

Dolomite and Micronutrient Fertilizer Affect Phosphorus Fate in Pine Bark Substrate used for Containerized Nursery Crop Production

Jacob H. Shreckhise

USDA, Agricultural Research Service
Application Technology Research Unit
1680 Madison Avenue
Wooster, OH 44691

Matthew J. Eick

School of Plant & Environmental
Sciences
185 Ag Quad Lane
236 Smyth Hall
Virginia Tech, Blacksburg, VA 24061

James E. Altland

USDA, Agricultural Research Service
Application Technology Research Unit
1680 Madison Avenue
Wooster, OH 44691

James S. Owen, Jr.*

School of Plant & Environmental
Sciences
Virginia Tech
Hampton Roads Agricultural Research
& Extension Center
1444 Diamond Springs Road
Virginia Beach, VA 23455

Alexander X. Niemiera

School of Plant & Environmental
Sciences
409 West Campus Drive
401-A Saunders Hall
Virginia Tech, Blacksburg, VA 24061

Sarah A. White

Department of Plant & Environmental
Sciences
Clemson Univ.
E-143 Poole Agricultural Center
Clemson, SC 29634

Dolomite and a micronutrient fertilizer are routinely incorporated into a pine bark-based soilless substrate when producing containerized nursery crops, yet the effect of these amendments on phosphorus (P) is not well understood. The objective of this research was to determine the effect of dolomite and micronutrient fertilizer amendments on P partitioning among four P fractions (i.e., orthophosphate-P [OP], non-orthophosphate dissolved P [NODP], total dissolved P [TDP], and particulate P [PP]) and to model potential P species in leachate of pine bark substrate. Amendment treatments incorporated into bark at experiment initiation included (1) a control (no fertilizer, dolomite, or micronutrient fertilizer), (2) controlled-release fertilizer (CRF), (3) CRF and dolomite, (4) CRF and micronutrient fertilizer, or (5) CRF, dolomite, and micronutrient fertilizer. Phosphorus fractions in leachate of irrigated pine bark columns were determined at eight sampling times over 48 days. Amending pine bark with dolomite and micronutrient fertilizer reduced leachate OP concentrations by 70% when averaged across sampling dates primarily due to retention of OP in the substrate by dolomite. The NODP fraction was unaffected by amendments, and the response of TDP was similar to that of OP. Particulate P was present throughout the study and was strongly correlated particulate Fe and DOC concentrations. Visual MINTEQ indicated MnHPO_4 and $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$ were consistently saturated with respect to their solid phase in treatments containing CRF. Results of this study suggest amending pine bark with dolomite and micronutrients is a best management practice for reducing P leaching from containerized nurseries.

Abbreviations: CRF, controlled-release fertilizer; DAI, days after initiation; DOC, dissolved organic carbon; EC, electrical conductivity; F, fertilizer-only; FL, fertilizer plus dolomite; FLM, fertilizer plus dolomite plus micronutrients; FM, fertilizer plus micronutrients; NODP, non-orthophosphate dissolved phosphorus; OP, orthophosphate-phosphorus; PP, particulate phosphorus; TDP, total dissolved phosphorus; TP, total phosphorus.

This publication is a portion of a dissertation submitted by Jacob H. Shreckhise in partial fulfillment of the degree of Doctor of Philosophy. The use of trade or brand names in this publication does not constitute a guarantee or warranty of the product by Virginia Tech and does not imply its approval to the exclusion of other products or vendors that also may be suitable.

Soil Sci. Soc. Am. J. 83:1410–1420

doi:10.2136/sssaj2018.12.0493

Received 21 Dec. 2018.

Accepted 1 July 2019.

*Corresponding author (jim.owen@vt.edu).

© 2019 The Author(s). Re-use requires permission from the publisher.

Core Ideas

- Addition of dolomite or micronutrients reduced leachate-P from bark-based substrates.
- Orthophosphate-P from controlled-release fertilizer contributed 74 to 86% of TDP.
- Dolomite reduced fertilizer orthophosphate-P in leachate by >50%.
- MnHPO_4 and $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$ were dominant modeled solid-phase species in leachate.
- Addition of dolomite and micronutrient amendments to bark may be considered a BMP.

Understanding the chemical fate of P fertilizers is critical for determining P bioavailability and its potential effect on down-stream ecosystems. Eutrophication and subsequent toxic algae blooms induced by elevated P concentrations is a global issue affecting surface water used for human consumption, irrigation, fisheries, and recreation (Carpenter et al., 1998). Agriculture is the leading source of P contamination in US surface waters (USEPA, 2017). In mineral soils, interactions between fertilizer-P and soil components have been thoroughly examined; however, little research has assessed the chemical fate of fertilizer P in organic substrates, specifically milled pine bark, used to produce container-grown crops.

In the United States, containerized woody crops contribute 63% of all nursery stock sales (USDA, 2014), and pine bark (*Pinus taeda* L.) is the predominant substrate utilized in the eastern USA for container-grown nursery crops (Bilderback et al., 2013b; Lu et al., 2006). Pine bark has limited ability to sorb P (Marconi and Nelson, 1984; Paradelo et al., 2017; Yeager and Wright, 1982); consequently, soluble P fertilizers rapidly leach from pine bark-filled containers during irrigation (Cole and Dole, 1997; Godoy and Cole, 2000; Yeager and Barrett, 1984, 1985a, 1985b). Phosphorus leaching from containers is exacerbated since container-grown crops are typically irrigated daily while the plants are actively growing. The use of polymer- or resin-coated controlled-release fertilizers (CRFs), a best management practice (Bilderback et al., 2013a) that reduces P leaching relative to soluble fertilizers (Broschat, 1995; Diara et al., 2014), has been widely adopted by the US nursery industry according to survey studies (Dennis et al., 2010; Fain et al., 2000; Mack et al., 2017). However, P uptake efficiency (percent of fertilizer-P absorbed by plants) of container-grown nursery crops remains between 7 and 62% when using CRFs (McGinnis et al., 2009; Owen et al., 2008; Tyler et al., 1996; Warren et al., 1995, 2001).

Pine bark has an initial pH range of 4.0 to 4.3; yet, optimal plant nutrient availability in organic substrates occurs when pH is 5.0 to 5.5 (Bunt, 1988). Accordingly, pine bark is routinely amended with a liming agent, such as dolomite [$\text{CaMg}(\text{CO}_3)_2$], to increase pH and supply plants with Ca and Mg. However, the effects of dolomite on P solubility in pine bark substrate has not been assessed. Multiple studies have reported dolomite sorption of OP in aqueous solution (Karaca et al., 2004, 2006; Mangwandi et al., 2014; Xu et al., 2014; Yuan et al., 2014, 2015). For example, in batch sorption experiments, Yuan et al. (2014, 2015) found that 10 g of dolomite removed up to 98% P from 1 L of solution containing 50 mg L⁻¹ P. Karaca et al. (2006) reported rapid sorption of OP by dolomite in solution, with equilibration occurring within 15 to 30 min depending on initial OP concentration. Xu et al. (2014) utilized attenuated total reflectance Fourier transform infrared (ATR-FTIR) spectroscopy, x-ray absorption near edge structure (XANES) spectroscopy, and diffuse layer modeling to investigate P speciation on calcite and dolomite. When solution pH values were varied from 5.0 to 7.0 and OP concentrations were 2 or 10 mg L⁻¹, which are common conditions in pore-water of dolomite-amended pine bark substrate for nursery-crop pro-

duction (Bilderback et al., 2013a), the authors determined that sorbed OP on the surface of dolomite was primarily in the form of Mg and Ca surface complexes and Ca-P secondary minerals.

Evidence of OP sorption by liming agents has also been observed in organic container-substrates. Amending peat-based substrates with varying rates of $\text{Ca}(\text{OH})_2$ was shown to reduce P leaching from containers by 20% (Haynes, 1982) to 88% (Havis and Baker, 1985) compared to substrates without $\text{Ca}(\text{OH})_2$. Two studies by Argo and Biernbaum (1996a, 1996b) reported that the amount of soluble OP in a peat-based substrate amended with various forms of dolomite was inversely related to the substrate pH. For example, Argo and Biernbaum (1996a) measured OP concentrations in saturated media extracts (Warncke, 1986) after growing *Impatiens walleriana* Hook. F. fertilized with 20 mg L⁻¹ P for 16 wk in a Sphagnum peat-based substrate containing 1.5 kg m⁻³ microfine $\text{Ca}(\text{OH})_2$ -MgO or 8.4 kg m⁻³ CaCO_3 ·MgCO₃. In both lime treatments, substrates with pH values of about 5.0 and 8.1 had OP concentrations of ~21 and 3 mg L⁻¹, respectively. Argo and Biernbaum (1996a) speculated that the reduction in soluble OP was due to precipitation of P as CaHPO_4 or $\text{CaH}_5\text{O}_6\text{P}$. However, the effect of liming agents, specifically dolomite, on P leaching from pine bark substrate has not been thoroughly investigated.

