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Ground Bovine Bone as a Perlite Alternative in Horticultural Substrates

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Summary. Two grades of ground bovine bone were evaluated as potential alternatives to perlite in horticultural substrates. The bulk density of small and large bone-amended substrates was significantly higher than equivalent perlite-amended substrates. Large and small bone increased the air-filled pore space of sphagnum peat. However, at 10% and 20% (v/v), neither size of bone resulted in as high an air-filled pore space as equivalent amounts of perlite. At 30% and 40%, incorporation of small bone resulted in a similar air-filled pore space as incorporation of equivalent amounts of perlite, and incorporation of large bone resulted in a higher air-filled pore space than incorporation of equivalent amounts of perlite. Water-filled pore space and water-holding capacities of substrates were inversely related to air-filled pore space. When placed in a moist substrate, mineral elements within the bone were able to leach into the substrate over time. Substrates amended with 40% large and small bone had significantly higher concentrations of ammonium (NH_4^+), phosphorus (P), potassium (K), calcium (Ca), sodium (Na), and chloride (Cl^-) than the 40% perlite-containing substrates. Substrates amended with 40% large bone had similar concentrations of magnesium (Mg), sulfur (S), iron (Fe), and copper (Cu) while substrates amended with 40% small bone had higher levels of these elements than perlite-amended substrates. Substrate concentrations of nitrate (NO_3^-), manganese (Mn), zinc (Zn), and boron (B) were not different among the substrates after 4 weeks in the greenhouse. The pH, electrical conductivity (EC) and NH_4^+ levels of bone-amended substrates increased to levels significantly higher than recommended and resulted in rapid mortality of 'Orbit Cardinal' geranium (*Pelargonium ×hortorum*), 'Cooler Blush' vinca (*Catharanthus roseus*), and 'Dazzler Rose Star' impatiens (*Impatiens walleriana*) plants grown in bone-amended substrates. Therefore, ground bovine bone was not a feasible alternative to perlite for use in horticultural substrates.

Artificial substrates are commonly used in the production of containerized greenhouse and nursery crops (Nelson, 1998). Substrates are formulated from various organic and inorganic components to provide suitable physical and chemical properties as required by the specific crop and growing conditions (Bunt, 1988). An important physical property of artificial substrates is air-filled pore space. Air-filled pores allow for drainage and gas exchange between the root environment and the outside atmosphere (Bunt, 1988). Various materials are used to at least partially provide for air-filled pore space in artificial substrates with the most common being perlite.

Perlite is an inorganic expanded aluminosilicate of volcanic origin (Nelson, 1998), and it is produced by mining the ore, grinding the crude ore to the desired particle size and heating it to temperatures of up to 982 °C (1799.6 °F). Heating causes the ore to expand from four to twenty times its original volume resulting in a lightweight white porous particle (Hanan, 1998). Because of the costs associated with mining, transportation and heating, perlite is a relatively expensive substrate component. In addition to its cost, in its dry state, perlite produces a siliceous dust that is classified as an eye and lung irritant. Substrate components that are lower in cost, do not have potential health issues and could provide for air-filled pore space in the substrate, which would be beneficial to the nursery and greenhouse crops industries.

Waste bovine bone is a byproduct of the meat processing industry. The material is ground, boiled and dried. Some of this material is processed into a powder to produce bone meal. Various particle sizes of bovine bone are available depending on initial grinding, and because it is a porous material, it is relatively light in weight. The objective of this research was to determine if ground bovine bone could be used in horticultural substrates as a low-cost alternative to perlite.

Materials and methods

Physical properties of perlite and bone-amended substrates. Two grades of ground bovine bone were obtained from Tyson Foods (Springdale, Ark.). Calcitic lime was added to a commercially-obtained sphagnum peat to adjust the initial pH to about 5.4 before mixing of the substrates. Twelve substrates were formulated by blending both grades of bone and a coarse horticultural perlite at 10%, 20%, 30%, and 40% (by volume) with the sphagnum peat. Particle size distributions were determined for both grades of bone and the perlite by sieving 100-g (3.5-oz) samples on a CSC Scientific (Fairfax, Va.) rotary shaker for 10 min using screens with pore diameters of 8.0, 6.3, 4.0, 2.0, 1.0, 0.5, and 0.25 mm (1.00 mm = 0.039 inch). Three random samples were screened for each material. The weight of the material collected in each screen was determined. The percent weight retained in each screen size and standard errors were plotted for perlite and both sizes of bone.

