An overview of new and revised methodologies and technologies in substrate analysis and characterization

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Abstract

Substrate research, analysis, development, and characterization remain critically important in the 21st century as horticulture and plant production continues to evolve and meet new challenges in today's ever-changing world. Three areas of substrate research in the Horticultural Substrates Lab at North Carolina State University have seen recent new methodologies and technologies introduced: root development and assessment, enhanced physical properties, and engineering and substrate construction. The mini-Horhizotron is a three-chambered device with clear sides (and detachable shade panels) that can be utilized to measure (manually or digitally) root growth and development, root branching and architecture, root hairs, root disease and overall health. The rhizometer is a clear cylinder apparatus that can be utilized to measure the influence of plant roots (undisturbed) on substrate physical properties over time. The clear cylinder design also offers the ability to observe and measure multiple root parameters over time without disturbance. Techniques to determine the wettability of substrates include the hydration efficiency and water capture method and contact angle measurements. The use of dewpoint potentiametry to directly measure substrate water potentials is providing new data on substrate unavailable water and permanent wilt ranges for horticultural crops. Advancements in organic matter processing and engineering (specifically bark and wood) have led to a better understanding of how various factors including moisture content, species, particle size, and machine type influence the consistency and reproducibility of these substrate components. New substrate fractioning and reconstruction techniques have been recently identified and studied to decrease weight/density and allow manufacturers to better be able to "design and build" substrates for specific purposes. The study of substrate particles is evolving to be able to measure surface area (external and internal) and identify specific particle shapes. Specific surface area and particle shape analysis can contribute to a greater understanding of how those parameters influence air and water relations, nitrogen drawdown potential, and shrinkage/decomposition in substrates. Advances in thermal imaging and near-infrared spectroscopy are also explored. These recent methods and techniques have the potential to further substrate science in the future and aid in our ability to design and construct substrates for maximum efficiency and productivity.

Keywords: mini-Horhizotron, particle dynamics, rhizometer, substrate hydration, substrate water potential, thermal analysis, wettability, wood processing

INTRODUCTION

Research on horticultural substrates has a long history around the world of providing critical answers to problems associated with growing plants in pot culture. Many researchers have contributed to the improvement of growing practices to make horticulture production more efficient, enhanced, economical, and sustainable. For over five decades scientists studying substrates have evolved and adapted their work around changing technologies, manufacturer and grower needs, environmental concerns, and governmental policies and regulations. Research first focused on finding suitable replacements for mineral



soils in containers. Today, studies include a broader scope of creating and managing the proper root environment for more precise and controlled plant production. This approach has spawned new efforts in quantifying and better understanding parameters that affect this root zone creation and management.

Three areas of substrate research in the Horticultural Substrates Laboratory (HSL) at North Carolina State University (Raleigh, NC, USA) have seen recent new methodologies and technologies introduced: root development and assessment, enhanced physical properties, and engineering and substrate construction. Their current and future use will be discussed here. Other methods have been recently identified that might have promise and are worth mentioning in this review. They are particle shape, thermal analysis and near-infrared spectroscopy. They will be mentioned as future possibilities. These recent methods and techniques have the potential to further substrate science in the future and aid in our ability to design and construct substrates for maximum efficiency and productivity.

RHIZOMETRICS

Currently, there are two methods of quantifying root growth that are used commonly in scientific literature: subjective ratings and root washing. Subjective root ratings can be a method to quantify root systems; however this method is completely dependent on the rater (Walters and Wehner, 1994). The person rating roots might have difficulty creating a well-defined rating system, and depending on the rating system and how broad/general it is, differences in amount of root hairs or root diameter would not be accounted for. Root washing is a destructive method involving removing all the substrate from the rootball in order to view/measure the roots unobstructed. Washing roots will reveal the root system; however this removes the roots from their natural position/architecture and 20-40% of the fine roots (including root hairs) can be lost in the washing process (Oliveira et al., 2000). This created a need for new non-destructive methods that can measure the whole root system in situ as well as root growth over time.

