Fertilizer Effects of Soy-plastic Containers during Crop Production and Transplant Establishment

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Abstract. As part of a project to develop and assess bio-based, biodegradable plastics for their potential to replace petroleum-based plastics in specialty-crop containers, we evaluated prototype containers made of protein-based polymers from soybean [Glycine max (L.) Merr.] for their effectiveness during production of plants in greenhouses and subsequent establishment of those plants outdoors. Our objective was to assess the function and biodegradation of soy-based plastic containers with special attention to whether a fertilizer effect results from degrading containers before and after plants are moved outdoors. In our first experiment, plants of tomato (Solanum lycopersicum L.) and pepper (Capsicum annuum L.) were grown in soy-plastic containers and control containers of petroleum-based (polypropylene) plastic under greenhouse conditions for 4 weeks. Each plant then was transplanted and grown in an outdoor garden plot for 5 weeks with the container removed, broken into pieces less than 4 cm in diameter, and installed beneath the roots of the transplant. Three additional experiments were performed: a greenhouse trial to quantify the relative concentration and form of plant-available nitrogen (N) released from soy-plastic containers of three types [soy plastic, soy plastic coated with polylactic acid (PLA), and soy–PLA polymer blended 50:50 by weight] during production; a greenhouse trial to evaluate the same three container types under production conditions with five container-crop species; and a field trial to assess the effects of the 50:50 soy–PLA container on transplant establishment. Plant-available N was released from soy-based plastic containers during greenhouse production, and transplant establishment was enhanced when the soy-based container was removed, crushed, and installed in the soil near plant roots. During greenhouse production, containers of high-percentage soy plastic released N at an excessive rate (623 mg L\(^-1\) in leachate) and predominantly in the form of NH\(_4\)\(^+\) (99.4% at 3 weeks of culture). Containers made by blending soy plastic with PLA released N at a favorable rate during production. In both field trials, growth and health of plants cultured in soy containers were better than those of controls. Although the design and material formulation of soy-plastic containers need to be improved to optimize container integrity and plant health during production, our results illustrate the potential to use soy-based plastics in biodegradable containers that release N at rates that promote growth and health of plants during greenhouse production and establishment of transplants outdoors.

An important obstacle to long-term sustainability in the container-crops industry is the nearly universal reliance on containers made from petroleum-based plastics. Although petroleum-plastic containers favor efficiency and profitability, their use comes at a large and increasing cost in terms of the non-renewable source of the materials, the rising price of petroleum, and the environmental damage caused by disposal of non-biodegradable plastics (Botts, 2007). Sustainability and environmental impacts are becoming more important to both producers and consumers of container crops (Hall et al., 2010; Yue et al., 2010), and although container performance and cost are important drivers of a grower’s choice of container, the common practice of using a container for only one production cycle does not require that containers be made of non-biodegradable materials that will last hundreds of years. Alternatives to petroleum plastic containers are commercially available (Kuehny et al., 2011), but many of these existing alternatives, most of which are made of natural fibers, perform poorly or have a poor water-use efficiency during crop production compared with plastic containers or do not degrade as well as suggested (Evans and Hensley, 2004; Evans and Karcher, 2004; Evans et al., 2010). The performance of many emerging bio-plastics and biocomposites compares well with petroleum plastics in trials evaluating their use in specialty-crop containers (Evans et al., 2010; Grewell et al., 2013). Although these new materials have potential to improve sustainability and to reduce environmental impact (Grewell et al., 2013), availability of materials is a critical issue that will influence implementation of the technology. In 2012, NatureWorks LLC (Minnetonka, MN), which specializes in PLA, was the largest producer of biopolymers in the world with an annual production capacity of 140 million kilograms (NatureWorks LLC, 2012), approximately double the capacity of any other biopolymer producer. With an annual industry-wide requirement of over 750 million kilograms of plastic for horticultural containers based on container-crop units produced in 2009 (Schrader, 2013; USDA, 2009), replacement of even a small percentage of the plastic materials needed by the container-crop industry will depend in part on the current and future production capacity of the biopolymer producers. Along with strong growth anticipated in the biopolymers industry (Neill, 2012), innovations in container design and material formulation will help resolve issues of material availability. The amount of a specific biopolymer required for a bioplastic application can be reduced by blending two or more biopolymers or by developing composites that incorporate low-cost natural fibers or fillers (Grewell et al., 2013). Trials evaluating the function and fertilizer effect of plastics made from blends of keratin protein indicate that protein-based plastics may be good replacements for petroleum-based plastics in plant containers because protein polymers could provide an inherent source of plant-available N (Choi and Nelson, 1996; Evans and Hensley, 2004; Roh et al., 2012). Soy-protein polymers and polymer blends have received little attention as replacements for conventional plastics in horticultural applications, but they may offer advantages. Soy feedstock is abundant and has an existing supply infrastructure, soy-based plastics may supply plant-available N as the soy protein degrades, and blending soy with other biopolymers may improve the biodegradability of the other biopolymer (Grewell et al., 2013).