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The bulk density ($g \cdot cm^{-3}$), water-filled pore space at container capacity (percent volume : volume), water-holding capacity at container capacity (percent weight : weight) and air-filled pore space at container capacity (percent volume:volume) were determined for each of the substrates using loose-packed cores and methods adapted from Byrne and Carty (1989) and Bilderback and Fonteno (1993). Three replications were conducted for each substrate. An analysis of variance and single-degree-of-freedom contrasts were conducted to determine if significant differences in physical properties occurred between equivalent bone-containing and perlite-containing substrates.

CHEMICAL PROPERTIES OF PERLITE AND BONE-AMENDED SUBSTRATES. Substrates containing 40% large bone, small bone or perlite were placed into 10.2-cm plastic containers [4-inch standard containers with a volume of 650 mL (39.7 $inch^3$)] and placed into a glass-glazed greenhouse. Greenhouse air temperatures were maintained between 20 and 25 °C (68.0 and 77.0 °F) and substrates were kept moist with deionized water. After 4 weeks, chemical properties were determined for a 2:1 (by volume) water extract of the substrates using methods adapted from the Southern Cooperative Research Service (1992). Electrical conductivity was determined using a Beckman (Cedar Grove, N.J.) solu-bridge and the pH was determined using an Orion (Cambridge, Mass.) pH meter. Nitrate concentration was determined using the copperized cadmium reduction procedure (Keeney and Nelson, 1982) and NH_4^+ was determined by the nitroprusside-salicylate procedure (Wall et al., 1975). The Cl^- concentration was estimated by the mercury thiocyanate procedure (Fixen et al., 1988). For P, K, Ca, Mg, B, Fe, Mn, Zn, Cu, and Na, the filtered extract was used for simultaneous inductively coupled argon plasma emission spectrometry (Jones, 1977; Munter and Grande, 1981). Additionally, the EC of water extracts of small and large bone were determined as described above except that both 30-min and 48-h incubation periods were conducted. For all chemical tests, five replications were tested for each substrate. An analysis of variance was conducted to determine if the substrates differed significantly in their chemical properties. Where significant differences occurred, a least significant difference mean separation test was conducted to establish significant differences between means.

PLANT GROWTH IN PERLITE AND BONE-AMENDED SUBSTRATES. Four-leaf plugs [seedlings in size 288 plug trays with 5 mL (0.17 fl oz) volume per plug cell] of 'Orbit Cardinal' geranium, 'Cooler Blush' vinca, and 'Dazzler Rose Star' impatiens were transplanted into 10.2-cm plastic containers filled with each of the test substrates described under experiment one.

Plants were placed into a glass-glazed greenhouse. Greenhouse air temperatures were maintained between 20 and 25 °C (68.0 and 77.0 °F), and ambient light levels were 250 to 300 $\mu mol \cdot s^{-1} \cdot m^{-2}$ at 1200 HR. Immediately after being placed into the greenhouse, plants were drenched with 50 mL (1.7 oz) of 15% etridiazole and 25% thiophanate-methyl [Banrot (Scotts, Marysville, Ohio)] fungicide according to label directions. Plants were fertilized at each irrigation with 200 $mg \cdot L^{-1}$ (ppm) N using a 15N–2.2P–12.5K fertilizer [Excel 15–5–15 Cal Mag (Scotts, Marysville, Ohio)].

After 6 weeks, the experiment was terminated. Dry shoot and root weights were determined. The experimental design was a complete randomized block with eight blocks and each treatment combination appearing once in each block. An analysis of variance was conducted to determine if significant differences in plant growth occurred among the different substrates. Single-degree-of-freedom contrasts were conducted to determine significant differences between substrates containing equivalent volumes of perlite and large or small bone.

Results and discussion

PHYSICAL PROPERTIES OF PERLITE AND BONE-AMENDED SUBSTRATES. The majority of perlite particles (Fig. 1) had a diameter of 2.0 to 6.3 mm (0.08 to 0.25 inch) with the highest portion of particles being 2.0 to 4.0 mm (0.08 to 0.16 inch). Most small bone particles had diameters from 2.0 to 6.3 mm (0.08 to 0.25 inch) with the highest portion of particles being 4.0 to 6.3 mm (0.16 to 0.25 inch). Although the particle size distribution of small bone overlapped with that of perlite, small bone particles were generally larger than the perlite used in this study. Most large bone particles had a diameter of 6.3 to 12.5 mm (0.25 to 0.49 inch) and were larger than perlite.