At the HSL at NC State University, the authors have been investigating different methods to observe and quantify root growth, in a project termed Rhizometrics (Judd et al., 2014a). Two new techniques to measure root growth of plants during production have been developed and/or explored as potential new methods of quantifying root growth: 1) the mini-Horhizotron, and 2) the Rhizometer. Both techniques allow for continual observations of intact root systems and non-destructive measurements.

Mini-Horhizotron

The Mini-Horhizotron (MHT) is a smaller version of a device (Horhizotron) developed to measure root growth in urban soils (Wright and Wright, 2004). The MHT was designed to have three arms and six flat surfaces to observe and measure root growth while maintaining the water holding and drainage characteristics of a 16-cm container (Figure 1). This flatsided design increases the surface area to volume ratio 2.5 times over the 16-cm round container. It includes shade panels to allow root development in the dark but can easily be removed to allow measurements. Comparisons with equivalent round containers showed no significant difference in root mass over a three to six week period (Judd et al., 2014b. In previous studies the MHT has been used to measure root length and root segments (Judd et al., 2015b). It has been used to determine the speed of root development over time, and the progression and severity of *Pithium* disease development non-destructively, as well as treatments that can reduce or suppress disease expression (Kaderabek et al., 2013). It has been used to observe the timing and location of root hair development. One of the best features of this rhizometric is that the device can be used for teaching and research under normal greenhouse production conditions. This allows for many traditional substrate treatments to be readily observed and measured. In addition to horticulture, it has also been used in botany and plant science courses as a teaching tool (Figure 2).

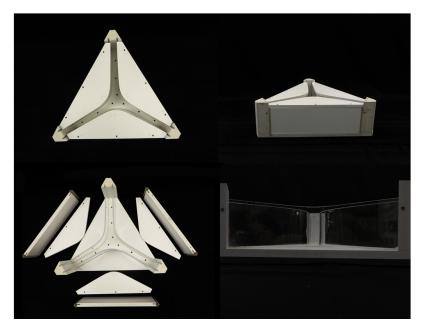


Figure 1. The mini-Horhizotron. Clockwise from top left: fully covered unit with shade panels in place (top view), unit from side view, unit from side view with shade panels removed, an exposed view of unit from top view.



Figure 2. The mini-Horhizotron. Clockwise from top left: water penetration, root exploration and branching, disease effects of healthy (circles) and damaged (arrow) roots, disease spread (inoculated from left side of image). (Image originally published in Judd et al., 2014b).

Rhizometer

The Rhizometer allows for the measurement of the effects of a growing root system on the physical properties of a substrate, i.e., total porosity, air space, water content, and bulk density. The rhizometer is basically a Porometer (NCSU Porometer) modified with a clear cylinder for root observation and measurement (Judd et al., 2014a). The cylinder is



fashioned with a short collar on top and a fine screen on the bottom (Figure 3). This allows for a small plant to be placed on the top collar portion and placed in a greenhouse for further growth. The bottom screen allows for normal water application in the greenhouse. The whole cylinder is enclosed with foil to prevent light and excessive heat from entering the root zone. The foil can be removed for root measurements and then replaced. The physical properties can be measured by removing the collar and bottom screen and placing the cylinder onto the Porometer base plate and tested using normal Porometer protocols (Fonteno et al., 1995). The root measurements may not be as descriptive as the MHT because the surface to volume ratio for the rhizotron is similar to a normal, round container and therefore one "sees" mostly root on the outside of the substrate. However, treatment comparisons including cropping time and root effects on physical properties can be easily made using this device.

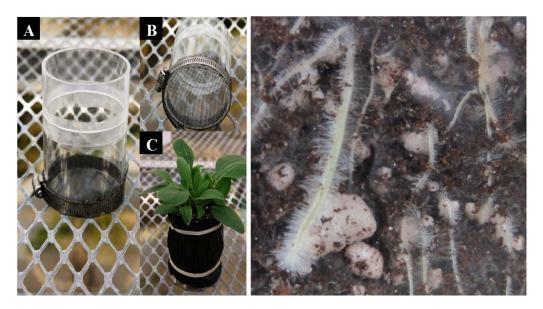


Figure 3. Left: Rhizometer clear cylinder A) set to receive plant, B) showing bottom screen designed for drainage during plant production, C) cylinder with growing plant and foil wrapped around cylinder to prevent light penetration; right: roots with extensive root hairs growing in rhizometer. (Images originally published and used with permission by Judd et al., 2015a).

