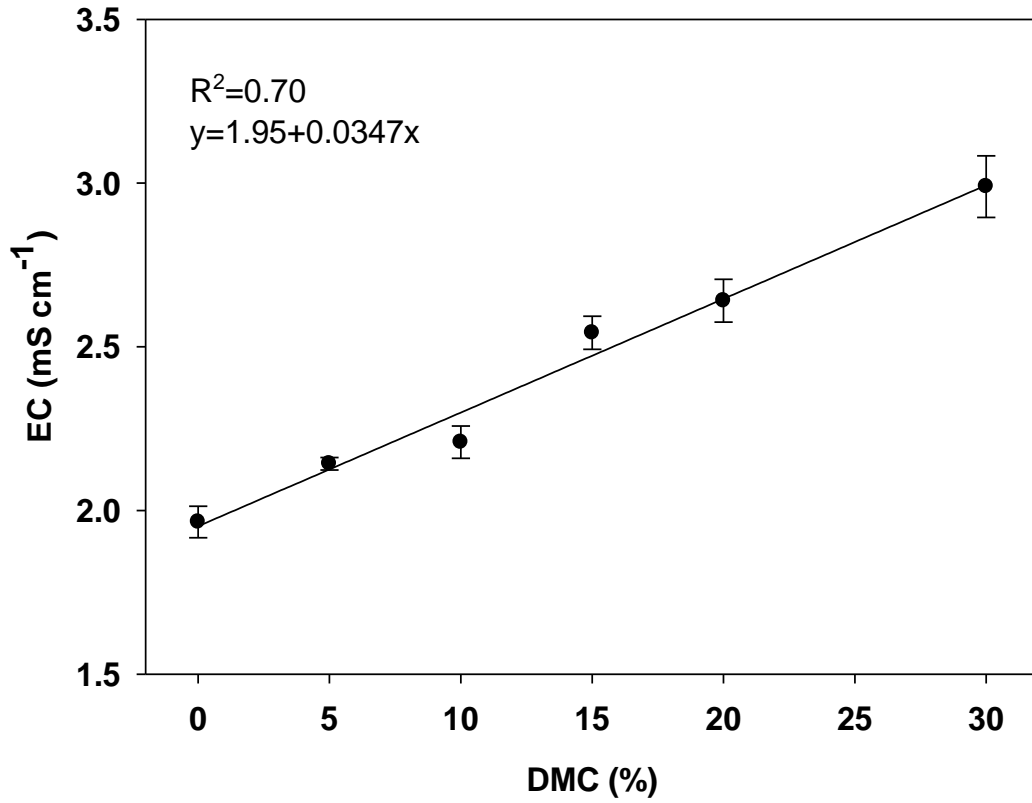


Figure 1.3. Average EC ( $\pm$ standard error, n=5) of substrates containing 0 to 30% dairy cow manure compost (DMC) in Expt. 1 after transplanting by the pour-through method.

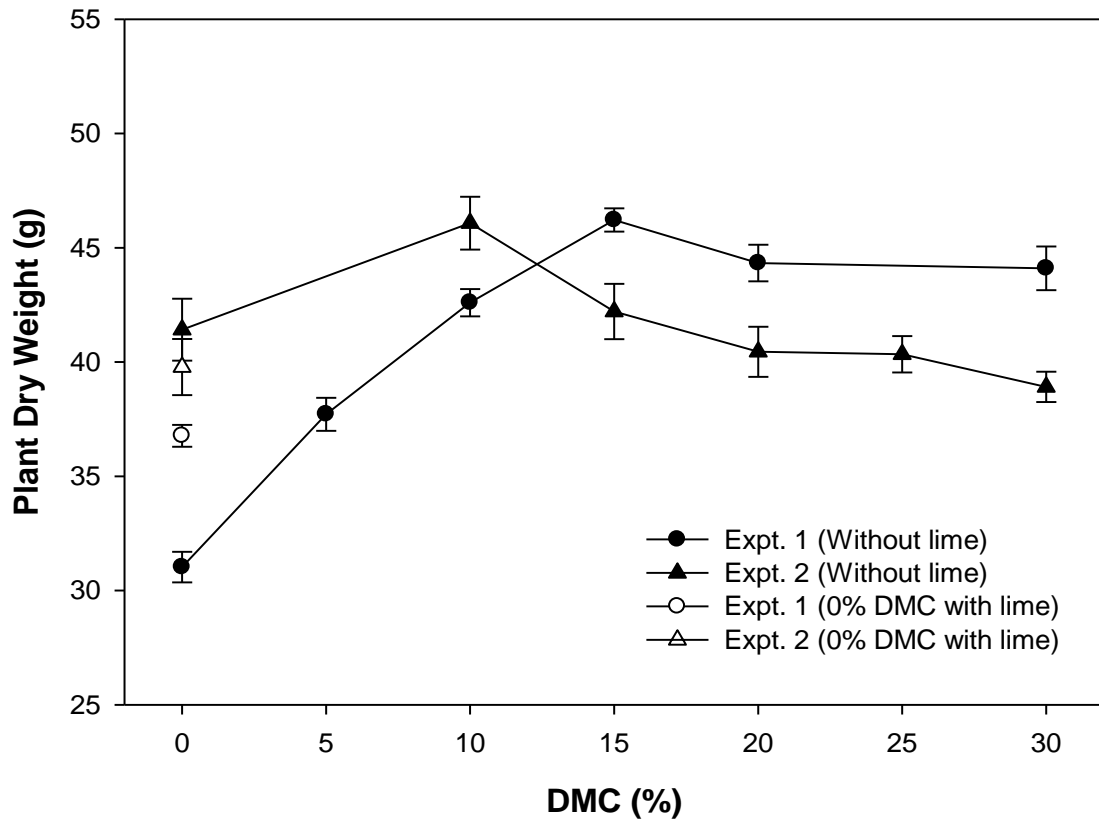


Figure 1.4. Plant shoot dry weight with increasing dairy cow manure compost (DMC) from 0 to 30% without lime and 0% DMC with lime.

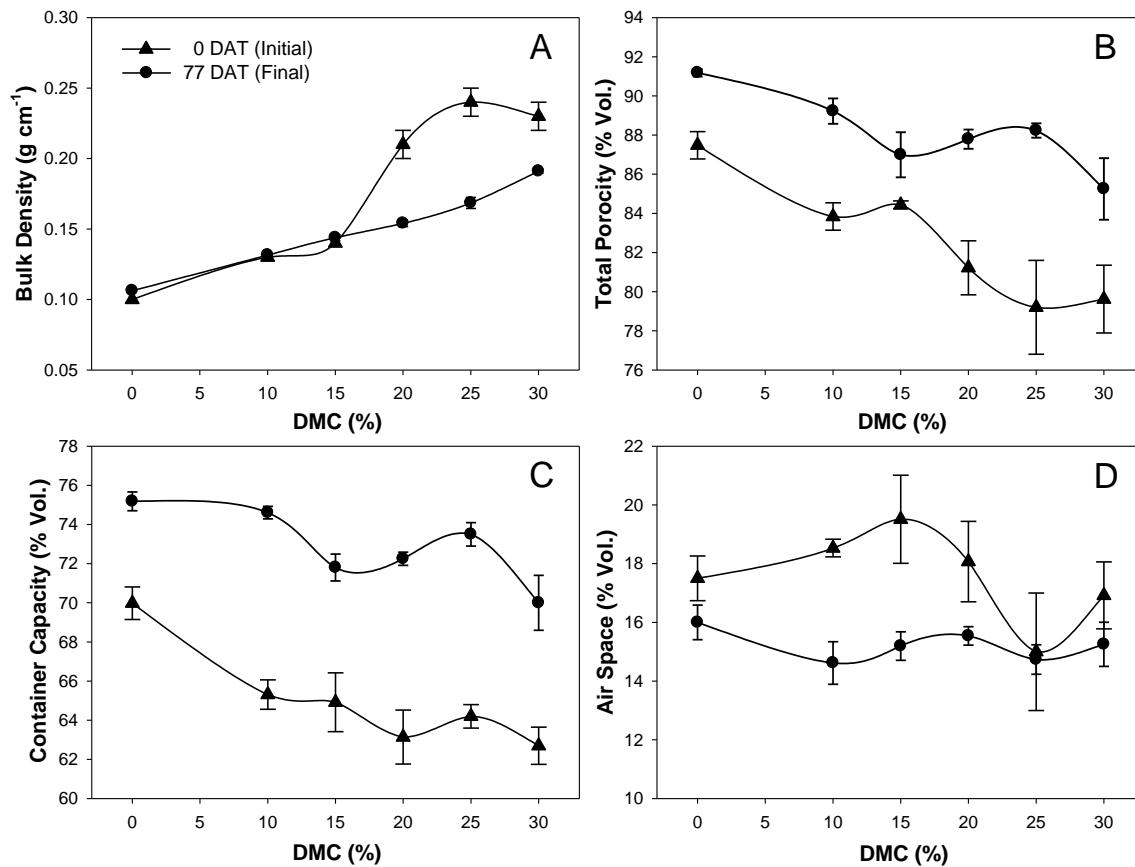


Figure 1.5. Average substrate physical properties in a 7.6 cm tall column at the beginning and the end of Expt. 2, including A) dry bulk density ( $D_b$ ), B) total porosity (TP), C) container capacity (CC), and D) air space (AS).

## CHAPTER 2

### **Predicting Calcite (CaCO<sub>3</sub>) Requirements of Sphagnum Peat Moss from pH Titration Curves**

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#### **Abstract**

Liming materials are required to neutralize acidity in peat moss to make it a suitable substrate for growing containerized plants. A series of time-consuming incubations of peat:lime mixtures are typically used to determine the liming rate to achieve a desired pH. Our objective was to develop a rapid, acid-base titration method for predicting calcite (CaCO<sub>3</sub>) requirement of sphagnum peat moss. Peat moss with an initial pH of 3.7 was titrated in aqueous suspension with 0.1 M NaOH or 0.1 M HCl solution from pH 3 to 11 (forward titration), then from pH 11 to 3 (backward titration). Because of hysteresis, the forward and backward titration curves were averaged to predict calcite requirements. For comparison, peat moss samples were incubated in plastic bags for 13 days after applying calcite at rates between 0 and 1.5 mol CaCO<sub>3</sub> kg<sup>-1</sup> peat moss (dry weight basis), and pH was monitored until

a steady-state was established. The pH achieved by calcite incubation could be predicted up to pH 6.2 by the averaged acid-base titration curve. Above pH 6.2, calcite solubility limits caused a deviation between the two methods as the pH in the incubation experiment reached a plateau with increasing calcite additions. Geochemical speciation calculations showed that calcite solubility limits imposed by atmospheric CO<sub>2</sub> and dissolved Ca<sup>2+</sup> activity restricts the maximum pH achievable. Nevertheless, the acid-base titration is a quick method for accurately predicting calcite requirements of acidic peat moss for target pH levels ≤ 6.2.

## **Introduction**

Sphagnum peat moss has been a key component of container root substrate for the past 60 years. The acidity of sphagnum peat moss, with pH values in the range 3.0 to 4.0 in water (Bailey, 1996; Bunt, 1976), significantly influences the final pH of peat-base substrate (Bailey, 1996). For most greenhouse crops, the optimal pH recommended for peat-base soilless substrate is between 5.4 to 6.5 (Bailey, 1996; Handreck, 2002; Nelson, 2003; Peterson, 1981). Therefore, liming is required for soilless substrates containing acidic organic components such as peat moss (Bishko, Fisher and Argo, 2002; Elliott, 1996; Rosenbaum and Sartain, 1982).

Since the pH acidity and buffering capacity varies depending on type and origin of peat moss (Bunt, 1976), the lime requirement for peat-based substrate is often assessed using titration curves. The incubation method – measuring the pH-response of peat or peat-base substrate after incubating with a liming source at incremental rates is commonly used. The

pH-response depends on the physical and chemical properties of the liming source as well as the application rate (Bishko et al., 2002; Huang et al., 2007; Rippy and Nelson, 2005; Rosenbaum and Sartain, 1982). The effect of limestone particle size on peat pH-response has been studied by Huang et al. (2007) and Rippy et al. (2007). With increasing particle diameter, initial solubility decreased while the residual component increased. Later dissolution of the residual component resulted in continual neutralization of peat moss acidity for extended periods of time. This residual effect of limestone resulted in under- or over-estimation of lime requirement and extended by two to three weeks the necessary incubation time (Fisher et al., 2006; Rosenbaum and Sartain, 1982) needed to complete react the lime and develop an accurate curve (Marshall, Young and Gregson, 1995).

The length of incubation time required for complete dissociation of added base is also affected by the strength of bases added (Rippy and Nelson, 2005). The pH-response to NaOH (strong base) was immediate, while an incubation period was required when peat moss was treated with  $\text{Ca}(\text{OH})_2$  (a weak base). The lime requirement should be determined on the basis of the desired increase in pH from the initial pH and on the pH buffering capacity of peat moss. The acidity of sphagnum peat moss consists of active and reserve (exchangeable) acidity, which is indicated by initial pH measurement and by pH buffering capacity, respectively (Bunt, 1976). The initial pH is related to hydrogen ions ( $\text{H}^+$ ) in the peat moss that readily enter solution when the peat is hydrated. The pH buffering capacity is derived from the pH-dependent dissociation of acidic functional groups in the peat.

The most abundant and significant acidic functional groups are carboxylic and phenolic groups, which bind proton and other cations (Aitken and Moody, 1994; Christl and Kretzschmar, 2001; Cooke et al., 2007; Gustafsson, 2005; Kinniburgh et al., 1996; Orsetti et al., 2009; Ramos et al., 1999). The carboxylic groups dissociate between pH 3 and 6, while the phenolic groups dissociate mainly in the neutral to alkaline pH range (Kam and Gregory, 2001; Swift, 1999).

The lengthy time requirements and inaccurate assessment of actual acidity of peat moss can be overcome by strong acid/base titration to determine the inherent pH buffering capacity of the peat moss. Several titration techniques and methods to measure the pH buffering capacity have been developed to determine the lime requirement for soil and organic matter (Aitken and Moody, 1994; Cooke et al., 2007; Follett, 1983; Godsey et al., 2007; Janos et al., 2008; Liu et al., 2005; E. O. McLean et al., 1966; Rippy and Nelson, 2005). Continuous potentiometric titration of organic soils was used to investigate the total buffering capacity (Kuznetsov, 2007). The potentiometric titration with incremental additions of acid or base has been investigated to test acid-base properties of humic substances (Christl and Kretzschmar, 2001; Cooke et al., 2007; Ritchie and Perdue, 2003; Ritchie and Perdue, 2008), because the acidic functional groups of humic substances in soil play a role in the soil pH buffering capacity. The objectives of this study were 1) develop a rapid, easy titration technique that accurately quantifies the acidity of sphagnum peat moss (pH buffering

capacity), and 2) apply the method to determine calcite ( $\text{CaCO}_3$ ) requirement for adjusting pH of sphagnum peat moss based on the strong acid/base acidity titration curve.

## **Materials and Methods**

The reserve acidity of sphagnum peat moss was quantified on a dry weight basis instead of a volume basis because the bulk density of sphagnum peat moss can be altered by handling and its water content. Therefore, the water content (W) of fresh peat moss sampled was measured as [(fresh weight-dry weight)/fresh weight]. Dry weight (g,  $P_d$ ) and initial water amount (ml,  $W_i$ ) of the given amount of fresh peat moss sample (g,  $P_f$ ) was estimated as  $P_d = P_f - (P_f \times W)$  and  $W_i = P_f - P_d$ , respectively.

### I. Strong Acid-Base Titration of Peat Moss

Standard acid (0.1 M HCl) and base (0.1 M NaOH) titrant solutions were prepared using de-gassed, de-ionized water prepared by bubbling  $\text{N}_2(\text{g})$  through the boiled deionized water. The potentiometric acid-base titration was performed by using an automatic titrator system (TitraLab 856, Radiometer Analytical SAS, Villeurbanne Cedex, France).

*Sample preparation.* Aqueous suspensions of sphagnum peat moss were prepared by stirring 1.5 g (dry weight basis) of fresh peat moss in 50 ml of de-gassed, de-ionized water at 500 rpm on a stir plate for 30 minutes in a 200 ml glass beaker covered with paraffin film on the top. Then, a pH electrode was inserted into the beaker. The initial pH of the peat moss suspension was measured ( $\text{pH}_i$ ). To accurately quantify the reserve (exchangeable) acidity of

peat moss, opening of the aggregated macro and micro structures of peat moss was critical to render the cation exchangeable sites available to react with added titrant solution. It was not possible to disperse the aggregates by physical methods such as sonification (Milne et al., 1994). According to the random coil model of humic substances, organic polymers are flocculated at low pH and dispersed at high pH due to the electrostatic repulsion between negatively charged acidic functional groups (McLean et al., 1996). To initially induce the conformational changes to disaggregate and hydrate the peat moss by electrostatic repulsion, the peat moss suspension sample was increased pH 11 by addition of 0.1M NaOH. The total volume of added NaOH solution ( $\text{NaOH}_t$ ) was recorded. Then the pH of the peat moss suspension sample was decreased to pH 3 by addition of 0.1M HCl. The total added 0.1M HCl solution was recorded as  $\text{HCl}_t$ .

*Forward- and Backward-Titration.* After the peat moss suspension cycled through the base-acid treatment to hydrate and disperse, it was forward titrated from pH 3 to 11 with 0.1M NaOH solution while stirring. The automatic titrator added NaOH solution at rate of 1/7 ml per minute as min./Max speed setting and recorded pH values ( $\text{pH}_f$ ) and volume of NaOH added (ml,  $\text{NaOH}_{\text{add}}$ ) after each addition until pH 11 was achieved. The total volume of NaOH solution added was recorded as  $\Sigma\text{NaOH}_{\text{add}}$  (ml). Then, the peat moss suspension was backward titrated from pH 11 to 3 by additions of 0.1 M HCl. Again, added volumes of HCl ( $\text{HCl}_{\text{add}}$ ) and pH levels ( $\text{pH}_b$ ) were recorded. And the total volume of HCl solution added was recorded as  $\Sigma\text{HCl}_{\text{add}}$  (ml).

