PHOSPHORUS NUTRITION OF *ILEX CRENATA* 'HELLERI' GROWN IN A PINE BARK MEDIUM

by

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Dissertation submitted to the Graduate Faculty of the
Virginia Polytechnic Institute and State University
in partial fulfillment of the requirements for the degree of
DOCTOR OF PHILOSOPHY
in
Horticulture

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August, 1981

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Dedication

This dissertation is dedicated to my wife, ,

and our daughter, .

Acknowledgements

My sincere thanks to Dr. Robert D. Wright for his continued guidance, friendship, and support during the last four years. Without his help and contribution this study would have been impossible. I also want to extend a special thanks to my committee members, Drs. A.J. Lewis III, D.G. Himelrick, M.M. Alley, and S.J. Donohue for their guidance and time through the course of this study, and during the preparation of this manuscript.

Finally, I want to thank for secretarial assistance, for technical assistance, for special assistance, and fellow graduate students for their support. A special thanks must also be extended to my parents who have been unfailing in their support of my endeavors.

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General Introduction

Fertilizer regimes are designed to maintain optimum nutritional levels in the growing medium. However, levels have not been established for most woody nursery plants and, as a result, many diverse fertilizer regimes are used commercially.

A common nursery practice is to amend growing media with superphosphate and subsequently fertilize with complete slow-release granular or water soluble fertilizers. Rates of P amendment and complete fertilizers vary, although, little consideration is given to the growing medium P level. Because P is fixed by soil and slowly available for plant growth, incorporation of P as ordinary superphosphate (9% P) has been a nursery practice dating from the time soil was used as a container medium. Soilless container mixes, such as pine bark, are currently used and research is needed regarding P requirements of woody nursery plants.

The purpose of this research was to determine if a P amendment is needed when woody nursery plants grown in pine bark are subsequently fertilized with complete slow-release granular or water soluble fertilizers and to establish pine bark P levels required for maximum plant growth. Once P levels resulting in maximum shoot dry weight have been established, and pine bark-P relationships characterized, rates of fertilizer P amendment needed to maintain this level can be determined.

Chapter I

Response of *Ilex crenata* Thunb. cv. Helleri to Superphosphate-amended Pine Bark

Published in *HortScience* 16:202-203

Abstract. Amending a pine bark growing medium with 270 g/m^3 of P did not increase the dry shoot weight of 'Helleri' holly if subsequently fertilized with a complete slow-release granular or water soluble fertilizer. Tissue P levels, except for the water soluble treatment, and pine bark P levels were higher as a result of the P amendment.

Additional Index Words. phosphorus, slow-release fertilizers, soluble fertilizers

Introduction

Peat media were predominate growing media for container plants for many years. These media contained small amounts of P thus necessitating supplemental P (4), typically supplied by amending the media with superphosphate (8, 9, 12, 14, 17, 19). Gouin and Link (11) grew 'Helleri' plants in a peat medium unamended with superphosphate, thus questioning the need to amend container media with P.

Presently, many container-grown landscape plants are grown in media containing mostly pine bark amended with 2.3-3.5 kg/m^3 of superphosphate (9% P). These plants are generally fertilized with a complete granular or water soluble fertilizer during the growing season. The purpose of this research was to compare shoot dry weights of 'Helleri' holly grown in 100% pine bark with or without P amendment and fertilized with either a complete slow-release granular or water soluble fertilizer.

Materials and Methods

Single stem cuttings of 'Helleri' holly were placed under intermittent mist (15 sec/10 min) on January 23, 1979, for 6 weeks in 6 cm x 8 cm plastic pots containing a medium (v/v) of 1 peat: 1 weblite (Webster Brick Company, Roanoke, VA 24012). Peters (20N-8.7P-16.7K) water soluble fertilizer was applied weekly at 300 ppm N after the plants were rooted.

The plants were potted June 29 in 3 liter plastic containers and subsequently grown in a greenhouse environment (28°C day, 21° night) for 4 months under natural photoperiod. The pine bark medium, which contained 0.02% total P, was amended with dolomitic limestone and Esmigran minor element mix (Mallinckrodt, Inc., St. Louis, MO 63147) at 4.7 and 2.4 kg/m^3 respectively. Half of the pine bark medium was amended with 3 kg/m^3 of superphosphate (9% P). The media contained 22% air space (15) and a particle analysis of 7% greater than 6.3 mm (U.S. Series seive #3), 71% between 6.3 mm and 0.5 mm (U.S. Series seive #35), and 22% less than 0.5 mm.

The experimental design was a completely randomized factorial which consisted of 2 groups (1 with and 1 without P amendment) and 3 fertilizers. Each fertilizer-group combination consisted of 3 plants replicated 4 times. Eleven days after potting, the following fertilizers were surface applied per container: 9.6 g of slow-release 18N-2.6P-10K fertilizer (Osmocote 18-6-12, a trademarked fertilizer of Sierra Chemical Co., Milpitas, CA 95035), 7.2 g of slow-release 24N-1.7P-8.3K fertilizer (Sulfurkote 24-4-10, a trademarked fertilizer of Sta-Green

Plant Food Co., Sylacauga, AL 35150) or a water soluble 20N-8.7P-16.7K fertilizer (Peters 20-20-20, a trademarked fertilizer of W.R. Grace & Co., Allentown, PA 18104). The granular fertilizers were applied once, while the soluble fertilizer was applied weekly as 500 ml of a 300 ppm N solution.

Six weeks after initiation of fertilization (August 21), 250 ml of distilled water were poured onto the surface of 2 of the 3 containers from each treatment and extracts (leachates) collected for P determination (18).

On November 8, leaf samples were taken from the uppermost mature foliage of all plants and plant stems were cut just above the upper roots. Leaf samples and shoot tissue were dried, weighed and leaf tissue analyzed for P (1).

Results

Plants grown in pine bark amended with P were not significantly larger than plants receiving the same fertilizer without P amendment (Table 1). Plants grown in pine bark amended with P and fertilized with the soluble fertilizer were significantly larger than plants fertilized with the granular fertilizers. The same trend occurred with plants grown in unamended pine bark but differences were not significant at the 5% level.

Extract P levels from pine bark amended with P were significantly higher than levels for unamended pine bark, except for the soluble fertilizer treatment. Extract P levels from the soluble fertilizer were significantly larger than extract P levels from the granular fertilizers. This is because more P was applied with the soluble fertilizer than with the granular fertilizers.

Leaf tissue P percentages reflect the pine bark extract P concentrations of the granular fertilizers with the P amendment resulting in significantly higher P concentrations. Tissue P concentrations were also significantly higher for plants receiving the soluble fertilizer, compared to granular irrespective of P amendment.

Table 1: Dry shoot weight, leaf tissue P and pine bark extract P when 'Helleri' holly was fertilized with 3 complete fertilizers in combination with or without superphosphate (9% P) amendment of 270 g of P per m^3 of pine bark medium.