In 2011 and 2012 we created prototype containers from blends of soy protein, soy
flour, and PLA. We used the prototype containers in greenhouse and field experiments with pepper, tomato, salvia (Salvia splendens Sellow ex Roem. & Schult.), marigold (Tagetes patula L.), and petunia (Petunia × hybridra Vilm.) to measure plant health, size, and visual quality during culture and to quantify the pH, electrical conductivity (EC), NH$_4^+$-N, NO$_3^-$-N, NO$_2^-$-N, and total N of leachate from plant/container units. These five plant species were chosen because they are popular container-grown crops, and N requirements vary among the five species. Preliminary trials suggested that containers made of high-percentage soy plastic (soy plastic that was not blended with other materials) degraded too quickly for use in greenhouse production and produced an excessive fertilizer effect. Therefore, our first two experiments were designed to evaluate containers made of high-percentage soy plastic dip-coated with paraffin wax. Results of these experiments led us to examine and compare the performance and fertilizer effects of containers made of high-percentage soy plastic, high-percentage soy plastic dip coated with bio-based PLA, and soy–PLA plastic blended 50:50 by weight. Our objectives were to determine if a beneficial fertilizer effect results from soy-based plastic containers during plant culture, to characterize the effects of blending or coating soy plastic with PLA, and to establish baseline data to guide the development of improved polymer formulations for bioplastic containers.

Materials and Methods

Evaluation of wax-coated soy-plastic containers

Expt. 1: Greenhouse trial with pepper and tomato. The performance, biodegradation, and fertilizer effect of wax-coated soy containers were evaluated. Plants of ‘California Wonder 300’ pepper and ‘Super Sweet 100’ tomato were grown from seeds in 288-celled plug trays (T.O. Plastics, Inc., Clearwater, MN) to 5–6 cm height, transplanted into soy-plastic containers and petroleum-plastic-control containers filled with Sunshine® LC-1 soilless substrate (SunGro Horticulture, Belleview, WA), and grown in a glass-glazed greenhouse for 4 weeks. Control containers were green 4-inch standard containers (Dillon Products, Middlefield, OH) made of petroleum-based polypropylene (top diameter = 11.4 cm, height = 9.7 cm, volume = 450 cm$^3$). The prototype soy containers were compression-molded, had the same volume and top diameter as controls, and were made of a soy-based polymer formulated with soy protein isolate (26%), soy flour (26%), water (31%), glycerin (12%), phthalic anhydride (4%), sodium sulfite (1%), and potassium sorbate (less than 1%). After compression molding, the soy-container prototypes were dip-coated with a thin layer (≈0.5 mm) of paraffin wax to slow degradation, because preliminary trials indicated that the fertilizer effect from uncoated soy containers was excessive. Container/plant units were arranged 10 cm apart on a greenhouse bench in a completely randomized design (n = 25 for each container × species factorial treatment) and were fertilized at the beginning of Week 3 with 400 mL of Peters Excel® Multi Purpose and Cal-Mag (Everris International B.V., The Netherlands) mixed to supply N at 150 mg L$^{-1}$ (16.6N–5P–16.3K). Fertilization was delayed until Week 3 to evaluate the potential of an early fertilizer effect from the soy-based containers. Air temperature was maintained at 25 ± 5 °C, relative humidity (RH) ranged from 47% to 88% (mean = 67%), and the mean photosynthetically active radiation (PAR) at 1200 s irradiances was 467 μmol m$^{-2}$ s$^{-1}$ during the trial. After 4 weeks, each container/plant unit was rated for plant health and container performance (blind ratings by two experienced horticulturists on a scale of 0 to 5 with 5 being best and 0 being worst). Plant health rating was based on visual assessment of relative leaf size and greenness and presence or absence of flowers, leaf burn, chlorosis, necrosis, wilting, and leaf or stem deformation (Nelson, 2012; OECD, 2006). Container rating was based on container structural durability, function, ease of handling, and presence or absence of mold or algae. Plants were measured for shoot height from the surface of the medium to the tip of the apex and were measured for foliar greenness of the newest fully expanded leaf by using a SPAD meter (Chlorophyll Meter: SPAD-502®; Minolta Camera Co. Ltd., Osaka, Japan) to quantify chlorophyll content indirectly as an indicator of plant nutritional status. Leachate was collected from each container/plant unit by using the PourThru extraction procedure (Cavins et al., 2008; LeBude and Bildbacher, 2009; Wright, 1986), and EC and pH of samples were measured by using a handheld pH–EC meter (HI 9813-6; Hanna Instruments, Smithfield, RI) to assess the nutrient (salt) concentration and pH of the root-zone environment inside the trial containers.

Expt. 2: Garden trial with pepper and tomato. After 4 weeks in the greenhouse, container/plant units were placed in a field trial at a research farm of Iowa State University near Gilbert, IA, to evaluate transpose establishment and growth for 5 weeks. The soil type was Clarion loam, a fine-loamy, medium-loam, arable, mesic, Typic Hapludolls. Experimental units were installed 45 cm on center in the garden plot in a completely randomized design (n = 25 for container × species factorial treatments) by removing the container, breaking it into pieces less than 4 cm in diameter, placing the container pieces in a non-degradable mesh bag (netting with 1 cm × 1 cm square openings; Associated Bag Co., Milwaukee, WI) for easy location and retrieval during harvest, and placing the container pieces beneath the plant roots with ≤5 cm of soil between the container pieces and the bottom of the plant root ball. This methodology was designed to allow degrada- tion of container materials by soil organisms within close proximity to the plant so that effects of material degradation on plant growth and health could be determined. During the trial, mean daily maximum air temperature, minimum air temperature, and 10-cm soil temperature were 20.4, 11.7, and 17.1 °C, respectively. The garden plot was irrigated uniformly once per week with ~2.5 cm of water. After 5 weeks, units were rated for health and measured for shoot height and leaf greenness as in the greenhouse experiment and then were harvested and measured for total plant dry weight (tissue dried at 33 ± 5 °C and mean RH = 27% for 9 d) and percentage container degradation (weight loss) during the 5 weeks in soil near plant roots. Tissue drying was performed in a hot greenhouse with low humidity because the large amount of tissue from the trial far exceeded the capacity of our drying oven. Weights of container remains at harvest were measured after drying samples at 33 ± 5 °C and mean RH of 27% for 9 d, then holding samples for 24 h in the same room where the initial weight was measured to reach equilib- rium with ambient humidity.