*Calculations.*

- 1) The forward titration curve - Added  $\text{OH}^-$  derived from each addition of 0.1M NaOH solution neutralized the acidity of the peat moss. The neutralized active acidity indicated as increase in pH was calculated by subtracting the amount of  $\text{H}^+$  ions at  $\text{pH}_f$  from that of at pH 3. Then, pH buffering was calculated as following when  $X(\text{ml})=50+W_i+\text{NaOH}_t+\text{HCl}_t$ :

$$\begin{aligned} & \text{pH buffering (forward, mol H}^+ \text{ kg}^{-1} \text{ of dry peat moss)} \\ & = \text{The reserve acidity between pH 3 and pH}_f \\ & = (\text{added NaOH} - \text{change in dissolved OH}^-) / \text{mass of peat} \\ & = [\{\text{added NaOH}\} - \{\text{neutralized active acidity}\}] / P_d \times 10^3 \\ & = [(0.1 \times \text{NaOH}_{\text{add}}) - \{10^{-3}(X) - 10^{-\text{pH}_f}(X + \text{NaOH}_{\text{add}})\}] / P_d \times 10^3 \\ & = [(0.1 \times \text{NaOH}_{\text{add}}) - \{(10^{-3} - 10^{-\text{pH}_f})(X) - 10^{-\text{pH}_f}(\text{NaOH}_{\text{add}})\}] / P_d \times 10^3 \\ & = \text{RA}_{f3} \end{aligned}$$

- 2) The backward titration - The peat moss suspension of pH 11 decreased by incremental additions of  $\text{H}^+$  derived from HCl solution. When the volume of peat moss suspension was  $Y(\text{ml})=X+\Sigma\text{NaOH}_{\text{add}}$  before the backward titration, the pH buffering at  $\text{pH}_b$  was calculated as following:

$$\begin{aligned} & \text{pH buffering (backward, mol H}^+ \text{ kg}^{-1} \text{ of dry peat moss)} \\ & = \text{The weak acid functional groups between pH 11 and pH}_b \\ & = (\text{added HCl} - \text{change in dissolved H}^+) / \text{mass of peat} \end{aligned}$$

$$\begin{aligned}
&= \{[(\text{added HCl}) - (\text{increase in active acidity})]\}/P_d \times 10^3 \\
&= [(0.1 \times \text{HCl}_{\text{add}}) - \{10^{-\text{pH}_b}(\text{Y} + \text{HCl}_{\text{add}}) - 10^{-11}(\text{Y})\}]/P_d \times 10^3 \\
&= [(0.1 \times \text{HCl}_{\text{add}}) - \{(10^{-\text{pH}_b} - 10^{-11})(\text{Y}) + 10^{-\text{pH}_b}(\text{HCl}_{\text{add}})\}]/P_d \times 10^3 \\
&= \text{RA}_{b11}
\end{aligned}$$

In order to plot forward- and backward- titration curves in one graph, the backward-titration curve was transformed by the following equation when  $Z(\text{ml}) = \text{Y} + \Sigma \text{HCl}_{\text{add}}$ :

$$\begin{aligned}
&\text{The reserve acidity (mol H}^+ \text{ kg}^{-1} \text{ of dry peat moss) between pH 3 and pH}_b \\
&= [(\text{total reserve acidity}) - \{(\text{added HCl}) - (\text{increase in active acidity})\}]/P_d \times 10^3 \\
&= [(0.1 \times \Sigma \text{HCl}_{\text{add}}) - (10^{-3} \times Z) + (10^{-11} \times \text{Y}) - (0.1 \times \text{HCl}_{\text{add}}) + 10^{-\text{pH}_b} \times (\text{Y} + \text{HCl}_{\text{add}}) - (10^{-11} \times \text{Y})]/P_d \times 10^3 \\
&= \{0.1(\Sigma \text{HCl}_{\text{add}} - \text{HCl}_{\text{add}}) - 10^{-3}(Z) + 10^{-\text{pH}_b}(\text{Y} + \text{HCl}_{\text{add}})\}/P_d \times 10^3 \\
&= \text{RA}_{b3}
\end{aligned}$$

3) Hysteresis between forward-backward titration curves - The reserve acidities of peat moss determined from the forward- and backward- titration curve did not agree with each other due to the well-know hysteresis phenomena between forward-backward curves for acid-base titrations of humic substances (Davis and Mott, 1981; Marshall et al., 1995; Paxeus and Wedborg, 1985; Ritchie and Perdue, 2003; Santos et al., 1999). The forward and backward titration results in this study agreed with Davis and Mott (1981) who reported that there were more acidic groups determined by the backward titration curve than by the forward titration curve.

The hysteresis exhibited from the forward and reverse titrations has been shown to decreased by repeating titrations of fulvic acid (Paxeus and Wedborg, 1985) and humic acid (Marshall et al., 1995). Also, Davis and Mott (1981) reported the difference between forward and reverse titration curves was smaller when less concentrated sample solution was used. In this study, the reserve acidity from forward ( $RA_{f3}$ ) and backward ( $RA_{b3}$ ) titration curves and the averaged reserve acidity from both titration curves,  $[(RA_{f3}+RA_{b3})/2]$  were tested. Amounts of  $CaCO_3$  required to achieve a target pH in peat moss were calculated from each of the three curves. When these amounts were incubated with peat moss, the quantity from the average curve came closest to achieving the target pH. Amounts calculated from the  $RA_{f3}$  and  $RA_{b3}$  curves resulted in lower and higher pH value than the targeted pH (data not shown). Therefore the reserve acidity of peat moss was recalculated by averaging acid-base titration curves as a function of pH.

4) Conversion from  $OH^-$  to  $CO_3^{2-}$  requirement - When agricultural lime or calcite is incorporated with peat moss, weak base carbonates ( $CO_3^{2-}$ ) rather than strong base  $OH^-$  neutralize the acidity of peat moss. While one mole of NaOH will neutralize one mole of acidity ( $H^+$ ), between 0.5 and 1 mol of  $CO_3^{2-}$  neutralizes 1 mol of  $H^+$ , depending on pH (Lindsay, 1979). The pKa values of  $CO_3$  are 6.36 ( $H_2CO_3^0 \rightleftharpoons HCO_3^-$ ) and 10.33 ( $HCO_3^- \rightleftharpoons CO_3^{2-}$ ). Below pH 6.36, the major carbonate species is  $H_2CO_3^0$ , which forms from  $CO_3^{2-}$  by combining 2  $H^+$  ions. However between pH 6.36 and 10.33 the major carbonate species is  $HCO_3^-$  which replaces only one  $H^+$  ion. The distributions of each carbonate species as a

function of pH as determined from the acid dissociation reaction were used to calculate calcite requirements. The mole fraction (MF) of  $\text{H}_2\text{CO}_3$ ,  $\text{HCO}_3^-$ , and  $\text{CO}_3^{2-}$  is represented as following: When  $[\text{H}^+]=10^{\text{pH}}$ ,  $[\text{H}_2\text{CO}_3^0]=10^{-1.46}$ ,  $[\text{HCO}_3^-]=10^{-7.82}/[\text{H}^+]$ , and  $[\text{CO}_3^{2-}]=10^{-18.15}/[\text{H}^+]^2$ ,

$$\text{MF}_{\text{H}_2\text{CO}_3^0} = \frac{[\text{H}_2\text{CO}_3^0]}{[\text{H}_2\text{CO}_3^0]+[\text{HCO}_3^-]+[\text{CO}_3^{2-}]}, \text{MF}_{\text{HCO}_3^-} = \frac{[\text{HCO}_3^-]}{[\text{H}_2\text{CO}_3^0]+[\text{HCO}_3^-]+[\text{CO}_3^{2-}]},$$

$$\text{MF}_{\text{CO}_3^{2-}} = \frac{[\text{CO}_3^{2-}]}{[\text{H}_2\text{CO}_3^0]+[\text{HCO}_3^-]+[\text{CO}_3^{2-}]}$$

The predicted  $\text{CaCO}_3$  requirement to bring peat moss to a given pH level is calculated as follows:

$$\text{CaCO}_3 \text{ requirement} = \text{TA} * (0.5 * \text{MF}_{\text{H}_2\text{CO}_3^0} + \text{MF}_{\text{HCO}_3^-})$$

## II. Peat Incubation Method

Fresh sphagnum peat moss, 20 g (dry weight equivalent), were mixed thoroughly in plastic bags with calcium carbonate ( $\text{CaCO}_3$  USP/FCC, Fisher Scientific, Pittsburgh, PA) at the rate of 0.3, 0.6, 0.9, 1.2, 1.5 mol per kg of peat moss (dry weight basis). Then, 140 mL of deionized water were added to each bag to saturate the peat moss. Substrate solution was subsequently extracted by squeezing the plastic bags of saturated peat moss at 1, 3, 4, 8, and 14 days after the start of incubation. Substrate solution pH was measured. The pH electrode

was calibrated with pH 4.0 and 7.0 standard buffers solutions after every 10 measurements. After measuring pH of the solution, it was added back into the plastic incubation bag. The incubation titration curve was developed based on the pH values measured over the 14 days of incubation.

## **Results and Discussions**

### *Hysteresis between forward- and backward titration curves*

The acid/base titration curves of sphagnum peat moss measured by forward- and backward- titrations showed hysteresis (Fig. 2.1). The total acidity ( $H^+$ ) between pH 3 and 6 was measured as 0.77, 1.45, and 1.11 mol · kg<sup>-1</sup> of sphagnum peat moss (dry weight basis) for the forward-, backward- and averaged-titration curves, respectively. Hysteresis has been proposed to result from physical and chemical factors affecting the protonation and deprotonation of acidic groups on humic substances during the forward- and backward-titration (Sposito et al., 1977; Varney et al., 1998; Paxeus and Wedborg, 1985; Marshall et al., 1995; Milne et al., 1995; Tombacz, 1999). During sample preparation, the pH of sphagnum peat moss suspension was increased by addition of 0.1M NaOH for the purpose of dispersing and hydrating the aggregated humic substance in sphagnum peat moss, then the pH was decreased by addition of 0.1M HCl to set the forward titration starting point of pH 3. After this procedure, it was possible that the opened structure of humic substance was not permanently fixed and could still respond to the pH level of the suspension. Also, Milne et al. (1995) reported that disaggregation or unfolding of the humic acid molecules occurs rapidly

during the forward titration (increasing pH) while aggregation or condensation occurs relatively slower during reverse titration. Therefore, re-aggregated of humic substance at pH 3 may have caused lower acidity measurement during the forward titration than during backward titration. From a chemical aspect, the higher acidity measured during backward-titration could be due to additional protons released from deprotonation of fulvic acid during forward-titration (Paxeus and Wedborg, 1985).

#### CaCO<sub>3</sub> Requirement Curve from acid-base titration curves

According to averaged titration curve of sphagnum peat moss (Fig. 2.1) the total acidity to adjust the peat moss pH 3.3 to targeted pH 6.0 was 0.96 mol of H<sup>+</sup> per 1 kg of peat moss (dry weight basis) calculated by subtracting the acidity (=0.15 mol H<sup>+</sup>) at pH 3.3 from the acidity (=1.11 mol H<sup>+</sup>) at pH 6.0. If CaCO<sub>3</sub> is only dissociated into Ca<sup>2+</sup> and carbonate (CO<sub>3</sub><sup>2-</sup>), 0.48 mol of CaCO<sub>3</sub> per 1 kg of peat moss (dry weight basis) is required to neutralize 0.96 mol of acidity (Fig. 2.2). However, the targeted pH 6.0 was not achieved by addition of 0.48 mol of CaCO<sub>3</sub> (Fig. 2.3) because of various carbonate species (H<sub>2</sub>CO<sub>3</sub><sup>0</sup>, HCO<sub>3</sub><sup>-</sup>, or CO<sub>3</sub><sup>2-</sup>) depending on pH in water solution (Lindsay, 1979). Lindsay (1979) presented the mole fraction distribution of the various carbonate species as function of pH (Equation 1). At pH 6.36, 1 mol of CaCO<sub>3</sub> can neutralize only 1.5 mol of H<sup>+</sup> instead of 2 moles, because the mole fraction of HCO<sub>3</sub><sup>-</sup> to H<sub>2</sub>CO<sub>3</sub><sup>0</sup> is 1. The distribution of CO<sub>3</sub><sup>2-</sup> species increases from pH 8.1, and the mole fraction of to HCO<sub>3</sub><sup>-</sup> to CO<sub>3</sub><sup>2-</sup> is 1 at pH 10.33 (Equation 1) (Lindsay, 1979). When the effect of pH on the mole fraction distribution of various carbonate species are

accounted (Fig. 2.2,  $\Delta$  curve), the  $\text{CaCO}_3$  requirement to achieve targeted pH 6.0 from original peat pH 3.3 was estimated at 0.51 mol of  $\text{CaCO}_3$  instead of 0.48 mol (Fig. 2.2). The estimated  $\text{CaCO}_3$  requirement,  $0.51 \text{ mol kg}^{-1}$  is in agreement with the result of incubation method test (Fig. 2.3). Therefore, the distribution of  $\text{H}_2\text{CO}_3^0$ ,  $\text{HCO}_3^-$ , or  $\text{CO}_3^{2-}$  as function of solution pH should be considered to determine accurate  $\text{CaCO}_3$  requirement.

*The comparison between estimated pH and measured pH level.*

From the average strong acid/base curve, quantities of  $\text{CaCO}_3$ , corrected for carbonate species, of 0.3, 0.6, 0.9, 1.2, and 1.5 mol of  $\text{CaCO}_3$  per kg of sphagnum peat moss (dry weight basis) were calculated to achieve pH levels of 4.9, 6.7, 8.8, 10.1, and 10.8. However, when these quantities of  $\text{CaCO}_3$  were incubated with peat moss the resulting pH levels were  $4.8(\pm 0.19)$ ,  $6.5(\pm 0.25)$ ,  $7.2(\pm 0.03)$ ,  $7.3(\pm 0.08)$ , and  $7.4(\pm 0.06)$  (Fig. 2.3 and 2.4). The predicted  $\text{CaCO}_3$  requirement curve based on the averaged titration curve agreed with the result of the incubation method only up to pH about 6.2 (Fig. 2.4), because the pH levels measured in the incubation method fitted into asymptotic curve as a function of  $\text{CaCO}_3$  addition.

Although the  $\text{CaCO}_3$  (fine powder, 98% purity) used in this study was anticipated to dissolve immediately without residual effect, at least 4-days incubation was required to achieve a stabilized pH level at  $0.6 \text{ mol kg}^{-1}$  and higher rates of added  $\text{CaCO}_3$  (Fig. 2.5). Undissolved  $\text{CaCO}_3$  was visually observed even after 14 days of incubation by its grayish color in the solution squeezed from peat moss treated with the higher rate of  $\text{CaCO}_3$  due to limited

solubility of  $\text{CaCO}_3$  in water ( $0.015 \text{ g}\cdot\text{L}^{-1}$ ). Solubility of  $\text{CaCO}_3$  is also suppressed by decreased concentration of hydrogen ion ( $\text{H}^+$ ), increased  $\text{Ca}^{2+}$  concentration, and increased partial pressure of  $\text{CO}_2$  (Equation 2). As added  $\text{CaCO}_3$  was dissolved in the incubation method, decreases in  $\text{H}^+$  and increases in  $\text{Ca}^{2+}$  concentrations lead to diminishing proportions of  $\text{CaCO}_3$  dissociating and thus, less  $\text{H}^+$  neutralization than predicted by the amount of  $\text{CaCO}_3$  applied.

Acid-base titration technique can quickly and accurately quantify the acidity of sphagnum peat moss compared with an incubation method. Based on the  $\text{CaCO}_3$  requirement curve converted from the acid-base titration curves, required  $\text{CaCO}_3$  to neutralize the sphagnum peat moss acidity to a given pH level can be determined up to pH 6.2.

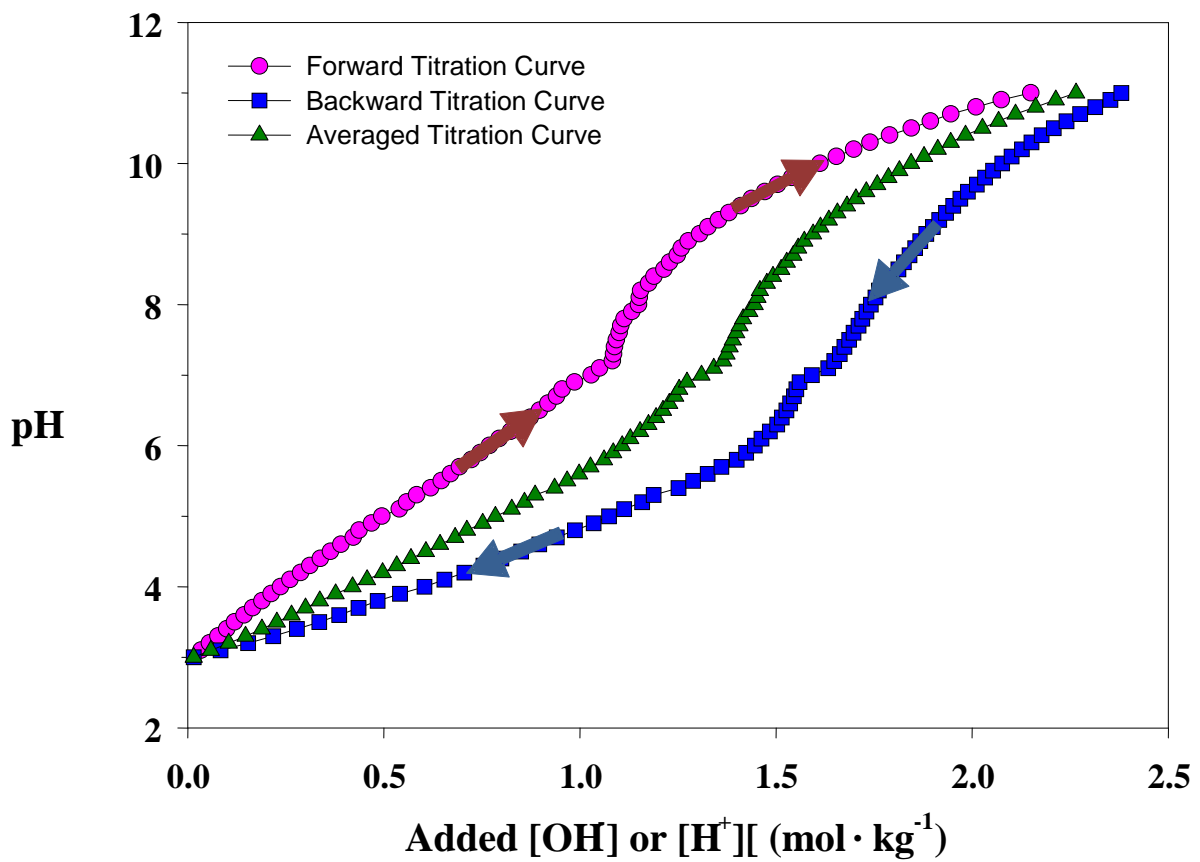


Figure 2.1. Acid/base titration curves of sphagnum peat moss from forward- and backward-titrations and the averaged curve.