Complete Fertilizer[z]	Superphosphate Amendment With	Superphosphate Amendment Without	Significance Within Complete Fertilizer
	Dry shoot weight (g)		
Osmocote	9.8a[y]	10.7a	ns[x]
Sulfurkote	8.9a	9.5a	ns
Peters	12.5b	11.0a	ns
	Leaf tissue P (%)		
Osmocote	0.4a	0.2a	*
Sulfurkote	0.4a	0.2a	*
Peters	0.9b	1.1b	*
	Extract P (ppm)		
Osmocote	30.3a	12.3a	*
Sulfurkote	40.0a	10.9a	*
Peters	71.0b	66.4b	ns

[z]Granular slow-release fertilizers, 9.6 g Osmocote (18N-2.6P-10K) and 7.2 g Sulfurkote (24N-1.7P-8.3K), were surface applied once to each container. Peters soluble fertilizer (20N-8.7P-16.7K) was applied weekly to each container as 500 ml of a 300 ppm N solution.

[y]Mean separation within columns by parameter by Duncan's multiple range test, 5% level.

[x]Mean separation within complete fertilizer by *t* test at 5% level (*) or nonsignificant (ns).

Discussion

Sanderson and Martin (14) grew Thuja occidentalis L. and Viburnum burkwoodii Hort. Burkw. and Skipw. in P amended media and fertilized with either Osmocote (18N-4.0P-7.5K or 14N-6.2P-11.6K) or a water soluble fertilizer (25N-4.4P-8.4K) applied biweekly as 615 ppm N or at each watering as 150 ppm N. Gouin and Link (11) grew 'Helleri' without P amendment and fertilized with 100 to 150 ppm N from a 25N-4.4P-8.4K water soluble fertilizer applied at each watering or fertilized with Osmocote 18N-2.6P-18K. The findings of these researchers agree with the results of this study. Plants grown in the P amended medium and receiving the soluble fertilizer were larger than plants fertilized with Osmocote; however, plants grown without P amendment and receiving the soluble fertilizer were not significantly larger than plants fertilized with Osmocote.

Extract P levels from the Osmocote, Sulfurkote, and soluble treatment increased 18, 29.1, and 4.6 ppm respectively with P amendment. This small increase in P for the soluble treatment, resulting from P amendment, may be due to an equilibrium established in the pine bark solution. The large amount of P supplied with the soluble fertilizer resulted in high solution P levels and consequently, release of phosphorus from the P amendment was inhibited. Thus, only a small increase in extract P levels resulted from the P amendment.

Tissue P levels for the granular treatments with P amendment were significantly higher than without P amendment indicating increased P absorption with increasing medium solution P levels (7, 10), which is

further exemplified by the high tissue P levels for the soluble treatment irrespective of P amendment. Brewster et al. (3) has suggested that P uptake may exceed the growth requirement which seems evident from these data. Although P levels of 0.9% and 1.1% for the soluble treatment with and without P amendment, respectively, are relatively high, it should be noted that woody plant tissue P levels vary with the species (6, 13) and within a species (2, 13) due to fertility regime (13). Deciduous woody plant leaves generally have higher P levels than broadleaf evergreen leaves (16). Cannon et al. (5) reported P levels of 0.72% for midshoot leaves of Gleditsia triacanthos inermis Wild. 'Moraine' while Smith (16) reported an average of 0.27% P for leaves of field-grown broadleaf evergreens in Ohio. Variability of P levels within a species is exemplified by the work of Boonstra et al. (2) where P levels in terminal shoots of Taxus media Rehd. 'Hicksii' ranged from 0.28% to 0.86%.

In conclusion, pine bark media should not be amended with superphosphate when 'Helleri' holly are fertilized with complete slow-release granular or water soluble fertilizers.

Acknowledgements. The author wishes to thank Klaus Hinkelmann for statistical assistance, and the Virginia Nurserymen's Association for their support of this research.

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Chapter II

Phosphorus Requirement of *Ilex crenata* Thunb. cv. Helleri Grown in a Pine Bark Medium

To be submitted for publication in *Journal of the American Society for Horticultural Science*

Abstract. Greenhouse grown branched liners of 'Helleri' holly were fertilized with either 0, 5, 10, 15, 20, 25, or 30 ppm P to establish a P level in the pine bark medium that resulted in maximum shoot dry weight. Shoot dry weight increased as the pine bark P level increased to 10 ppm, while root dry weight decreased with increasing pine bark P levels. Total mg of P in shoot tissues continued to increase with P treatments higher than 10 ppm, indicating luxury consumption of P. Total mg of P in root tissues increased to the 10 ppm P treatment. Total µg of Fe, Cu, and Zn in shoot tissues followed the dry weight response, increasing to the 5-10 ppm P treatment, then tending to decrease as pine bark P levels increased. Root tissue Fe was erratic while Cu did not vary with treatment and Zn decreased with increasing pine bark P levels. Total µg of Mn in shoot tissues increased with P treatments while total µg of Mn in root tissues decreased with increasing pine bark P levels. In a subsequent experiment, dry shoot weights of 'Helleri' holly grown in a pine bark medium amended with either 270, 540, or 810 g/m^3 of P or fertilized with 10 ppm P were not different while root dry weights decreased with increasing P rate. Water extractable P for the 810 g/m^3 treatment decreased 245 ppm during the experiment and by week 5 was below 10 ppm P.

Additional Index Words. superphosphate, plant nutrition, micronutrient cations, holly

Introduction

The P requirement of many agronomic and forest crops has been studied extensively. Optimum soil solution P levels have been determined even though sources of P and methods of application vary. Estes and Bruetsch (14) grew Zea mays L. hydroponically and obtained best shoot growth at 44 ppm P and best root growth at 10 ppm P. Rin'kis (26) grew barley (Hordeum sp. L.), oats (Avena sp. L.), and lettuce (Lactuca sp. L.) in sand cultures and determined 40 to 60 ppm P resulted in maximum productivity. Einspahr (13) grew aspen (Populus sp. L.) in sand culture in which the greatest fresh weight occurred at 22 ppm P. Ingestad determined that Picea abies Karst. grew best in a nutrient solution containing 10 ppm P based on needle dry weight (21) and Betula verrucosa Ehrh. grew best in a nutrient solution containing 100 ppm P based on leaf and root dry weights (20).

Research is needed to establish pine bark solution P levels resulting in maximum shoot dry weight of woody nursery crops. Media amended with superphosphate (9% P) have been used extensively as a P source for container-grown woody plants (15, 16, 27, 30, 33), yet little consideration has been given to the resulting media solution P levels. Dissemination of soluble P through the irrigation water enables the grower to maintain a stable P level in the container. Flint (15) grew Forsythia intermedia Zab. 'Lynwood Gold', Weigela sp. Thunb. 'Vanicekii', Taxus cuspidata Siebold and Zucc. 'Densiformis', and Pieris japonica (Thunb.) D. Don ex G. Don in a 1 peat:1 perlite medium (v/v) and compared superphosphate (9% P) amendment with soluble $NH_4H_2PO_4$ as P

sources. Optimum acetic acid (0.018N) extractable P levels (Spurway Test) ranged from 1 ppm for 540 g/m^3 of P to 10 ppm when 135 g/m^3 of P were supplemented with 100 ppm P from $NH_4H_2PO_4$. Brewer (8) grew Ilex crenata Thunb. 'Green Island' in sand culture and recorded the greatest stem length at 8 ppm P. Sanderson and Martin (27) found that irrespective of the media studied, constant applications of 26 ppm P or biweekly applications of 108 ppm P resulted in the greatest dry weight of Viburnum burkwoodii Burkw. and Skipw. and Thuja occidentalis L. Dickey et al. (12) determined that applying more than 30 kg/ha of P did not affect visual grade or growth index of container-grown Rhododendron indicum (L.) Sweet 'Formosa' and Viburnum suspensum Lindl. Yeager and Wright (35) grew 'Helleri' holly in a pine bark medium with weekly applications of P in the irrigation water ranging from 17 to 500 ppm P and found no influence on shoot dry weight.