In addition to amending organic substrates with a liming agent, growers commonly add a granular micronutrient fertilizer. Iron is a primary constituent of many micronutrient fertilizers and is often supplied in chelated form or as FeSO_4 . Ferrous sulfate, a compound used to remove P from wastewater (Strickland, 1998), can reduce OP solubility via Fe-PO_4 precipitation (Moore and Miller, 1994; Rich, 2005; Tasistro and Kissel, 2006) and OP sorption to humic substances via Fe bridges (Gerke and Hermann, 1992; Weir and Soper, 1963). Many Fe-PO_4 precipitates are water-insoluble (Sanyal and De Datta, 1991) and thus unavailable for plant-uptake. Current understanding of Fe-P interactions in pine bark-based substrates is based on pioneering research conducted by Handreck (1991a, 1991b, 1992, 1996) who analyzed the effect of FeSO_4 application rate on P concentrations in diethylenetriaminepentaacetic acid (DTPA) extracts. Although results from all four studies indicated that FeSO_4 ·7H₂O reduces DTPA-extractable P concentrations in pine bark-based substrates, these results may have been confounded by the CaCO_3 -based liming agent used to maintain a constant pH across FeSO_4 treatments. Similar to dolomite, CaCO_3 can sorb OP (Xu et al., 2014); presumably, treatments containing more FeSO_4 , which acidifies pore-water during hydrolysis, would have required more CaCO_3 to achieve the same substrate pH as treatments with less FeSO_4 . Accordingly, the reported lower P concentrations in pine bark containing more FeSO_4 may have been partially due to P sorption by CaCO_3 . Hence, reinvestigating the effect of sulfated micronutrient fertilizer on water-extractable P species and concentration in pine bark would provide insight into determining P fertilization recommendations and environmental implications of P leached from containerized crops.

In natural waters, P fractions (e.g., total P [TP], total dissolved P [TDP], particulate P [PP], and OP) are commonly de-

terminated to assess the threat of P to water quality (Worsfold et al., 2016). Since only OP is available for plant uptake, fractioning P in soil-water extracts provides insight into P lability. Despite the importance of P fractions on surface water quality and plant availability of P, to our knowledge, PP, TDP, OP, and non-orthophosphate dissolved P (NODP) concentrations and the impact of dolomite and micronutrient fertilizer on these P fractions has not been studied in effluent or water-extracts of pine bark substrate with incorporated CRF. Accordingly, the objectives of our study were to (i) determine the effect of CRF, dolomite, and micronutrient fertilizer amendments on OP, NODP, TDP, and PP concentrations in leachate of pine bark substrate; and (ii) examine potential precipitated P species in pine bark leachate using the geochemical speciation software, Visual MINTEQ.

MATERIALS AND METHODS

Stabilized pine bark (milled through a 12.7-mm screen) was attained from Pacific Organics (Henderson, NC) on 15 Jan. 2016. Measured air space, container capacity, and available water (by volume) of the substrate were 40, 40, and 27%, respectively, and bulk density was 0.15 g cm^{-3} (NCSU porometer method; Fonteno et al., 1995). Substrate particle size distribution (by weight) was 72% coarse ($>2 \text{ mm}$), 16% medium (2.0 to 0.5 mm), and 12% fine ($<0.5 \text{ mm}$). Pine bark elemental analysis was determined by Brookside Laboratories (New Bremen, OH) using a Thermal 6500 Duo inductively coupled plasma optical emission spectrometer (ICP-OES) following microwave-assisted, nitric acid digestion (Peters et al., 2003). The substrate was comprised of $0.29 \text{ g kg}^{-1} \text{ N}$, $0.01 \text{ g kg}^{-1} \text{ P}$, $0.14 \text{ g kg}^{-1} \text{ K}$, $0.29 \text{ g kg}^{-1} \text{ Ca}$, $0.05 \text{ g kg}^{-1} \text{ Mg}$, $0.03 \text{ g kg}^{-1} \text{ S}$, $6.2 \text{ mg kg}^{-1} \text{ B}$, $1321 \text{ mg kg}^{-1} \text{ Fe}$, $117.4 \text{ mg kg}^{-1} \text{ Mn}$, $6.3 \text{ mg kg}^{-1} \text{ Cu}$, $27.5 \text{ mg kg}^{-1} \text{ Zn}$, and the C to N ratio was 176.

One day prior to experiment initiation, pine bark was amended with 4.75 kg m^{-3} of a homogeneous, 5 to 6 mo (release based on a 27°C) CRF (Polyon 19–2.6–10 [as $\text{N-P}_2\text{O}_5\text{-K}_2\text{O}$], Harrell's, Lakeland, FL) and one of four amendment treatments: (1) 4.15 kg m^{-3} dolomite (FL), (2) 0.89 kg m^{-3} granular micronutrient fertilizer (FM), (3) both dolomite and micronutrient fertilizer (FLM), or (4) neither dolomite nor micronutrient fertilizer (F; i.e., CRF-only). In addition, non-amended pine bark without CRF was included as a control. The CRF was composed of NH_4NO_3 , $\text{NH}_4\text{H}_2\text{PO}_4$, and K_2SO_4 . For the FL and FLM substrate treatments, dolomite was supplied as 50% pulverized dolomite (94% CaCO_3 equivalent [CCE], Old Castle Lawn and Garden, Thomasville, PA) and 50% ground dolomite (97% CCE, Rockydale Quarries Corporation, Roanoke, VA). The pulverized dolomite had 100, 95, 72, and 54% and the ground dolomite had 100, 90, 50 and 35% passing through 2.00-, 0.84-, 0.25-, and 0.15-mm mesh screens, respectively. Collectively, the dolomite mixture contained 21% Ca and 22% Mg by weight. The granular micronutrient fertilizer (Micromax, Everris, Dublin, OH) contained 6.00% Ca, 3.00% Mg, 12.00% S, 0.10% B, 1.00% Cu, 17.00% Fe, 2.50% Mn, 0.05% Mo, and 1.00% Zn derived from $\text{CaMg}(\text{CO}_3)_2$, $\text{FeSO}_4 \cdot 1\text{H}_2\text{O}$, MnSO_4 , ZnSO_4 , $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, $\text{Na}_2\text{B}_4\text{O}_7$, and $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$. The CRF and substrate amendments were in-

corporated into the pine bark by hand-mixing for 2 min to ensure homogeneity without damaging CRF granules.

Twenty columns were constructed of 30-cm sections of polyvinyl chloride (PVC) pipe (7.8 cm i.d.; 8.9 cm o.d.), and the bottom of each column was fastened with a PVC flat cap with 20 evenly distributed, 8-mm, circular holes. Precision-woven, monofilament, 500- μm mesh fabric (Sefar Propyltex 05-500/36, Heiden, Switzerland) was placed between the bottom end-cap and column to prevent substrate from falling through or clogging the 8-mm holes. The top of each column was affixed to a PVC coupler which was used to elevate diffusers 5 cm above the substrate surface. Diffusers were PVC flat caps with 16 evenly distributed, 2-mm holes and were used to evenly disperse irrigation water over the substrate surface, minimizing preferential flow. A diagram of the column assembly can be found in Hoskins et al. (2014), labeled "unsaturated column". A high-density polyethylene (HDPE) funnel was positioned beneath each column to direct leachate into individual 250-mL HDPE bottles.

For each substrate treatment and the control, four columns (i.e., replicates) were packed with 1400 cm^3 substrate to achieve a mean bulk density of 0.145 g cm^{-3} ($\sim 0.02 \text{ g cm}^{-1}$ vertical variation within a column) and mounted vertically in a randomized complete block design with two rows of ten columns. The experiment was blocked to account for filtration time within a sampling date and laboratory climate differences (mean air temperature of $22.4 \pm 0.33^\circ\text{C}$ [SE]), which could affect CRF release of nutrients. Using a GEVA 75 controller with a latch solenoid and hydraulic valve (G75-C-1W-61, Baccara Automation Control, Bayswater, Victoria, AU), each column received a single daily aliquot of tap water from a pressure compensating emitter ($2.2 \pm 0.01 \text{ mL s}^{-1}$; 01WPCLL8, Netafim, Fresno, CA) fastened to 127 mm polyethylene tubing 2 cm above each diffuser. At experiment initiation (Day 0), each column was irrigated for 228 s ($\sim 500 \text{ mL}$); for 48 d thereafter, columns were automatically irrigated every 24 h for 57 s ($\sim 125 \text{ mL}$ or 2.6 cm). The intent of the initial high-volume irrigation event was to settle substrate particles and mimic standard nursery practices following pot-up of a crop. Irrigation water, collected periodically over the course of the experiment ($n = 8$), contained ($\text{mg L}^{-1} \pm \text{SE}$): $0.21 \pm 0.01 \text{ P}$, $13.5 \pm 0.49 \text{ Ca}$, $5.77 \pm 0.20 \text{ Mg}$, $1.33 \pm 0.12 \text{ K}$, $11.5 \pm 0.49 \text{ Na}$, $<0.01 \text{ Fe}$, $<0.20 \text{ Al}$, $18.7 \pm 0.54 \text{ Cl}$, $1.99 \pm 0.09 \text{ S}$, $0.38 \pm 0.04 \text{ Zn}$, and $<0.02 \text{ B, Mn, and Cu}$. Alkalinity of the irrigation water at experiment termination ($n = 3$) was $55.9 \text{ mg L}^{-1} \pm 0.7$ (Brookside Laboratories). The daily irrigation volume of 125 mL was determined after conducting a preliminary study which showed 125 mL displaced enough substrate pore-water for subsequent analyses while minimally diluting leachate with the fresh tap water. Average leachate-volume ($100 \pm 0.05 \text{ mL}$) was measured from each column 12 h after irrigating to ensure leaching was consistent across treatments.