The bulk density of small and large bone-amended substrates was higher than equivalent perlite-amended substrates (Table 1). Water-filled pore space was higher in 10% and 20% small and large bone than in 10% and 20% perlite. Water-filled pore space for 30% and 40% small bone was not different from that of 30% and 40% perlite, but water-filled pore space of 30% and 40% large bone was lower than that of 30% and 40% perlite.

Substrates amended with 10% and 20% large and small bone had higher water-holding capacities than those amended with comparable amounts of perlite, but substrates amended with 30% and 40% large and small bone had similar water-holding capacities as those amended with comparable amounts of perlite. Substrates amended with 10% and 20% small and large bone had lower air-filled pore space than those amended with comparable amounts of perlite. Substrates amended with 30% and 40% small bone had similar air-filled pore space as those amended with comparable amounts of perlite. Substrates amended with 30% and 40% large bone had higher air-filled pore space than those amended with comparable amounts of perlite.

No specific standards exist for acceptable bulk density of horticultural substrates, and substrates or substrate components successfully used in horticultural production can varying widely in bulk density. For example sphagnum peat, 3-mm fir bark and pumice had bulk densities of 0.11, 0.23, and 0.46 $g \cdot cm^{-3}$ ($1.00\ g \cdot cm^{-3} = 0.578\ oz/inch^3$), respectively (Hanan, 1998). Bulk density is primarily important because in many, although not all, cases the higher the bulk density, the lower the low air-filled pore space. Additionally, the higher the bulk density, the higher the shipping costs of the substrate. Although bone-amended substrates had bulk densities within the range of other common substrate components, bone-amended substrates had higher bulk densities than perlite-containing substrates and shipping costs would thus be expected to be higher.

At 10% and 20%, large and small bone-amended substrates had higher water-filled pore space and lower air-filled pore space than perlite. At 30% and 40%, small bone-amended substrates had similar water-holding capacities and air-filled pore space as perlite-amended substrates, but large

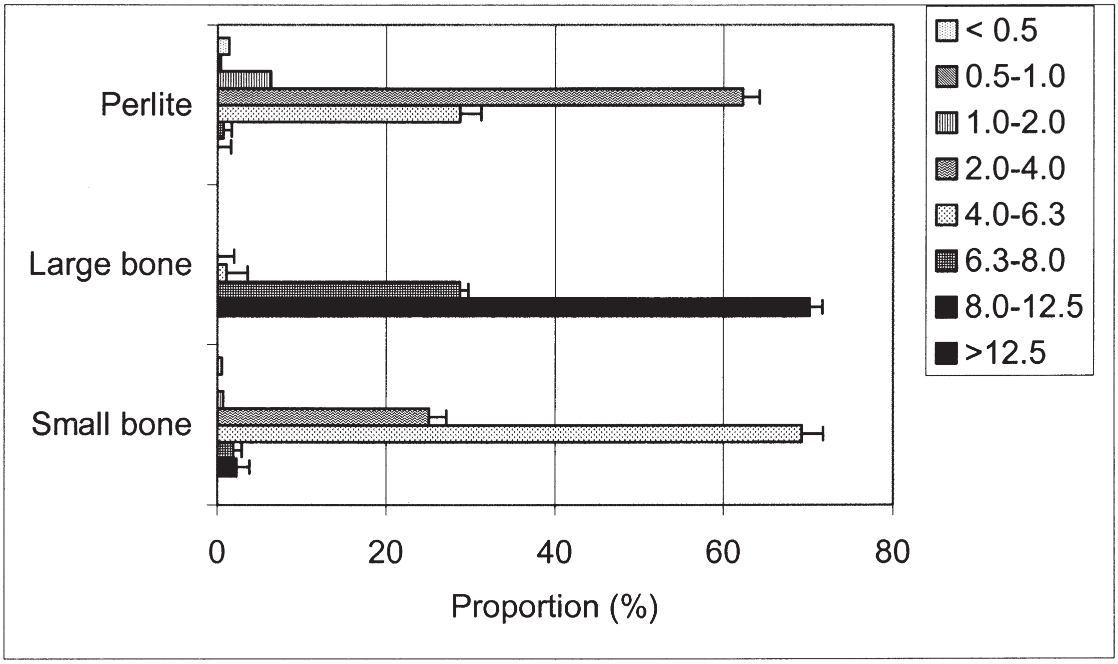

Fig. 1. Particle size (mm) distribution for large bone, small bone and perlite used to formulate test substrates. Error bars represent standard errors. 1 mm = 0.039 inch.