Few studies have been conducted maintaining constant P levels in the growing medium. The purpose of this study was to determine the sustained P level of a pine bark medium for maximal shoot dry weight of 'Helleri' holly and to determine the rate of P fertilizer necessary to maintain this level.

Materials and Methods

Experiment 1: Branched liners of 'Helleri' holly were potted June 12, 1980, in 1 liter plastic containers and subsequently grown in a greenhouse environment (28°C day, 21°C night) for 10 weeks under natural photoperiod. The pine bark medium, which contained 0.02% total P was amended with 4.7 kg/m^3 of dolomitic limestone. The media contained 16% air space (27) and a particle analysis of 38% less than 0.50 mm (U.S. Series seive #35), 28% between 0.50 mm and 1.19 mm (U.S. Series seive #16), 20% between 1.19 mm and 2.38 mm (U.S. Series seive #8), 14% between 2.38 mm and 6.35 mm (U.S. Series seive #3).

Each plant was fertilized daily with 500 ml (pH 6.3) of either 0, 5, 10, 15, or 20 ppm P as H_3PO_4, 200 ppm N as NH_4NO_3, 150 ppm K as KCl, 150 ppm Ca as $CaSO_4$, and 150 ppm Mg as $MgSO_4$, respectively. Minor elements were supplied according to Hoagland and Arnon (19) with 5 ppm Fe supplied as NaFeEDTA. Distilled water and reagent grade chemicals were used for all treatments. The experimental design was a randomized complete block design with 4 replications and 3 plants per treatment per replicate. Every 2 weeks, 50 ml of distilled water were poured onto the surface of each container and approximately 30 ml of extract (leachate) were collected. Extracts were filtered and analyzed for P (32). On August 21, roots and shoots were separated by cutting the stem just above the upper roots. Root and shoot tissue were rinsed in distilled water, dried, weighed, ashed in a muffle furnace at 450°C for 4 hr, then analyzed for total P, Cu, Fe, Mn, and Zn. P was determined colorimetrically (32) and Cu, Fe, Mn and Zn by atomic absorption spectroscopy.

Experiment 2: Experiment 2 was similar to Experiment 1 with the following exceptions. Branched liners of 'Helleri' holly were potted September 3, 1980 and grown for 12 weeks with the dark periods interrupted (11 PM to 2AM) by incadescent lighting (14 $\mu Em^{-2}s^{-1}$) to promote vegetative growth. Each plant was fertilized every other day with 500 ml (pH 6.3) of either 0, 5, 10, 15, 20, 25, or 30 ppm P as H_3PO_4, 125 ppm N as NH_4NO_3, 150 ppm K as KCl, 80 ppm Ca as $CaSO_4$, and 40 ppm Mg as $MgSO_4$, respectively. Extracts from each container were collected every 3 weeks and analyzed for P (32). Total root and shoot P, Cu, Fe, Mn, and Zn were determined.

Experiment 3: Branched liners of 'Helleri' holly were potted February 7, 1981 in pine bark amended with either 0, 270, 540, or 810 g/m^3 of P supplied as superphosphate (9% P). Every other day each plant received 500 ml of the fertilizer solution used in Experiment 2 without P. Another group of plants without P amendment received 500 ml of the same solution, but containing 10 ppm P. The experimental design and environmental conditions were like those of Experiment 2. Extracts were collected every 2 weeks as previously described and analyzed for P (32). Extract soluble salts were determined using a Barnstead conductivity bridge (Barnstead Company, Boston, MA 02132).

On May 2, leaf samples were taken from the uppermost mature foliage of all plants and plant stems were cut just above the upper roots. Leaf samples, root tissue, and shoot tissue were rinsed in distilled water, dried, weighed, and leaf tissue analyzed for P (32).

Results

Dry shoot weights of plants in Experiment 1 increased sequentially to the 10 ppm P treatment with no statistical difference in dry weights from 5 to 15 ppm P (Table 1). Dry shoot weights in Experiment 2 also increased sequentially to the 10 ppm P treatment with no statistical difference in weights for treatments higher than 5 ppm P. Dry root weights decreased with increasing P treatments in Experiments 1 and 2.

Water extractable P levels in Experiments 1 and 2 were similar to applied P levels. During the 10 week duration of Experiment 1, extract P levels for the 10 ppm treatment, which resulted in maximum shoot dry weight, ranged from 9.4 to 10.8 ppm (Table 2), while during the 12 week duration of Experiment 2, extract P levels for the 10 ppm treatment ranged from 10.2 to 10.7 ppm. The extract P levels of 0.8 and 2.2 ppm for the 0 treatment of Experiments 1 and 2 respectively, were recorded during the first extraction.

Total amounts of P, Cu, Mn, Fe, and Zn in shoot and root tissues of plants from Experiment 2 are shown in Table 3. Total mg of P increased in shoot tissue with increasing pine bark P levels while root tissue P increased to the 10 ppm P treatment. Total µg of Fe, Cu, and Zn in shoot tissues followed the dry weight response, increasing to the 5-10 ppm P treatment then tending to decrease as pine bark P levels increased. Root tissue Fe was erratic while Cu did not vary with treatment and Zn decreased with increasing pine bark P levels. Total µg of Mn in shoot tissues continued to increase with P treatments higher than 10 ppm while

root tissue Mn generally decreased with increasing pine bark P levels. Data from Experiment 1 (not shown) are similar to that of Experiment 2.

Dry shoot weight of 'Helleri' holly in Experiment 3 increased sequentially with increasing levels of P amendment although statistically no difference in dry weight was observed between 270, 540, or 810 g/m^3 of P or the 10 ppm P (phosphoric acid) treatment (Table 4). Root dry weight decreased with increasing P amendment. Extract P levels at week 1 (Figure 1) were 248, 169, and 97 ppm for the 810, 540, and 270 g/m^3 treatments, respectively and declined rapidly during the first 3 weeks. By week 5, the extract P levels for all treatments were below 10 ppm (Figure 2, expanded portion of Figure 1). Extract soluble salts (Figure 3) exhibited the same trend as P with soluble salt levels declining rapidly during the first 3 weeks. Leaf tissue P percentages (Table 5) increased statistically with increasing P amendment, while the 10 ppm P treatment resulted in the highest leaf tissue P percentage.

Table 1: Dry weight of 'Helleri' holly fertilized with different levels of P (phosphoric acid).