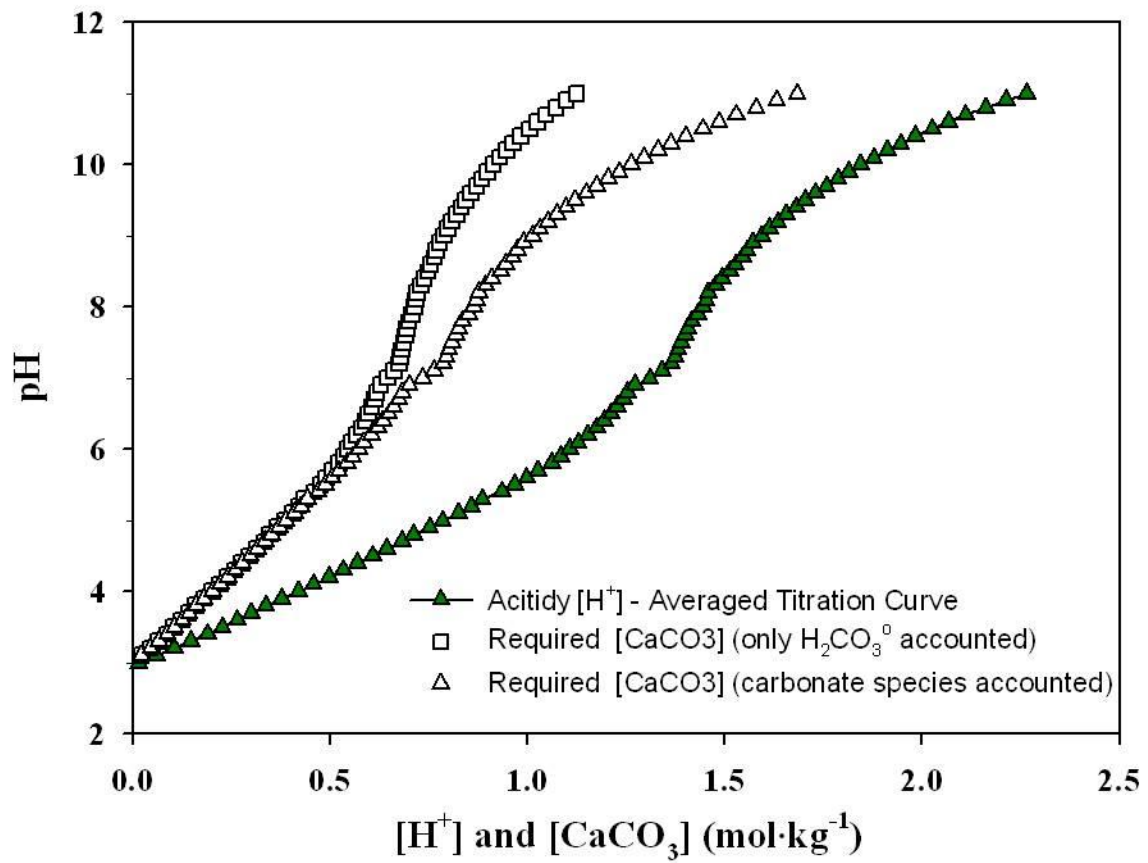


Figure 2.2. Predicted  $CaCO_3$  requirement based on the acidity of sphagnum peat moss from the averaged titration curve only.

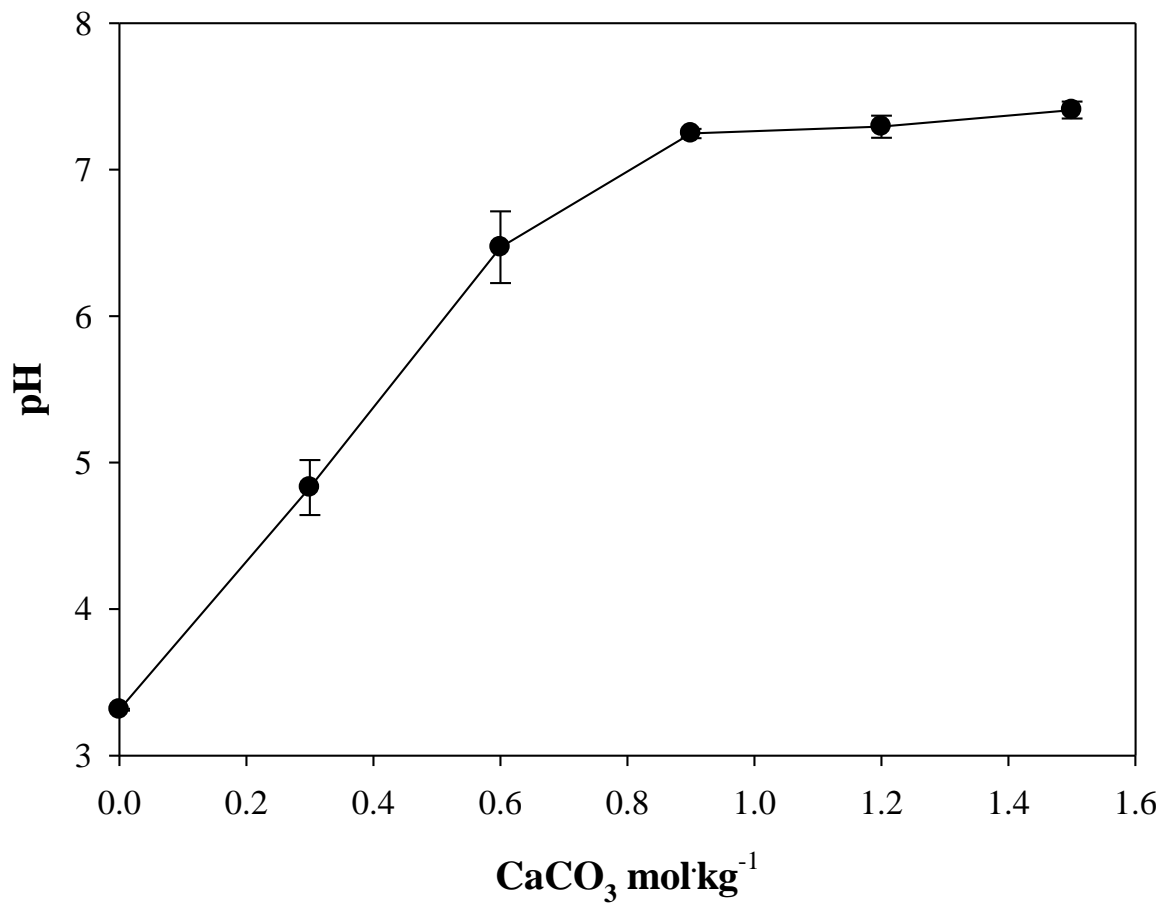


Figure 2.3. The pH response curve of sphagnum peat moss as a function of CaCO<sub>3</sub> addition.

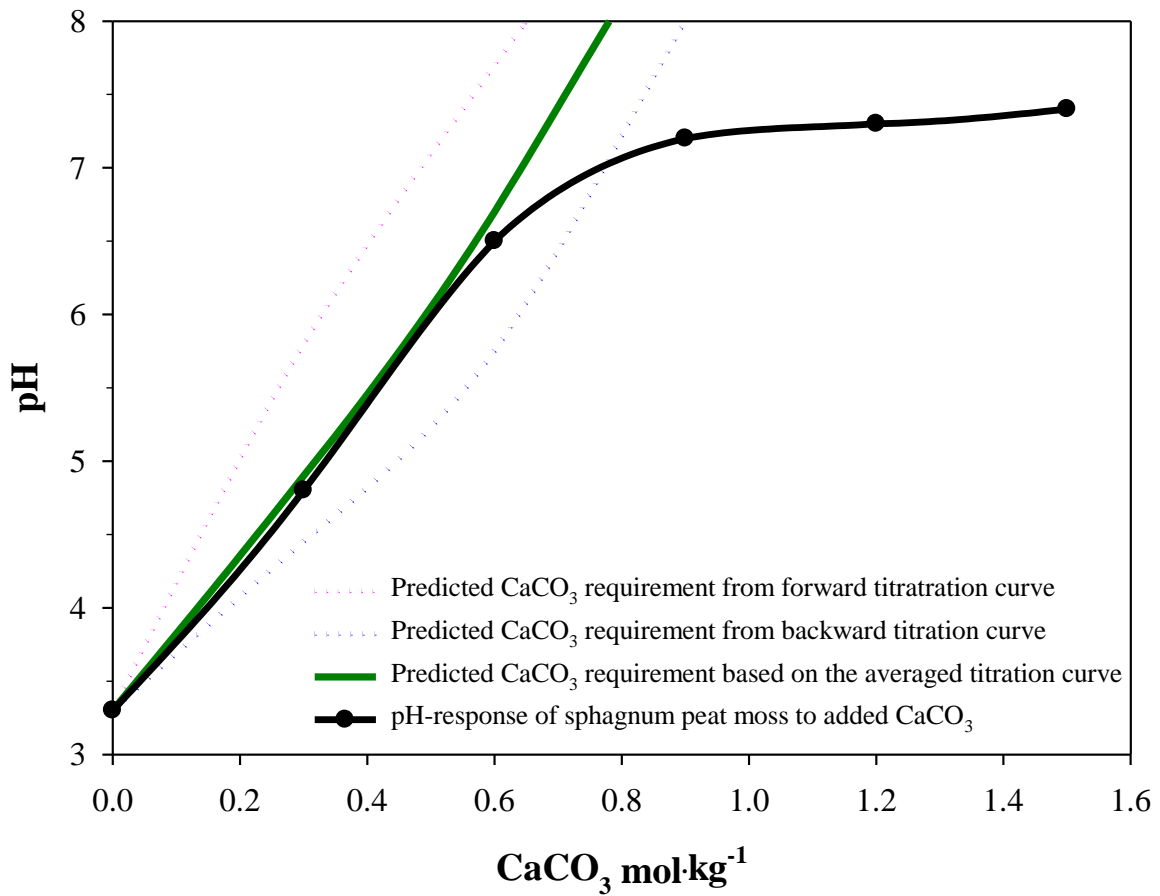


Figure 2.4. Comparison between predicted CaCO<sub>3</sub> requirement curves based on the acid-base titration curves of sphagnum peat moss and the effect of added CaCO<sub>3</sub> on the pH of sphagnum peat moss.

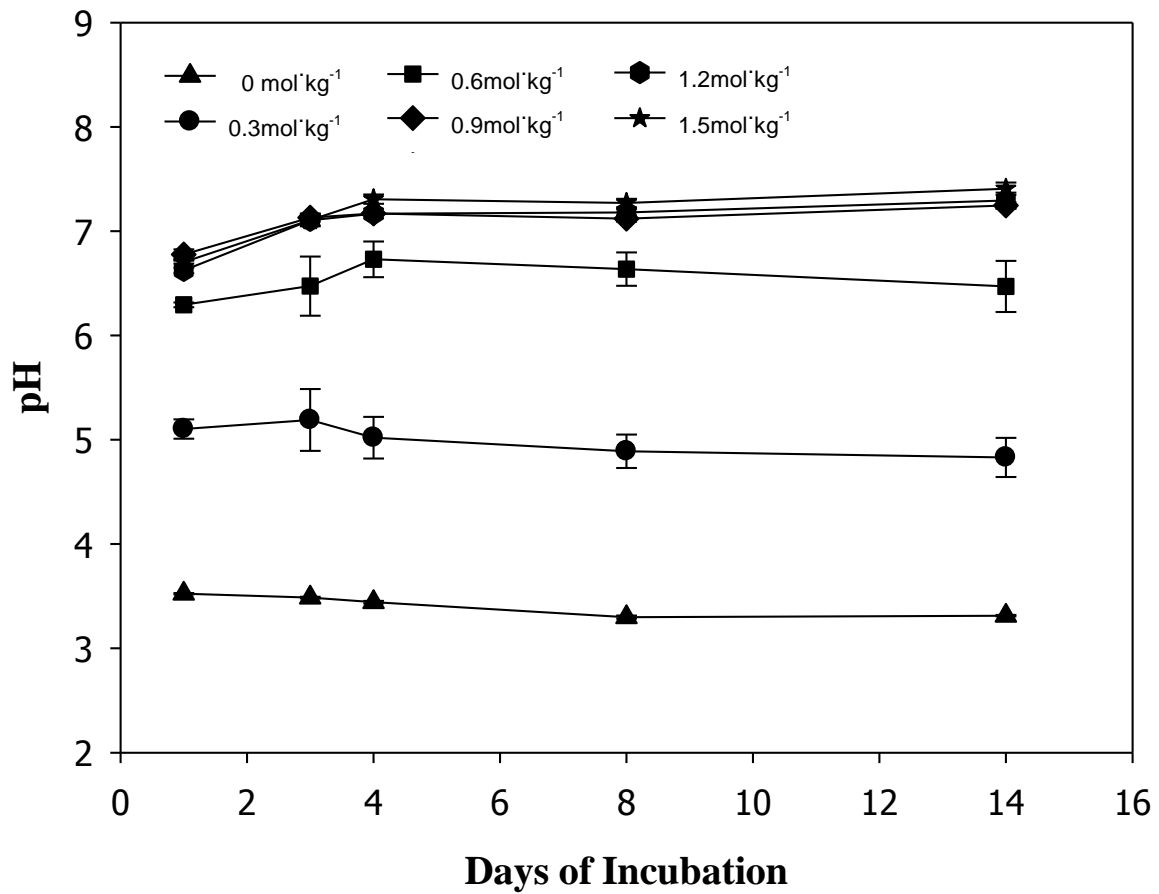
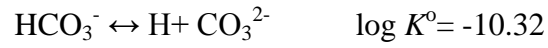


Figure 2.5. The pH response of sphagnum peat moss treated with addition of 0, 0.3, 0.6, 0.9, 1.2, and 1.5 mol of CaCO<sub>3</sub> per kg of sphagnum peat moss (dry weight basis) over 14 days. Each symbol is the averaged pH of 5 replications; and the vertical bar is standard error.

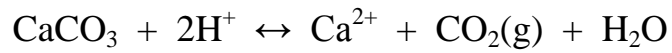
Equation 1. Dissociation of carbonic acid ( $\text{H}_2\text{CO}_3^0$ ) and bicarbonate ion ( $\text{HCO}_3^-$ )



$$\log \frac{(\text{HCO}_3^-)}{\text{H}_2\text{CO}_3^0} = \text{pH} - 6.36$$

$$\log \frac{(\text{CO}_3^{2-})}{\text{HCO}_3^-} = \text{pH} - 10.33$$

Equation 2. Dissociation of  $\text{CaCO}_3$  in equilibrium with hydrogen ion.



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## CHAPTER 3

### **Comparison of Rhizon Soil Moisture Sampler, Pour-through, and Saturated Media Extract Extractions of Container Root Substrate**

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Key words: substrate pH, substrate EC, soil testing

#### **Abstract**

The Rhizon soil moisture sampler (RS) provides a non-destructive method for extracting soil solution. The RS with a 2.5 mm outer diameter draws solution from a small area and raises the question whether it can represent the whole pot. Also, when placed diagonally or vertically in a pot more solution would be expected to be collected from the base relative to the top of the pot due to lower water container capacity at the top. When 5 RS samplers were placed horizontally at 1, 3, 5, 7, and 9 cm above the bottom of 16.5 cm diameter, 10 cm substrate depth, 1.8 L volume pots containing lettuce plants, the following average gradients from bottom to top of pot were found: EC 1.1 – 1.8 dS m<sup>-1</sup>, pH 6.3 – 5.6, and relative rate of solution extracted 1 : 0.32. When the solution from a Rhizon sampler

oriented diagonally (D-RS) from top to bottom of the pot was compared to the five horizontally and equally spaced Rhizon samplers (H-RS), pH level in the D-RS extract was equivalent to that in the lower 20% of the substrate column (H-RS<sub>1</sub>). EC in the lower three H-RS and D-RS extracts were within standard error of each other. A comparison of extracts from D-RS, pour-through (PT), and saturated media extract (SME) showed equivalent EC levels for the D-RS (1.21) and PT (1.26) but lower values for SME (0.69). The pH levels were significantly different for all three methods and were 6.6, 6.2, and 5.5 for the PT, D-RS, and SME methods, respectively. Overall, separate EC and pH standards are required for interpreting SME results compared to the other two methods which is reasonable because SME alters the solution by dilution whereas the other two do not. Similar EC standards can be used for PT and D-RS but pH levels can be lower in the D-RS procedure. Data indicated that the D-RS drew solution primarily from its lower end, far in excess of the proportion suggested by the five H-RSs.

## **Introduction**

Sampling and analysis of root substrate is necessary for fertility management in greenhouses and nurseries. Saturated media extraction (SME) (Warncke, 1986) and pour-through (PT) (Wright, 1986) methods have been predominantly used for container substrate solution extraction. Monitoring of substrate pH and soluble salt [electrical conductivity (EC)] levels is strongly recommended as a regular practice in container production (Bilderback, 2001; Cavins, 2000; Whipker et al., 2001). Substrate nutritional status and nitrogen form

requirements can be estimated from these measurements (Nelson and Faber, 1986; Warncke, 1986).

The RS sampler has been studied as a tool for monitoring nutrition in container substrates during crop production and research (Argo and Biernbaum, 1997; Bergman et al., 1994; Biernbaum et al., 1999; Cabrera, 1998). Cabrera (1998) compared the RS with the SME and PT methods for testing pH and EC in fallow peat-based container substrates. He recommended the RS method as a quick, simple and nondestructive extraction method for horticultural container production and nutrition research. The RS, due to its small size (5-10 cm length and 2.5mm diameter), draws a small amount of solution from a very limited volume of substrate. The PT and SME procedures typically provide larger volumes of solution and access much greater volumes of substrate. During crop production, EC and pH gradients can develop along the vertical substrate profile due to differences in root density, aeration, water content, and evaporation of water from the substrate surface. This suggests that positioning of the RS in the substrate profile could have a large effect on the pH, EC and nutritional values in the extract. (Handreck, 1994) indicated that a vertical pH gradient could also have an impact on the PT method since more of the lower rootball is sampled in this procedure. A further complication is the expectation that a vertically oriented RS would be expected to draw more substrate solution per unit of time from its lower than top end due to higher container capacity at the bottom of the substrate profile. To assess these potential problems and compare the RS to the PT and SME procedures, objectives undertaken in this

study included 1) quantification of the pH and EC gradients in the vertical substrate profile during plant production, 2) determination of the proportions of a sample volume that are drawn from each of five depths in a 6.5 inch azalea pot by a Rhizon sampler placed diagonally in the pot, and 3) comparison of the substrate pH and EC measurements from the D-RS, PT, and SME extraction methods.