Evaluation of soy plastic, PLA-coated soy plastic, and blended soy–PLA plastic containers

The performance, N release, and fertilizer effect of injection-molded containers made of soy plastic, plastic dip-coated with PLA, soy–PLA plastic blended, and a petroleum-based polypropylene (top diameter = 11.4 cm, height = 9.4 cm, volume = 655 cm$^3$). The prototype soy-based containers (top diameter = 11.4 cm, height = 9.7 cm, and volume = 680 cm$^3$) were molded in collaboration with R&D/Leverage Company, Lee’s Summit, MO, and were designed to match commercially available 4.5-inch containers as closely as possible. The material formulation for the soy-plastic containers was soy protein isolate (26%), soy flour (26%), water (31%), glycerin (5%), phthalic anhydride (4%), adipic acid (4%), sodium sulfite (1%), and potassium sorbate (less than 1%). The PLA-coated soy-plastic containers were made of the same injection-molded material as the soy-plastic containers, but were dip-coated after molding with a thin layer (≈0.5 mm) of Ingeo™ PLA 3001D (NatureWorks LLC) by using chloroform as the organic solvent to liquefy the PLA for processing. The material for the soy–PLA blended containers was a 50:50 blend by weight of Ingeo™ PLA 3001D and soy plastic with the same formulation used in the uncoated soy containers. Blending of the soy and PLA materials was facilitated by first blending PLA with polyethylene glycol (80:20 weight) to lower the melt temper-ature of PLA. This adjustment in melt temperature was required to avoid thermal degradation of the soy resin during compounding.

Expt. 3: Greenhouse nutrition trial. Seedlings of salvia ‘St. John’s Fire’ and tomato
‘Rutgers’ were started in 288-celled plug trays to \( \approx 5 \) cm height. Seedlings were transplanted into trial pots containing 655 cm\(^3\) of Sunshine\textsuperscript{8} LC-2 soilless substrate, and units were arranged 15 cm apart in a completely randomized design (\( n = 5 \) for each container \( \times \) species factorial treatment) in a glass-glazed greenhouse. Air temperature was maintained at 24 ± 3 °C, RH ranged from 17% to 80% (mean = 46%), and the mean PAR at 1200 HR was 410 \( \mu \text{mol}\text{m}^{-2}\text{s}^{-1} \) during the trial. Containter/plant units were cultured for 7 weeks and were fertigated once per week with 200 mL of Peters Excel\textsuperscript{9} Multi Purpose and Cal-Mag mixed to supply N at 150 mg L\(^{-1}\) (16.6N–5P–16.3K). After 3 weeks of culture, leachate was collected from each container/plant unit by using the Pour-Thru extraction procedure (Cavins et al., 2008; LeBude and Bilderkamp, 2009; Wright, 1986). Additional samples were collected for two of the treatments, the soy–PLA blend and polypropylene control, after 7 weeks. Additional samples were not collected for the soy plastic and PLA-coated soy treatments because those two container types had failed structurally before the end of the seventh week. Leachate samples were measured for EC and pH by using a handheld pH–EC meter (HI 9813-6; Hanna Instruments) and were analyzed for concentrations of NH\(_4\)\(^+\), NO\(_3\)\(^-\), NO\(_2\)\(^-\), and total N at the Soil and Plant Analysis Laboratory, Iowa State University. Results reported for N concentrations represent the mean for container type across the two species (\( n = 10 \)).

Expt. 4: Greenhouse production trial with five species. Marigold ‘Honeycomb’, petunia ‘Madness Red’, salvia ‘St. John’s Fire’, pepper ‘Autumn Bell’, and tomato ‘Rutgers’ were grown under standard container-production conditions for 5 weeks. Seedlings were started in 288-celled plug trays to \( \approx 5 \) cm height, were transplanted into trial pots containing \( \approx 655 \) cm\(^3\) of Sunshine\textsuperscript{8} LC-1 soilless substrate, and units were arranged 15 cm apart in a randomized complete block design (\( n = 14 \) for each container \( \times \) species factorial treatment) in a glass-glazed greenhouse. Container/plant units were fertigated once per week with \( \approx 200 \) mL of Peters Excel\textsuperscript{9} Multi Purpose and Cal-Mag mixed to supply N at 150 mg L\(^{-1}\) (16.6N–5P–16.3K). Fertilizer was applied at a rate to ensure healthy production of plants in control containers. Air temperature was maintained at 26 ± 5 °C, RH ranged from 25% to 87% (mean = 59%), and the mean PAR at 1200 HR was 530 \( \mu \text{mol}\text{m}^{-2}\text{s}^{-1} \) during the trial. After 5 weeks, all container/plant units were rated for plant health and container performance as described in the methods for Expt. 1. Plants were measured for shoot height from the surface of the root medium to the tip of the apex and were measured for shoot width in two perpendicular directions. These three measurements were used to calculate the three-dimensional shoot volume (height \( \times \) width \( \times \) width). Five randomly selected replications of each container \( \times \) species factorial treatment were harvested and assessed for presence of root circling.