Treatments PPM-P Applied	Experiment 1 Dry Weight (g) Shoots	Experiment 1 Dry Weight (g) Roots	Experiment 2 Dry Weight (g) Shoots	Experiment 2 Dry Weight (g) Roots
0	4.3a[z]	0.84a	1.4a	0.39a
5	4.6ab	0.76a	1.7b	0.33b
10	5.3bc	0.80a	1.8b	0.31bc
15	4.8ab	0.77a	1.7b	0.31bc
20	4.1a	0.72a	1.7b	0.29cd
25	-	-	1.6b	0.27d
30	-	-	1.7b	0.29cd

[z]Mean separation within columns by Duncan's multiple range test, 5% level.

Table 2: Water extractable P from a pine bark medium fertilized with different levels of P (phosphoric acid).

Treatments PPM-P Applied	Experiment 1	Experiment 2
	Water Extractable P (ppm)	
0	0[z] - 0.8	0[y] - 2.2
5	4.5 - 5.2	4.2 - 5.9
10	9.4 - 10.8	10.2 - 10.7
15	15.5 - 16.9	15.1 - 17.1
20	20.2 - 22.0	19.7 - 22.3
25	--	25.6 - 28.9
30	--	30.1 - 34.3

[z]Smallest and largest mean from 4 extractions, each 2 weeks apart.

[y]Smallest and largest mean from 4 extractions, each 3 weeks apart.

Table 3: Effect of pine bark P level on mineral uptake of 'Helleri' holly (Experiment 2).

Treatments PPM-P Applied	Dry Weight (g)	P (mg/plant)	Fe	Cu	Zn	Mn
			µg/plant			
Shoot Tissue						
0	1.4a[z]	0.77a	113a	6.7a	265ab	249a
5	1.7b	4.75b	152bc	11.3ab	302c	399b
10	1.8b	7.75c	160c	12.3b	297bc	420bc
15	1.7b	9.45d	146bc	10.5ab	293bc	449bc
20	1.7b	10.08de	142b	9.9ab	265ab	425bc
25	1.6b	10.84e	143b	10.2ab	276abc	474bc
30	1.7b	11.41e	143b	9.2ab	255a	490c
Root Tissue						
0	0.39a	0.27a	37ab	3.4a	68a	33a
5	0.33b	1.48b	31a	2.8a	51b	25b
10	0.31bc	1.69b	36ab	3.0a	46bc	24bc
15	0.31bc	1.67b	34ab	2.7a	45bc	23bc
20	0.29cd	1.63b	34ab	3.3a	42c	21bc
25	0.27d	1.63b	35ab	2.9a	41c	18c
30	0.29cd	1.69b	40b	3.3a	38c	20bc

[z]Mean separation within columns, by shoots and roots, by Duncan's multiple range text, 5% level.

Table 4: Dry weight of 'Helleri' holly grown in a pine bark medium amended with superphosphate (9% P) or fertilized with 10 ppm P (Experiment 3).

Treatments	Dry Weight (g)	
	Shoots	Roots
P Amendment (g/m^3)		
0	0.94a[z]	0.22a
270	1.39b	0.19ab
540	1.48b	0.19ab
810	1.54b	0.18b
Phosphoric Acid (ppm P)		
10	1.53b	0.18b

[z]Mean separation within columns by Duncan's multiple range test, 5% level.

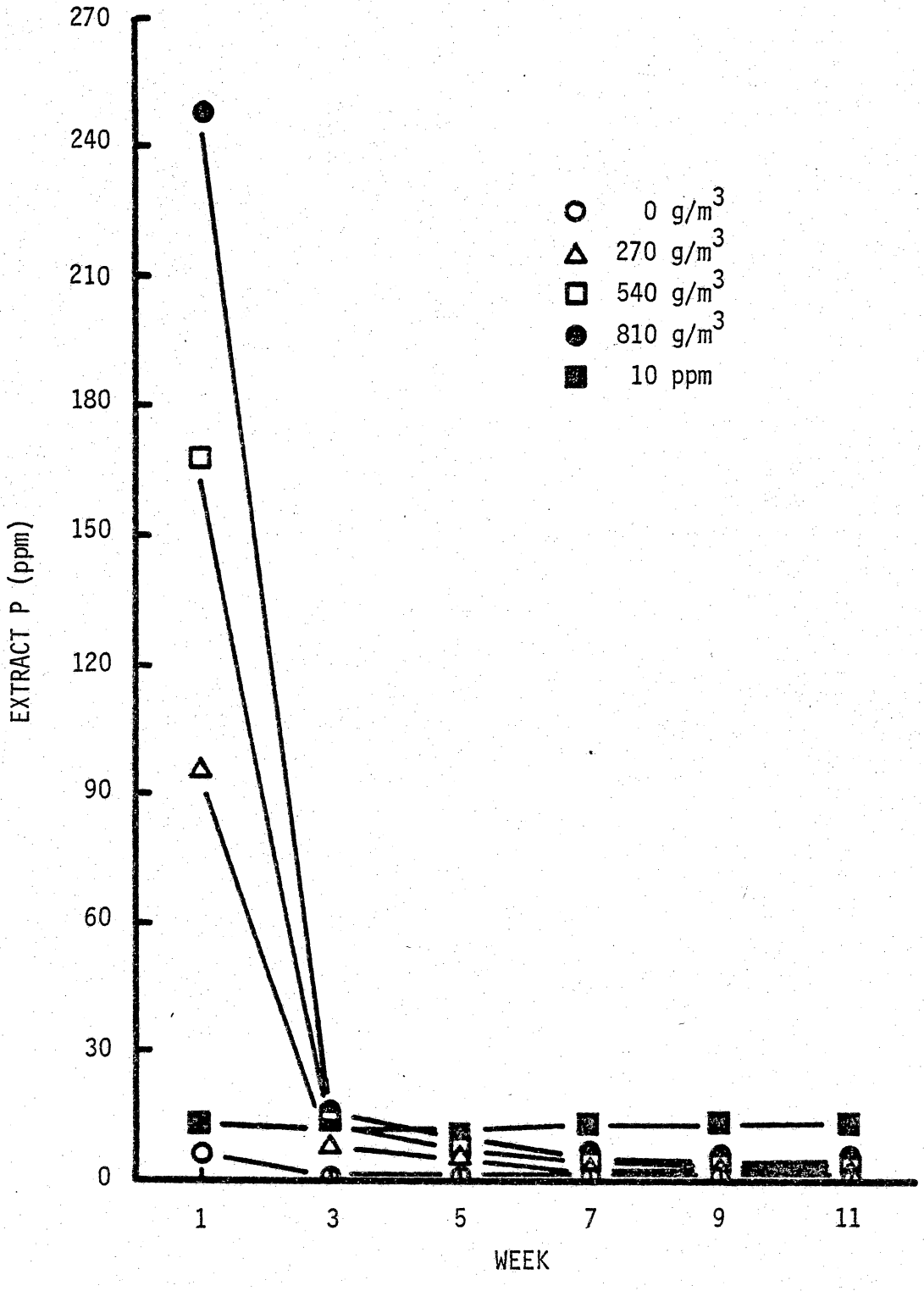

Figure 1: Extract P from a pine bark medium amended with either 0, 270, 540, or 810 g/m^3 of P supplied as superphosphate (9% P) or fertilized with 10 ppm P as phosphoric acid (Experiment 3).