## **Materials and Methods**

Lettuce 'Ideal COS' seedlings were transplanted on 19 Nov. into 16.5 cm diameter, 1.8L volume pots (two plants per pot) containing Fafard Growing Mix 2 (Fafard Inc., Agawam, Mass.). Plants were fertilized at each irrigation with 180 mg·L<sup>-1</sup> N solutions of neutral 17N-2.2P-14.1K water-soluble fertilizer (Greencare 17N-5P<sub>2</sub>O<sub>5</sub>-17K<sub>2</sub>O, Kankakee, IL) and were grown until roots filled the substrate profile. Plants were grown in a glass greenhouse at day and night temperature settings of 15 and 24 °C. A randomized complete block design was used with eight extraction methods, three sampling dates, and 5 replications. Since SME sampling disrupts the substrate profile and the pour-thru method alters the substrate solution, a separate set of 5 pots were used for each of the three sampling dates for each of these sampling techniques. Only one set of 5 pots was used for each of 6 RS positions, because the sampling procedure did not alter the substrate profile or solution. A total of 60 pots of lettuce were grown. The Rhizon soil-moisture sampler (Soilmoisture Equipment Corp., Santa Barbara, CA) tested consisted of a porous polymer tube (10 cm length and 2.5 mm diameter). The RS was connected to a 5 cm long by 2.5 mm outer

diameter and 1.0 mm inside diameter flexible PVC tube fitted with a Luer-Lock male connector at the distal end from the RS. Six Rhizon sampler positions in a pot were tested. For the diagonal position (D-RS), one RS was inserted diagonally at a 45° angle from the top rim of root substrate to the bottom of the root substrate. An additional five RSs were positioned horizontally, one per pot, at cm distances from the bottom of the pot of 1 (H-RS<sub>1</sub>), 3 (H-RS<sub>3</sub>), 5 (H-RS<sub>5</sub>), 7 (H-RS<sub>7</sub>), and 9 cm (H-RS<sub>9</sub>). The horizontal RSs were inserted into the substrate at the time of transplanting through holes drilled through the wall of the pots. At each sample time, a Luer-Lock needle (20G×3.8cm) (BD PrecisionGlide™, Franklin Lakes, NJ) was connected to the Luer-Lock male connector of the RS. This needle was inserted through the silicon septum of a 24 ml glass screw-thread sample vial fitted with a silicon septum and an open-top polypropylene closure cap (Kimble Chase Kontes, Vineland, NJ). A vacuum line was installed along the greenhouse bench holding the experiment. The vacuum line was connected to the sample vial via a 18G×2.5 cm needle (Kimble Chase Kontes, Vineland, NJ) inserted into the septum to create a vacuum in the vial when needed which would draw substrate solution from the RS. RS samples were always collected one hour after irrigation at a vacuum in the sample vial of 3.4 kPa (15 inches Hg) for 20 minutes.

Pour-through extraction method was also carried out one hour after irrigation by pouring 100 ml deionized water over the substrate surface of assigned pots and collecting leached solution from the bottom of the pot (Whipker et al., 2001; Wright, 1986). For the SME extraction method, the substrate profiles were obtained one hour after irrigation by

coring the substrate from the top to bottom in the pot using a soil profile sampler (1.6 cm inner diameter, 33 cm length). Five cores were taken from each pot and were combined in a 200 ml beaker. Deionized water was stirred into the substrate until a glistening paste formed at the surface. The final volume of the saturated substrate was  $\approx 60$  mL. After standing for 1.5 hour, the substrate was pressed to obtain solution (Warncke, 1986).

Substrate pH was measured using a pH meter (Accumet pH Meter 50, Fisher Scientific, Pittsburgh, PA). The pH electrode was standardized with pH 4 and 7 buffering solution for every 5 sample measurements and the EC electrode was standardized with  $1.412 \text{ mS cm}^{-1}$  standard solution. The measured pH, EC, and volume data were subjected to analysis of variance (ANOVA) using the GLM Procedure of the Statistical Analysis Software, SAS 9.0 (SAS Institute, Cary, NC). The comparison among DR, PT, and SME methods was carried out by using Duncan's Multiple Range test at  $\alpha=0.05$ .

## **Results and Discussion**

There was no significant interaction between the five H-RS positions in the substrate profile and three sampling dates on the substrate pH and EC,  $P=0.9648$  and  $0.5101$ , respectively. The pH values of substrate solution drawn from each H-RS position did not significantly change over three sampling dates ( $P=0.061$ ), while the measured EC levels were significantly affected by sampling date ( $P<0.0001$ ). On the first sampling date, all the measured EC values at each horizontal media profile at 1, 3, 5, 7, and 9 cm height from the bottom of the pot decreased on the second sampling date, and increased on the third sampling

date. The EC change in substrate across dates was less than  $0.6 \text{ mS}\cdot\text{cm}^{-1}$ , and could be due to different irrigation frequency between sampling dates, cumulative leaching, and plant nutrient requirement.

Gradients in pH and EC in substrate profile were determined from values averaged over the three-dates for each H-RS position (Fig. 3.1 and 3.2). Substrate pH decreased within the range of pH 6.25 to 5.46 (Fig. 3.1), and substrate EC increased within the range of EC 1.10 to  $1.81 \text{ mS}\cdot\text{cm}^{-1}$  (Fig. 3.2) from the bottom to the top of the substrate in the pot. Handreck (1994) found similar trends in substrate pH and EC gradients in horizontal slices of *Petunia* rootballs as determined by SME. The low pH in the uppermost part of the substrate was due to application of fertilizer solution to the surfaces of the rootballs (Handreck, 1994), and high EC value in the upper part of the substrate was due to salt accumulation as a result of water evaporation in the surface of substrate (Handreck and Black, 2002).

The solution volume extracted by a RS was affected only by position of the H-RS in the vertical profile of the container substrate ( $P<0.0001$ ), while the volume was consistent over three sampling dates for each H-RS level ( $P=0.4$ ). Therefore, the volumes of substrate solution at each H-RS position, averaged over the three dates, are presented (Fig. 3.3). The largest volume of substrate solution (11.2 mL) was collected by H-RS<sub>1</sub> in the lowest portion of the substrate, while only 3.5 mL of substrate solution was drawn at H-RS<sub>9</sub>, in the upmost part. This is consistent with the existence of a higher container capacity volume percent of water in the lower horizon of the substrate than in the top horizon.

The pH, EC, and extracted solution volume values of the D-RS were compared to these values from the five horizontal RSs (HR<sub>1 to 9</sub>). Based on standard errors of the means, the pH of the D-RS extraction was equivalent to the pH value of the lowest H-RS<sub>1</sub> (Fig. 3.1). The EC level of the D-RS and lowest three H-RSs were within standard error of each other (Fig. 3.2), indicating that the D-RS extract could have been drawn from the mid and/or lower layers of the substrate profile. The volume of substrate solution extracted by the D-RS was equivalent to that drawn by the mid position H-RS<sub>3, 5, and 7</sub> (Fig. 3.3).

To further explore the origin of extract sampled by the D-RS, the quantities of H<sup>+</sup> sampled by each of the five H-RSs was computed from the measured pH values, summed, and converted back to a pH value. The resulting pH value was 5.70. It was not possible to simply average pH values since they are in a log scale. When similar calculations were applied to the combinations of the lower three H-RSs, the lower two H-RSs, and the lowest H-RS, pH values of 5.96, 6.03, and 6.16 were obtained. A pH value of 6.17 was obtained when these calculations were applied to the D-RS, indicating that the D-RS extract solution was drawn from the lowest layer of substrate and was not obtained from all five layers in the proportion found in the five H-RSs.

When the three substrate extraction techniques were compared, pH was lowest in the SME solution (5.46). White, (1969) reported the pH of a suspension is typically lower than that of the supernatant liquid in soil pH measurement. The extracted solution from SME was a dark brownish-black suspension, while the extractions from D-RS and PT were much

clearer, which could explain the lower pH level in SME. Substrate pH was higher in the D-RS (6.24) and PT (6.60) tests. In each extraction a clearer extract was obtained, which would have a higher pH level. Also, extract came proportionately more from the lower portion of the substrate profile where pH was higher (Fig. 3.1). Cabrera (1998) reported significantly higher pH values of D-RS compared to pH values of PT and SME samples in peat:perlite medium without plants. When a porous sampling system such as the ceramic suction cup or RS is used in the field, an increase in pH (>0.5 units) has been reported due to changes in gaseous equilibrium, particularly by degassing of CO<sub>2</sub> (Cabrera, 1998; Grossmann and Udluft, 1991). However, in our study, plant growth and continuous top-irrigation with fertilizer solution generated a large pH gradient in the shallow container substrate depth compared to the smaller gradient in the deep field soil profile. Compared to this large pH gradient in our substrate profile, the effect of changes in CO<sub>2</sub> equilibrium by degassing-CO<sub>2</sub> on pH during D-RS sampling was insignificant. The result of  $pH_{PT} > pH_{D-RS} > pH_{SME}$  (Table 3.1) is more likely due to the different proportional distribution of extract from each level of the substrate profile. For the SME procedure, the substrate extract was collected equally from the top to the bottom of the substrate profile. Since the SME extract had proportionately more solution from the upper levels of the substrate profile than the PT and D-RS extracts, it would contain more acidity, thus a lower pH level.

The EC values from the D-RS and PT methods were not significantly different (Table 3.1). (Cabrera, 1998) suggested that EC interpretation guidelines for PT can be used for RS

extracts since over the measured EC range of 0 to 8 dS m<sup>-1</sup> their PT and RS values were not statistically different. Although we found an EC gradient along the vertical substrate profile (Fig. 3.2), EC values in the lower three substrate levels were within standard error of each other. Most of the volumes of our D-RS and PT samples were drawn from the lowest layers which would cause them to have similar EC levels. The lower EC<sub>SME</sub> (0.69 mS cm<sup>-1</sup>) than both EC<sub>D-RS</sub> (1.21 mS cm<sup>-1</sup>) and EC<sub>PT</sub> (1.26 mS cm<sup>-1</sup>) (Table 3.1) was most likely due to a dilution effect caused by saturating the substrate with deionized water in the SME procedure (Cabrera, 1998; Cavins et al., 2004; Wright et al., 1990; Yeager et al., 1983). To arrive at the saturated paste state required in the SME procedure, water must be added beyond the point of container capacity. Bergman et al. (1994) also reported approximately 30% lower EC values from SME solutions compared to the EC values of substrate solutions extracted by a suction lysimeter tube while EC<sub>SME</sub> was 43% lower than EC<sub>D-RS</sub>. The difference between studies may have resulted from the different magnitudes of EC gradients caused by fertilization and irrigation methods as well as subjective determination of the amount of additional water to saturate the substrate.

In summary, large pH and EC gradients were found to exist along the vertical profile of substrate supporting plant growth. These gradients will affect pH and EC test results, depending on where in the profile the extract is obtained. When the pH of the D-RS was computed based on pH and extract volumes in the five substrate layers, a pH value of 5.70 was obtained. However, the measured pH was 5.17, essentially the same as that in the lowest

layer. The D-RS draws solution from the lowest layer where water is held at the lowest soil moisture tension. This points out the importance of placing the lower end of the RS at the level where the test analysis is desired. Rhizon samplers can also be placed horizontally at the desired level. While RS and PT extractions provide unaltered substrate solution, the RS has the advantage of minimal alteration of the substrate solution for future sampling and impact on growth. The RS procedure does not entail replacement of substrate solution with water and withdraws a smaller amount of sample. The RS technique also requires less mechanical handling of plants. Different, increasing levels of pH were found in the extracts obtained by SME, D-RS, and PT, respectively. These require three sets of interpretation. The EC levels were similar in the D-RS and PT extracts but lower in the SME extract, thus requiring two sets of interpretative values.

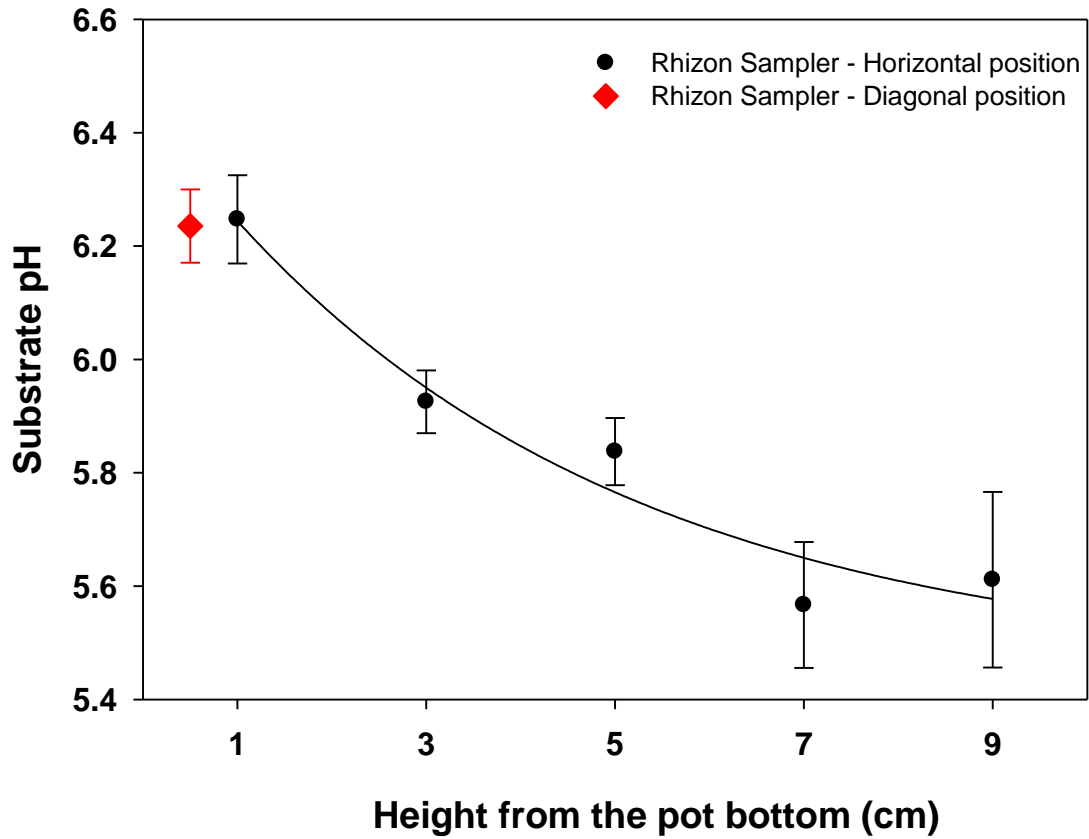


Figure 3.1. Average substrate pH values with standard error bar (n=15) measured using five horizontally placed Rhizon samplers (H-RS) (-●-) and one diagonally inserted Rhizon sampler (D-RS) (◆) from top to bottom of substrate profile. (a fitting curve equation:  $y=5.46+\exp^{(-0.23x)}$ ,  $R^2=0.29$ )

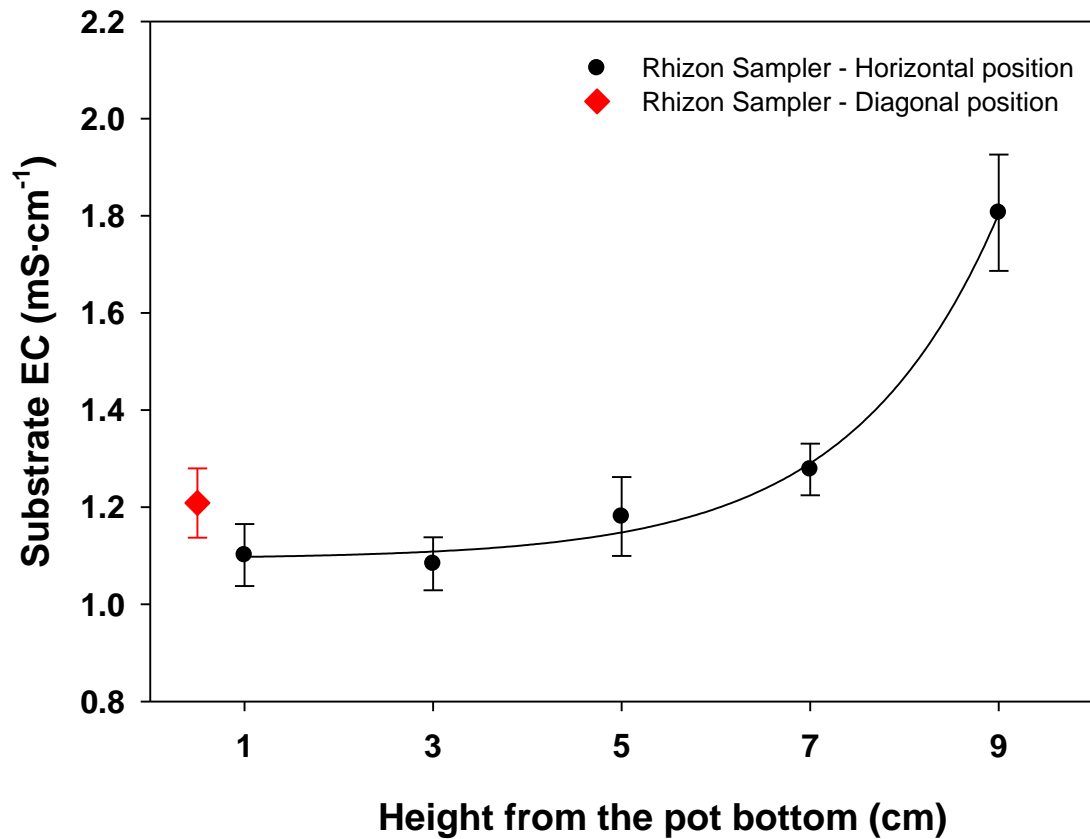


Figure 3.2. Average substrate EC values with standard error bar (n=15) measured using five horizontally placed Rhizon samplers (H-RS) (-●-) and one diagonally inserted Rhizon sampler (D-RS) (◆) from top to bottom of substrate profile. (a fitting curve equation:  $y=1.09+0.0022\exp^{(0.64x)}$ ,  $R^2=0.45$ )

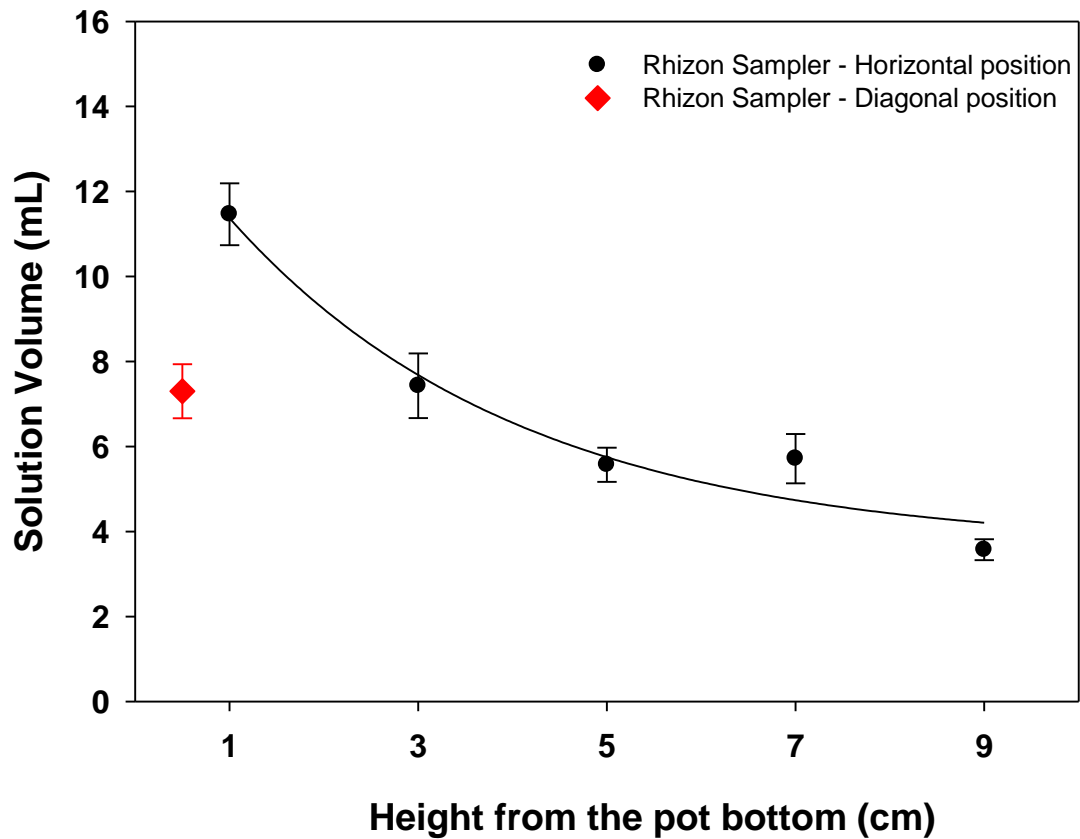


Figure 3.3. Average volume of collected substrate solution with standard error bar (n=15) measured using five horizontally placed Rhizon samplers (H-RS) (-●-) and one diagonally inserted Rhizon sampler (D-RS) (◆) from top to bottom of substrate profile. (a fitting curve equation:  $y=3.63+10.71\exp^{(-0.32x)}$ ,  $R^2=0.59$ ).