Presence of root circling was a visual assessment of the presence or absence of roots that were touching the inside of the container and had growth that had turned and followed the contour of the container wall. Root circling was considered to be present if 20% or greater of the root system exhibited these characteristics. After harvest, plant samples were prepared and dried at 33 ± 5 °C and mean RH of 26% for 9 d as in Expt. 2, and the samples were measured for plant dry weight and root:shoot dry weight ratio.

Expt. 5: Garden trial with five species. Three of the remaining units from each species in the control treatment and soy–PLA blended-plastic treatment were cultured in a garden plot at a research farm of Iowa State University near Gilbert, IA. The soil type was the same as that described for Expt. 2. The soy-plastic and PLA-coated soy treatments were not included in the field trial because they performed poorly in the greenhouse production trial (Expt. 4). Soy–PLA plant/container units were installed without the container, breaking it into pieces less than 4 cm in diameter, and placing the container pieces beneath the plant roots with \( \approx 5 \) cm of soil between the plastic pieces and the bottom of the plant root ball. Plants from the control treatment were installed without the container to represent standard gardening protocol for plants grown in plastic containers. Plant/container units were spaced 1 m on center in a randomized complete block design. A separate trial was established adjacent to the plant/container trial in the same garden plot to assess the degradation of the container materials in soil. The degradation portion of the experiment included samples of soy plastic and PLA-coated soy plastic for comparison although these material types were not included in the full garden trial. Samples for the degradation assessment were new, one-fourth-sized container pieces that were weighed, placed in a non-degradable mesh bag for easy retrieval, and buried 10 cm below the soil surface. During the trial, mean daily maximum air temperature, minimum air temperature, and 10-cm soil temperature were 29.2, 18.2, and 25.0 °C, respectively. The garden plot was irrigated uniformly once per week with \( \approx 2.5 \) cm of water. Fruits of pepper and tomato 5 cm or greater in diameter were harvested after Week 6, after Week 7, and after Week 8 (final harvest) and were measured for fresh weight the same day after each harvest. After 8 weeks, units were rated for health and measured for shoot height and width in perpendicular directions as in Expt. 4 and then harvested and measured for shoot dry weight (tissue dried in a greenhouse at 33 ± 5 °C and mean RH = 26% for 9 d). Samples of container materials were collected from the soil, dried in a greenhouse at 33 ± 5 °C and mean RH of 26% for 9 d, and measured for percentage degradation (weight loss) after 8 weeks in soil. Shoots of tomato were not measured for dry weight because their large size prevented them from drying consistently.

**Statistical analysis**

Data for all experiments were analyzed for main effects, interactions, and mean separation statistics by using the general linear models procedure and the least significant difference option of SAS/STAT\textsuperscript{10}, Version 6.12 (SAS Institute, Cary, NC). Data sets were tested for homogeneity of variance by using Levene’s test, and non-homogeneous data were transformed by a log or square-root function. Means were calculated from raw data, and the mean separation statistics were calculated from raw or transformed data as necessary. In Expts. 1, 2, and 3, all results were analyzed and reported as the simple numeric means of the measurement or rating parameter. In the greenhouse production trial evaluating soy plastic, PLA-coated soy plastic, and soy–PLA blended plastic containers (Expt. 4), and in the field trial evaluating soy–PLA blended plastic containers (Expt. 5), raw data for plant dry weight, shoot volume, shoot dry weight, and fruit production were normalized to a range from 0 to 100 for results of each species. Normalization was performed separately for each species to achieve a common range for data of the five species for each variable (Pyle, 1999), enabling consistent comparison of container-material treatments across the five plant species. Normalization of individual values was accomplished by using the formula: \( a = (X - A)(b - a)/(B - A) \) where \( A \) = minimum of original data set, \( B \) = maximum of original data set, \( a \) = minimum of normalized data set (0), \( b \) = maximum of normalized data set (100), and \( X \) = the original individual value. The parameter of plant quality index was calculated as the product of the plant health rating and normalized shoot volume and was calculated separately for each experimental unit.

**Results**

**Evaluation of wax-coated soy-plastic containers**

Expt. 1: Greenhouse trial with pepper and tomato. Plants of pepper and tomato were healthier, taller, and greener when grown in control containers compared with those of the same species grown in soy-plastic containers (Table 1; Fig. 1). Both pepper and tomato plants grown in soy containers were chlorotic (Fig. 1). Container rating (based on structure, function, and durability) was poor for soy-plastic containers compared with control containers regardless of species (Table 1). Leachate pH and EC were higher for container/plant units in the soy-plastic treatment for both species. There were interaction effects between container type and species for plant health rating (\( P = 0.0041 \)), shoot height (\( P < 0.0001 \)), and leachate EC (\( P = 0.0318 \)). The interaction effect for plant health rating was evident in the lower health rating received for tomato in the control containers compared with peppers in the same container treatment, and the interaction for shoot height reflects the greater reduction in height of pepper grown in soy containers compared with the reduction shown for tomato. The
interaction between container type and species treatments for EC was evident in the results for tomato, which showed the highest EC for units in the soy-container treatment and the lowest EC for units in the control treatment.