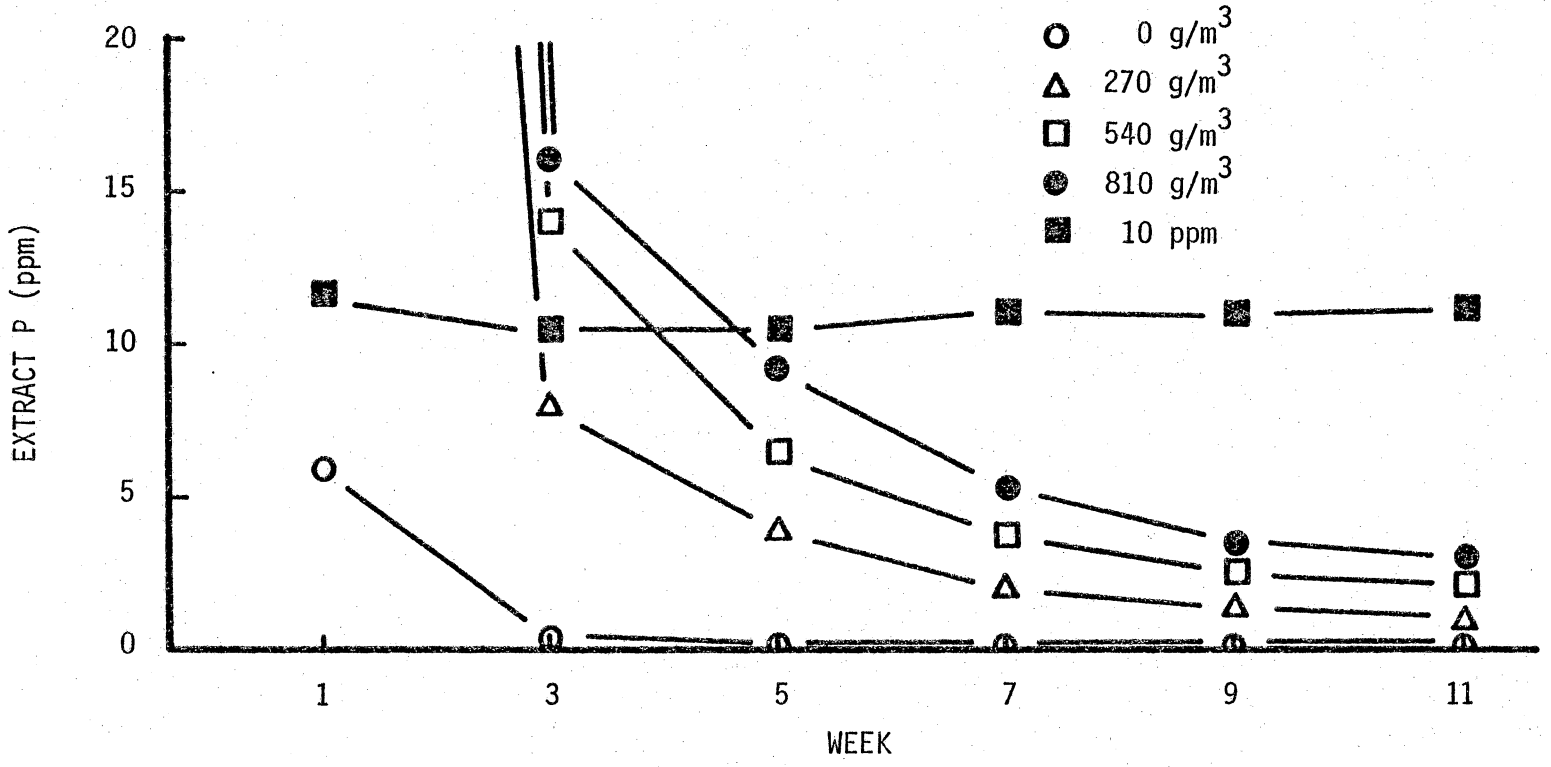

Figure 2: Extract P from a pine bark medium amended with either 0, 270, 540, or 810 g/m^3 of P supplied as superphosphate (9% P) or fertilized with 10 ppm P as phosphoric acid (Experiment 3).

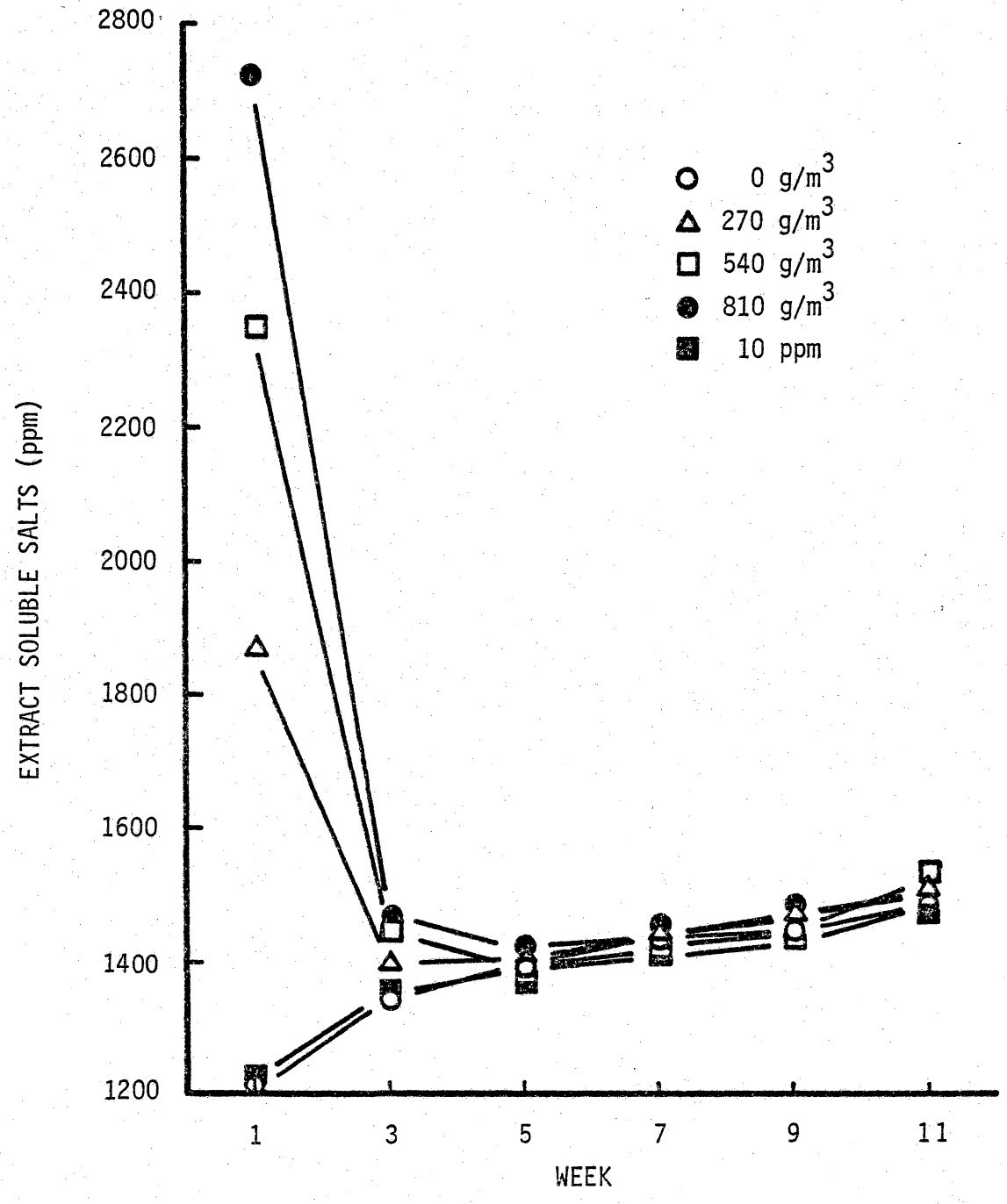

Figure 3: Extract soluble salts from a pine bark medium amended with either 0, 270, 540, or 810 g/m^3 of P supplied as superphosphate (9% P) or fertilized with 10 ppm P as phosphoric acid (Experiment 3).