On Days 1, 2, 5, 6, 9, 10, 15, 16, 23, 24, 31, 32, 41, 42, 47, and 48, a 20-mL aliquot of leachate from each column and the irrigation water was analyzed for pH and electrical conductivity (EC) using a benchtop meter (HI5521-01, Hanna Instruments, Woonsocket, RI) within 4 h of sample collection (i.e., within 16 h

of irrigation). Concentrations of ions (i.e., PO_4^{3-} , NO_3^- , NO_2^- , Cl^- and NH_4^+), dissolved ($<0.45 \mu\text{m}$) organic carbon (DOC), and total and dissolved ($<0.45 \mu\text{m}$) elements (i.e., B, Ca, Cu, Fe, K, Mg, Mn, Mo, Na, Ni, P, S, and Zn) were determined from 2-d composite samples. The first half of each composite sample was collected on Days 1, 5, 9, 15, 23, 31, 41, and 47, and were prepared as follows. For samples in which leachate ion concentrations were determined, 3 mL leachate were syringe-filtered within 12 h of collection into a 10-mL glass vial using a 60-mL Luer-Lok syringe (26300, Exel International Medical Products, St. Petersburg, FL) and 0.2- μm hydrophilic polyvinylidene fluoride (PVDF) disposable syringe-filters. Filters of 0.2 μm pore size were used to reduce possible sorption of OP to Fe colloids in the 0.45 to 0.2 μm size range (Sinaj et al., 1998). The 0.2- μm filters were then replaced with 25 mm Swinnex filter holders (SX0002500, Millipore Sigma, Burlington, MS), each containing a 0.45 μm PVDF membrane (HVL02500, Millipore Sigma), to filter 13.5 mL and 10 mL leachate into respective 60-mL HDPE bottles. The 13.5 mL filtered sample (27 mL composite), which was ultimately analyzed for dissolved organic carbon (DOC) concentration, was acidified to a $\text{pH} < 3$ with two drops of 2 mol L^{-1} HCl to remove inorganic carbon from samples via volatilization. The filtered 10-mL sample (20-mL composite) was later analyzed for dissolved element concentrations. Ten mL of non-filtered leachate was dispensed into a 60-mL HDPE bottle within 12 h of collection for eventual determination of total concentrations of elements. Samples were refrigerated at 8°C until the remainder of the composite sample was added the following day (Days 2, 6, 10, 16, 24, 32, 42, or 48). The 6-mL, 27-mL and two 20-mL composite samples were then stored at -30°C . Non-filtered samples were later thawed and digested according to the method described in QuikChem Method 10-115-01-4-B, except sample volume and amount of digestion reagent added to samples was reduced by half. After digestion, white sediment was present in some samples; thus, digested samples were filtered through a 0.2- μm PVDF filter before being stored at 8°C .

Analysis of leachate ion concentrations was accomplished using two ion chromatography (IC) systems. The IC systems used to determine anion concentrations (ICS-2100, Thermo-Fisher Scientific) and cation concentrations (ICS-1600, Thermo-Fisher Scientific) utilized respective 4 mm by 250 mm (i.d. \times length) anion- and cation-exchange columns (AS19 and CS12A, respectively, Thermo Fisher Scientific) at 35°C and an autosampler (AS-AP, Thermo Fisher Scientific) on a 25 μL sample loop. The ICS-2100 was equipped with a metal trap column (MFC-1, Thermo Fisher Scientific) to facilitate detection of the anions. Total and dissolved element concentrations were determined with optical emission spectrometry (iCAP 6300 Duo View ICP-OES Spectrometer; Thermo Fisher Scientific). Dissolved organic carbon (non-purgeable organic carbon from organic sources that is available for microbial metabolic functions) concentrations were analyzed via NPOC/TN analysis using a Shimadzu TOC-V_{CPH} total organic carbon analyzer with TNM-1 TN measuring unit (Shimadzu Scientific Instruments, Kyoto, Japan).

Model Analysis

Theoretical chemical P speciation in leachate was modeled using Visual MINTEQ, a geochemical speciation software (Gustafsson, 2013). Input parameters included pH, DOC (NICA-Donnan model), PO_4^{3-} , NH_4^+ , NO_2^- , NO_3^- , B(III), Ca^{2+} , Cl^- , Cu^{2+} , Fe^{3+} , K^+ , Mg^{2+} , Mn^{2+} , Mo(VI), Na^+ , Ni^{2+} , SO_4^{2-} , and Zn^{2+} concentrations. Element input values were based on concentrations measured in previously described filtered samples. Metals were assumed to be in their oxidized state. Carbonate concentrations were estimated based on measured irrigation water alkalinity as well as Ca and Mg concentrations in leachate of substrates containing dolomite and/or micro-nutrient fertilizer. Electrical conductivity was equated to ionic strength using the formula proposed by Alva et al. (1991; i.e., ionic strength = $[\text{EC} \times 0.012] - 0.0002$) and compared to ionic strengths calculated by Visual MINTEQ to ensure consistency. Saturation indices provided in output were used to interpret degree of saturation in solutions with respect to solid phases.

Statistics

Phosphorus fraction partitioning in the particulate versus dissolved phases was analyzed via three-way analysis of variance (ANOVA) with a repeated measures factor (time) and two between-subjects factors, substrate (F, FL, FM, and FLM) and fraction (PP [i.e., TP – TDP] and TDP). Dissolved Fe ($<0.45 \mu\text{m}$) and particulate Fe (i.e., total Fe – dissolved Fe) concentrations were analyzed in the same manner. The dissolved P phase was further assessed in a separate, but similar, analysis that included fractions, OP and NODP (i.e., TDP – OP) instead of PP and TDP. For all three-way ANOVA, the three-way interaction (time \times P fraction \times substrate), three two-way interactions (time \times fraction, time \times substrate, and fraction \times substrate), and three main effects were assessed. Concentrations of other elements were equivalent between filtered and non-filtered samples ($p > 0.05$); thus, analyses were performed using dissolved (i.e., filterable) concentration values. Effects of substrate, time, and the substrate \times time interaction on Ca, EC, DOC, and pH were analyzed via two-way repeated measures ANOVA. The block effect (random) was non-significant ($p > 0.05$) in all ANOVA and therefore removed from analysis to simplify the model.

Prior to analysis, data were transformed to correct for heteroscedasticity and non-normality. The log-transformation was used for Ca, Mg, EC, and DOC values, and the normalized Johnson's transformation (Johnson, 1949) was used for all Fe and P data. Repeated measures analysis was executed by modeling covariance structures (Wolfinger, 1993). The most appropriate covariance structure was selected by fitting data to various homogeneous and heterogeneous covariance structures available in JMP Pro 14 (SAS Institute Inc., Cary, NC) and comparing corrected Akaike information criterion (AIC_c) values. According to AIC_c values, the following covariance structures were selected for repeated measures analyses: homogeneous antidependent for P (PP vs. TDP fractions), Fe, Ca, and Mg; heterogeneous antidependent for P (OP vs. NODP fractions); first-order autoregressive

(AR[1]) for EC and DOC; and unequal variance for pH data.

When interactions were significant ($p < 0.05$), simple effects were analyzed using Tukey's Honest Significant Difference (HSD) to compare fertilized substrates. The control was excluded from the multiple-comparison procedure to ensure power of the test to accurately detect differences between the effect of amendments when a CRF was present. Dunnett's test was used to contrast values of the non-fertilized control to those of each substrate treatment containing CRF. Correlations were assessed using the Pearson correlation coefficient (r). Percent reduction in element concentrations by amended substrates, discussed hereafter, were calculated on the basis of concentrations in the F substrate unless otherwise noted. All data were processed using JMP Pro 14 (SAS Institute Inc., Cary, NC).

RESULTS AND DISCUSSION

Calcium

Calcium concentrations were affected by a substrate \times time interaction ($p < 0.0001$). Solution Ca concentrations in FM and FLM were 91 mg L^{-1} at 1 d after experiment initiation (DAI), over twice as high as concentrations in FL (Fig. 1). Thereafter, Ca concentrations in FM and FLM sharply decreased to a minimum concentration of 5 mg L^{-1} (23 DAI) or 15 mg L^{-1} (15 DAI), respectively. The relatively high initial Ca concentrations in FM and FLM compared to FL suggests that the dolomite component in the micronutrient fertilizer was highly soluble relative to the separately added dolomite amendment in FL and FLM. Rapid dissolution of the dolomite component in the micronutrient fertilizer is likely partially attributed to the acidic pH of the FM substrate at 1 to 9 DAI (pH = 3.8 to 4.3) relative to that in FL (pH = 6.4 to 7.3; Fig. 1). Solubility of $\text{CaMg}(\text{CO}_3)_2$ is well known to increase with decreasing pH (Gautelier et al., 1999; Lindsay, 1979). Calcium concentrations in FLM were equivalent to those in FL from 15 to 48 DAI, whereas Ca concentrations in FM were consistently 6 to 11 mg L^{-1} lower than those in FL during this period. From 31 to 48 DAI, Ca concentrations in FM were equivalent to those in F. These data suggest that dolomite was the primary source of soluble Ca in FLM after 15 DAI or receiving $\sim 39 \text{ cm}$ total irrigation. The increase in Ca concentration in F between 23 and 41 DAI corresponded with increased leachate K concentrations due to CRF release (data not shown). Displacement of Ca from exchange sites in pine bark by K has been reported and may explain the increasing leachate Ca concentration in F between 23 and 48 DAI (Hoskins et al.,