Expt. 2: Garden trial with pepper and tomato. After 5 weeks in garden conditions, plants of tomato cultured with pieces of soy containers degrading near roots were greener and healthier than controls but were shorter and showed no difference in plant dry weight. Degradation of soy-plastic containers under tomato plants exceeded that of containers under pepper plants, although degradation was greater than 50% regardless of species (Table 1). As expected, the petroleum-plastic containers showed no degradation over the 5 weeks of the field trial. There were interaction effects for shoot height ($P < 0.0147$), leaf greenness ($P < 0.0001$), plant dry weight ($P = 0.0009$), and container degradation ($P = 0.0057$). These interactions are reflected in the different effects of container treatments shown for the two species (Table 1; Fig. 2).

### Evaluation of soy plastic, PLA-coated soy plastic, and blended soy–PLA plastic containers

Expt. 3: Greenhouse nutrition trial. After 3 weeks of culture, pH and EC were highest for leachate from soy-plastic containers (Table 2). Leachate pH was lowest for the PLA-coated soy container treatment, but this treatment had the second highest EC of the four treatments. Total N concentrations at 3 weeks of culture were highest for leachate from the soy-plastic containers, with concentrations of total N that were more than double that of the other three container types (Table 2). Total N concentration in leachate from each of the three soy-based containers was greater than that of the control container at 3 weeks of culture, but leachate from the control container had the highest concentration of NO$_3$-N (Table 2). The N in leachate from soy-plastic and PLA-coated soy containers was predominantly (greater than 99%) in the form of NH$_4^+$, but N in leachate from the soy–PLA blended plastic containers was a mixture of three measured forms: NH$_4^+$, NO$_3^-$, and NO$_2^-$ (33%, 18%, and 49%, respectively). There were no interactions between container treatment and species.

### Measures of pH, EC, NH$_4^+$, NO$_3^-$, NO$_2^-$, and total N of leachate from control containers after 7 weeks were similar to those of controls at 3 weeks, but values for the soy–PLA blend treatment at 7 weeks differed from those measured at 3 weeks (Table 2). The pH of leachate from soy–PLA blended containers decreased between Weeks 3 and 7, whereas the EC increased. Consequently, the pH at 7 weeks was lower and the EC was higher for leachate from the soy–PLA blended containers compared with leachate from controls. The N of leachate from soy–PLA blended plastic containers showed a decrease in NH$_4^+$, an increase in NO$_3^-$, and an increase in total N between 3 weeks and 7 weeks, and values from the soy–PLA blended container treatment at 7 weeks were greater than those of controls for each of the three forms of N and for total N (Table 2).

Expt. 4: Greenhouse production trial with five species. Analyses of container/plant units grown under greenhouse-production conditions for 5 weeks showed main effects for container type and species ($P < 0.0001$) and interaction effects between container type and species for all parameters ($P < 0.01$). However, analyses of data separated into full factorial treatment categories (four container treatments $\times$ five species $= 20$ factorial treatments) revealed only slight variations in responses of the five species to container treatments and showed no meaningful trends that warranted presentation of data in such detail. Across species, plants grown in the field trial.
Fig. 2. Shoots of pepper (A) and tomato (B) plants grown in containers made of soy plastic and polypropylene plastic (control) and then transplanted and grown in a garden plot for 5 weeks with the production container crushed and buried in the soil near the plant root zone (see scale bar).

Table 2. Results of the greenhouse nutrition trial (Expt. 3) with prototypes of soy-based plant containers and a polypropylene control.

| Leachate properties after 3 weeks | Soy plastic | PLA-coated soy | Soy–PLA blend | Polypropylene control |
|-----------------------------------|-------------|----------------|---------------|-----------------------|
| pH                                | 8.0 a*      | 6.6 c          | 7.2 b         | 6.9 b                 |
| 0.05 l                  | 5.5 a      | 4.0 b          | 2.0 a         | 1.1 d                 |
| NO3-N (mg L−1)                | 0.4 c      | 0.3 c          | 31.4 b        | 57.1a                 |
| NH4-N (mg L−1)                | 619 a      | 273 b          | 56 c          | 8 d                   |
| NO2-N (mg L−1)                | 1.4 b      | 2.1 b          | 83.2 a        | 1.1 b                 |
| Total N (mg L−1)              | 623 a      | 275 b          | 169 c         | 68 d                  |

| Leachate properties after 7 weeks | Soy plastic | PLA-coated soy | Soy–PLA blend | Polypropylene control |
|-----------------------------------|-------------|----------------|---------------|-----------------------|
| pH                                | 6.3 b**     | 6.9 a          |               |                       |
| EC (mS cm−1)                     | 2.8 a*      | 0.8 b          |               |                       |
| NO3-N (mg L−1)                   | 331 a*     | 62 b           |               |                       |
| NH4-N (mg L−1)                   | 38 b       | 8 b            |               |                       |
| NO2-N (mg L−1)                   | 863 a      | 1.4 b          |               |                       |
| Total N (mg L−1)                 | 457 a*     | 71 b           |               |                       |

*Results describe the pH, electrical conductivity (EC), and nitrogen (N) profile of leachate collected after 3 and 7 weeks of greenhouse culture of salvia and tomato. No samples were collected for soy-plastic and PLA-coated soy treatments after Week 7, because those container types had failed structurally before the end of the seventh week. Results represent the mean for container type across the two species (n = 10).