Table 5: Leaf tissue P (%) of 'Helleri' holly grown in a pine bark medium amended with superphosphate (9% P) or fertilized with 10 ppm P (Experiment 3).

Treatments	% P Leaves
P Amendment (g/m^3)	
0	0.04a[z]
270	0.15b
540	0.17b
810	0.20c
Phosphoric Acid (ppm P)	
10	0.32d

[z]Mean separation within columns by Duncan's multiple range test, 5% level.

Discussion

Maintaining 10 ppm P in the pine bark medium resulted in the greatest dry weight of 'Helleri' holly. This agrees with the results of Ingestad (21) for Picea which grew best, based on needle dry weight, in a nutrient solution containing 10 ppm P and Brewer (8) where 8 ppm P resulted in the greatest stem length of Ilex grown in sand culture. The results from Experiments 1 and 2 also concur with those of Flint (15) who determined 10 ppm acetic acid (0.018N) extractable P is the upper limit of an optimum range for Forsythia, Weigela and Pieris when grown in a medium (v/v) of 1 peat:1 perlite.

Root dry weight numerically decreased in Experiment 1 and decreased statistically in Experiment 2 with increasing pine bark P levels. A response of this type was observed by Yeager and Wright (35) for 'Helleri' holly where N treatments that increased shoot growth caused a corresponding decrease in root growth. This is thought to be due to competition by expanding shoots for nutrients (10). Adriano et al. (1) also noted a reduction in root growth of Zea mays when nutrient solution P levels increased from 4 to 155 ppm. Gerloff (17) observed increased root growth for beans (Phaseolus vulgaris L.) grown with inadequate P while Atkinson (3) observed the same response for over 20 species of herbs.

Total mg of P in shoot tissues (Experiment 2) increased statistically with increasing pine bark P levels (Table 3). Other investigators (7, 18, 22, 23, 24) have reported increased absorption with increasing growing medium P levels or availability while Asher and

Loneragan (2) noted that species differ in ability to absorb P. In the present investigation, total mg of P in shoot tissues continued to increase with P treatments greater than 10 ppm, even though dry shoot weights decreased, indicating luxury consumption of P. Brewster et al. (9) also noted that P uptake may exceed the plant's growth requirement.

In Experiments 1 and 2, shoot dry weight increased sequentially to the 10 ppm P treatment then decreased statistically in Experiment 1 as pine bark P levels increased. A response of this type was observed by Moose (25) working with onions (Allium sp. L. var. James Keeping). This decrease in dry weight at high P levels may be explained by P-micronutrient cation relationships. In Experiment 3, total µg of Fe, Cu, and Zn in shoot tissues followed the dry weight response, increasing to the 5-10 ppm treatment then tending to decrease as pine bark P levels increased. Root tissue Fe was erratic while Cu did not vary with treatment and Zn decreased with increasing pine bark P levels (Table 3). Mathanan and Amberger (23) working with maize (Zea sp. L) and Cumbus et al. (11) with watercress (Rorippa nasturtium - aquaticum (L.) Hayek) indicated Fe was inactivated in roots as growing medium P levels increased, thus reducing shoot tissue Fe. Watanabe (31) observed decreases in Fe of Phaseolus vulgaris (Univ. Idaho III) shoots with increasing nutrient solution P levels. A similar response for Cu has been reported with sour orange (Poncirus sp. Raf.) where increasing growing medium P levels reduce Cu uptake or availability (5, 6). Data from Experiment 3 do not indicate that similar Fe-P or Cu-P responses occurred with 'Helleri' holly.

Shoot tissue Zn decreased as treatments increased from 5 to 30 ppm P, while root tissue Zn decreased for treatments 0 to 30 ppm P (Table 3). Reduced Zn uptake with increasing growing medium P (4, 5) supports the theory of P-Zn precipitation in the growing medium; however, precipitate formation has been refuted (5, 29). Stukenholtz (29) suggests this reduction in Zn uptake occurs as a physiological change in the root or root surface. However, in the present investigation, reduced Zn uptake is not evident in view of the fact that root and shoot dry weights decreased with treatments higher than 10 ppm P.

Total µg of Mn in shoot tissues (Table 3) continued to increase with P treatments higher than 10 ppm, a possible explanation for the decrease in dry shoot weight. However, it should be noted that plant responses to Mn vary with species. Bingham (4) observed an increase in Mn uptake for kidney beans (Phaseolus sp.) when nutrient solution P increased from 1 to 100 ppm and decreases in Mn uptake for Pearson tomatoes Lycopersicon esculentum Mill. and sour orange (Poncirus sp.). Bingham et al. (6) noted that for sour orange a Cu-P relationship was important in reducing growth; whereas, for Trifolium subterraneum L. a P-Zn relationship may be important in altering growth (24) thus emphasizing relationship variation between species.

Dry shoot weights (Experiment 3) for plants growing in pine bark amended with 270, 540, or 810 g/m^3 of P supplied as superphosphate (9% P) were not statistically different from those of plants in which 10 ppm P was maintained in the pine bark medium (Table 4). The extract P levels for all treatments declined rapidly (34) and were below 10 ppm by

week 5 (Figure 2), thus, indicating that superphosphate should not be used as a P source. These data agree with Flint (15) in that a critical P concentration range is not maintained in a superphosphate-amended medium.

The fact that shoot dry weight differences were not evident when comparing the 270, 540, and 810 g/m^3 treatments with the soluble treatment, could be due to mobilization (9) and utilization of P absorbed during the initial part of the experiment when pine bark P levels were high. With a longer experimental period, shoot dry weight differences might be found since extract P levels were below 10 ppm by week 5. During the first 3 weeks of the experiment extract soluble salts for the 540 and 810 g/m^3 treatment were above 2100 ppm, a high salt level for 'Helleri' holly (personal communication, R.D. Wright). Salt burn could result if the medium were allowed to dry. Leaf tissue P percentages (Table 5) reflect the extract P levels after week 5, with increasing pine bark P levels resulting in increasing leaf tissue P percentages which concurs with the work of others (6, 14, 20, 21).

These studies indicate that maintaining 10 ppm P in the pine bark medium results in the greatest dry weight of 'Helleri' holly. A stable pine bark P level was not attained with superphosphate-amended pine bark, thus superphosphate is not recommended as a P source.