Table 3.1. Averaged pH, EC, and solution volume (mean  $\pm$  standard error, n=5).

	pH	EC (mS cm <sup>-1</sup> )	Solution Volume (mL)
Rhizon Sampler (Diagonal)	6.24 $\pm$ 0.06 <sup>b</sup>	1.21 $\pm$ 0.07 <sup>a</sup>	7.3 $\pm$ 0.6 <sup>a</sup>
Pour-through	6.60 $\pm$ 0.07 <sup>a</sup>	1.26 $\pm$ 0.05 <sup>a</sup>	39.6 $\pm$ 3.0 <sup>b</sup>
Saturated media extract	5.46 $\pm$ 0.07 <sup>c</sup>	0.69 $\pm$ 0.04 <sup>b</sup>	

Letter after each mean  $\pm$  S.E. represents mean separation within a column using Duncan's Multiple Range test at  $\alpha=0.05$ .

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## CHAPTER 4

### Impact of 13 Floricultural Crops on Soilless Substrate pH

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#### Abstract

Impacts of 13 floricultural crops on substrate pH were assessed. Plants were grown in 16.5 cm diameter (1.8L) plastic pots filled with 3 sphagnum peat moss: 1 perlite (v:v) substrate amended with CaCO<sub>3</sub> powder to establish an initial target pH of 6.5, and were fertilized at each irrigation with neutral 17N-2.2P-14.1K fertilizer dissolved in deionized water at 100 or 200 mg L<sup>-1</sup> N. Substrate ΔpH was determined as the difference in pH between 4 days after transplant (DAT) and 78 DAT. Petunia (ΔpH=0.14), fibrous begonia (0.19), and osteospermum (0.43) were very light acidifier-species (ΔpH ≤0.5). Pansy (ΔpH=0.51), impatiens (0.79), New Guinea impatiens (0.89), and geranium (0.97) were light acidifier-species (ΔpH=0.5-1.99). Vinca (ΔpH=1.00), tomato (1.17), and Reiger begonia (1.46) were medium acidifier-species (ΔpH=1.0-1.49). Pot mum (ΔpH=1.56), sunflower (2.44), and kalanchoe (2.45) were heavy acidifier-species (ΔpH ≥1.50). The former three acidification

groups of plants were fertilized at the rate of  $100 \text{ mg} \cdot \text{L}^{-1} \text{ N}$  and the final group with  $200 \text{ mg} \cdot \text{L}^{-1} \text{ N}$  to comply with fertilization recommendations. For the first 25 days, pH declined for all crops and then rose, leveled, or declined moderately or heavily in the four groups listed above, respectively. One species was selected from each group for a repeat experiment. The pH outcome was in the same order. When kalanchoe was fertilized at  $100 \text{ mg} \cdot \text{L}^{-1} \text{ N}$ , substrate EC continually declined from 2.1 to  $0.4 \text{ dS} \cdot \text{m}^{-1}$ . Simultaneously, pH declined from 6.5 to 5.4 over the first 35 DAT and then rose slightly to 5.6 by 56 DAT. When kalanchoe was fertilized at  $200 \text{ mg} \cdot \text{L}^{-1} \text{ N}$ , EC increased over crop time to 3.4. At the same time pH declined continuously from 6.5 to 4.2. The pH of fertilizer solutions at  $100$  and  $200 \text{ mg} \cdot \text{L}^{-1} \text{ N}$  were 5.0 and 4.6, respectively. During both experiments, when fertilizer was fully utilized by plants, the potential neutrality rating of the fertilizer was expressed in substrate pH levels that held constant or rose slightly. When fertilizer accumulated in the substrate, the acidic chemistry of the fertilizer had the predominant effect with substrate pH levels declining. Data indicate that the quantitative impact of potential acidity/basicity ratings assigned to fertilizers is highly dependent upon amount of fertilizer applied relative to plant requirement.

## **Introduction**

It is relatively easy to set the initial target pH of substrate. The challenge lies in maintaining this target pH throughout crop production. Forces that impact pH over time include irrigation water alkalinity, residual content of liming materials, potential acidity/basicity of fertilizer, and plant system acidification. A moderate level of alkalinity in

irrigation water is desired to neutralize acids from fertilizers (Bailey, 1996; Handreck, 1983; Handreck, 2002) as well as acidity from the plant system. Handreck (2002) suggested incorporating coarse dolomite (0.5-2mm) in potting mixes to maintain desirable substrate pH for several months by providing buffering capacity (residual lime) in the root substrate when irrigation water has low alkalinity.

Rhizosphere pH is affected by uptake of all cationic and anionic nutrients. A pH decline is associated with cation uptake, whereas a rise occurs with anion uptake (Marschner, 1995a). Since more nitrogen ions are often taken up than other types combined, form of nitrogen has a large effect on substrate pH. Plant species vary according to preference for ammonium and nitrate (Tabatabaei et al., 2006) and cultural conditions can shift the preference further (Taylor, et al., 2010). Thus, there is the potential for plant species to play a role in substrate pH maintenance. Release of CO<sub>2</sub> by roots and rhizosphere microorganisms during respiration has an acidifying effect on rhizosphere pH through generation of carbonic acid (Marschner, 1995b). Again, there is a potential for variation in the level of impact of respiration on substrate pH across plant species. The significant effect of species on substrate pH during germination and early seedling growth was reported by Huang, et al. (2001). Some species tend to change substrate pH to an undesired pH range for their growth. Therefore, understanding the species effects on pH is very important for maintaining substrate pH within an optimal range. The objectives of this study were 1) to evaluate the level of

substrate acidification by 13 floricultural crops during the growing cycle, and 2) to test the correlation between fertilizer utilization and substrate pH.

## **Materials and Methods**

**2008 Expt.** Seeds of fibrous begonia (*Begonia x semperflorens-cultorum* 'Encore III Pink Bicolor'), impatiens (*Impatiens* 'Taboo Mix'), pansy (*Viola x wittrockiana* 'Ultima Radiance Red'), petunia (*Petunia x hybrid* 'Petunia Easy Wave Blue'), sunflower (*Helianthus* 'Ballad F1 Helianthus') and vinca (*Catharanthus rossus* 'Pacifica XP White and Really Red') were sown on 5 Nov. 2008, and tomato (*Lycopersicon esculentum* 'Early Girl Hybrid') on 19 Nov. 2008. Cuttings of New Guinea impatiens (*Impatiens x hawkeri* 'Super Sonic White') and geranium (*Pelargonium zonale-hybrid* 'Tango') were taken from stock plants and inserted into 51 cell trays on 18 Nov. 2008. Rooted cuttings of pot mum (chrysanthemum, *Dendranthema x grandiflora* 'Kory') and 51-cell tray liners of Osteospermum (*Osteospermum ecklonis* 'Astra White'), kalanchoe (*Kalanchoe blossfeldiana* 'kerinci') and Reiger begonia (*Begonia x hiemalis* 'Amstel Blitz') were obtained from propagators. The following numbers of the above established plants were transplanted on 23 Dec. into each 16.5 cm diameter, 1.8L green, plastic pot: geranium, New Guinea impatiens, reiger begonia, sunflower, and tomato – 1; impatiens, pansy, petunia – 2; begonia, kalanchoe, and vinca – 3; osteospermum – 4; and pot mum – 5. A root substrate consisting of 25% perlite and 75% sphagnum peat moss by volume (Sun Gro Horticulture, Bellevue, WA) was formulated with calcium carbonate powder (CaCO<sub>3</sub> USP/FCC, Fisher Scientific, Pittsburgh, PA) at the rate of

65g per kg of peat moss (dry weight basis) instead of agricultural dolomitic limestone to avoid any residual lime effect over time.

Plants were watered as needed with deionized water from transplanting through the first day of substrate solution extraction, and then fertilized at each irrigation with water-soluble fertilizer solutions of 17N-2.2P-14.1K (Greencare 17N-5P<sub>2</sub>O<sub>5</sub>-17K<sub>2</sub>O, Kankakee, IL) dissolved in deionized water (no alkalinity). In accordance with crop requirements, fertilizer solutions were applied at a rate of 200 mg·L<sup>-1</sup> N for sunflower, pot mum, and kalanchoe, and 100 mg·L<sup>-1</sup> N for other species. Fertilizer solutions were applied to the top of each pot using a drip system supplied by sump-pumps (model 1A, Little Giant Pump Co., Oklahoma City, Oklahoma) in the bottom of barrels of single strength solution. Substrate pH was measured periodically in substrate solutions extracted with a Rhizon soil-moisture sampler (Soilmoisture Equipment Corp., Santa Barbara, CA) consisting of a porous polymer tube (10 cm length and 2.5 mm outside diameter). One sampler was placed diagonally in each pot by inserting the sampler at an angle of 45 degrees from the top rim of root substrate to the bottom of the root substrate. Substrate solution was extracted one hour after irrigation by connecting the sampler to a sample vial to which a vacuum was applied. Substrate pH was measured using a pH meter (accumet pH meter 50, Fisher Scientific, Pittsburgh, PA). The pH electrode was standardized with pH 4 and 7 buffering solution for every 5 sample measurements. Plants were harvested on 78 DAT. The ΔpH was determined as the difference between initial and final substrate pH measured on 4 DAT and 78 DAT. The acidification

level of species was categorized as very light ( $\Delta\text{pH}\leq 0.5$ ), light ( $\Delta\text{pH} 0.50-0.99$ ), medium (pH 1.00-1.49), and heavy ( $\Delta\text{pH}\geq 1.50$ ). The experimental plan consisted of a randomized complete block design with 13 species and 5 replications.

**2010 Expt.** Based on the results from the *2008 Expt.*, a same cultivar of each fibrous begonia, New Guinea impatiens, Reiger begonia, and pot mum was selected to represent the very light, light, medium, and heavy acidifier-species, respectively, as a replication of the *2008 Expt.* In addition, the correlation between substrate pH and proportion of applied fertilizer consumed was tested. Plants, propagated in a similar manner to those in the *2008 Expt.*, were transplanted on 5 April, 2010 using the same type fertilizer, substrate, pots, and numbers of plants as employed in the *2008 Expt.* The rate of calcium carbonate powder added into the substrate was adjusted to 60.1g per kg of peat moss (dry weight basis) to avoid high initial pH ( $>6.5$ ) and gypsum ( $\text{CaSO}_4$ ) was incorporated at 0.9g per L. Fibrous begonia, new Guinea impatiens, and Rieger begonia were fertilized at each irrigation with  $100 \text{ mg}\cdot\text{L}^{-1}$  fertilizer solution the same as in the *2008 Expt.* The effect of fertilizer concentration on the substrate pH was tested with kalanchoe. Kalanchoe I and kalanchoe II were fertilized at each irrigation at rates of 100 and  $200 \text{ mg}\cdot\text{L}^{-1}$  N, respectively. All fertilization was started after the first measurement of substrate pH and EC on 3 DAT. This experiment was arranged in a randomized complete block design with five treatments and 5 replications.

## Results and Discussions

Based on the 2008 *Expt.* data, the 13 species were categorized into four groups according to the acidification level of each species. Petunia ( $\Delta\text{pH}=0.14$ ), fibrous begonia (0.19), and osteospermum (0.43) were very light acidifier-species ( $\Delta\text{pH}\leq 0.5$ ) (Fig. 4.1A). Pansy ( $\Delta\text{pH}=0.51$ ), impatiens (0.79), New Guinea impatiens (0.89), and geranium (0.97) were light acidifier-species ( $\Delta\text{pH} 0.50-0.99$ ) (Fig.4.1B). Vinca ( $\Delta\text{pH}=1.00$ ), tomato (1.17), and Reiger begonia (1.46) were medium acidifier-species ( $\Delta\text{pH} 1.00-1.49$ ) (Fig.4.1C). Pot mum ( $\Delta\text{pH}=1.56$ ), sunflower (2.44), and kalanchoe (2.45) were heavy acidifier-species ( $\Delta\text{pH}\geq 1.50$ ) (Fig. 4.1D). In the 2010 *Expt.*, the  $\Delta\text{pH}$  of fibrous begonia, New Guinea impatiens, Rieger begonia, kalanchoe I and II was 0.18, 0.66, 0.76, 0.94, and 2.39, respectively (Fig. 4.2). The moderately lower  $\Delta\text{pH}$  values compared to those from the 2008 *Expt.* for each species may have been caused by the intentionally lower initial substrate pH and shorter growth period, 8 weeks compared to 10 weeks in the 2008 *Expt.* However, the arrangement of species according to the magnitude of their  $\Delta\text{pH}$  values was the same as in the 2008 *Expt.* Similarly, Argo et al. (1997) found that the substrate pH levels of pansy and petunia were the highest, while geranium was the lowest among seven bedding plant species sampled four weeks after planting when all plants were grown under the same system. Although we used a different cultivar of each species than Argo et al., similar results were obtained with substrate pH levels of  $6.3\pm 0.04$ ,  $6.4\pm 0.05$ , and  $6.05\pm 0.1$  for pansy, petunia, and geranium, respectively at 25 DAT.

In the 2010 Expt., the  $\Delta$ pH of kalanchoe grown with either 100 or 200 mg  $\cdot$  L<sup>-1</sup> N fertilizer solution was larger than values for the other three species (Fig. 4.2). However, the decrease in substrate pH of kalanchoe II over 8 weeks was significantly greater than that of kalanchoe I. Although the fertilizer applied in this study was classified as a physiologically neutral fertilizer (potential acidity/basicity of 0), this neutral impact on root substrate would only apply during plant uptake. The fertilizer solution itself had an acid pH level of 4.6-4.8, depending on concentration. Therefore, when fertilizer was applied during a period of light growth, the acidic chemical effect of fertilizer on substrate pH would be expected to predominate while during a period of extensive growth, the neutral physiological effect of fertilizer on pH would be anticipated. For kalanchoe, the recommended EC range (PourThru) is 2.0 to 3.5 mS $\cdot$ cm<sup>-1</sup> (Whipker et al., 2001). Our 100 mg  $\cdot$  L<sup>-1</sup> N fertilizer rate did not provide sufficient nutrients as seen in the low EC levels that continually declined to 0.38 $\pm$ 0.05 mS $\cdot$ cm<sup>-1</sup> by the end of the crop (Fig. 4.3). This situation would be expected to allow the physiological effect of fertilizer to predominate over the chemical effect. This did happen as seen in the fairly level pH values after 14 DAT (Fig. 4.2). The 200 mg  $\cdot$  L<sup>-1</sup> N fertilizer rate provided more than adequate nutrients and resulted in continually rising EC levels after 14 DAT that reached 3.38 $\pm$ 0.20 mS $\cdot$ cm<sup>-1</sup>, all within the recommended range (Fig.4.3). Since excess fertilizer was accumulating in the substrate, the chemical effect of fertilizer on pH would be anticipated to predominate. This turned out to be the situation as seen in the rapidly declining substrate pH (Fig. 4.2). Argo et al. (1997) also found a fertilizer concentration effect on substrate pH. When they fertilized kalanchoe plants with three concentrations of 50,

100, and 200 mg L<sup>-1</sup> N from acidic reaction commercial water soluble fertilizer, 20N-4.3P-16.6K peatlite special (Scotts, Marysville, Ohio), the substrate pH was 6.08, 5.95, and 5.56, respectively at week 12. Therefore, our low substrate pH levels of pot mum, sunflower, and kalanchoe appear to be due to high fertilizer concentration as well as species effects.

The substrate pH curves for the 13 crops in Expt. 2008 (Fig. 4.1) can also be explained on the basis of the chemical and physiological effects of fertilizer on pH. Overall, substrate pH declined rapidly for the initial 25 days in all crops. This can be explained by the acidic chemical effect of the fertilizer during the period of light biomass accumulation. During the post 25 DAT period, the physiologically neutral impact of the fertilizer became more apparent. The substrate pH of fibrous begonia and petunia rose from 6.24 and 6.41 (25 DAT) to 6.65 and 6.70 (78 DAT), respectively while pH remained essentially constant for osteospermum. The net pH effect in the light and medium acidifier crops was a decline, but there was a substrate pH plateau after 39 DAT for New Guinea impatiens and pansy and a similar plateau between 25 and 55 DAT for tomato and vinca. The rapid pH decline after 25 DAT for the heavy acidifier crops are likely due to the luxuriant level of fertilizer applied, as explained earlier for kalanchoe in Expt. 2010.