PLA = polylactic acid.

containers made of soy–PLA blended plastic were the healthiest, had the largest shoot volume, and were the best quality after 5 weeks of culture, but plants in this treatment were not different from controls in dry weight and had a lower root:shoot dry weight ratio than plants in the other container treatments (Table 3). Plants grown in PLA-coated soy containers were less healthy and had lower dry weights than controls but were similar to controls in root:shoot ratio, shoot volume, and plant quality. Plants grown in the high-percentage soy-plastic containers showed the poorest health and the lowest plant dry weight, shoot volume, and quality of the four container treatments (Table 3). Plants grown in control containers exhibited root circling after 5 weeks of culture, but there was no root circling present for plants grown in the three soy-based container types (Table 3). Container ratings were highest for petroleum-plastic controls but were only slightly lower for the containers made of soy–PLA blended plastic. Containers of soy–PLA blended plastic were rated much higher than containers made of soy plastic or PLA-coated soy plastic (Table 3). Differences in the condition of containers and differences in plant health, size, and quality of marigold after 5 weeks of greenhouse production are illustrated in Figure 3 and are representative of the general trends observed across the five species.

Expt. 5: Garden trial with five species. Plants grown in a garden plot for 8 weeks with pieces of soy–PLA blended plastic containers degrading near their roots showed health ratings similar to those of control plants for which the petroleum-plastic containers were removed and discarded and the plant installed without the container (Table 3). Although the health rating was similar for plants in the two container/planting treatments, plants grown with soy–PLA blended plastic near their roots had greater shoot dry weight, shoot volume, plant quality, and fruit production after 8 weeks than did plants in the control treatment. Degradation of soy–PLA blended plastic in soil was substantial, with samples of soy–PLA plastic losing an average of 41% of their original weight over the 8 weeks of the trial (Table 3). Results showed very high degradation rates for soy plastic and PLA-coated soy plastic. Analyses showed main effects for container and species treatments (P < 0.0001), but there were no interactions between container and species treatments (P > 0.05 for all variables).

Discussion

If soy-based plastics and composites are to be used as materials to replace petroleum-based plastics in specialty crop containers, they must be developed to fulfill all of the current strengths of petroleum plastics, be available in quantities that satisfy the demand, and must either be competitively priced with petroleum-plastic containers or supply an added function that will justify a higher price. Compared with the availability of other emerging bioplastics, soy-based raw materials are abundant, and the infrastructure that supplies them can be upscaled more easily than that of many other bioplastic feedstocks (Srinivasan, 2010). In addition to being biorenewable and biodegradable, containers made of soy-based plastics and composites have strong potential to supply an added function over that of petroleum-based plastics; specifically, they possess an intrinsic capacity to release plant-available N during production and after transplant (Tables 1, 2, and 3; Figs. 2 and 3). Our results indicate that high-percentage soy plastics (labeled “soy plastic” and “PLA-coated soy” in our trials) that are not blended with other materials are not sufficiently durable or functional for use in specialty crop containers (Tables 3; Figs. 2 and 3). Coating high-percentage soy plastic with wax or a bio-based polymer like PLA improves the function of containers in some ways (e.g., helps moderate the fertilizer effect; Tables 1, 2, and 3) but fails to improve durability and functionality.
Blending soy-based polymer with PLA strongly improves the durability and functionality of the plastic over that of the high-percentage soy polymer (Table 3; Fig. 3) and moderates the fertilizer effect of soy-plastic better than does coating the material with PLA (Table 2; Fig. 3).

### Table 3. Results of the greenhouse production trial (Expt. 4) and garden field trial (Expt. 5) with prototypes of soy-based plant containers and a polypropylene control.

|                              | Soy plastic | PL-coated soy | Soy–PL blend | Polypropylene control |
|------------------------------|-------------|---------------|--------------|-----------------------|
| **Greenhouse production trial (Expt. 4)** |             |               |              |                       |
| Plant health rating \(^{a}\) | 1.1 \(\text{d}^a\) | 4.4 \(\text{c}^a\) | 5.0 \(\text{a}^a\) | 4.7 \(\text{b}^a\) |
| Plant dry weight \(^{a}\)     | 3 \(\text{c}^a\) | 68 \(\text{b}^a\) | 78 \(\text{a}^a\) | 80 \(\text{a}^a\) |
| Root:shoot dry weight ratio   | 0.39 \(\text{a}^a\) | 0.43 \(\text{a}^a\) | 0.23 \(\text{b}^a\) | 0.43 \(\text{a}^a\) |
| Presence of root circling (%) | 0 \(\text{c}^a\) | 0 | 92 |                       |
| Shoot volume \(^{a}\)         | 2 \(\text{c}^a\) | 59 \(\text{b}^a\) | 76 \(\text{a}^a\) | 62 \(\text{b}^a\) |
| Plant quality index \(^{a}\)   | 2 \(\text{c}^a\) | 264 \(\text{b}^a\) | 378 \(\text{a}^a\) | 295 \(\text{b}^a\) |
| Container rating \(^{a}\)      | 0.7 \(\text{c}^a\) | 0.5 \(\text{c}^a\) | 4.6 \(\text{b}^a\) | 5.0 \(\text{a}^a\) |
| **Garden field trial (Expt. 5)** |             |               |              |                       |
| Plant health rating \(^{a}\)   |             |               | 4.7 \(\text{a}^a\) | 4.7 \(\text{a}^a\) |
| Shoot dry weight \(^{a}\)      |             | 63 \(\text{a}^a\) | 39 \(\text{b}^a\) |                       |
| Shoot volume \(^{a}\)          |             | 60 \(\text{a}^a\) | 44 \(\text{b}^a\) |                       |
| Plant quality index \(^{a}\)   |             | 292 \(\text{a}^a\) | 207 \(\text{b}^a\) |                       |
| Fruit production \(^{a}\)      |             | 68 \(\text{a}^a\) | 25 \(\text{b}^a\) |                       |
| Container degradation (%)      | 100 \(\text{a}^a\) | 98 \(\text{a}^a\) | 41 \(\text{b}^a\) | 0 \(\text{c}^a\) |