Acknowledgement. The author wishes to thank the Virginia Nurserymen's Association for their support of this research.

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Chapter III

Pine Bark - Phosphorus Relationships

To be submitted for publication in

Communications in Soil Science and Plant Analysis

Abstract. Columns of milled pine bark were leached weekly with 576 ml of distilled water for 6 weeks and leachates collected. Leachate analysis indicated that significant amounts of indigenous P were leachable. When 'Helleri' holly were grown in pine bark with no P applied, 0.08 mg of indigenous P was absorbed. Studies were also conducted to determine if pine bark adsorbs P. The amount of P adsorbed by pine bark, as determined by equilibrium solution analysis, ranged from 0 to 9.7 µg per g of pine bark when solutions of P ranging from 0 to 15 ppm were equilibrated with pine bark for 24 hours. P adsorbed, as determined by analysis of the above ashed pine bark following distilled water leaching, ranged from 0 to 5.3 µg per g of pine bark for the 0 to 15 ppm P treatments, respectively.

Additional Index Words. indigenous phosphorus, adsorption, leaching, absorption, effluxing, Ilex crenata Thunb. cv. Helleri

Introduction

Container nursery plants in the southeast are grown in a pine bark medium or a medium that contains a large percentage of pine bark. However, most fertilization practices and regimes for container plants are based on a mineral soil system. Phosphorus applied to mineral soil is generally fixed (2, 3, 9, 11, 13), thus leading to the practice of amending container mixes with superphosphate. However, P leaches from organic soils (12) and peat mixes (2, 13). The extent of P leaching or adsorption by a pine bark medium is not known. Yeager and Wright (17) found P in the water extract of a pine bark medium not previously fertilized with P. Thus, pine bark may possess indigenous P available for plant growth. Research is needed to investigate the relationships of indigenous and applied P with pine bark and the availability of indigenous P for plant growth. The following studies were conducted to determine if pine bark contains leachable indigenous P, whether it is available for plant growth and if pine bark adsorbs applied P.

Materials and Methods

Experiment 1. Two PVC columns (4 x 13.5 cm) were filled with 35 g of milled pine bark with an ashable P content of .02%. Particle analysis of the pine bark revealed 38% less than 0.50 mm (U.S. Series seive #35), 28% between 0.50 mm and 1.19 mm (U.S. Series seive #16), 20% between 1.19 mm and 2.38 mm (U.S. Series seive #8), 14% between 2.38 mm and 6.35 mm (U.S. Series seive #3). Six pieces of #42 filter paper were placed on the surface of each column to disperse applied water and cheese cloth was secured around the bottom of each column by a rubber band to retain the pine bark. The columns remained in the laboratory for 6 weeks (21-27°C).

Each week, 576 mls of distilled water (pH 6.3) were applied by peristaltic pump to the surface of each column at a rate of 32 ml/hr. Leachates were collected, analyzed for P (15) and total mg of P leached were calculated.

Experiment 2. Branched liners of 'Helleri' holly (Ilex crenata Thunb.) were potted in 1 liter plastic containers and subsequently grown in a greenhouse environment (28°C day, 21°C night). Initially, 10 representative plants were sacrificed for dry weight and P analysis (15). The pine bark medium which contained .02% total P, was amended with 4.7 kg/m^3 of dolomitic limestone. Each plant was fertilized daily with 500 ml (pH 6.3) of either 0, 5, 10, 15, or 20 ppm P as H_3PO_4, 200 ppm N as NH_4NO_3, 150 ppm K as KCl, 150 ppm Ca as $CaSO_4$, and 150 ppm Mg as $MgSO_4$. Minor elements were supplied according to Hoagland and Arnon (10) with 5 ppm Fe supplied as NaFeEDTA. Distilled water and reagent

grade chemicals were used for all treatments. The experimental design was a completely randomized block design with 4 replications and 3 plants per treatment per replicate. After 10 weeks, plant stems were cut just above the upper roots. Root and shoot tissue were rinsed in distilled water, dried, weighed and analyzed for P (15).

Experiment 3. Branched liners of 'Helleri' holly were handled as in Experiment 2 with the following exceptions. The plants were grown for 12 weeks with the dark periods interrupted (11 PM to 2 AM) by incadescent lighting (14 $\mu Em^{-2}s^{-1}$) to promote vegetative growth. Each plant was fertilized every other day with 500 ml (pH 6.3) of either 0, 5, 10, 15, 20, 25, or 30 ppm P as H_3PO_4, 125 ppm N as NH_4NO_3, 150 ppm K as KCl, 80 ppm Ca as $CaSO_4$, and 40 ppm Mg as $MgSO_4$. The total amount of P accumulated in shoot and root tissues of plants from Experiments 2 and 3 was calculated by subtracting the nutrient content of plants initially from that determined at the end of each experiment.

Experiment 4. Milled pine bark was incorporated with 4.7 kg/m^3 of dolomitic limestone, placed in a plastic bag at 80% moisture and incubated at 25°C for 2 weeks to adjust pH. Distilled water was added daily to maintain 80% moisture. Eighteen g of the above pine bark were placed in each of 12, 125 ml Erlenmeyer flasks. Thirty-two ml of nutrient solution, like that of Experiment 3, but 1.25 times more concentrated were added to each flask so that 3 flasks contained either 0, 5, 10, or 15 ppm P. Two drops of toluene were added to each flask, then stoppered and placed in a shaker for 24 hr (25°C, 60 oscillations/min).

The equilibrium solution (pH 5.3) from each flask was filtered and analyzed for P (15).

The pine bark from each flask was placed in a PVC column like that of Experiment 1, but with a 2.5 cm diameter. Each column was leached with distilled water (pH 6.3) for 12 hr (1.5 ml/min). The pine bark was then ashed (450°C) and analyzed for P (15). The experiment was repeated.

Results

Total mg of indigenous P leached from the pine bark columns are shown in Table 1. The mg of P leached decreased each week and after 6 weeks 3.5 mg of indigenous P had leached from 35 g of pine bark. The initial total ashable P was 7 mg/35 g of pine bark, thus 51% of the indigenous P was leached.

Total mg of P accumulated per plant are shown in Table 2. In Experiment 2, 0.17 mg of P was effluxed when 0 ppm P were applied; whereas, in Experiment 3, 0.08 mg of P was accumulated when 0 ppm P were applied. Plant P accumulation in Experiment 2 and Experiment 3 increased as the amount of P applied increased. Total mg of P in shoot and root tissues of plants before treatment and at experiment termination for Experiments 2 and 3 are shown in Table 3. Root tissue P for Experiment 2 decreased from 2.31 to 0.71 mg during the experiment while shoot tissue P increased from 1.96 to 3.39 mg. The same trend was evident for Experiment 3. Root tissue P decreased from 0.65 to 0.27 mg while shoot tissue P increased from 0.31 to 0.77 mg.

The amount of P adsorbed by pine bark (Experiment 4) increased with increasing P treatment and was greater for equilibrium solution analysis than that determined by analysis of ashed pine bark after leaching (Figure 1). Adsorption for the 0 to 15 ppm P treatment ranged from 0 to 5.3 µg and 0 to 9.7 µg of P per g of pine bark for ashed and equilibrium solution analyses, respectively.