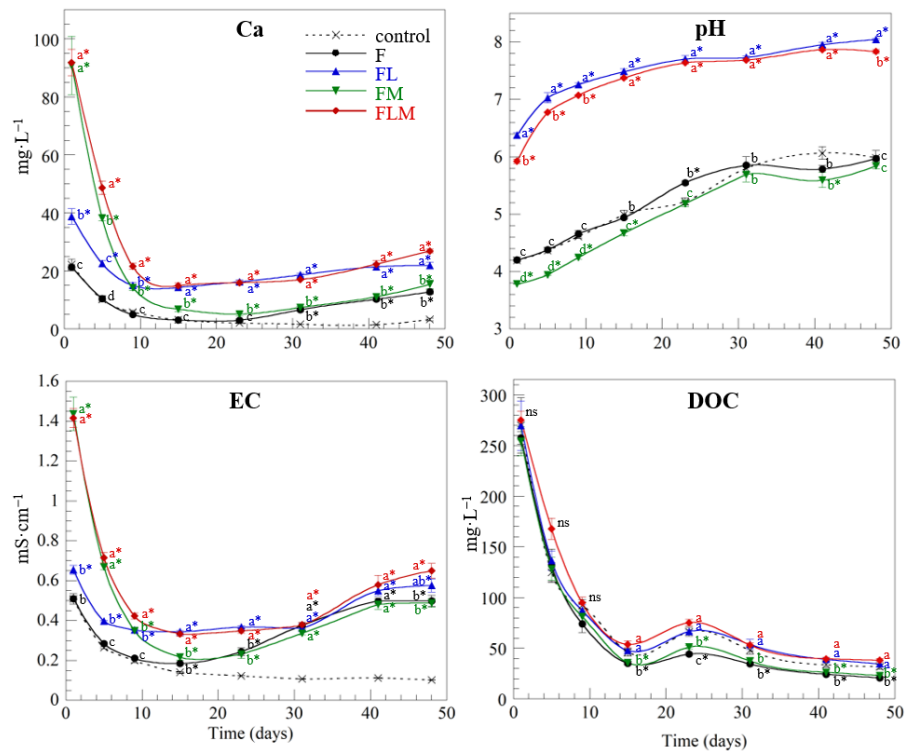


Fig. 1. Effect of substrate amendments on Ca concentration, pH, electrical conductivity (EC), and dissolved organic carbon (DOC) concentration in leachate over time of daily-irrigated pine bark columns containing 4.75 kg m^{-3} controlled-release fertilizer (F; 19N–2.6P–10K). Substrate treatments consisted of 4.15 kg m^{-3} dolomite (FL), 0.89 kg m^{-3} micronutrient fertilizer (FM), the combination of dolomite and micronutrient fertilizer (FLM), or substrate with no controlled-release fertilizer or amendments (control). Different vertically aligned letters next to means indicate significant difference among substrate treatments (F, FL, FM, and FLM) within each sampling date via Tukey's HSD ($p < 0.05$), and non-significant differences are denoted by "ns". Asterisks (*) indicate significant difference from the control using Dunnett's test ($p < 0.05$). Vertical bars represent SE of the mean.

2014). Magnesium concentrations in leachate (data not shown) had a strong, positive correlation with Ca for all substrate treatments ($r \geq 0.940$) due to the coinciding presence of Mg in all sources of Ca (e.g., dolomite, irrigation water, pine bark).

Iron

Iron concentrations were affected by a fraction \times substrate \times time interaction ($p < 0.0001$; Table 1). Among substrates, the highest dissolved Fe concentrations were in FM at 1 to 15 DAI and in the control from 31 to 48 DAI (Table 2). The relatively high dissolved Fe concentrations in FM at 1 to 15 DAI were a result of the Fe supplied by the micronutrient fertilizer in combination with low substrate pH (i.e., 3.8 to 4.7) which maintained Fe in solution. At 31 to 48 DAI, higher dissolved Fe concentrations in the control compared to all other treatments may have been a result of lower OP concentrations in the control to precipitate or coprecipitate with dissolved Fe (Table 3). The observed relatively low dissolved Fe concentrations in substrates containing the micronutrient fertilizer at 31 to 48 DAI corresponds with Wright and Hinesley (1991) who reported that granular micronutrient fertilizer amendment rates of up to 1.5 kg m^{-3} in a pine bark substrate had no effect on water-extractable Fe concentrations when measured 60 d after potting *Juniperus virginiana* L. The authors also reported that liming the substrate reduced Fe in water extracts, regardless

Table 1. Degrees of freedom (df), F values, and p values for three-way analysis of variance (ANOVA) to determine significant effects of substrate treatment, fraction, time, and their interactions on P and Fe concentrations in leachate of irrigated pine bark. Two analyses were performed for P, one included total P fractions (particulate P [PP] and total dissolved P [TDP]), and the other included dissolved P fractions (orthophosphate-P [OP] and non-orthophosphate dissolved P [NODP]). Fractions in Fe analysis included particulate Fe and dissolved Fe.

ANOVA source	df	Total P fractions		Dissolved P fractions		Fe fractions	
		F value	p value	F value	p value	F value	p value
3-way ANOVA							
Time (T)	7	273.4	<0.0001	309.8	<0.0001	735.5	<0.0001
Substrate (Sub)†	3	9.9	<0.0001	28.9	<0.0001	93.3	<0.0001
Fraction (Frac)	1	355.2	<0.0001	34.9	<0.0001	1032.4	<0.0001
T × Sub	21	5.8	<0.0001	6.5	<0.0001	13.0	<0.0001
T × Frac	7	45.5	<0.0001	45.8	<0.0001	74.4	<0.0001
Sub × Frac	3	11.6	<0.0001	14.2	<0.0001	26.1	<0.0001
T × Sub × Frac	21	3.0	0.0002	4.3	<0.0001	16.6	<0.0001
Simple interactions‡							
T × Sub (in each Frac)							
PP, NODP, particulate Fe	21	4.8	<0.0001	1.2	0.3186	13.0	<0.0001
TDP, OP-P, dissolved Fe	21	4.3	<0.0001	10.7	<0.0001	11.8	<0.0001
T × Frac (in each Sub)							
F	7	34.8	<0.0001	10.1	0.0026	58.9	<0.0001
FL	7	14.0	<0.0001	3.8	0.0480	46.7	<0.0001
FM	7	3.3	0.0370	10.9	0.0032	12.6	<0.0001
FLM	7	18.0	<0.0001	60.9	<0.0001	36.8	<0.0001
Sub × Frac (at each T)							
1	3	6.5	0.0025	27.5	<0.0001	120.4	<0.0001
5	3	5.7	0.0042	18.6	<0.0001	37.3	<0.0001
9	3	2.1	0.1336	3.5	0.0296	5.0	0.0081
15	3	2.5	0.0812	4.4	0.0130	1.2	0.3340
23	3	3.0	0.0496	0.2	0.8897	2.0	0.1481
31	3	15.3	<0.0001	4.0	0.0193	2.8	0.0596
41	3	8.7	0.0004	3.3	0.0369	1.1	0.3795
48	3	8.6	0.0005	2.6	0.0740	1.0	0.4272

† Four substrate amendment treatments each contained 4.75 kg m⁻³ controlled-release fertilizer (CRF; 19N–2.6P–10K) and either 4.15 kg m⁻³ dolomitic limestone (FL), 0.89 kg m⁻³ micronutrient fertilizer (FM), both amendments (FLM), or no amendments (F).

‡ Since 3-way interactions were significant, 2-way interactions were analyzed at each level of the third factor (i.e., simple interactions).

of the presence of micronutrient fertilizer. Similarly, Abreu et al. (2006) found that water-extractable Fe concentrations were similar in limed pine bark with and without added micronutrients. In a study that used the same micronutrient fertilizer product used in the current study, Handreck (1989) found that <1% of 111 mg Fe added to pine bark was recovered in effluent after 50 d of daily leaching, most of which was found within the first 6 d. Our results agree with these studies in that liming agents and the subsequent increase in pH reduces Fe solubility in pine bark.

Particulate-Fe concentrations were highest in the control, F, and FL at 1 DAI and in the control and F at 5 DAI (Table 2). From 9 to 48 DAI, substrates had a similar effect on particulate Fe as that described for dissolved Fe. At 1 DAI, the lower particulate Fe concentrations in substrates containing micronutrient fertilizer suggests Fe-containing particles may have flocculated due to relatively high solution EC (Jirsa et al., 2013; Sholkovitz, 1976). Similarly, at 31 to 48 DAI, low particulate Fe in fertilized substrates relative to the control corresponded with higher EC.

pH

Leachate pH was affected by a substrate × time interaction ($p < 0.0001$), which is explained by the more rapid initial increase in pH of substrates containing dolomite compared to those without dolomite (Fig. 1). Leachate pH in all five substrate treatments increased by $\sim 1.8 \pm 0.07$ units over 48 d. Substrates containing dolomite reached pH values of ~ 8.0 by 41 DAI, which are higher than values reported in studies in which a comparable dolomite rate was used. In a comprehensive review on the effects of dolomite additions in pine bark, Altland and Jeong (2016) proposed that dolomite can increase substrate pH up to ~ 6.5 before pH limits its solubility and concomitant effects on pH. A possible explanation for the observed high pH values is that the inherent acidity of the pine bark was leached due to daily irrigation with ~ 2.6 cm water; however, this amount of water can be applied in a conventional nursery setting via daily irrigation and rainfall. Assuming the inherent acidity of the pine bark was reduced, substrate pH would have been more responsive to the lime reaction and the pH of the irrigation water, which increased from 6.9 to 7.8 over the course of the experiment (data not shown). Leachate pH values of the control and F

were generally equivalent; hence, the CRF had a minor effect on leachate pH. From 1 to 23 DAI, pH values of FM were 0.38 ± 0.03 units lower than those of F. Similarly, pH of FLM was 0.2 to 0.5 units lower than that of FL at 1 to 9 DAI, suggesting the micronutrient fertilizer acidified the substrate. Wright et al. (1999) observed a similar magnitude decrease in pH (0.5 units) when pine bark was amended with micronutrient fertilizer.