HYDRATION EFFICIENCY

In the last five years new efforts have been made to measure hydration efficiency. Two main areas have been explored- the capture and retention of water by substrates and a more direct measurement of substrate water potentials. The capture and retention of water is also known as wettability.

Wettability

Wettability is affected by hydrophobicity, moisture content and hysteresis (Fields et al., 2014a; Michel et al., 2001). The HSL approach is to perform an irrigation event on a substrate sample and measure the water retained. By producing a sequence of irrigation events a hydration curve can be established to show how quickly and to what degree a substrate can be wetted. The equipment for the hydration unit consists of a transparent cylinder, 5 cm diameter × 15 cm height, with a mesh screen on the bottom; a 100 mL plastic vial (4 cm dia); a 250 mL separatory funnel; and a 250 mL beaker on the bottom (Fonteno et al., 2013). The vial is fitted with a large O-ring and placed in the top the transparent cylinder (Figure 4). The O-ring allows for exact positioning of the vial above the substrate surface to control the hydraulic head. The vial has 5 holes in the bottom and acts as a diffuser for the force and even spread of the water as it contacts the substrate.



Figure 4. The parts of the wettability unit. Left/ A whole unit assembled; center: the diffuser fitted with the O-ring on top of the substrate; right: the bottom of the diffuser with 5 holes drilled to allow for more equal flow across the substrate surface.

Because moisture content affects water retention and hydrophobicity, samples are tested at two moisture contents: 50 and 25% by weight (Figure 5). As examples, hydrations curves were performed on peat with four levels of wetting agents applied. With 50% initial moisture before testing, only the peat with zero wetting agent failed to hydrate completely at the first hydration (Figure 6). And all but the zero rate reached the container capacity for that material (solid horizontal lines). At 25% moisture, progressively higher rates of wetting agent did improve wettability, however, none of the treatments were able to reach container capacity for their respective materials (Figure 7). Additional work was performed in 2014-2015 comparing wettability measurement techniques above with those employing contact angle measurements.



Figure 5. Wettability units being used to generate a hydration curve for three different materials.



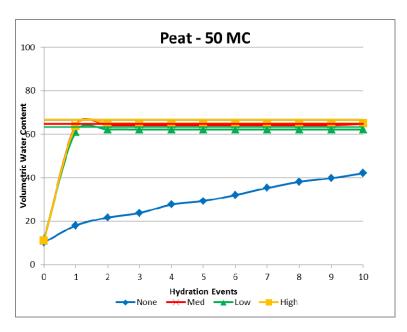


Figure 6. Hydration curves for peat moss at four wetting agent concentrations at 50% initial moisture. (Figure used with permission from Fields et al., 2014a).

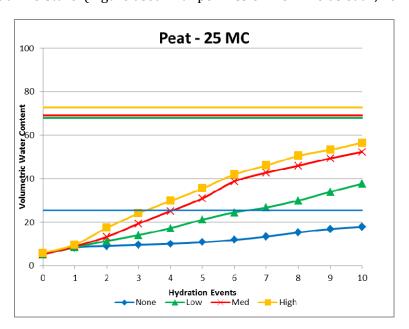


Figure 7. Hydration curves for peat moss with four levels of wetting agent at 25% initial moisture. (Figure used with permission from Fields et al., 2014a).

Water potential measurements

Another area of hydration efficiency recently explored was to measure water-holding capacities. This involves water held from container capacity (CC) to the permanent wilting percentage (PWP). Several procedures are capable of measuring CC with useful results. However, measuring substrate water potentials near permanent wilt have proven difficult. Bouyoucos (1929) described an apparatus that produced a suction equal to -1.5 MPa when drawn upon a soil sample. This idea was refined by Richards and Fireman (1943) who applied 1.5 MPa of pressure, and employed the use of porous plates which soil samples are placed upon to allow water to be moved out of the samples until equilibrium is reached with the 1.5 MPa pressure that has been applied. A modified version of Richards and Fireman's