In summary, a commercially important effect of species on substrate pH level was found. This suggests the need to categorize more greenhouse crops according to their impact on substrate pH. It further indicates a need in commercial settings to analyze separately the substrate from light and heavy acidifier species. Data also suggest that the quantitative

impact of potential acidity/basicity ratings assigned to fertilizers is highly dependent upon amount of fertilizer applied relative to plant requirement.

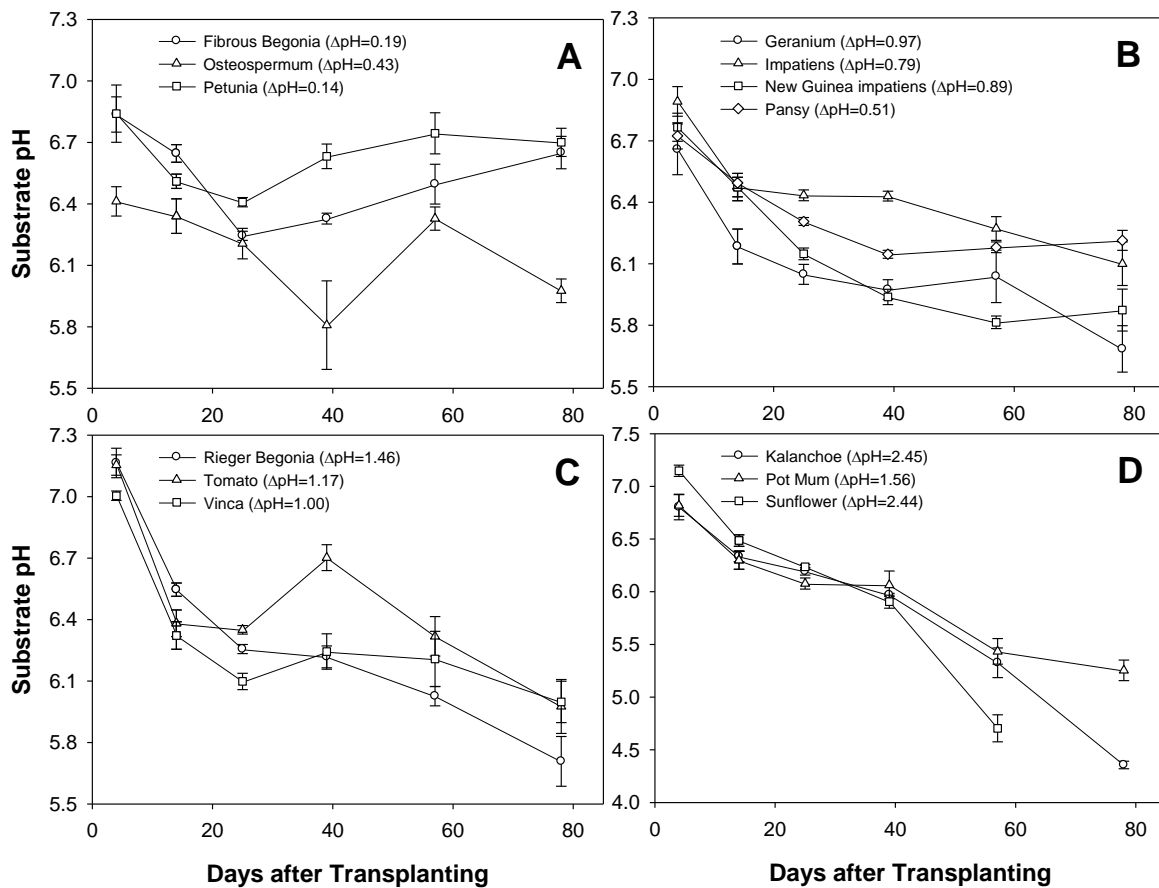


Figure 4.1. Substrate pH levels measured from 4 to 78 DAT (mean  $\pm$  standard error,  $n=5$ ) for 13 species of plants fertilized with neutral reaction fertilizer solution, 17N-2.2P-14.1K. The acidification levels of the thirteen species categorized by the difference ( $\Delta\text{pH}$ ) between initial (4 DAT) and final pH (78 DAT) were very light (A,  $\Delta\text{pH} < 0.50$ ), light (B,  $\Delta\text{pH} 0.50-0.99$ ), medium (C,  $\Delta\text{pH} 1.00-1.49$ ), and heavy (D,  $\Delta\text{pH} 1.5-2.49$ ) acidifier-species.

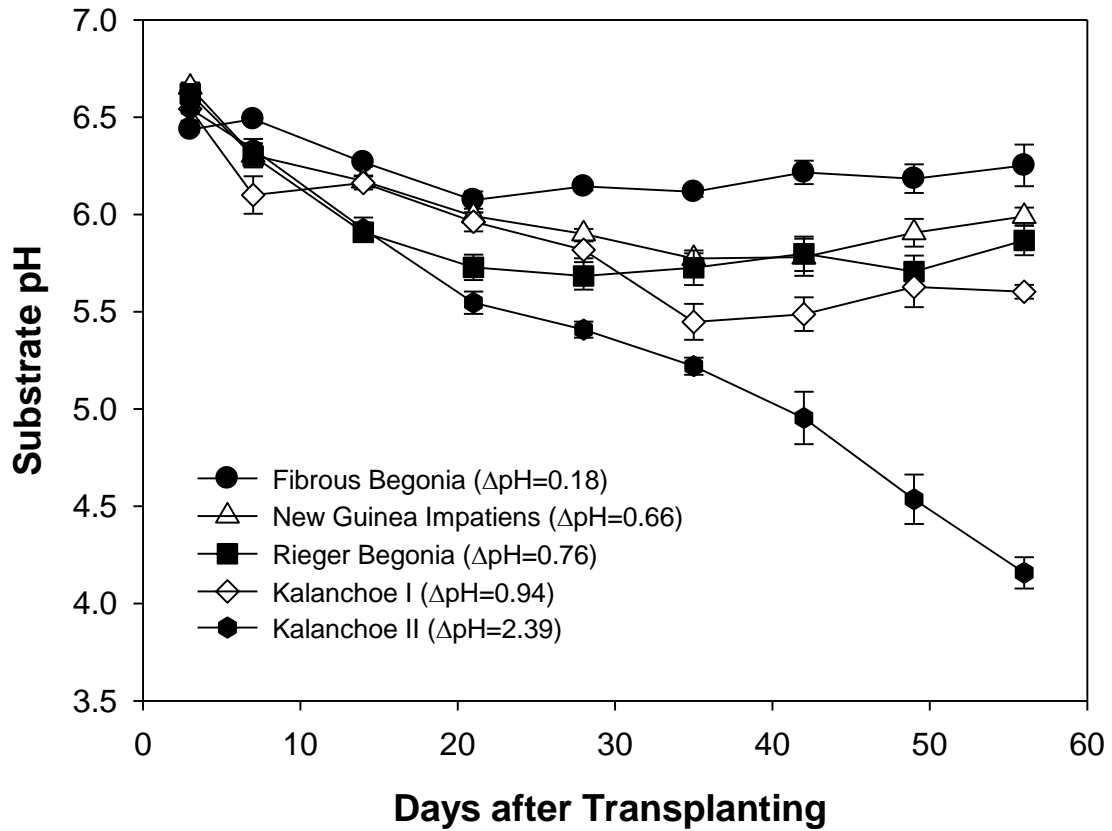


Figure 4.2. The averaged substrate pH of four representative species. The  $\Delta\text{pH}$  was calculated as the difference between initial (3 DAT) and final pH (56 DAT) (mean  $\pm$  standard error,  $n=5$ ). Fibrous begonia, New Guinea impatiens, Rieger begonia, and kalanchoe I were fertigated with  $100 \text{ mg}\cdot\text{L}^{-1}$  N from neutral water soluble fertilizer solution, 17N-2.2P-14.1K, while kalanchoe II plants were fertigated with the same fertilizer at  $200 \text{ mg}\cdot\text{L}^{-1}$  N.

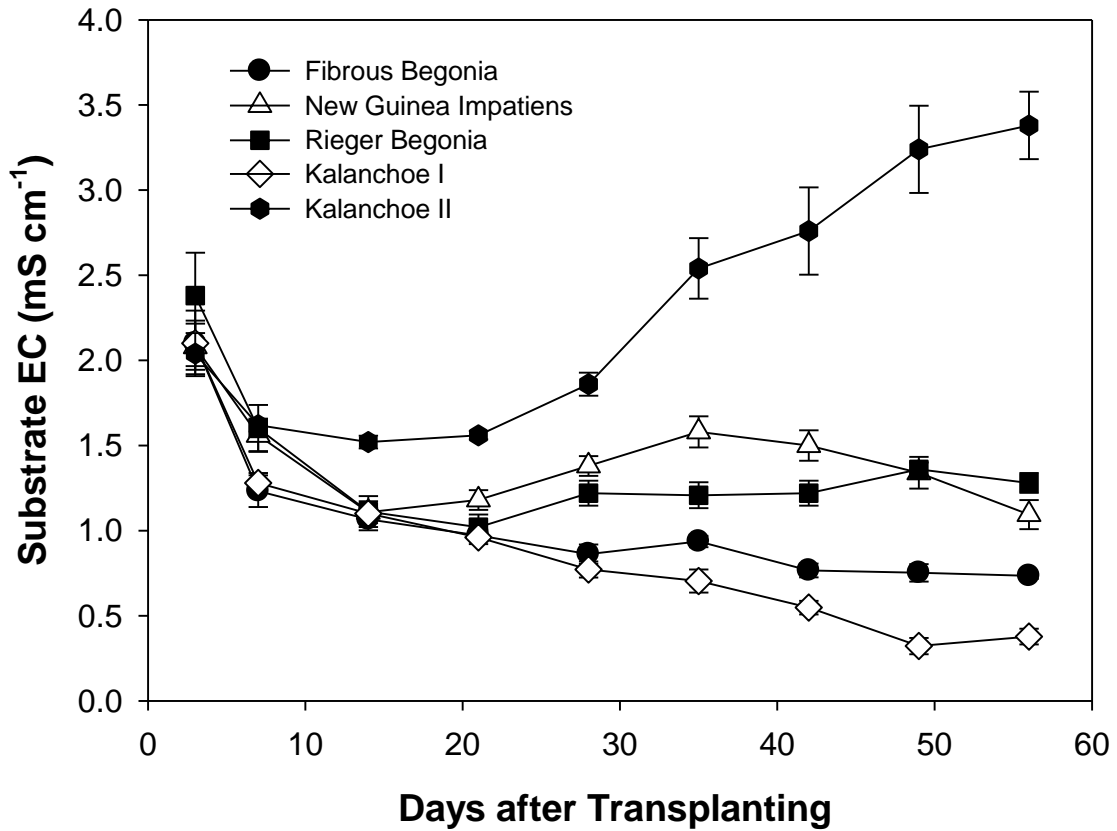


Figure 4.3. The averaged substrate EC of the four species. Fibrous begonia, New Guinea impatiens, rieger begonia, and kalanchoe I were fertigated with  $100 \text{ mg}\cdot\text{L}^{-1}$  N from neutral water soluble fertilizer solution, 17N-2.2P-14.1K, while kalanchoe II plants were fertigated with the same fertilizer at  $200 \text{ mg}\cdot\text{L}^{-1}$  N. (mean  $\pm$  standard error, n=5)

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## CHAPTER 5

### **Fertilizer Acidity/Basicity and Concentration Effect on Substrate pH and EC**

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#### **Abstract**

This study assessed the interactive effect of fertilizer concentration and potential acidity/basicity rating on substrate pH during plant growth. A root substrate consisting of 75% sphagnum peat moss and 25% perlite by volume was formulated with fine calcium carbonate to achieve substrate pH 6.5 without any residual lime effect during plant cultivation. Three commercial water soluble fertilizers were selected; an acidic fertilizer (AF), 20N-4.4P-16.6K (a potential acidity: 193 kg), a neutral fertilizer (NF), 17N-2.2P-14.1K with a 0 potential acidity/basicity, and a basic fertilizer (BF), 13N-0.88P-10.8K (a potential basicity: 150 kg). The AF, NF, and BF were dissolved in deionized water at rate of 100 and 200N mg L<sup>-1</sup> N. Liners of kalanchoe ‘Kerinci’ were grown in a 16.5 cm diameter, 1.8L volume pots (2 plants per pot). Plants were fertigated using one of the prepared water-soluble fertilizer solutions for 9 weeks. Fertilizers applied at the 100 mg·L<sup>-1</sup> N rate resulted in sub-optimal substrate EC levels that declined to below 0.54 dS·m<sup>-1</sup>. Associated substrate pH

levels expressed the physiological fertilizer effect (potential acidity/basicity rating) with a decline in the BF and NF treatments to 6.0 and 5.4 at 35 days, followed by steady levels. Substrate pH in the AF treatment declined steadily to 4.4. The 200 mg L<sup>-1</sup> N treatments resulted in substrate EC levels that rose to between 2.5 and 2.8 dS m<sup>-1</sup>, indicating luxuriant fertilization. Associated substrate pH levels declined steadily throughout crop time to 4.6, 3.9, and 3.9 in the BF, NF, and AF treatments, respectively. The pH levels for the actual fertilizer solutions for the 200 mg L<sup>-1</sup> N BF, NF, and AF fertilizers were 5.8, 4.6, and 6.0, respectively. Results indicated that addition of fertilizer solution, regardless of potential acidity/basicity, caused an initial decrease in substrate pH. This was followed by a rise or fall in substrate pH, depending on potential acidity/basicity of the fertilizer, when a large proportion of the applied fertilizer was taken up by plants.

### **Introduction**

Fertilizers are rated on their labels or technical sheets according to their potential acidity or potential basicity. The equivalent (potential) acidity and basicity of fertilizers have been defined as required calcium carbonate to neutralize the acidity generated by the fertilizers, and the basic residue left in the soil by applied fertilizers, measured in terms of equivalent calcium carbonate, respectively (Pierre, 1933). The numeric rating value of a potentially acid fertilizer indicates the kilograms of CaCO<sub>3</sub> required to neutralize the acidifying effect of 1,000 kg of the fertilizer to a crop. Conversely, the numeric value of a potentially basic fertilizer indicates the kilograms of CaCO<sub>3</sub> that would be equivalent to the

alkalinity generated by application of 1,000 kg of the fertilizer to a crop. Potential acidity/basicity depends on physiological processes of microbes and plants. The potential acidity effect can be realized during microbial nitrification of ammonium to nitrate, where two protons are generated for each ammonium ion nitrified, and during microbial or plant uptake of cationic nutrients where a proton is released to the rhizosphere in exchange for uptake of positive cation charges (Havlin et al., 2005; Kafkafi, 2008; Marschner, 1995; Nelson, 2003; Zhu et al., 2009). The potential basicity effect occurs when microbes or plants take up protons along with anionic nutrients or release  $\text{OH}^-$  or  $\text{HCO}_3^-$  to the rhizosphere in exchange for anionic nutrients taken up (Pertusatti and Prado, 2007). In this paper we refer to these processes as the physiological effect of fertilizer on substrate pH.

Independent of the physiological pH effect of fertilizers, they also have a chemical effect. The pH levels of fertilizer solutions are generally different from their physiological (potential) acidity or basicity ratings. Most fertilizer solutions are acidic, even when they are physiologically basic. Early in a container substrate crop, when there is minimal microbial and plant growth, application of a chemically acid fertilizer solution would be expected to lower substrate pH. Later when microbial and plant biomass accumulation is greater, the chemical fertilizer effect would be expected to be proportionately low compared to the physiological fertilizer effect. A fertilizer solution that is chemically acidic but physiologically basic would be expected to lower substrate pH early in a crop and then raise pH later in the crop. The relative chemical to physiological effects of fertilizer solutions has

not been clearly assessed. The objective of this study was to assess the interactive effect of fertilizer concentration and potential acidity/basicity on substrate pH during plant growth.

### **Materials and Methods**

A root substrate consisting of 75% sphagnum peat moss and 25% perlite by volume (Sun Gro Horticulture, Bellevue, WA) was formulated with fine calcium carbonate ( $\text{CaCO}_3$  USP/FCC, Fisher Scientific, Pittsburgh, PA) at the rate of 60.1g per kg of peat moss (dry weight basis) instead of agricultural dolomitic limestone to achieve substrate pH 6.5 without any residual lime effect during plant cultivation. The substrate also included wetting agent (AquaGro 2000 G, Aquatrols, Paulsboro, NJ) at the label rate of  $0.6 \text{ g L}^{-1}$  and anhydrous calcium sulfate ( $\text{CaSO}_4$ ) at  $0.9 \text{ g L}^{-1}$ .

Three commercial water soluble fertilizers were selected. These were an acidic fertilizer (AF), 20N-4.4P-16.6K (Greencare 20N-10P<sub>2</sub>O<sub>5</sub>-20K<sub>2</sub>O, Kankakee, IL), with a potential acidity of 193 kg  $\text{CaCO}_3$  equivalent per 1000 kg of fertilizer, a neutral fertilizer (NF), 17N-2.2P-14.1K (Greencare 17N-5P<sub>2</sub>O<sub>5</sub>-17K<sub>2</sub>O-3Ca-1Mg, Kankakee, IL), with 0 potential acidity/basicity, and a basic fertilizer (BF), 13N-0.88P-10.8K (Greencare 13N-2 P<sub>2</sub>O<sub>5</sub>-13K<sub>2</sub>O-6Ca-3Mg PlugCare Plus, Kankakee, IL), with a potential basicity of 150 kg  $\text{CaCO}_3$  equivalent per 1000 kg. The percentage of ammoniacal nitrogen ( $\text{NH}_4\text{-N}$ ) was 8.0, 4.2, and 0.6, while nitrate nitrogen ( $\text{NO}_3\text{-N}$ ) 12.8, 12.8, and 12.4%, respectively for AF, NF, and BF. The AF, NF, and BF were dissolved in deionized water at rates of 100 and 200 mg  $\text{N}\cdot\text{L}^{-1}$ .

Liners of kalanchoe ‘Kerinci’ were transplanted from 51 cell trays into a 16.5 cm diameter, 1.8L volume pots (2 plants per pot) on 5 April, 2010. Plants were grown in a glass greenhouse in Raleigh, NC at the latitude of 35°N under natural photoperiod. Until collecting substrate solution to measure the initial substrate pH and EC on 2 DAT, plants were irrigated using deionized water, and then plants were fertilized at each irrigation using one of the prepared water-soluble fertilizer solutions. The fertilizer solutions were applied to the top of each pot using a drip system supplied by sump-pumps (model 1A, Little Giant Pump Co., Oklahoma City, Oklahoma) in the bottom of barrels of each fertilizer solution. Substrate pH was measured periodically in substrate solutions extracted with a Rhizon soil-moisture sampler (Soilmoisture Equipment Corp., Santa Barbara, CA) consisting of a porous polymer tube 10 cm length and 2.5 mm diameter. One sampler was placed diagonally in each pot by inserting the sampler at an angle of 45 degrees from the top rim of root substrate to the bottom of the root substrate. Substrate solution was extracted one hour after irrigation by connecting the sampler to a sample vial to which a vacuum was applied. Substrate pH was measured using a pH meter (Accumet pH Meter 50, Fisher Scientific, Pittsburgh, PA). The pH electrode was standardized with pH 4 and 7 buffering solution for every 5 sample measurements.