bone-amended substrates had lower water-filled pore space and higher air-filled pore space than perlite-amended substrates. This occurred because, unlike perlite that tended to be angular and rounded, bone was angular and flattened. Small amounts of bone layered within the sphagnum peat and tended to not result in the creation of large pores. However, as the amount of bone reached a certain threshold level (30% in this study), the particles of bone tended to lay against one another and created large pores that drained after irrigation and thus reduced water-filled pore space and increased air-filled pore space. Although no specific recommendations exist for the water-holding capacity of substrates, 10% air-filled pore space is typically recommended for 10.2-cm containers (Bunt, 1988). To achieve this air-filled pore space, the substrates in this study required 30% small bone or about 20% large bone. This air-filled pore space was nearly achieved with only 10% perlite. Therefore, to achieve the recommended air-filled pore, more bone than perlite would be required in the substrate.

CHEMICAL PROPERTIES OF PERLITE AND BONE-AMENDED SUBSTRATES. Substrates amended with 40% large and small bone had a higher pH and EC after 4 weeks than the perlite-amended substrates (Table 2). The substrate amended with 40% small bone had a higher pH and EC than the substrate amended with 40% large bone. The pH of both small and large bone-amended substrates was higher than recommended levels, and the EC of the small bone-amended substrate was higher than recommended levels for seedlings. Substrates amended with 40% large and small bone had higher concentrations of NH_4^+, P, K, Ca, Na, and Cl^- than the 40% perlite-amended substrates. Substrates amended with 40% large bone had similar concentrations of Mg, S, Fe, and Cu while substrates amended with 40% small bone had higher levels of these elements than perlite-amended substrates. Substrate concentrations of NO_3^-, Mn, Zn and B were not different among the substrates after 4 weeks in the greenhouse (data not shown). All of these mineral elements occurred within commonly recommended ranges for actively growing crops with the exception of NH_4^+ in small bone amended substrates, which was higher than the recommended level.

The EC of the small and large bone used in this study were 0.76 and 0.23 $mS\cdot cm^{-1}$, respectively, after a standard 30-min incubation period in deionized water. The EC of the water extract from small bone was higher than that from large bone. However, after a 48-h incubation period in deionized water, the EC of small and large bone were 2.9 and 1.3 $mS\cdot cm^{-1}$, respectively, and the EC of the water extract from small bone was higher than that from large bone. As water penetrated the bone during the 48-h incubation, it became soft and mineral elements within the structure were released into solution. Therefore, using a standard 30-min incubation period, the EC levels and mineral element content of bone were within acceptable ranges. However, a 30-min incubation period did not provide an accurate indication of how ground bone would affect substrate pH and EC over time. As mineral elements were released from the bone, into the substrate solution the pH and EC of the substrate increased, and differences in chemical properties of the small bone, large bone and perlite-amended substrates resulted.

PLANT GROWTH IN PERLITE AND BONE-AMENDED SUBSTRATES. For substrates amended with small bone, only geranium plants grown in 10% small bone survived for 6 weeks (Table 3). All plants grown in the substrate amended with 40% large bone failed to grow and died within 2 weeks after planting.

All vinca grown in substrates amended with 20%, 30% and 40% large bone died within 2 weeks. All geraniums grown in 40% large bone died within 2 weeks, and all impatiens planted in substrates amended with 30% and 40% large bone died within 2 weeks. In all cases where plants survived, all species grown in substrates amended with small or large bone had lower dry shoot and root weights than plants in comparable perlite-amended substrates.

The high degree of mortality in seedlings grown in bone-amended substrates may have been due to a combination of factors. As the bone softened and allowed mineral elements to be released into the substrate solution, pH of the substrate increased to levels that would be significantly higher than optimal but not likely to cause rapid seedling death. However, the EC of small bone-amended substrates increased to levels that were significantly above recommended levels and that would have been injurious to seedlings. With the increased surface area of small bone, the EC of small bone-amended substrates was higher than that of equivalent large-bone amended substrates. Although only the 40% bone-amended substrates were tested, it is also reasonable to assume that substrates amended with lower levels of bone would have had lower EC levels. This would explain why mortality increased as the amount of bone in the substrate increased. In addition to having a high EC, substrates amended with 40% bone had high NH_4^+

Table 1. Physical properties of sphagnum peat-based substrates amended with small bone, large bone, or perlite[z].