pressure plates is currently the most common method of measuring PWP (Cassel and Nielsen, 1986). However, inaccuracies have been reported with the use of pressure plates at pressures as high as 1.5 MPa (Stevenson, 1982; Fonteno and Bilderback, 1993; Gee et al., 2002). This seems to be more pronounced in coarse, highly porous substrates. Research by Curtis and Claassen (2008) has shown the effectiveness of using dewpoint potentiometry (WP4C, Decagon, Pullman, WA) to measure the water potential of inorganic amendments with higher precision. This was verified in organic materials by measuring peat moss, pine bark and perlite, along with a clay mineral soil. The pressure system appeared to overestimate the water held at 1.5 MPa, presumably by losing proper connectivity with the pressure plate (Fields et al., 2014b).

Water content of organic substrates using pressure plates at -1.5 MPa have been reported as high as 30% by volume and were typically reported from 15 to 25% (Bilderback et al., 2013). However, data obtained from the dewpoint potentiometer for peat and coir at -1.0 to -2.0 MPa were approximately 5% (v/v) and 7-9% for pine bark (Figure 8). The dewpoint method allows a more direct measurement of substrate water potentials and avoids the overestimation due to loss of water connectivity. The WP4C Dewpoint Potentiameter can measure water potential ranges down to -100 MPa. Therefore another area was explored to measure water potentials of plants as they wilt.

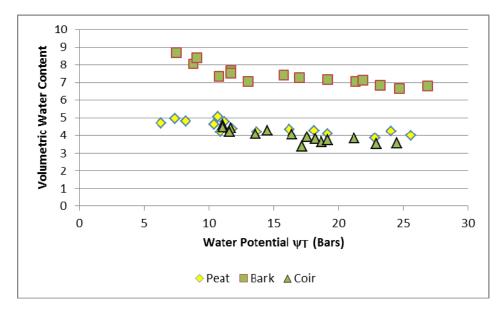


Figure 8. Water content developed from water potentials between -1.0 to -2.0 MPa (-10 to -20 bars) for peat moss, pine bark and coir. (Figure used with permission from Fields et al., 2014b).

Characterizing water potentials during plant wilt

A plant-based method relating water potentials to moisture content was first proposed by Furr and Reeve (1945) and involved growing sunflower seedlings and allowing them to wilt until PWP is reached, and then measuring soil water content. The sunflowers were not re-irrigated upon wilt, but were removed and allowed recover on their own in a room at 20-22°C for 24 h. If they did not recover, the plant was considered to be in permanent wilt and the resulting water content was considered to be at -1.5 MPa.

NCSU researchers have taken a more pragmatic approach. They took plants that had been grown in a greenhouse under standard irrigation practices and began to wilt them. Each container was saturated, allowed to drain and to begin the wilting process. The plants were observed for wilting until the plant reached one of the three progressive stages of visible wilt (Figure 9): Stage 1 – initial flagging; Stage 2 – leaves wilted with stems drooping to an angle of 45°; and Stage 3 – all leaves completely wilted. Once the plant was at the



appropriate wilt stage, substrate samples from each container were removed and analyzed for water potential in the WP4C and water content determined. After samples were removed, the plants were rewatered at each wilt stage and observed for visible recovery. The data for these studies show plants routinely tolerate wilting far below -1.0 to -2.0 MPa, down to -5, -10, even -20 MPa and recover when irrigated. Some plants even have recovered at potentials of -60 MPa. Granted, the plants were rewatered, but clearly plants can survive at substrate water potentials well below -1.5 MPa and recover very well. Work is continuing in this area to better develop these parameters.



Figure 9. Visible wilting stages for water potential measurements. Left: Stage 1 (initial wilt), Center: Stage 2 (leaves wilted with drooping stems to 45°, Right: Stage 3 (severe wilt) all leaves completely wilted.