## **Results and Discussion**

All fertilizer solutions used in this study were chemically acidic ( $<7.0$ ). At a rate of  $100 \text{ mg}\cdot\text{L}^{-1} \text{ N}$ , the averaged pH level of AF (AF100), NF (NF100), and BF (BF100) solutions were  $6.07\pm 0.11$ ,  $5.02\pm 0.06$ , and  $6.19\pm 0.08$ , while at a rate of  $200 \text{ mg}\cdot\text{L}^{-1} \text{ N}$  the pH of AF (AF200), NF (NF200), and BF (BF200), and solutions were  $6.02\pm 0.08$ ,  $4.55\pm 0.14$ , and  $5.82\pm 0.06$ , respectively. The NF solution was the most acidic among the three types of fertilizers at both concentrations. The averaged EC of AF100, NF100, and BF100 solutions were  $0.79\pm 0.03$ ,  $0.75\pm 0.02$ , and  $0.86\pm 0.01 \text{ mS}\cdot\text{cm}^{-1}$ , and the averaged EC of AF200, NF200, and BF200 solutions were  $1.53\pm 0.05$ ,  $1.39\pm 0.03$ , and  $1.65\pm 0.03$ , respectively.

All substrate EC levels declined for the first week because fertilization did not begin until 7 days (Fig. 5.1). Beyond 7 days, substrate EC levels for the three  $100 \text{ mg}\cdot\text{L}^{-1} \text{ N}$  treatments underwent a slow steady decline reaching low levels of  $0.38$  to  $0.54 \text{ dS}\cdot\text{m}^{-1}$  at 63 DAT. This indicated insufficient fertilizer supply and suggested that a high proportion of fertilizer was taken up by plants. Whipker, et al. (2001) indicated that the recommended pour-through extract EC range for kalanchoe is  $2.0$  to  $3.5 \text{ mS}\cdot\text{cm}^{-1}$ . Substrate pH levels associated with these three treatments are presented in Figure 5.2. Solution AF100 resulted in a steady pH decline from  $6.4$  to  $4.4$  at 63 DAT. The physiologically neutral, NF100, solution was associated with a pH decline for 35 days to  $5.4$  after which it held fairly constant. Substrate pH in the physiologically basic, BF100, treatment declined to a level around  $6.0$  by 35 days and was nearly constant after that time. The potential acidity/basicity ratings for

these AF, NF, and BF were expressed in the final substrate pH levels of 4.4, 5.5, and 6.1, respectively.

Applications with either AF, NF, or BF solution at  $200 \text{ mg L}^{-1}$  N rate resulted in increase in substrate EC, reaching levels around  $2.7 \text{ dS m}^{-1}$  at 42 DAT (Fig. 5.1). After that time the levels remained relatively constant. Substrate pH declined continuously throughout the experiment in the three treatments (Fig. 5.2). The physiologically acid and neutral fertilizers had similar responses to each other, with the final pH 3.9 at 63 DAT. Although the basic fertilizer resulted in higher pH levels, the levels declined continuously to 4.6. The potential acidity/basicity ratings of the fertilizers were best expressed in substrate pH when the  $100 \text{ mg L}^{-1}$  N rate was applied. Substrate EC levels in these treatments indicated inadequate nutrition, thus a high proportion of fertilizer was taken up by plants. Under this condition the physiological fertilizer uptake effect (the potential acidity/basicity rating) would be strong relative to the chemical fertilizer effect. Fertilizer BF100 counteracted chemical acidity of the fertilizer to a stronger degree than NF100, as would be expected. The acidic physiological effect of fertilizer AF100 enhanced the chemical acidity of the fertilizer as seen in the rapid decline in substrate pH throughout the 63 days. Accumulation of fertilizer salts in substrate of the  $200 \text{ mg L}^{-1}$  N series of fertilizer treatments, as seen in rising substrate EC levels, resulted in the physiological fertilizer effect being proportionately smaller than the chemical fertilizer effect on substrate pH. Consequently, all substrate pH values were lower in this fertilizer series than in the  $100 \text{ mg L}^{-1}$  N series. The basic BF200 fertilizer had the

highest pH values. Substrate pH in this treatment did not plateau as in BF100, but continued to decline to a value of 4.6 compared to 6.1 in the BF100 treatment. The physiological neutral effect of NF200 was insufficient to counteract the chemical fertilizer acidity effect, thus the pH curves for NF200 and AF200 were essentially the same.

In summary, magnitude of the physiological fertilizer uptake effects on substrate pH, predicted by the potential acidity/basicity rating of fertilizer, depends on the proportion of applied fertilizer taken up by the plant. Basic and neutral reaction fertilizers can have an acidic effect on substrate when applied to crops in super-optimal quantity.

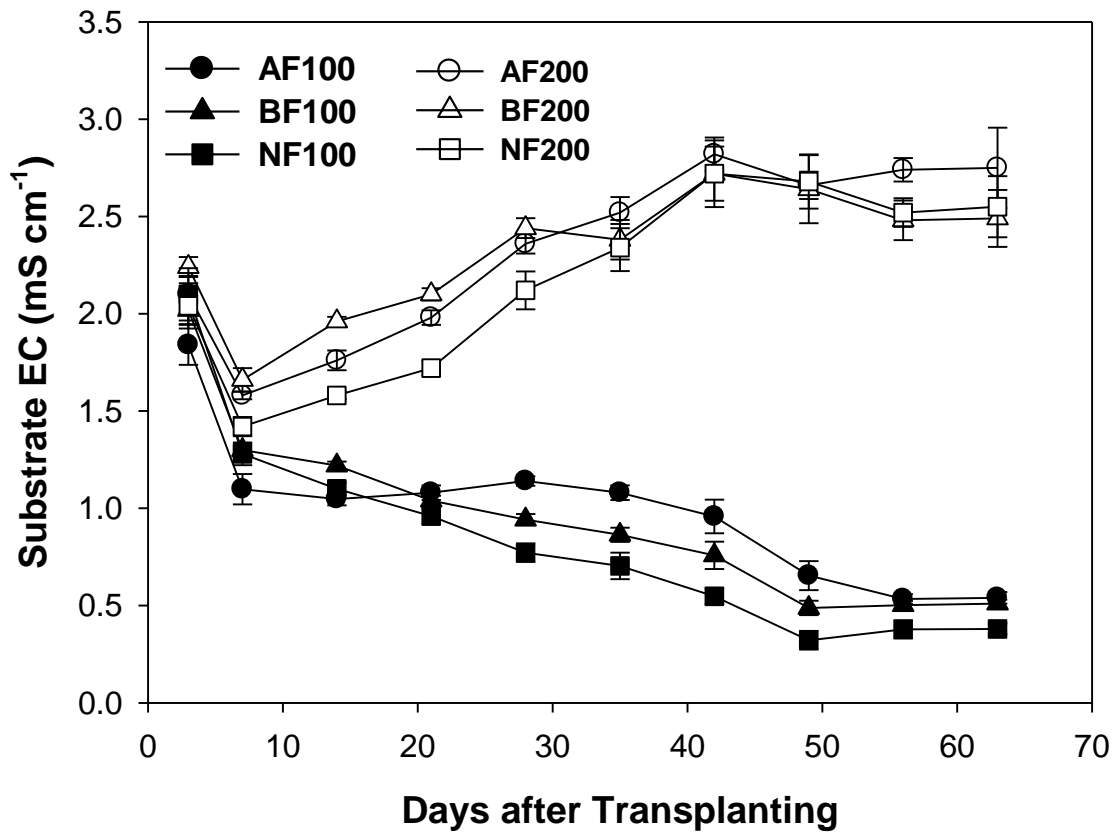


Figure 5.1. Averaged EC of the substrate irrigated with one of the acidic (AF), basic (BF), neutral (NF) fertilizer solutions at either 100 or 200mg·L<sup>-1</sup>. (A – 100 mg·L<sup>-1</sup> fertilizer solution of AF (AF100), BF (BF100), and NF (NF100); B – 200mg·L<sup>-1</sup> fertilizer solution of AF (AF200), BF (BF200), and NF (NF200)). (mean ± standard error, n=5)

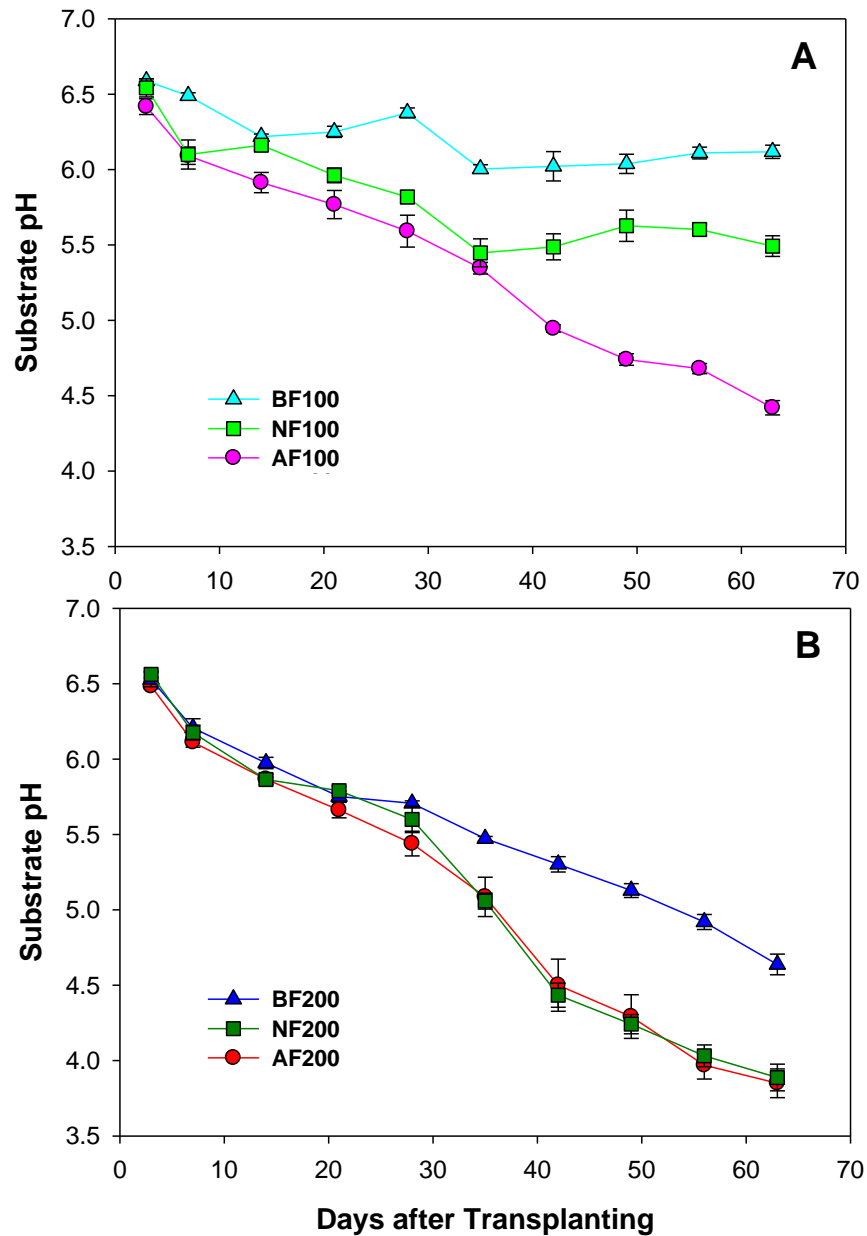


Figure 5.2. Averaged pH of root substrate irrigated with one of the acidic (AF), basic (BF), or neutral (NF) fertilizer solutions at either 100 or 200mg·L<sup>-1</sup>. (A – 100 mg·L<sup>-1</sup> fertilizer solution of AF (AF100), BF (BF100), and NF (NF100); B – 200 mg·L<sup>-1</sup> fertilizer solution of AF (AF200), BF (BF200), and NF (NF200)). (mean ± standard error, n=5)

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## CHAPTER 6

### **Effects of mature dairy manure compost on physical properties in a peat moss:perlite root substrate before and after plant cultivation**

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#### **Abstract**

The effects of mature dairy manure compost (DMC) on physical properties of a peat moss-perlite substrate before and after 12 weeks of plant growth were evaluated. In four substrates, DMC<sub>0, 1, 2, and 3</sub>, the ratio of DMC to peat moss was 0, 1, 2, or 3 on a dry weight basis, equivalent to DMC at 0, 16, 26, or 33% on a volume basis. In all mixes, the peat moss plus DMC was held constant at 75% volume and perlite at 25%. Two sets of cylinders with an inside diameter of 7.6 cm and height of 15.2 cm were filled with the test substrates. The first set remained fallow and after three top irrigations was used for the initial physical property tests. In the second set of cylinders, one rooted cutting of pot chrysanthemum 'Macumba' (*Dendranthema x grandiflora* (Ramat.) Kitam) was transplanted into each cylinder and grown for 12 weeks after which the substrates were tested for physical properties. The effects of addition of DMC to peat moss:perlite substrate were as follows. Addition of DMC to

DMC<sub>0</sub> nearly doubled initial and final dry bulk density ( $D_b$ ) from  $0.08 \text{ g} \cdot \text{mL}^{-1}$  to  $0.14 - 0.15 \text{ g} \cdot \text{mL}^{-1}$  in all three DMC containing substrates. Compared to DMC<sub>0</sub>, total porosity (TP) in DMC<sub>1</sub> was similar whereas in DMC<sub>2 and 3</sub> TP was lower. Addition of DMC generally resulted in a rise in initial and final container capacity (CC). The increase in CC was greatest in initial and final DMC<sub>1</sub> followed by initial and final DMC<sub>2</sub> and then initial DMC<sub>3</sub>. The CC in final DMC<sub>3</sub> was similar to DMC<sub>0</sub>. Addition of DMC resulted in decreased air space (AS) at all levels of DMC. The effects of 12 weeks of crop cultivation on substrate physical properties were as follows. No change in  $D_b$  occurred in any substrate; TP was similar in DMC<sub>0 and 1</sub> but lower in final DMC<sub>2 and 3</sub> by 3 and 5% of substrate volume, respectively, however all values were above 84%; CC was higher in all final substrates by 4-7% of substrate volume, with the highest rise in DMC<sub>0</sub>; and AS was lower in all final substrates by 5-7% of substrate volume. The distribution of particle sizes comprising the DMC<sub>0</sub> substrate showed an increase in size by the end of the crop time while the DMC containing substrates showed no change in particle size indicating. This indicated that little or no decomposition of DMC occurred during crop time. Plant shoot dry weight increased with additions of DMC to a maximum at DMC<sub>2</sub> and then decreased to the second highest level at DMC<sub>3</sub> which indicated that the DMC effects on physical properties were not adverse.

## **Introduction**

Use of dairy cow manure compost (DMC) has contributed to sustainable agricultural production by reducing animal waste and improving soil quality (Butler et al., 2008; Butler and Muir, 2006; Eghball, 1999; Klausner et al., 1998). DMC and other composted materials such as fiber from digested cattle slurry or composted cattle slurry fiber have been used as a substitute for peat moss in a container root substrate for a number of crops (Bradley et al., 1996; Chen et al., 1986; Prasad, 2008). However, various results have been attributed to use of composted materials in container root substrate. One concern has been reduction in substrate bulk volume during crop production due to the possible decomposition of secondary degradable organic matters (Chen et al., 2002; Nash and Pokorny, 1990). Decomposition becomes less of a problem as the compost matures during longer composting time. Dairy manure compost has a higher dry bulk density than peat moss. When DMC was substituted for 30 percent of the peat moss in a 3 peat moss:1 perlite (v/v) substrate, the dry bulk density changed from 0.10 to 0.23 g cm<sup>-3</sup> (Jeong, et al. 2010). High substrate bulk density causes an increase in transportation costs and was reported to cause a decrease in porosity and air capacity (Corti et al., 1998).

Information on impact of DMC on immediate physical properties in container root substrates is limited. Even less is known about the effect of DMC on physical properties over crop production time. This study was undertaken to address these immediate and long-term effects of DMC in a peat moss-perlite container substrate. Specific objectives were 1) to quantify the impact of DMC on immediate physical properties of peat-based substrates and

2) to test changes in physical properties of the substrates between the beginning and end of a crop cultivation period.

### **Materials and Methods**

A stable mature compost of dairy cow manure plus spoiled-silage (DMC) adjusted initially to a C:N of 30, (Woods End Laboratories, Inc, Mt Vernon, Maine) was prepared by turned-pile method, using a tractor front-end loader to lift and mix a conical-shaped pile (dimensions 4.3m d x 1.8m H) 5 times in the course of 90-days. Temperature in the core of the pile rose within 7-days of mixing to 57-60°C and remained very warm (49 – 57°C) for 6-weeks. After cooling to less than 30°C piles were stored outdoors by covering with Compostex® compost fabric, a polypropylene spun fabric permeable to air but which sheds water. Prior to use, a cubic meter sample of DMC was sieved through a 13 mm screen and mixed in a Twister™ II Batch Mixer, (Bouldin & Lawson, McMinnville, TN).

Four substrates were prepared at the ratio 0 (DMC<sub>0</sub>, control), 1 (DMC<sub>1</sub>), 2 (DMC<sub>2</sub>), and 3 (DMC<sub>3</sub>) of DMC to peat moss by dry weight basis, as a partial substitute of peat moss. This resulted in DMC occupying 0, 16, 26, or 33% of the total mix on a volume basis. The peat moss plus DMC for each substrate was held constant at 75% and perlite at 25% on a volume basis. The substrate pH was adjusted to pH 6.5 by incorporating agricultural dolomitic limestone at 180, 240, 140, and 40 g kg<sup>-1</sup> dry peat moss for DMC<sub>0</sub>, 1, 2, and 3, respectively. All treatments included granular wetting agent (AquaGro 2000 G, Aquatrols, Paulsboro, NJ) at the label rate of 0.6 g L<sup>-1</sup>.