\(^{a}\)Results of the greenhouse production trial describe plant health, size, root:shoot ratio, quality, and presence of root circling after 5 weeks of greenhouse production of marigold, pepper, petunia, salvia, and tomato. Results of the garden field trial describe plant health, size, and quality of those five species and fruit production of pepper and tomato after 8 weeks in a garden plot with the container crushed and buried in the soil near the plant root zone. The soy–PLA-coated soy treatments were not included in the field trial because they performed poorly in the greenhouse production trial.

\(^{b}\)Means within a row followed by the same letter are not different according to Fisher’s least significant difference test \((P \leq 0.05)\).

\(^{c}\)Plant health and container ratings were on a scale of 0 to 5 with 5 being best and 0 being worst. Each datum was the mean of blind ratings by two horticulturists.

\(^{d}\)Values describe the percentage of units for each container type that exhibited root circling. Presence of root circling was considered to be present if 20% or greater of the root system exhibited these characteristics.

\(^{e}\)Plant quality index was the product of the plant health rating and normalized shoot volume calculated separately for each experimental unit.

The fertilizer effect of soy-based plastic containers ranged from beneficial to excessive, depending on the formulation of the materials (whether it was blended with PLA) and on the presence or absence of a coating. Based on recommendations for N nutrition and monitoring of leachate by the PourThru method for actively growing container crops, acceptable values for pH range from 5.8 to 6.8, EC from 2.6 to 4.6 mS·cm\(^{-1}\), NO\(_3\)-N from 180 to 320 mg·L\(^{-1}\), NH\(_4\)-N from 60 to 110 mg·L\(^{-1}\), and total N from 240 to 420 mg·L\(^{-1}\) (Cavins et al., 2000, 2008; Jones, 2005; Yeager et al., 1997). All of these values, except pH, should be lower when plants are young and have not yet reached the stage of active growth (Cavins et al., 2000; Warncke and Krauskopf, 1983). In our greenhouse trials (Expts. 1, 3, and 4), the fertilizer effect from high-percentage soy-plastic containers was excessive for young seedlings in both coated and uncoated soy containers, and N was present almost exclusively in the form of NH\(_4\) (Tables 1, 2, and 3; Figs. 1 and 3), which made the fertilizer effect toxic to plants (Jones, 2005; Mengel and Kirkby, 2001). The fertilizer effect from uncoated containers of high-percentage soy was accompanied by a pH much higher than that desired for healthy plant growth (Table 2), a factor that is also believed to increase the toxic effect caused by a high ratio of NH\(_4\) to NO\(_3\) (Mengel and Kirkby, 2001).

Blending soy plastic with PLA improves the fertilizer effect of the containers and increases their durability to a level appropriate for greenhouse production cycles of at least 7 weeks. The fertilizer effect of containers made from a 50:50 blend of soy and PLA was beneficial for plant health, size, and quality and helped produce plants that were superior to controls produced by standard production protocols (Table 3; Fig. 3). In terms of plant health and growth in the greenhouse, the only potentially negative outcome that we observed of units grown in soy–PL blended containers was a low root:shoot ratio compared with controls (Table 3). However, this
result appeared to have no effect on shoot size and quality in the greenhouse and had no impact on establishment and growth of plants transplanted into the garden, where plants from the soy–PLA container treatment were larger and of better quality and produced more fruit after 8 weeks than controls (Table 3; Fig. 3). A potentially beneficial impact of the root characteristics of plants in the soy–PLA blended containers is that root systems did not circle the periiphery of the root ball at the time of transplant as did the roots of most plants in the control treatment (Table 3). Root circling can be detrimental to container crops and is particularly troublesome for growers of nursery crops (Warren and Blazich, 1991). Recent advances in container design provide some options to help reduce the problem of root circling, but many of these options increase water use during production. The potential benefit of root-pruning by protein-based plastic containers has been discussed for containers made of zein, a protein from corn (Helgeson et al., 2010). Compared with the root-pruning effect described for zein, an effect that is accompanied by severe reduction in plant health and growth similar to that seen for high-percentage soy, the mild root-pruning effect of soy–PLA blended containers is a strong improvement. These results suggest potential for developing a blended soy-plastic container that will reduce root circling without sacrificing water use efficiency and may indicate the potential for effectively blending other proteins with carbohydrate-based polymers to optimize those materials for use in horticultural products.