SUBSTRATE ENGINEERING

Advancements in organic matter processing and engineering (specifically bark and wood) have led to a better understanding of how various factors including moisture content, species, particle size, and machine type influence the consistency and reproducibility of these substrate components (Figure 10). New substrate fractioning and re-construction techniques have been recently identified and studied to decrease weight/density and allow manufacturers to "design and build" substrates for specific purposes. The study of substrate particles is evolving to be able to measure surface area (external and internal) and identify specific particle shapes. Surface area and particle shape analysis can contribute to a greater understanding of how those parameters influence air and water relations, nitrogen drawdown potential, and shrinkage/decomposition in substrates.



Figure 10. Some of the many different forms/types of wood substrate components that have been engineered around the world and tested at NC State University.

Specific surface

Specific surface is the total surface area contained in a unit mass of soil or substrate. It is closely tied to particle size in mineral soils but has yet to be fully explored in substrates. Traditional soils methods involve the use of nitrogen or EGME (ethylene glycol monoethyl ether) to coat or penetrate the soil sample (Carter et al., 1986). Most mineral soils are quite dense and resist absorption, but many organic components used for substrates, such as peat, coir and bark can readily absorb water and other materials that can skew the results. The dewpoint potentiometer (WP4C) can be used to obtain water potentials at low water contents and used to estimate surface area of soils (Tuller and Or, 2005). The NCSU team is currently evaluating traditional surface area measurements with the WP4C method to determine its utility.

Particle analysis

Direct measurement of a substrate's solid fraction has been limited to bulk and particle densities and particle size distributions through a series of sieve openings. These were used to characterize the particles as they were extracted from their raw form or composted to a specific process as a way of improving consistency and predictability of performance. Today, many organic materials, especially wood, are being engineered into specific shapes and sizes. Never before has there been the ability to purposefully reengineer these organic materials into specific shapes. The new questions are what should these shapes look like and how can we measure them?

There are new measurements to be made for these issues. Particle shapes and sizes need new measures. Instrumentation has recently become available which can analyze particle shapes quickly and accurately (Haver CPA 2 Conveyor, Haver and Boecker, Germany) from 36 μm to 45 mm. These units were originally developed for the pharmaceutical industry but have great potential in substrate research. The reengineering of bark and wood products may be better scrutinized and measured to determine how effective these engineering procedures can be in creating specific materials for substrates in the future.

Thermography

Thermography involves the use of thermal imagining cameras that operate in infrared wavelengths from 2,000 to 14,000 nm. Thermography has been used extensive in building construction and other industrial applications and has seen success in determining plant water relations status (Jones and Leinonen, 2003). Cameras used today (FLIR, Santa Barbera, CA) employ technology that allows for a digital image and thermal image to be taken simultaneously, providing better temperature data of the photo subject. Software allows for further exploration of the image with temperature values available for all areas of the thermal image (Figure 11). The NCSU team is currently using thermal imagery to expand temperature information associated with composting organic matter, particularly pine bark.

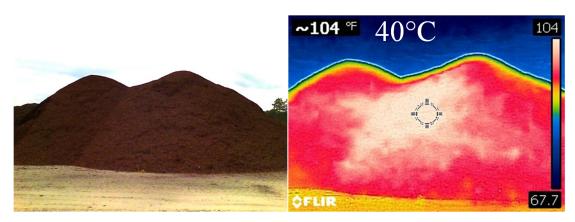


Figure 11. A normal digital image (left) and thermal image (right) of a pile of pine bark undergoing aging/composting. Note the variation in pile surface temperature.



Near-infrared spectroscopy

Near-infrared spectroscopy (NIRS) uses wavelengths of \sim 700-2500 nm to produce spectra and images of a multitude of materials and applications in agriculture, soils, pharmaceuticals, materials science, medicine and astronomy. (Burns and Ciurczak, 2007). In agriculture, it is used to determine quality of many foodstuffs, such as grains, fruits, vegetables, dairy and animal products and coffee. In soils it is used for chemical and biological analyses. It seems particularly useful on bulk materials and requires little sample preparation.