Substrate components (% by vol)								
Small bone	Large bone	Perlite	Sphagnum peat	df	Bulk density $(g \cdot cm^{-3})$[y]	Water-filled pore space at container capacity (% by vol)	Water-holding capacity (% by vol)	Air-filled pore space at container capacity (% by vol)
10	0	0	90		0.19	78.3	61	5.4
20	0	0	80		0.27	77.6	60	6.2
30	0	0	70		0.34	70.1	52	11.2
40	0	0	60		0.42	63.5	50	14.5
0	10	0	90		0.19	79.7	61	5.6
0	20	0	80		0.28	77.7	60	9.7
0	30	0	70		0.39	64.5	51	15.6
0	40	0	60		0.44	60.1	48	18.4
0	0	10	90		0.11	76.2	57	9.2
0	0	20	80		0.09	70.7	56	11.4
0	0	30	70		0.09	68.5	53	13.9
0	0	40	60		0.09	65.5	49	14.8
Significance								
Treatment				11	***	***	**	***
10% small bone vs. 10% perlite				1	**	*	*	**
20% small bone vs. 20% perlite				1	***	**	*	***
30% small bone vs. 30% perlite				1	***	NS	NS	NS
40% small bone vs. 40% perlite				1	***	NS	NS	NS
10% large bone vs. 10% perlite				1	**	**	*	***
20% large bone vs. 20% perlite				1	***	**	*	*
30% large bone vs. 30% perlite				1	***	**	NS	*
40% large bone vs. 40% perlite				1	***	**	NS	***

[z] Analyses performed using aluminum cylinders [7.6 cm (3 inches) inside diameter × 7.6 cm height].

[y] 1.00 $g \cdot cm^{-3}$ = 0.578 oz/inch^3.

[NS,*,**,***] Nonsignificant or significant at $P < 0.05$, 0.01, or 0.01, respectively.

Table 2. Chemical properties of a 2:1 water extract of perlite, large bone and small bone-amended sphagnum peat-based substrates[z].

Parameter	Perlite	Large bone	Small bone	Significance	LSD[y] (α = 0.05)
pH	5.3	7.8	8.2	***	0.20
Electrical conductivity $(mS \cdot cm^{-1})$	0.22	0.95	1.9	***	0.17
Ammonium $[mg \cdot L^{-1}$ (ppm)]	6.8	59	173	***	29
Phosphorus $(mg \cdot L^{-1})$	2.6	31	52	***	5.0
Potassium $(mg \cdot L^{-1})$	7.3	30	52	***	7.0
Calcium $(mg \cdot L^{-1})$	7.6	27	54	***	8.3
Magnesium $(mg \cdot L^{-1})$	1.1	1.2	2.6	***	0.45
Sodium $(mg \cdot L^{-1})$	10	82	97	***	5.6
Sulfur $(mg \cdot L^{-1})$	9.5	16	62	***	9.5
Iron $(mg \cdot L^{-1})$	0.46	0.47	0.91	***	0.12
Copper $(mg \cdot L^{-1})$	0.04	0.04	0.08	**	0.03
Chloride $(mg \cdot L^{-1})$	12	63	136	***	18

[z] Each substrate contained 60% (v/v) sphagnum peat amended with either 40% perlite, large bone, or small bone. Substrate samples were tested after 4 weeks in the greenhouse.

[y] LSD = least significant difference.

[**,***] Significant at $P < 0.01$ or 0.001, respectively.

Table 3. Impatiens, geranium, and vinca growth in sphagnum peat-based growing media amended with large bone, small bone, and perlite.

Substrate components (% by volume)					Dry shoot wt (g)^z			Dry root wt (g)		
Small bone	Large bone	Perlite	Sphagnum peat	df	Geranium	Vinca	Impatiens	Geranium	Vinca	Impatiens
10	0	0	90		0.1	0	0	0	0	0
20	0	0	80		0	0	0	0	0	0
30	0	0	70		0	0	0	0	0	0
40	0	0	60		0	0	0	0	0	0
0	10	0	90		1.5	0.1	0.9	0.2	0	0.3
0	20	0	80		0.7	0	0.4	0	0	0.1
0	30	0	70		0.1	0	0	0	0	0
0	40	0	60		0	0	0	0	0	0
0	0	10	90		2.2	0.6	0.9	0.5	0.2	0.4
0	0	20	80		1.9	0.7	0.9	0.4	0.2	0.4
0	0	30	70		1.7	0.7	0.8	0.4	0.2	0.3
0	0	40	60		1.8	0.5	0.9	0.4	0.2	0.4
Significance										
Substrate				11	***	***	***	***	***	***
Block				7	NS	NS	NS	***	NS	NS
10% small bone vs. 10% perlite				1	***	***	***	***	***	***
20% small bone vs. 20% perlite				1	***	***	***	***	***	***
30% small bone vs. 30% perlite				1	***	***	***	***	***	***
40% small bone vs. 40% perlite				1	***	***	***	***	***	***
10% large bone vs. 10% perlite				1	***	***	NS	***	***	NS
20% large bone vs. 20% perlite				1	***	***	***	***	***	***
30% large bone vs. 30% perlite				1	***	***	***	***	***	***
40% large bone vs. 40% perlite				1	***	***	***	***	***	***