Electrical Conductivity

Leachate EC was affected by a substrate \times time interaction ($p < 0.0001$) due to the more rapid decrease in EC in substrates containing micronutrient fertilizer compared to those with no micronutrient fertilizer during the first 9 DAI. Electrical conductivity values in F were the same as those in the control at 1, 5, and 9 DAI during which EC values in both substrates decreased from 0.51 to 0.21 mS cm^{-1} . At 15 DAI, EC values in F were 29% higher than those in the control, and from 15 to 48 DAI, EC values in F and the control increased 178% and decreased 29%, respectively. Accordingly, these data suggest that the fertilizer began releasing salts between 9 and 15 DAI. Electrical conductivity values in FM and FLM were equivalent at 1 and 5 DAI during which EC values in these two treatments decreased $\sim 50\%$. From 1 to 23 DAI, EC values in FL were between 27 and 89% higher than those in F. Observed higher EC values in FL compared to F were likely due to dolomite dissolution and subsequent presence of Mg and Ca in leachate.

Dissolved Organic Carbon

Dissolved organic carbon was influenced by a substrate \times time interaction ($p = 0.0043$; Fig. 1). The DOC concentrations were the same across treatments at 1, 5, and 9 DAI during which DOC decreased from ~ 265 to $\sim 85 \text{ mg L}^{-1}$. Thereafter, DOC concentrations increased by ~ 10 to 20 mg L^{-1} between 15 and 23 DAI then decreased for the remainder of the study. A substrate effect on DOC concentrations was first apparent at 15 DAI, during which the control or substrates containing dolomite had highest DOC. This relationship among treatments remained the same until experiment termination, with DOC concentrations 35 to 45% higher in limed treatments or the control compared to F or FM. The higher DOC concentrations of limed treatments at 15 to 48 DAI was likely due to the relatively high pH and thus greater solubility of humic acids in these treatments (Kipton and Town, 1992). However, DOC concentrations in the control were higher than those in F and FM despite having similar pH values. Higher DOC concentrations of the control compared to the non-limed treatments may be due to the substantially lower EC

Table 2. Effect of substrate amendments ($n = 4$) on Fe concentrations (mg L^{-1}) and partitioning of Fe in the particulate (i.e., total Fe – dissolved Fe) and dissolved ($<0.45 \mu\text{m}$) fractions over time in leachate of daily-irrigated pine bark columns containing 4.75 kg m^{-3} controlled-release fertilizer (F; 19N–2.6P–10K). Substrate treatments consisted of 4.15 kg m^{-3} dolomite (FL), 0.89 kg m^{-3} micronutrient fertilizer (FM), the combination of dolomite and micronutrient fertilizer (FLM), or pine bark with no fertilizer or amendments (control).

	Time, days after initiation							
	1	5	9	15	23	31	41	48
	Dissolved Fe							
	mg L^{-1}							
Control	1.3	0.85	0.56	0.27	0.22	0.22	0.12	0.12
F	1.10 b†	0.79 b	0.38 b	0.19 b	0.05 b*	0.04 b*	0.02 b*	0.01 b*
FL	0.46 c*	0.23 c*	0.19 b*	0.07 b*	0.04 b*	0.06 b*	0.03 ab*	0.01 b*
FM	8.30 a*	3.15 a*	1.30 a*	0.63 a*	0.23 a	0.10 a*	0.04 ab*	0.04 a*
FLM	0.47 c*	0.20 c*	0.20 b*	0.13 b*	0.08 b*	0.10 a*	0.05 a*	0.02 b*
<i>p</i> value	<0.0001	<0.0001	<0.0001	<0.0001	0.0003	0.0014	0.0089	0.0051
	Particulate Fe							
	mg L^{-1}							
Control	22.84	7.67	4.33	1.06	0.71	0.52	0.43	0.27
F	23.64 a	8.00 a	4.11 b	0.87 bc	0.16 b*	0.08 b*	0.08 *	0.04 *
FL	20.96 a	3.70 b*	2.56 c*	0.50 c*	0.16 b*	0.10 b*	0.06 *	0.06 *
FM	7.00 b*	4.00 b*	6.73 a*	2.29 a*	0.92 a	0.22 a*	0.10 *	0.07 *
FLM	9.33 b*	2.53 b*	3.17 bc	1.07 b	0.35 b	0.30 a*	0.14 *	0.08 *
<i>p</i> value	<0.0001	0.0006	<0.0001	0.0002	0.0005	0.0005	0.1016	0.5428

* Significantly different from control according to Dunnett's test at the 0.05 probability level.

† Within columns for dissolve or particulate Fe, means followed by the same letter are not significantly different according to Tukey's HSD (0.05).

values of the control and corresponding degree of DOC flocculation. According to findings by Tiemeyer et al. (2017), pH has less impact on DOC when EC values are low (i.e., $94 \mu\text{S cm}^{-1}$). Hence, the low EC values in the control between 15 and 48 DAI may have nullified the effect of relatively low pH values on DOC.

Phosphorus

The time \times substrate \times treatment interaction was significant for both P-fraction analyses, that is, PP versus TDP and OP versus NODP ($p < 0.0001$; Table 1). For the PP versus TDP analysis, all simple interactions were significant ($p < 0.05$) except the substrate \times fraction interaction at 9 and 15 DAI ($p \geq 0.05$).

Particulate P concentrations were affected by substrate treatments at all sampling dates except 9, 15, and 41 DAI (Table 3). In all substrates, PP concentrations were highest at 1 DAI, decreased rapidly between 1 and 5 DAI, then remained $<1.5 \text{ mg L}^{-1}$. At 1 DAI, highest PP was found in the control, F, and FL, whereas at 5 DAI, PP concentrations were highest in the control, F, and FM. A portion of the PP present at 1 and 5 DAI may be attributed to MnHPO_4 and $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$ precipitates, as both compounds were saturated with respect to their solid phases in all substrates during this period (Table 4). When pooled across all substrates and sampling dates, PP had a strong, positive correlation with particulate Fe ($r = 0.94$) and DOC ($r = 0.86$). These parallel changes in PP, particulate Fe, and DOC suggest possible formation of humic-Fe- PO_4 precipitates (Petrovic and Kastelan-Macan, 1996). However, further investigation is needed to confirm the existence of humic-Fe- PO_4 precipitates in pine bark substrates. At

Table 3. Effect of substrate amendments ($n = 4$) on P concentrations (mg L^{-1} P) and distribution of P concentration across fractions over time in leachate of daily-irrigated pine bark columns containing 4.75 kg m^{-3} controlled-release fertilizer (F; 19N–2.6P–10K). Substrate treatments consisted of 4.15 kg m^{-3} dolomite (FL), 0.89 kg m^{-3} micronutrient fertilizer (FM), the combination of dolomite and micronutrient fertilizer (FLM), or pine bark with no fertilizer or amendments (control). Total P (TP) was fractionated into total dissolved P (TDP; $<0.45 \mu\text{m}$) and particulate P (PP; TP – TDP), and TDP was further divided into orthophosphate-P (OP) and non-orthophosphate dissolved P (NODP; TDP – OP) for separate statistical analysis.

Time, days	Substrate	PP	TDP	p value†	NODP	OP	p value‡
1	Control	6.34	8.60	0.0235	4.08	4.52	0.3479
	F	6.67 a§	8.86 a	0.0097	4.28	4.58 a	0.3554
	FL	5.94 ab	6.57 b*	0.2384	3.54	3.03 b*	0.1264
	FM	3.07 c*	6.24 b*	0.0072	4.29	1.96 c*	0.0053
	FLM	4.04 bc*	4.48 c*	0.2400	3.45	1.03 d*	<0.0001
	p value	0.0003	<0.0001		0.0911	<0.0001	
5	Control	0.97	7.53	<0.0001	3.57	3.97	0.0486
	F	1.28 a	8.06 a	<0.0001	3.65 a	4.42 a	0.0810
	FL	0.56 b	5.46 b*	<0.0001	2.44 b*	3.02 b*	0.0662
	FM	1.04 ab	4.80 b*	0.0003	2.95 ab	1.85 c*	0.0240
	FLM	0.45 b	3.30 c*	<0.0001	2.38 b*	0.92 d*	<0.0001
	p value	0.0125	<0.0001		0.0131	<0.0001	
9	Control	1.21	5.15	<0.0001	1.68	3.47	0.0002
	F	1.28	4.20 a	0.0004	1.31	2.89 a	0.0043
	FL	1.15	3.33 a*	0.0023	1.10 *	2.23 a*	0.0156
	FM	1.37	3.24 a*	0.0021	1.30	1.94 ab*	0.1034
	FLM	0.82	1.98 b*	0.0025	0.90 *	1.08 b*	0.0548
	p value	0.0895	0.0026		0.1631	0.0028	
15	Control	0.25	2.17	0.0008	0.86	1.31	0.5276
	F	0.53	1.15 *	0.0002	0.32	0.84	0.2086
	FL	0.58	1.06 *	0.1272	0.63	0.43	0.4733
	FM	0.43	1.64	0.0014	0.37	1.27	0.1596
	FLM	0.65	0.91 *	0.2607	0.84	0.07	<0.0001
	p value	0.7876	0.0513		0.2437	0.0750	
23	Control	0.26	1.20	<0.0001	0.90	0.30	0.1024
	F	0.09 b	0.60	0.0017	0.20	0.40	0.1732
	FL	0.20 ab	0.69	0.1923	0.37	0.32	0.9330
	FM	0.18 ab	1.11	0.0011	0.45	0.66	0.5723
	FLM	0.39 a	0.50	0.2906	0.21	0.29	0.5760
	p value	0.0279	0.1952		0.8770	0.4832	
31	Control	0.27	0.56	0.0123	0.26	0.30	0.7567
	F	0.06 b*	0.99 ab	0.0008	0.17 ab	0.82 a*	0.0086
	FL	0.22 a	0.27 c	0.7162	0.07 b	0.20 b	0.1191
	FM	0.07 b*	1.25 a*	0.0002	0.23 a	1.02 a*	0.0036
	FLM	0.28 a	0.53 bc	0.0923	0.10 ab	0.43 ab	0.0117
	p value	0.0025	0.0018		0.0453	0.0036	
41	Control	0.20	0.30	0.0740	0.09	0.21	0.0291
	F	0.08 *	1.77 a*	<0.0001	0.27 *	1.50 a*	0.0002
	FL	0.13	0.81 bc*	0.0003	0.12	0.69 c*	0.0016
	FM	0.10	1.57 ab*	<0.0001	0.22	1.35 ab*	0.0003
	FLM	0.19	0.83 c*	0.0089	0.13	0.70 bc*	0.0123
	p value	0.1781	0.0046		0.0910	0.0050	
48	Control	0.34	0.23	0.8684	0.08	0.15	0.0921
	F	0.16 bc	2.47 a*	<0.0001	0.36 *	2.11 a*	<0.0001
	FL	0.36 ab	1.20 b*	0.2441	0.14	1.06 b*	0.0011
	FM	0.09 c	1.98 ab*	0.0003	0.32	1.66 ab*	0.0032
	FLM	0.40 a	1.17 b*	0.0133	0.23	0.94 b*	0.0101
	p value	0.0022	0.0138		0.2534	0.0069	