For substrate purposes, NIRS has been used to predict the decomposition of organic matter, total organic carbon, total nitrogen, ammonium, and nitrate. It has been used to determine C:N ratios, pH and electrical conductivity and volatile compounds. It has been useful to determine biological properties of carbon mineralization, microbial biomass, nitrogen immobilization and nitrogen mineralization (Burns and Ciurczak, 2007). New research at NCSU has been initiated to use NIRS for monitoring the composting and stabilization of organic components for substrate use, particularly the degree and activity of biological agents that affect stabilization. This is being compared to traditional chemical and biological analyses to correlate spectra with traditional testing. The goal is to develop spectral and imaging protocols for remote monitoring of potential organic materials.

CONCLUSIONS

Recent changes in available equipment and technologies have sparked a major effort of revamping techniques in substrate analysis and characterization. The data presented here are largely from the research team at NC State University. However, several researchers are also engaged in advancing new and revised protocols for substrate analysis. These techniques can have multiple uses. The MHT can be used for root length, time and degree of rooting, disease progression, nutritional stress, and more. The MHT is also being evaluated for plant science activities in elementary and secondary schools. The wettability system can be used to investigate new components, wetting agent rates or moisture content needed for better irrigation during production. The dewpoint system can be used to determine, moisture retention curves, "unavailable water" content for substrates, water potentials during wilt or any other type of stress, and surface area determinations. Measuring particle size and shape can be expanded to better define changes in engineering new components for substrates. Two remote sensing techniques, thermography and NIRS, also have promise for substrate analysis. Thermal imaging can provide a wider measure of temperature information for processing substrates, as well as identify temperature anomalies during plant production. NIRS may actually have the largest impact by providing non-destructive spectra and images that correlate well with traditional destructive techniques. The main deterrent for these two techniques have been the expense of the technology. Now the costs of these technologies are more suitable for use in many more investigations in the future.

Literature cited

Bilderback, T.E., Riley, E.D., Jackson, B.E., Kraus, H.T., Fonteno, Owen, J.S., Jr., W.C., Altland, J., and Fain, G.B. (2013). Strategies for developing sustainable substrates in nursery crop production. Acta Hortic. *1013*, 43–56 http://dx.doi.org/10.17660/ActaHortic.2013.1013.2.

Bouyoucos, G.J. (1929). A new, simple, and rapid method for determining the moisture equivalent of soils, and the role of soil colloids in this moisture equivalent. Soil Sci. 27 (3), 233–242 http://dx.doi.org/10.1097/00010694-192903000-00004.

Burns, D., and Ciurczak, E. (2007). Handbook of Near-Infrared Analysis, 3rd edn (Practical Spectroscopy). p.349–369

Carter, D.L., Mortland, M.M., and Kemper, W.D. (1986). Specific surface. In Methods of Soil Analysis, Part 1 – Physical and Mineralogical Methods, 2nd edn, A. Klute, ed. Agron.. Mono. 9 (Madison, WI: Soiol Sci. Soc. Amer.), p.413–423.

Cassel, D.K., and Nielsen, D.R. (1986). Field capacity and available water capacity. In Methods of Soil Analysis, Part 1 – Physical and Mineralogical Methods, 2nd edn, A. Klute, ed., Agron. Mono. 9. (Madison, WI: Soil Sci. Soc. Amer.), p.901–926.

Curtis, M.J., and Claassen, V.P. (2008). An alternative method for measuring plant available water in inorganic amendments. Crop Sci. 48 (6), 2447–2452 http://dx.doi.org/10.2135/cropsci2008.01.0069.

Fields, J.S., Fonteno, W.C., and Jackson, B.E. (2014a). Hydration efficiency of traditional and alternative greenhouse substrate components. HortScience 49 (3), 336–342.

Fields, J.S., Fonteno, W.C., and Jackson, B.E. (2014b). Plant available and unavailable water in greenhouse substrates: assessment and considerations. Acta Hortic. 1034, 341–346 http://dx.doi.org/10.17660/ActaHortic.2014.1034.42.

Fonteno, W.C., and Bilderback, T.E. (1993). Impact of hydrogel on physical properties of coarse-structured horticultural substrates. J. Am. Soc. Hortic. Sci. 118, 217–222.

Fonteno, W.C., Hardin, C.T., and Brewster, J.P. (1995). Procedures for Determining Physical Properties of Horticultural Substrates using the NCSU Porometer (Horticultural Substrates Laboratory, North Carolina State University).