^{z}1.0 g = 0.035 oz.

^{NS,***}Nonsignificant or significant at $P < 0.05$ or 0.01, respectively.

levels. These levels were above commonly recommended levels and high enough to be injurious to seedlings. Additionally, under high pH such as what occurred in bone-amended substrates, NH_4^+ is converted to ammonia (NH_3), which is toxic to plants (Mengel and Kirkby, 2001). Although, ammonia was not determined in this study, both high NH_4^+ levels and high pH occurred. The symptoms of poor root growth and rapid plant death observed in this study were also similar to those described for NH_3 toxicity (Mengel and Kirkby, 2001). Therefore, NH_3 toxicity may have contributed to the rapid plant death observed in this study.

Conclusions

Large and small bone increased the air-filled pore space of sphagnum peat. However, at 10% and 20% (by volume), neither bone size resulted in as high an air-filled pore space as equivalent amounts of perlite. At 30% and 40%, small bone resulted in a similar air-filled pore space as equivalent amounts of perlite, and large bone resulted in a higher air-filled pore space than equivalent amounts of perlite. Therefore, higher amounts of bone would be required to achieve a similar air-filled pore space as a given amount of perlite. When placed in a moist substrate salts within the bone were able to leach over time into the substrate causing the pH, EC, and NH_4^+ to increase significantly resulting in a high plant mortality rate in bone-amended substrates. Therefore, the ground bovine bone used in this study was not a suitable alternative to perlite. Leaching of the bone once or multiple times before use might remove much of the salt content within the bone. However, even if successful, this would increase processing and handling costs and make ground bone a less desirable alternative to perlite.

Literature cited

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

Bunt, A.C. 1988. Media and mixes for container grown plants. Unwin Hyman, Ltd., London.

Byrne, P.J. and B. Carty. 1989. Developments in the measurement of air filled porosity of peat substrates. Acta Hort. 238:37–44.

Fixen, P.E., R.H. Gelderman, and J.L. Denning. 1988. Chloride tests, p. 26–28. In: W.C. Dahnke (ed.). Recommended chemical soil tests procedures for the north central region. N. Dak. Agr. Expt. Sta. Bul. 499.

Hanan, J.J. 1998. Greenhouses—Advanced technology for protected horticulture. CRC Press, Boca Raton, Fla.

Jones, Jr., J.B. 1977. Elemental analysis of soil extracts and plant tissue ash by plasma emission spectroscopy. Commun. Soil Sci. Plant Anal. 8:345–365.

Keeney, D.R. and D.W. Nelson. 1982. Nitrogen—Inorganic forms, p. 643–698. In: A.L. Page (ed.). Methods of soil analysis. Part 2. 2nd ed. Amer. Soc. Agron., Madison, Wis.

Mengel, K. and E.A. Kirkby. 2001. Principles of plant nutrition. Kluwer Academic Publ., Dordrecht, The Netherlands.

Munter, R.C. and R.A. Grande. 1981. Plant tissue and soil extract analysis by ICP-atomic emission spectrometry, p. 653–672. In: R.M. Barnes (ed.). Developments in atomic plasma spectro-chemical analysis. Heydon and Sons, London.

Nelson, P.V. 1998. Greenhouse operation and management. 5th ed. Prentice Hall, Upper Saddle River, N.J.

Southern Cooperative Research Sevice. 1992. Plant analysis reference procedures for the southern region of the United States. S. Coop. Res. Serv. Bul. 368.

Wall, L.L., C.W. Gehrke, T.E. Neuner, R.D. Cathey, and P.R. Rexroad. 1975. Total protein nitrogen: Evaluation and comparison of four different methods. J. Assn. Official Anal. Chem. 58:811–817.