* Significantly different from control according to Dunnett's test at the 0.05 probability level.

† p values < 0.05 indicate TDP and PP concentrations are significantly different.

‡ p values < 0.05 indicate OP and NODP concentrations are significantly different.

§ Within columns for each sampling date, means followed by the same letter are not significantly different according to Tukey's HSD (0.05).

31 and 48 DAI, highest PP concentrations were in the control and substrates containing dolomite (Table 3). Relatively high PP concentrations in substrates containing dolomite at 31 and 48 DAI, during which pH values were favorable for Ca-PO₄ precipitation, may have been due to formation of Ca-PO₄ precipitates. Formation of Ca-PO₄ precipitates is supported by data from Visual MINTEQ modeling which indicated leachate solutions from FL and FLM were saturated with respect to the Ca₅(PO₄)₃(OH) solid phase at 41 and 48 DAI (Table 4). The solid phase of Ca₃(PO₄)₂ was also saturated in FL and FLM at 48 DAI; however, saturation index values of Ca₃(PO₄)₂ were low relative to those of Ca₅(PO₄)₃(OH). Kinetics, metastable fast formers, and the presence of DOC can limit precipitation and thus interpretation of this simulation output. Relatively high DOC concentrations, such as those observed in the current study make interpretation of Ca-PO₄ precipitate formation particularly challenging, as DOC has been shown to impede nucleation of Ca-PO₄ precipitates (Song et al., 2006).

Total dissolved P concentrations in F were similar to those in the control until 41 DAI (Table 3), suggesting indigenous P in the pine bark was the primary source of TDP until the CRF began releasing P between 31 and 41 DAI. Delayed P release from CRF is commonly reported in nursery crop production research (Broschat, 2005; Broschat and Moore, 2007; Du et al., 2006; Newman et al., 2006). Total dissolved P concentrations in the control decreased 75% between 1 and 15 DAI. Yeager and Wright (1982) also reported relatively high amounts of soluble P in a pine bark substrate. At 1, 5, 41, and 48 DAI, TDP concentrations were lower in FL and FLM than in F. Total dissolved P concentrations were lowest in FLM at 1 and 5 DAI, whereas at 41 and 48 DAI, FL and FLM had equivalent TDP concentrations. The FM substrate reduced TDP at 1 and 5 DAI in similar amounts as FL; however, TDP concentrations in FM were the same as those in F at 9 to 48 DAI. These data suggest that while micronutrient fertilizer reduced TDP concentrations by 30 and 40% at 1 and 5 DAI, respectively, this effect was short-lived. The greater effectiveness of FLM at reducing TDP concentrations at the beginning of the study, relative to the end of the study, can therefore be explained by initial TDP reductions by the micronutrient fertilizer. Decreases in TDP by dolomite and the micronutrient fertilizer did not necessarily correspond with a proportional increase in PP, suggesting dolomite and micronutrient fertilizer amendments improved total P retention in the substrate. To further investigate the effect of substrate amendments on P retention in pine bark, an additional repeated measures ANOVA was performed on total P (TDP + PP; data not shown). Assessment of total P concentrations revealed a similar response to substrate treatments as was described for TDP. The effect of substrate on TP was significant at 1 DAI ($p < 0.0001$), 5 DAI ($p < 0.0001$), 9 DAI ($p = 0.0004$), 31 DAI ($p = 0.0050$), and 41 DAI ($p = 0.0076$). The FLM substrate reduced TP concentrations, on average, by 51% at 1, 5, and 9 DAI and by 45% at 41 DAI. The FL substrate reduced TP concentrations by 19 and 36% at 1 and 5 DAI, respectively, and by ~52% at 31 and 41 DAI. In contrast,

Table 4. Saturation indices calculated by Visual MINTEQ for P species saturated with respect to the solid phase (positive values) in leachate at 1, 5, 40, and 48 d after experiment initiation of irrigated pine bark columns containing no amendments (control) or 4.75 kg m⁻³ of 19N-2.6P-10K controlled-release fertilizer (F) with 4.15 kg m⁻³ dolomite (FL), 0.89 kg m⁻³ micronutrient fertilizer (FM), or the combination of dolomite and micronutrient fertilizer (FLM).

Substrate	Species	Time, days after initiation			
		1	5	41	48
Control	MnHPO ₄	1.32	0.90	-1.09	-0.87
F	MnHPO ₄	1.32	0.98	2.21	2.41
FL	Ca ₅ (PO ₄) ₃ (OH)	2.01	4.09	7.00	7.70
	MnHPO ₄	2.95	2.97	2.11	2.09
FM	Ca ₃ (PO ₄) ₂ (beta)	-2.43	-1.37	-0.12	0.30
	MnHPO ₄	1.94	1.65	2.22	2.61
FLM	Ca ₅ (PO ₄) ₃ (OH)	0.23	3.51	6.59	7.26
	MnHPO ₄	3.42	3.53	2.53	2.50
	Ca ₃ (PO ₄) ₂ (beta)	-3.44	-1.70	-0.33	0.10

effects of FM on TP concentrations were apparent only at 1 and 5 DAI, during which TP concentrations were ~36% lower than those in F. Thus, TP retention in FLM was initially controlled by the combination of dolomite and micronutrient fertilizer and later by dolomite only. The short-term effect of micronutrient fertilizer on P sorption may be related to its solubility. The rapid decrease in Ca and Mg to concentrations equivalent to or nearly equivalent to those in F by 23 DAI suggests that most of the dolomite in the micronutrient fertilizer had dissolved by 31 DAI. The component in the micronutrient fertilizer controlling P sorption is unclear however since the dolomite, Mn, and Fe constituents in the micronutrient fertilizer may have had a role. Spectroscopic evidence has shown that retention of P by dolomite at relatively low P concentrations (i.e., <2 mg L⁻¹) is primarily due to PO₄ sorption to Ca and Mg sorption complexes (Xu et al., 2014).

The substrate × time simple interaction was significant for OP ($p < 0.0001$), but not for NODP ($p = 0.3186$; Table 1). The substrate × time interaction for OP was due to the decreasing effect of micronutrient fertilizer on OP concentrations over time. At 1 and 5 DAI, OP concentrations were lower in FM than in FL, and lowest concentrations were in FLM. In contrast, at 31 to 48 DAI, OP concentrations in FM were the same as those in F, and lowest concentrations were found in FL to FLM. Hence, the response of OP to substrate amendments was similar to that of TDP; however, percentage reductions in OP in response to amendments were generally greater than those of TDP. Orthophosphate-P was 23 to 55% of TDP in all substrates at 1 and 5 DAI, with generally lower proportions found in FLM. Thirty-one to 48 DAI, when the CRF was releasing P, OP contributed 74 to 86% of TDP in fertilized treatments. The relatively high initial NODP concentrations (i.e., 3.9 mg L⁻¹) decreased until 31 DAI, then remained relatively low compared to OP. The NODP fraction may have been associated with dissolved metal-humic complexes (Gerke, 2010). This fate of P is supported by Visual MINTEQ aqueous speciation output which predicted between 95% and 99% of Fe in solution was associated organic acids (data not shown). In addition, the positive correlation between NODP and DOC ($r = 0.90$) was stronger

that that between OP and DOC ($r = 0.58$), which supports the possibility of an association between NODP and organic substances. Van Moorlehem et al. (2011) determined that organic Fe colloids present in filtered solutions ($<0.45 \mu\text{m}$) reduced detection of OP by ion chromatography by as much as 51%, whereas TDP (determined via ICP–OES) was unaffected. Additional research is needed to investigate the OP to TDP ratio in pine bark once CRF release increases OP concentrations to 5 to 10 mg L^{-1} , the current recommended range according to best management practices for container-grown crops (Bilderback et al., 2013a).