Fonteno, W.C., Fields, J.S., and Jackson, B.E. (2013). A pragmatic approach to wettability and hydration of horticultural substrates. Acta Hortic. 1013, 139–146 http://dx.doi.org/10.17660/ActaHortic.2013.1013.15.

Furr, J.R., and Reeve, J.O. (1945). The range of soil moisture percentages through which plants undergo permanent wilting in some soils from semi-arid irrigated areas. J. Agric. Res. 71, 149–170.

Gee, G.W., Ward, A.L., Zhang, Z.F., Campbell, G.S., and Mathison, J. (2002). Influence of hydraulic nonequilibrium on pressure plate data. Vadose Zone J. 1 (1), 172–178 http://dx.doi.org/10.2136/vzj2002.1720.

Jones, G.H., and Leinonen, I. (2003). Thermal imaging for the study of plant water relations. Journal of Agricultural Meteorology *59* (3), 205–217 http://dx.doi.org/10.2480/agrmet.59.205.

Judd, L.A., Jackson, B.E., and Fonteno, W.C. (2014a). Rhizometrics: A review of three in situ techniques for observation and measurement of plant root systems in containers. Acta Hortic. *1034*, 389–397 http://dx.doi.org/10.17660/ActaHortic.2014.1034.48.

Judd, L.A., Jackson, B.E., Yap, T.C., and Fonteno, W.C. (2014b). Mini-horizotron: an apparatus for observing and measuring root growth of container-grown plant material *in situ*. HortScience *49* (*11*), 1424–1431.

Judd, L.A., Jackson, B.E., and Fonteno, W.C. (2015a). Rhizometer: A new technique to observe and measure root growth and its effects on container substrate physical properties over time. HortScience 50 (2), 288–294.

Judd, L.A., Jackson, B.E., and Fonteno, W.C. (2015b). Advancements in root growth measurement technologies and observation capabilities for container-grown plants. Plants (Basel) 4 (3), 369–392. PubMed http://dx.doi.org/10.3390/plants4030369

Kaderabek, L., Jackson, B., and Fonteno, W. (2013). Measuring disease severity of *Pythium* spp. and *Rhizoctonia* solani in substrates containing pine wood chips. Paper presented at: SNA Research Conference (Atlanta, GA).

Michel, J.C., Riviere, L.M., and Bellon-Fontaine, M.N. (2001). Measurement of the wettability of organic materials in relation to water content by the capillary rise method. Eur. J. Soil Sci. 52 (3), 459-467 http://dx.doi.org/10.1046/j.1365-2389.2001.00392.x.

Oliveira, M.R.G., Van Noordwijk, M., Gaze, S.R., Brouwer, G., Bona, S., Mosca, G., and Hairiah, K. (2000). Auger sampling, ingrowth cores and pinboard methods. In Root Methods: A Handbook, A.L. Smit, A.G. Bengough, C. Engels, M. van Noordwijk, S. Pellerin and S.C. van de Geijn, eds. (Berlin, Germany: Springer-Verlag), p.175–210.

Richards, L.A., and Fireman, M. (1943). Pressure-plate apparatus for measuring moisture sorption and transmission by soils. Soil Sci. *56* (*6*), 395–404 http://dx.doi.org/10.1097/00010694-194312000-00001.

Stevenson, D.S. (1982). Unreliability's of pressure plate 1500 kilopascal data in predicting soil water contents at which plants become wilted in soil-peat mixes. Can. J. Soil Sci. 62 (2), 415–419 http://dx.doi.org/10.4141/cjss82-045

Tuller, M., and Or, D. (2005). Water films and scaling of soil characteristic curves at low water contents. Water Resour. Res. 41 (9), W09403 http://dx.doi.org/10.1029/2005WR004142.

Walters, S.A., and Wehner, T.C. (1994). Evaluation of the U.S. cucumber germplasm collection for root size using subjective rating technique. Euphytica *79* (*1-2*), 39–43 http://dx.doi.org/10.1007/BF00023574.

Wright, A.N., and Wright, R.D. (2004). The Horhizotron^m: A new instrument for measuring root growth. Horttechnology 14, 560–563.