Two sets of columns were assembled and filled with treatment substrates. Cylinders used for the initial substrate physical properties tests (I-cylinders) at the start of the crop remained fallow, while those used for physical property tests at the end of the crop production period (F-cylinders) had one chrysanthemum cutting planted in each. Cylinders for the initial substrate tests had an inside diameter of 7.6 cm and consisted from the bottom up of a 2.5 cm tall PVC ring, a 7.6 cm tall aluminum core to be used in the NCSU porometer test, and finally a 5.1 cm tall PVC ring. The three parts were taped together with duct tape. Cylinders for final substrate tests at week 12 consisted of a PVC pipe with an inside diameter of 7.6 cm and a height of 15.2cm. The PVC rings were cut from three-inch diameter, schedule 40 PVC pipe. A plastic screen was attached to the base of each cylinder. All cylinders were fully filled with one of the DMC<sub>0</sub>, <sub>1</sub>, <sub>2</sub>, and <sub>3</sub> substrates and then compacted by dropping the cylinders 3 times from a height of 15 cm.

All I and F cylinders were placed on a greenhouse bench on January 11 and one rooted cutting of pot chrysanthemum 'Macumba' (*Dendranthema x grandiflora* (Ramat.) Kitam) was transplanted into each F-cylinder. The plants were grown for 12 weeks in a glass greenhouse at 35°N latitude in Raleigh, NC. Neutral water soluble fertilizer, 17N-2.2P-14.1K (Greencare 17N-5P<sub>2</sub>O<sub>5</sub>-17K<sub>2</sub>O, Kankakee, IL) was formulated in deionized water, and applied at a concentration of 200 mg·L<sup>-1</sup> N to the top of the substrate in the F-cylinders at each irrigation with approximately 20% leaching. The frequency of irrigation ranged from twice a week at the beginning to daily at the end of the experiment. Incandescence light was

applied at an intensity of  $2 \mu\text{mol m}^{-2} \text{s}^{-1}$  for the first two weeks from 10:00pm until 2:00am daily to retard floral initiation.

The I-cylinders were watered with deionized water at the same time that fertilizer was applied to the F-cylinders for the first three irrigations. After the 3<sup>rd</sup> irrigation (one week) with tap water, the aluminum core within each I-cylinder was recovered by removing duct tape and cutting off the PVC rings above and below the aluminum core with a serrated knife. Five of the ten I-cylinders for each DMC level were used in the NCSU porometer test for dry bulk density ( $D_b$ ,  $\text{g cm}^{-3}$ ), total porosity (TP, % substrate volume), container capacity (CC, % substrate volume), and air space at CC (AS, % substrate volume) (Fonteno, 1996) and the other five in a test for water content at 300cm tension. The amount of available water in media was defined as the difference in water content between CC and unavailable water defined as 300cm tension in a 7.6cm tall container. Extra substrates for each treatment were used to conduct a particle size distribution test. Particles were separated in a Ro Tap Testing Sieve Shaker, Model B (W. S. Tyler, Inc., Mentor Calif.) using 6.2, 2.0, 0.71, 0.5, 0.25, and 0.106mm screens.

At the end of crop cultivation, plants in the F-cylinders were harvested at the substrate surface level and dried in a forced draft oven at  $70^\circ\text{C}$  for 48 hours for determination of shoot dry weight. After harvesting the plant from each F-cylinder, the screen was removed from the base of the column. The substrate column was pushed out of the cylinder into a 7.3 cm inside diameter by 7.6 cm tall aluminum core. The substrate core was positioned in the aluminum core with 2.5 cm protruding from the bottom. The bottom 2.5 cm and the top

portion of substrate were cut away with a serrated knife to prepare the aluminum core for the porometer and 300 cm tension tests. In this way, the height at which substrate was drawn from the I- and F-cylinders for the porometer and 300 cm tension test was identical. Five of the aluminum cores of substrate transferred from F-cylinders were used in the porometer tests, five in the 300cm tension test, and three for the particle distribution test.

The experiment was arranged in a randomized complete block design with five blocks. Each plot consisted of two I-cylinders and three F-cylinders for each of four substrates. The porometer and particle size distribution data were subjected to analysis of variance (ANOVA) using the GLM procedure of Enterprise Guide 3.0 of SAS 9.1 Statistical Analysis Software (SAS Institute, Cary, NC). When cubic regression fitting curve for  $D_b$ , TP, CC, and AS was significant, regression equation was provided with  $R^2$  value. The effects of an increase in the ratio of DMC to peat moss and plant cultivation time on the percentage of particle size distribution were tested using Duncan's multiple range test at  $\alpha=0.05$ . When the quadratic fitting curve of plant growth (dry weight) was significant at  $P \leq 0.05$ , regression equation and  $R^2$  were provided.

### **Results and Discussions**

The  $D_b$  of  $DMC_0$  was  $0.08 \text{ g}\cdot\text{mL}^{-1}$  which is significantly lower than that of  $DMC_{1, 2, \text{ or } 3}$  (Fig. 6.1A). Addition of DMC to peat-based substrate resulted in increased  $D_b$  due to much higher  $D_b$  of DMC ( $0.31 \text{ g}\cdot\text{mL}^{-1}$ ) compared to peat moss  $D_b$  ( $0.07 \text{ g}\cdot\text{mL}^{-1}$ ). However, increasing the ratio of DMC to peat moss from 1 to 3 (15.6, 25.9, and 33.2% of DMC by

volume) did not affect  $D_b$  which was 0.15, 0.14, and 0.15  $\text{g}\cdot\text{mL}^{-1}$  for  $\text{DMC}_{1, 2, \text{ and } 3}$ , respectively. Since the initial and final  $D_b$  were identical for each treatment, there was no significant plant cultivation effect such as irrigation, plant growth, and secondary degradation of organic materials on the  $D_b$  (Fig. 6.1A).

Initial TP was slightly lower in the DMC containing treatments than in  $\text{DMC}_0$  (Fig. 6.1B). TP values for  $\text{DMC}_{0, 1, 2, \text{ and } 3}$  were 89, 88, 87, and 88%. After cultivation for 12 weeks, TP of  $\text{DMC}_{2 \text{ and } 3}$  were lower at 86 and 84% than  $\text{DMC}_0$  at 89%. The TP of  $\text{DMC}_1$  was similar to  $\text{DMC}_0$ . The impact of 12 weeks of cultivation time on TP was seen only in DMC 2 and 3 where TP decreased 1 and 4% respectively. However, the decreased percentages of TP were still higher than the TP of 78% for pine bark and peat based container substrate with 25 to 100% of biosolids compost (Bugbee, 2002). Although the TP was reduced to 78% from 85% by adding biosolids compost, Bugbee reported greater plant growth in media containing compost. Therefore, the TP of  $\text{DMC}_{2 \text{ and } 3}$  were still within an acceptable range.

Initial and final CC was at its highest level in the  $\text{DMC}_1$  substrate (Fig. 6.1C). As DMC was increased in the  $\text{DMC}_{2 \text{ and } 3}$  substrates, CC decreased. At the beginning of the crop CC was lowest in the  $\text{DMC}_0$  substrate while at the end of the crop CC was only slightly higher than in  $\text{DMC}_3$ . The effect of 12 weeks of cultivation time was seen in a 4 to 7% rise in CC, with the highest rise occurring in the  $\text{DMC}_0$  substrate.

Air space was higher at the beginning and the end of the crop in the  $\text{DMC}_0$  substrate compared to all of the substrates containing DMC (Fig. 6.1D). At both times, DMC resulted

in lower AS with only small differences between the levels of DMC. The impact of 12 weeks of cultivation time was seen in large reductions of 5 to 7% AS in all four substrate treatments. The increase in CC and decrease in AS found in all four substrates during cultivation was likely due in part to plant root growth in the container because these changes occurred in the DMC<sub>0</sub> treatment which is a physically stable substrate. Pore sizes are categorized into three groups that include macropores, mesopores, and micropores (Drzal et al., 1999). Water in macropores is easily drained under the tension of gravity. As plant roots grew in this study, many macropores filled with roots, resulting in decreased AS, and increased CC as more water was retained on the additional root surface area within the macropores.

The distribution of particle sizes within each substrate is presented in Table 1. At the beginning of the crop, the control (DMC<sub>0</sub>) treatment had a similar amount of large and fine particles and smaller amount of medium particles than the DMC treatments. This explained, in part, the higher initial AS in DMC<sub>0</sub>. In general, at the end of the crop, the DMC<sub>0</sub> substrate contained more large, a similar amount of medium, and less fine particles than the DMC containing substrates. This could partially explain the higher final AS and low CC in the DMC<sub>0</sub> treatment at the end of the experiment. Changes over time in the DMC<sub>0</sub> treatment were favorable with a shift upward in particle size, as seen in less fine and more medium size particles at the end. Changes over time did not occur within any of the three DMC containing substrates. Again, this attests to the stability of DMC.

Plant growth in DMC<sub>1, 2, and 3</sub> was significantly higher than that of DMC<sub>0</sub> (Fig.2). Bugbee (2002) also found that greater growth of flowering annuals, herbaceous perennials and woody shrubs occurred in a softwood bark, peat and sand medium with biosolids compost compared to media without the compost. Based on growth, there was no adverse effect of altered physical properties, including increased  $D_b$  and decreased TP and AS, caused by increasing the ratio of DMC to peat moss in peat-based substrate. Moreover, the degree of change in the physical properties of substrate with or without DMC during plant cultivation was similar because DMC is a highly stabilized compost material. Therefore, use of DMC in a peat-based container substrate can be recommended up to the dry weight ratio of 3 DMC to 1 peat moss (33.2% by volume) for enhancing plant growth without negative physical properties of substrates under greenhouse conditions.

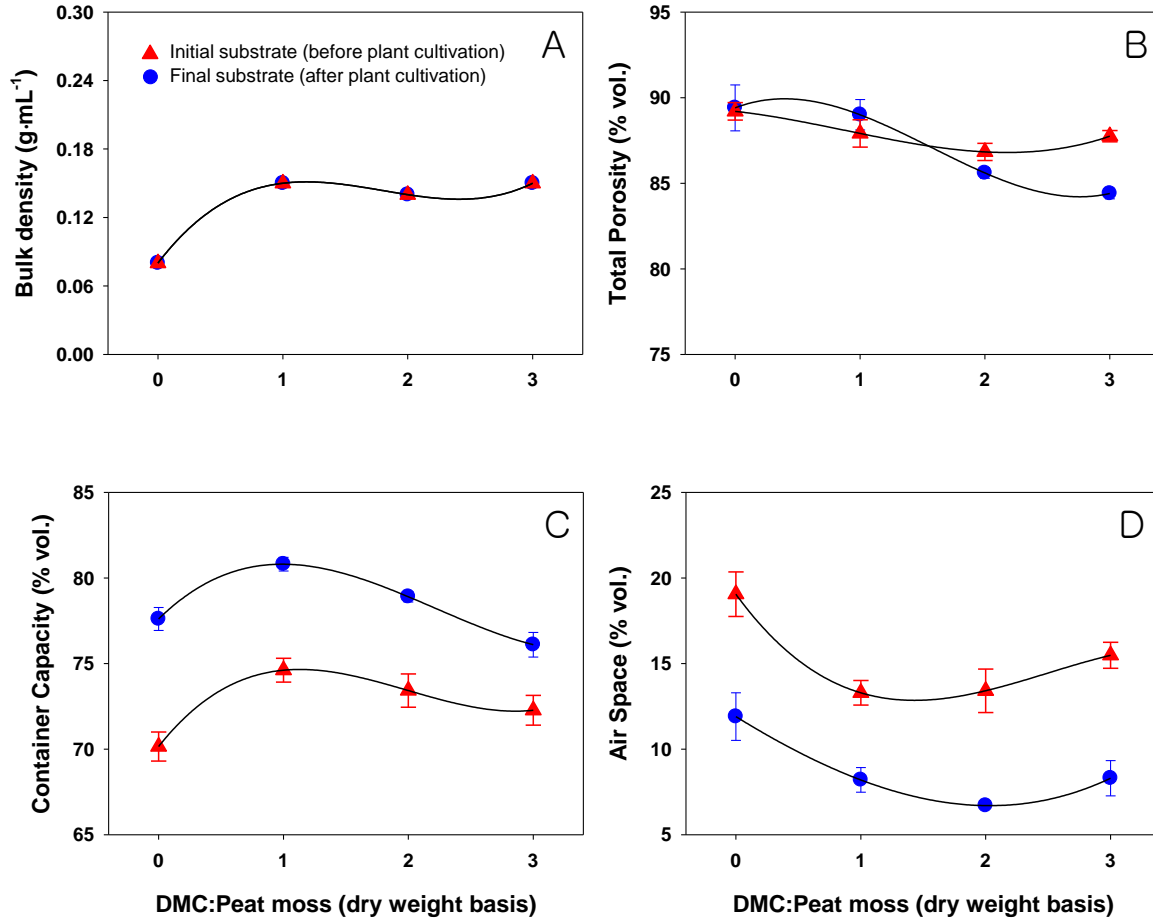


Figure 6.1. The substrate physical properties in a 7.6 cm tall column at the beginning and the end of plant cultivation, including A) average dry bulk density ( $D_b$ ), B) total porosity (TP), C) container capacity (CC), D) air space (AS), and E) available water (AW) with standard error bar (n=5).

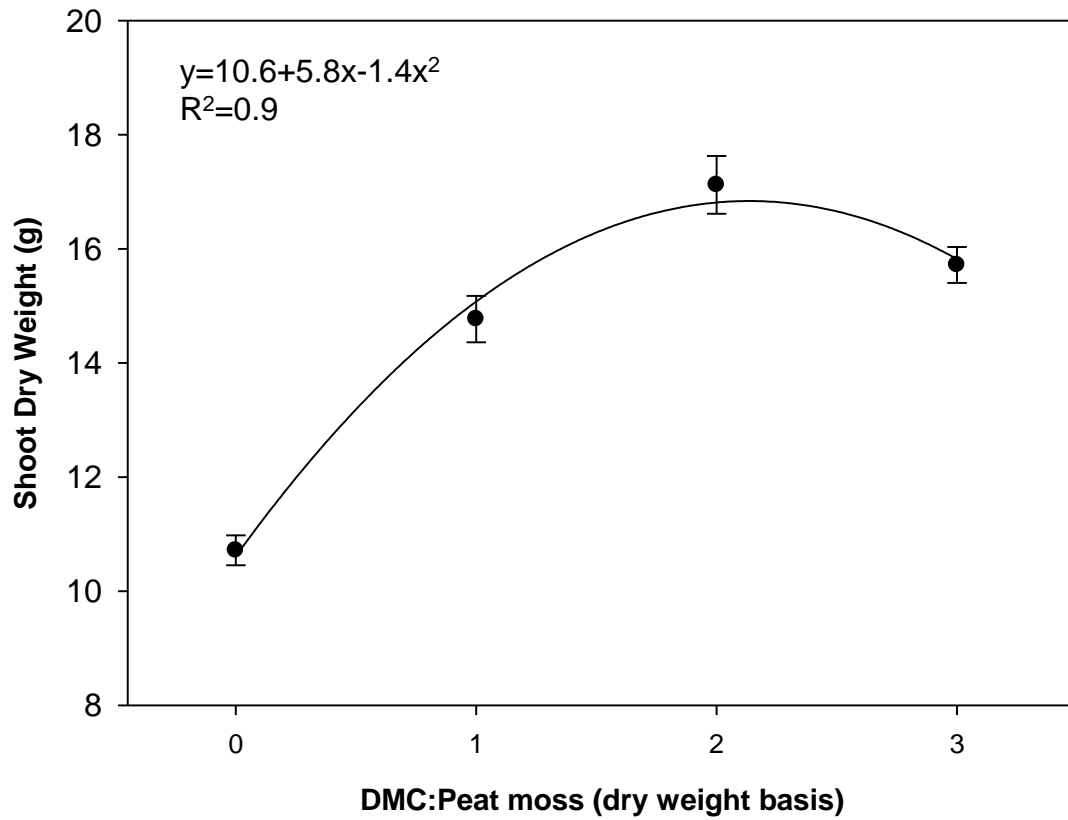


Figure 6.2. Plant shoot dry weight response to increasing ratio of dairy cow manure compost (DMC) to peat moss (dry weight basis) in peat-based soilless substrate from 0 to 3. (mean  $\pm$  standard error, n=5).

Table 6.1. Average percentage ( $\pm$  standard error, n=3) of particles size distribution of substrates consisting of 75% of DMC plus peat moss at ratio of 0 to 3, dry weight basis, and 25% of perlite at the beginning (initial) and end (final) of 12 weeks of plant cultivation. (Capitalized letters after each mean represents mean separation within a column, and small letters represents mean separation between initial and final substrate values within a row and a particle size category using Duncan's multiple range test at  $\alpha=0.05$ .)

DMC:Peat moss (dry weight)	Particle Size (%)					
	Large (>2mm)		Medium (0.5-0.2mm)		Fine (<0.5mm)	
	Initial Substrate	Final Substrate	Initial Substrate	Final Substrate	Initial Substrate	Final Substrate
0 (DMC <sub>0</sub> )	31.4 $\pm$ 1.4 <sup>A</sup> <sub>a</sub>	34.4 $\pm$ 1.3 <sup>A</sup> <sub>a</sub>	38.8 $\pm$ 0.2 <sup>C</sup> <sub>b</sub>	43.8 $\pm$ 0.1 <sup>B</sup> <sub>a</sub>	29.8 $\pm$ 1.3 <sup>A</sup> <sub>a</sub>	21.9 $\pm$ 1.2 <sup>C</sup> <sub>b</sub>
1 (DMC <sub>1</sub> )	29.5 $\pm$ 0.6 <sup>A</sup> <sub>a</sub>	29.4 $\pm$ 0.4 <sup>BC</sup> <sub>a</sub>	44.3 $\pm$ 0.7 <sup>AB</sup> <sub>a</sub>	45.1 $\pm$ 0.6 <sup>A</sup> <sub>a</sub>	26.3 $\pm$ 0.3 <sup>AB</sup> <sub>a</sub>	25.5 $\pm$ 0.3 <sup>AB</sup> <sub>a</sub>
2 (DMC <sub>2</sub> )	31.8 $\pm$ 4.1 <sup>A</sup> <sub>a</sub>	28.4 $\pm$ 0.6 <sup>C</sup> <sub>a</sub>	42.4 $\pm$ 2.0 <sup>BC</sup> <sub>a</sub>	43.8 $\pm$ 0.1 <sup>B</sup> <sub>a</sub>	25.8 $\pm$ 2.3 <sup>AB</sup> <sub>a</sub>	27.8 $\pm$ 0.5 <sup>A</sup> <sub>a</sub>
3 (DMC <sub>3</sub> )	27.9 $\pm$ 1.8 <sup>A</sup> <sub>a</sub>	31.3 $\pm$ 0.5 <sup>B</sup> <sub>a</sub>	47.7 $\pm$ 1.4 <sup>A</sup> <sub>a</sub>	44.9 $\pm$ 0.4 <sup>AB</sup> <sub>a</sub>	24.5 $\pm$ 0.8 <sup>B</sup> <sub>a</sub>	24.1 $\pm$ 0.6 <sup>BC</sup> <sub>a</sub>

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