CONCLUSIONS

Results demonstrate that dolomite and micronutrient fertilizer improve P retention and therefore reduce P leaching from a pine bark substrate commonly used for production of containerized ornamentals. Compared to the fertilized substrate containing no amendments, the pine bark with dolomite and micronutrient fertilizer reduced OP concentrations by 70% when averaged across the sampling dates. The effects of micronutrient fertilizer on P retention were diminished by the ninth irrigation event (23 cm), whereas dolomite reduced OP leaching throughout the 48-d study. Longevity and extent of P-retention of dolomite when baseline P concentrations are $>5 \text{ mg L}^{-1}$ need to be further investigated. If dolomite additions to pine bark can reduce P leaching throughout a growing season, dolomite amendments may be considered a best management practice for containerized crop production.

ACKNOWLEDGMENTS

The authors thank Julie Brindley, Velva Groover, Anna Birnbaum, and Doug Sturtz for technical assistance, as well as Pacific Organics for donating pine bark for this research. Funding was provided by the Virginia Agricultural Experiment Station and the Hatch Program of the National Institute of Food and Agriculture (including SCRI 2014-51181-22372), the US Dep. of Agriculture, the Horticultural Research Institute, and the Virginia Nursery and Landscape Association.

REFERENCES

- Abreu, C.A., A.M.C. Furlani, P.R. Furlani, M.F. Abreu, O.C. Bataglia, and A. Paz-Gonzalez. 2006. Quest of water extract analysis of micronutrients in soilless organic substrates. *Commun. Soil Sci. Plant Anal.* 37:2327–2338. doi:10.1080/00103620600819602
- Altland, J.E., and K.Y. Jeong. 2016. Dolomitic lime amendment affects pine bark substrate pH, nutrient availability, and plant growth: A review. *Horttechnology* 26:565–573. doi:10.21273/HORTTECH03465-16
- Alva, A.K., M.E. Sumner, and W.P. Miller. 1991. Relationship between ionic strength and electrical conductivity for soil solutions. *Soil Sci.* 152:239–242. doi:10.1097/00010694-199110000-00001
- Argo, W.R., and J.A. Biernbaum. 1996a. The effect of lime, irrigation-water source, and water-soluble fertilizer on root-zone pH, electrical conductivity, and macronutrient management of container root media with impatiens. *J. Am. Soc. Hortic. Sci.* 121:442–452. doi:10.21273/JASHS.121.3.442
- Argo, W.R., and J.A. Biernbaum. 1996b. Availability and persistence of macronutrients from lime and preplant nutrient charge fertilizers in peat-based root media. *J. Am. Soc. Hortic. Sci.* 121:453–460. doi:10.21273/JASHS.121.3.453
- Bilderback, T., C. Boyer, M. Chappell, G. Fain, D. Fare, C. Gilliam, B.E. Jackson, J. Lea-Cox, A.V. LeBude, A. Niemiera, J. Owen, J. Ruter, K. Tilt, S. Warren, S. White, T. Whitwell, R. Wright, and T. Yeager. 2013a. Best management practices: Guide for producing nursery crops. 3rd ed. Southern Nursery Association, Acworth, GA.
- Bilderback, T.E., E.D. Riley, B.E. Jackson, H.T. Kraus, W.C. Fonteno, J.S. Owen, Jr., J.E. Altland, and G.B. Fain. 2013b. Strategies for developing sustainable substrates in nursery crop production. *Acta Hortic.* 1013:43–56. doi:10.17660/ActaHortic.2013.1013.2
- Broschat, T.K., and K.K. Moore. 2007. Release rates of ammonium-nitrogen, nitrate-nitrogen, phosphorus, potassium, magnesium, iron, and manganese from seven controlled-release fertilizers. *Commun. Soil Sci. Plant Anal.* 38:843–850. doi:10.1080/00103620701260946
- Broschat, T. 1995. Nitrate, phosphate, and potassium leaching from container-grown plants fertilized by several methods. *HortScience* 30:74–77. doi:10.21273/HORTSCI.30.1.74
- Broschat, T. 2005. Rates of ammonium-nitrogen, nitrate-nitrogen, phosphorus, and potassium from two controlled-release fertilizers under different substrate environments. *Horttechnology* 15:332–335. doi:10.21273/HORTTECH.15.2.0332
- Bunt, A.C. 1988. Media and mixes for container-grown plants: A manual on the preparation and use of growing media for pot plants. 2nd ed. Unwin Hyman, London. doi:10.1007/978-94-011-7904-1
- Carpenter, S.R., N.F. Caraco, D.L. Correll, R.W. Howarth, A.N. Sharpley, and V.H. Smith. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. *Ecol. Appl.* 8:559–568. doi:10.1890/1051-0761(1998)008[0559:NPOSWW]2.0.CO;2
- Cole, J.C., and J.M. Dole. 1997. Temperature and phosphorus source affect phosphorus retention by pine bark-based container medium. *HortScience* 32:236–240. doi:10.21273/HORTSCI.32.2.236
- Dennis, J.H., R.G. Lopez, B.K. Behe, C.R. Hall, C. Yue, and B.L. Campbell. 2010. Sustainable production practices adopted by greenhouse and nursery plant growers. *HortScience* 45:1232–1237. doi:10.21273/HORTSCI.45.8.1232
- Diara, C., L. Incrocci, G. Incrocci, G. Carmassi, A. Pardossi, P. Marzalletti, P. Cozzi, F. Fibbi, and R. Clemens. 2014. Reduction of nutrient run-off by the use of coated slow-release fertilizers on two container-grown nursery crops. *Acta Hortic.* 1034:85–91. doi:10.17660/ActaHortic.2014.1034.9
- Du, C., J. Zhou, and A. Shaviv. 2006. Release characteristics of nutrients from polymer-coated compound controlled release fertilizers. *J. Polym. Environ.* 14:223–230. doi:10.1007/s10924-006-0025-4
- Fain, G.B., C.H. Gilliam, K.M. Tilt, J.W. Olive, and B. Wallace. 2000. Survey of best management practices in container production nurseries. *J. Environ. Hortic.* 18:142–144.
- Fonteno, W.C., C.T. Hardin, and J.P. Brewster. 1995. Procedures for determining physical properties of horticultural substrates using the NCSU porometer. Horticultural Substrates Laboratory, North Carolina State University, Raleigh, NC. https://projects.ncsu.edu/project/hortsublab/pdf/porometer_manual.pdf.
- Gautelier, M., E.H. Oelkers, and J. Schott. 1999. An experimental study of dolomite dissolution rates as a function of pH from -0.5 to 5 and temperature from 25 to 80°C. *Chem. Geol.* 157:13–26. doi:10.1016/S0009-2541(98)00193-4
- Gerke, J. 2010. Humic (organic matter)-Al(Fe)-phosphate complexes: An underestimated phosphate form in soils and source of plant-available phosphate. *Soil Sci.* 175:417–425. doi:10.1097/SS.0b013e3181f1b4dd
- Gerke, J., and R. Hermann. 1992. Adsorption of orthophosphate to Humic-Fe-Complexes and to amorphous Fe-Oxide. *Z. Pflanzenernähr. Boden.* 155:233–236. doi:10.1002/jpln.19921550313
- Godoy, A., and J.C. Cole. 2000. Phosphorous source affects phosphorous leaching and growth of containerized spirea. *HortScience* 35:1249–1252. doi:10.21273/HORTSCI.35.7.1249
- Gustafsson, J.P. 2013. Visual MINTEQ ver. 3.1. KTH, Stockholm. <https://vminteq.lwr.kth.se/> (Verified 15 Aug. 2019).
- Handreck, K.A. 1989. Assessment of iron availability in soilless potting media. *Commun. Soil Sci. Plant Anal.* 20:1297–1320. doi:10.1080/00103628909368152
- Handreck, K.A. 1991a. Interactions between iron and phosphorus in the nutrition of *Banksia ericifolia* L.f. var. *ericifolia* (Proteaceae) in soil-less potting media. *Aust. J. Bot.* 39:373–384. doi:10.1071/BT9910373
- Handreck, K.A. 1991b. Available phosphorus in potting media: Extractants and interpretation of extract data. *Commun. Soil Sci. Plant Anal.* 22:529–557. doi:10.1080/00103629109368436
- Handreck, K.A. 1992. Iron phosphorous interactions in the nutrition of seedling macadamia in organic potting media. *Australian J. Exp. Hort.* 32:773–779.
- Handreck, K.A. 1996. Comparison of phosphate-phosphorous and total phosphorus in DTPA extracts for assessing plant-available phosphorus in soilless potting media. *Commun. Soil Sci. Plant Anal.* 27:2125–2135. doi:10.1080/00103629609369692