With the exception of the high pH measured during Week 3, a value that was not different from the pH of the control, values for pH, EC, and N nutrients in leachate from containers made of soy–PLA blended plastic were within or near acceptable ranges for effective production of most container crops (Table 2). At Week 3, the concentration of total N in leachate from soy–PLA blended containers was within the acceptable range for young, establishing plants (~140 to 215 mg L−1) (Cavins et al., 2008; Jones, 2005; Warncke and Krauskopf, 1983]), but the ratio of NH₄⁺ to NO₃⁻ (1.8) was higher than recommended [1.0 or less (Jones, 2005)], and the concentration of NO₃⁻ was much higher than in leachate of the other container treatments (Table 2). By Week 7, the concentration of NO₃⁻ had increased markedly and the ratio of NH₄⁺ to NO₃⁻ was well within proportions recommended for good plant health and growth (Jones, 2005). The high concentrations of NH₄⁺ and NO₃⁻ at Week 3 and the increase in NO₃⁻ between Weeks 3 and 7 is consistent with models describing the natural microbial conversion of N from polyamides into forms available to plants (Mengel and Kirkby, 2001). In the case of soy plastic, N conversion likely begins with microbial hydrolysis of soy polyamides to peptide monomers that are converted to NH₄⁺/NH₃ by a different set of microbes (heterotrophic bacteria) that promote ammonification. During nitrification, NH₄⁺ is converted by oxidation to NO₂⁻ by another group of bacteria, then NO₂⁻ is converted by oxidation to NO₃⁻ by yet another specialized group of bacteria (Mengel and Kirkby, 2001). The presence in soy–PLA containers of NH₃, NO₂⁻, and NO₃⁻ in greater concentrations than in controls confirms that the organisms required for N conversion from amino-N to NO₃⁻ are available under conditions of greenhouse production, even when soilless medium is used. Efforts to apply this technology to commercial greenhouse production will need to acknowledge the lag time that takes place before sufficient levels of NO₃⁻ are available to support healthy plant growth. Therefore, production protocols developed to utilize the intrinsic N from soy–PLA containers will likely need to include a fertilizer regimen that can meet the N requirements of plants until NO₃⁻ from the containers becomes available.

The excessive NH₄⁺-N compared with other forms of N in high-percentage soy containers at Week 3 was likely the result of a combination of factors. The carbon:nitrogen (C:N) ratio of the high-percentage soy plastic may be so low that microbes must mineralize and release relatively large amounts of N to obtain enough C from the soy material to facilitate their own growth. When the C:N ratio of the substrate is low, the form of N released by microbes is predominately NH₃/NH₄⁺ (Mengel and Kirkby, 2001). Although NH₄⁺-N was abundant, little or no nitrification had taken place in high-percentage soy containers by Week 3 (Table 2), and the low concentration of both NO₂⁻ and NO₃⁻ suggests that nitrification had failed early in the process, before conversion of NH₄⁺ to NO₂⁻. The most plausible explanation for lack of nitrification in high-percentage soy containers is inhibition of nitrifying bacteria by unfavorable chemical characteristics (e.g., salinity or pH) of the medium that accompanied the rapid accumulation of NH₃ (Gandhi and Paliwal, 1976; Groeneweg et al., 1994; Mengel and Kirkby, 2001). Regardless of the specific cause, the problem of excessive NH₃ accumulation in soy-based plastic containers can be effectively corrected by blending soy polymer with a carbohydrate-based polymer like PLA (Table 2).

In contrast to common practice in which petroleum-plastic containers are discarded for recycling or disposal when plants are transplanted into the garden or landscape, there is strong potential to utilize used soy-based containers as a soil amendment that can provide a fertilizer effect as the container material degrades and the container completes its life cycle. In our first garden trial (Expt. 2), transplants from coated high-percentage soy-plastic containers benefitted strongly from the fertilizer capacity of the container pieces buried near their roots. All plants in the soy-plastic treatments grew faster and showed a greater increase in height and greenness than did controls with pieces of petroleum containers near roots (Table 1). Our first garden trial also demonstrated the capacity for soy-based plastic materials to biodegrade in soil at normal garden temperature and moisture levels (mean daily 10-cm soil temperature = 17.1 °C and irrigation = 2.5 cm per week in our trial). Blending soy plastic with PLA greatly improved the performance of containers during greenhouse plant production (Table 3; Fig. 3) without eliminating the capacity for a beneficial fertilizer effect when container pieces were installed with plants in the garden (Table 3). The improvement in material durability of blended soy–PLA plastic compared with high-percentage soy plastic is accompanied by slower degradation (Table 3), but the rate of degradation of 50:50 soy–PLA plastic appears sufficient for containers to end their life cycle as indistinct organic matter in soil within one growing season.

This report provides proof of concept that bio-based plastics containing soy protein have strong potential as sustainable alternatives to petroleum-based plastics in specialty crop containers and that soy-based plastics can provide an additional function over petroleum plastic by delivering a beneficial fertilizer effect during greenhouse production and transplant establishment in the landscape. Continued development of composites and blends of soy plastics with other bio-based materials should help optimize the fertilizer effect, performance, durability, and degradability of soy-based plastics for use in plant containers for horticultural crops. Our results with soy-protein-based polymers confirm the need for research examining other bio-based plastics, blends, and composites to ensure that the best sustainable technologies are discovered and adopted by the specialty crops industry.

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