- Havis, J.R., and J.H. Baker. 1985. Influence of liming rate and phosphorus leaching from a peat-sand medium. *J. Environ. Hortic.* 3:74–76.
- Haynes, J.R. 1982. Leaching losses of nutrients and yield and nutrient uptake by container-grown begonia as affected by lime and fertilizer applications to a peat medium. *J. Sci. Food Agric.* 33:407–413. doi:10.1002/jfsa.2740330503
- Hoskins, T.C., J.S. Owen, Jr., and A.X. Niemiera. 2014. Controlled-release fertilizer placement affects the leaching pattern of nutrients from nursery containers during irrigation. *HortScience* 49:1341–1345. doi:10.21273/HORTSCI.49.10.1341
- Jirsa, F., E. Neubauer, R. Kittinger, T. Hofmann, R. Krachler, F. Kammer, and B.K. Keppler. 2013. Natural organic matter and iron export from the Tanner Moor, Austria. *Limnologia* 43:239–244.
- Johnson, N.L. 1949. Systems of frequency curves generated by methods of translation. *Biometrika* 36:149–176. doi:10.1093/biomet/36.1-2.149
- Karaca, S., A. Gürses, M. Edjer, and M. Açıkıldız. 2004. Kinetic modeling of liquid-phase adsorption and phosphate on dolomite. *J. Colloid Interface Sci.* 277:257–263. doi:10.1016/j.jcis.2004.04.042
- Karaca, S., A. Gürses, M. Edjer, and M. Açıkıldız. 2006. Adsorptive removal of phosphate from aqueous solutions using raw and calcinated dolomite. *J. Hazard. Mater.* 128:273–279. doi:10.1016/j.jhazmat.2005.08.003
- Kipton, H.J.P., and R.M. Town. 1992. Solubility and fractionation of humic acid; effect of pH and ionic medium. *Anal. Chim. Acta* 267:47–54. doi:10.1016/0003-2670(92)85005-Q
- Lindsay, W.L. 1979. *Chemical equilibria in soils*. John Wiley & Sons, New York.
- Lu, W., J.L. Sibley, C.H. Gilliam, J.S. Bannon, and Y. Zhang. 2006. Estimation of US bark generation and implications for horticultural industries. *J. Environ. Hortic.* 24:29–34.
- Mack, R., J.S. Owen, A.X. Niemiera, and J. Latimer. 2017. Virginia nursery and greenhouse grower survey of best management practices. *HortTechnology* 27:386–392. doi:10.21273/HORTTECH03664-17
- Mangwandi, C., A.B. Albadarin, Y. Glocheux, and G.M. Walker. 2014. Removal of ortho-phosphate form aqueous solution by adsorption onto dolomite. *J. Environ. Chem. Eng.* 2:1123–1130. doi:10.1016/j.jece.2014.04.010
- Marconi, D.J., and P.V. Nelson. 1984. Leaching of applied phosphorus in container media. *Sci. Hortic. (Amsterdam)* 22:275–285. doi:10.1016/0304-4238(84)90061-X
- McGinnis, M.S., S.L. Warren, and T.E. Bilderback. 2009. Replacing conventional nursery crop nutrient inputs with vermicompost for container production of *Hibiscus moscheutos* L. 'Luna Blush'. *HortScience* 44:1690–1703.
- Moore, P.A., Jr., and D.M. Miller. 1994. Decreasing phosphorus solubility in poultry litter with aluminum, calcium, and iron amendments. *J. Environ. Qual.* 23:325–330. doi:10.2134/jeq1994.00472425002300020016x
- Newman, J.P., J.P. Albano, D.J. Merhaut, and E.K. Blythe. 2006. Nutrient release from controlled-release fertilizers in a neutral-pH substrate in an outdoor environment: I. Leachate electrical conductivity, pH, and nitrogen, phosphorus, and potassium concentrations. *HortScience* 41:1674–1682. doi:10.21273/HORTSCI.41.7.1674
- Owen, J.S., S.L. Warren, T.E. Bilderback, and J.P. Albano. 2008. Phosphorus rate, leaching fraction, and substrate influence on influent quantity, effluent nutrient content, and response of a containerized woody ornamental crop. *HortScience* 43:906–912. doi:10.21273/HORTSCI.43.3.906
- Paradelo, R., M. Conde-Cid, M. Arias-Estévez, J.C. Nóvoa-Muñoz, E. Álvarez-Rodríguez, M.J. Fernández-Sanjurjo, and A. Núñez-Delgado. 2017. Removal of anionic pollutants by pine bark influenced by the mechanism of retention. *Chemosphere* 167:139–145. doi:10.1016/j.chemosphere.2016.09.158
- Peters, J., S. Combs, B. Hoskins, J. Jarman, J. Kovar, M. Watson, A. Wolf, and N. Wolf. 2003. Recommended methods of manure analysis. Univ. of Wisconsin Coop. Ext. Publ. A3769. Coop. Ext. Publishing, Madison, WI.
- Petrovic, M., and M. Kastelan-Macan. 1996. The uptake of inorganic phosphorus by insoluble metal-humic complexes. *Water Sci. Technol.* 34:253–258. doi:10.2166/wst.1996.0629
- Rich, D. 2005. The removal of total phosphorus from natural waters by precipitation. Ph.D. diss., Northwestern International Univ., Evanston, IL.
- Sanyal, S.K., and S.K. De Datta. 1991. Chemistry of phosphorous transformations in soil. *Adv. Soil Sci.* 16:1–94.
- Sholkovitz, E.R. 1976. Flocculation of dissolved organic and inorganic matter during the mixing of river water and seawater. *Geochim. Cosmochim. Ac.* 40:831–845.
- Sinaj, S., F. Mächler, E. Frossard, C. Faisse, A. Oberson, and C. Morel. 1998. Interference of colloidal particles in the determination of orthophosphate concentrations in soil water extracts. *Commun. Soil Sci. Plant Anal.* 29:1091–1105. doi:10.1080/00103629809370011
- Song, Y., H.H. Hahn, E. Hoffmann, and P.G. Weidler. 2006. Effect of humic substances on the precipitation of calcium phosphate. *J. Environ. Sci. (China)* 18:852–857. doi:10.1016/S1001-0742(06)60004-1
- Strickland, J. 1998. The development and application of phosphorus removal from wastewater using biological and metal precipitation techniques. *Water Environ. J.* 12:30–37. doi:10.1111/j.1747-6593.1998.tb00143.x
- Tasistro, A.S., and D.E. Kissel. 2006. Composition of aqueous extracts of broiler litter treated with aluminum sulfate, ferrous sulfate, ferric chloride and gypsum. *J. Environ. Sci. Health B* 41:989–1000. doi:10.1080/03601230600808620
- Tiemeyer, B., N. Pfaffner, S. Frank, K. Kaiser, and S. Fieldler. 2017. Pore water velocity and ionic strength effects on DOC release from peat-sand mixtures: Results from laboratory and field experiments. *Geoderma* 296:86–97. doi:10.1016/j.geoderma.2017.02.024
- Tyler, H.H., S.L. Warren, and T.E. Bilderback. 1996. Reduced leaching fractions improve irrigation use efficiency and nutrient efficacy. *J. Environ. Hortic.* 14:199–204.
- USDA. 2014. 2012 Census of agriculture: Census of horticultural specialties. USDA, Washington, DC.
- USEPA. 2017. Nutrient pollution: Sources and solutions. US Environmental Protection Agency, Washington, DC. <https://www.epa.gov/nutrientpollution/sources-and-solutions> (Accessed 15 May 2018).
- Van Moorleghe, C., L. Six, F. Degryse, E. Smolders, and R. Merckx. 2011. Effect of organic P forms and P present in inorganic colloids on the determination of dissolved P in environmental samples by the diffusive gradient in thin films technique, ion chromatography, and colorimetry. *Anal. Chem.* 83:5317–5323. doi:10.1021/ac200748e
- Warncke, D.D. 1986. Analyzing greenhouse growth media by the saturation extraction method. *HortScience* 21:223–225.
- Warren, S.L., T.E. Bilderback, and H.H. Kraus. 2001. Method of fertilizer application affects nutrient losses of controlled-release fertilizer. *Acta Hortic.* 548:349–356. doi:10.17660/ActaHortic.2001.548.40
- Warren, S.L., T.E. Bilderback, and H.H. Tyler. 1995. Efficacy of three nitrogen and phosphorus sources in container-grown azalea production. *J. Environ. Hortic.* 13:147–151.
- Weir, C.C., and R.J. Soper. 1963. Interaction of phosphates with ferric organic complexes. *Can. J. Soil Sci.* 43:393–399. doi:10.4141/cjss63-046
- Wolfinger, R. 1993. Covariance structure selection in general mixed models. *Commun. Statist. Simula.* 22:1079–1106.
- Worsfold, P., I. McKelvie, and P. Monbet. 2016. Determination of phosphorus in natural waters: A historical review. *Anal. Chim. Acta* 918:8–20. doi:10.1016/j.aca.2016.02.047
- Wright, A.N., A.X. Niemiera, J.R. Harris, and R.D. Wright. 1999. Micronutrient fertilization of woody seedlings essential regardless of pine bark pH. *J. Environ. Hortic.* 17:69–72.
- Wright, R.D., and L.E. Hinesley. 1991. Growth of containerized Eastern red cedar amended with dolomitic limestone and micronutrients. *HortScience* 26:143–145. doi:10.21273/HORTSCI.26.2.143
- Xu, N., M. Chen, K. Zhou, Y. Wang, H. Yin, and Z. Chen. 2014. Retention of phosphorus on calcite and dolomite: Speciation and modeling. *RSC Advances* 4:35205–35214. doi:10.1039/C4RA05461J
- Yeager, T.H., and J.E. Barrett. 1984. Phosphorus leaching from ³²P-superphosphate-amended soilless container media. *HortScience* 19:216–217.
- Yeager, T.H., and J.E. Barrett. 1985a. Phosphorous and sulfur leaching from an incubated superphosphate-amended soilless container medium. *HortScience* 20:671–672.
- Yeager, T.H., and J.E. Barrett. 1985b. Influence of incubation time on phosphorous leaching from a container medium. *J. Environ. Hortic.* 3:186–187.
- Yeager, T.H., and R.D. Wright. 1982. Pine bark-phosphorus relationships. *Commun. Soil Sci. Plant Anal.* 13:57–66. doi:10.1080/00103628209367244
- Yuan, X., W. Xia, J. An, W. Yang, and J. Yin. 2014. Removal of phosphate anions from aqueous solutions using dolomite as adsorbent. *Adv. Mat. Res.* 864–867:1454–1457.
- Yuan, X., X. Wentag, J. An, J. Yin, X. Zhou, and W. Yang. 2015. Kinetic and thermodynamic studies on the phosphate adsorption removal by dolomite mineral. *J. Chem.* 2015:1–8.