Table 1: Leachable indigenous P from 35 g of pine bark.

Day[z]	P Leached (mg/35 g)
1	2.37
8	0.50
15	0.33
22	0.12
29	0.12
36	0.10
Total	3.54

[z]576 ml of distilled water applied each day as 32 ml/hr.

Table 2: Total mg of P accumulated by 'Helleri' holly grown at different levels of P.

PPM-P Applied	P Accumulated (mg/plant)	
	Experiment 2	Experiment 3
0	-0.17a[z]	0.08a
5	6.09b	5.27b
10	13.37c	8.48c
15	16.93d	10.15d
20	17.26d	10.75de
25	--	11.51de
30	--	12.14e

[z]Mean separation within columns by Duncan's multiple range test, 5% level.

Table 3: Total mg of P in 'Helleri' holly shoots and roots before and after fertilization with a nutrient solution without P.

	Experiment 2 (mg/plant)		Experiment 3 (mg/plant)	
	Shoots	Roots	Shoots	Roots
Before Treatment	1.96a[z]	2.31a	0.31a	0.65a
After Treatment	3.39b	0.71b	0.77b	0.27b
Difference	+1.43	-1.60	+0.46	-0.38

[z]Mean separation within columns by *t* test, 5% level.

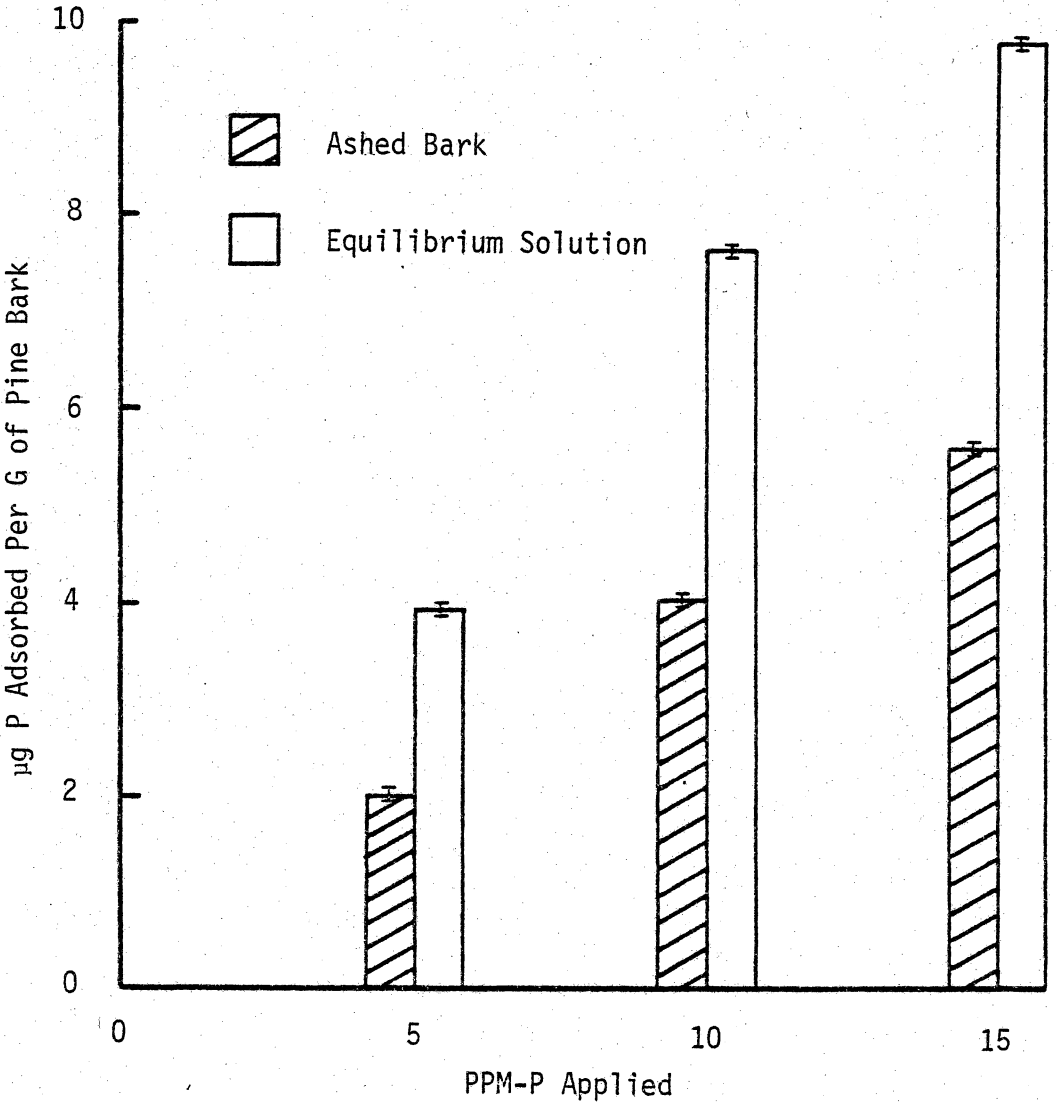

Figure 1: P adsorbed by pine bark as determined by equilibrium solution analysis and ashed pine bark analysis. Vertical bars indicate standard errors.

Discussion

Approximately half of the indigenous P was leached from the pine bark columns (Table 1). This means for a 1 liter container of pine bark, 16.7 mg of P would be leached. 'Helleri' holly grown in a 1 liter container would contain on the average 24 mg of P; thus, more than half of the P needed by the plant is initially contained by the pine bark medium. Data for the 0 treatment of Experiment 3 indicate that 'Helleri' holly absorbs indigenous P from pine bark (Table 2). However, the 0.08 mg of P absorbed is only a fraction of the P utilized during a growing season. This is because indigenous P leaches from pine bark at a very rapid rate initially, thus reducing the potential for indigenous P uptake. This point was illustrated in Experiment 1 where 67% of the indigenous P was leached the first day.

Data from Experiment 2 (Table 2) indicate that 'Helleri' holly lost 0.17 mg of P during the experimental period when 0 ppm P were applied. This does not agree with Experiment 3; however, in both experiments P was lost from root tissues and accumulated in shoot tissues (Table 3). In Experiment 2, root tissue P decreased by 1.60 mg while shoot tissue P increased by 1.43 mg. Assuming translocation of root P to shoots a deficiency of 0.17 mg of P results. Applying the same reasoning to Experiment 3, a net accumulation of 0.08 mg of P occurs.

This deficiency of 0.17 mg of P in Experiment 2 could be due to P effluxed from the roots. Effluxing of P from roots has been shown by many investigators (1, 5, 6, 7, 14); however, neither conditions causing effluxing nor the mechanisms are understood. Moore (14) working with

maize (Zea sp. L.) found P was absorbed by roots, translocated to shoots, then to roots where effluxing occurred. Emmert (5) observed more effluxing from Phaseolus vulgaris L. roots when grown in demineralized water compared to tap water. However, effluxing increased when P was added to the nutrient solution. Divalent cations in solution also increase effluxing (6). Emmert (6) has reported that effluxed P may be about 1% of root P based on the activity of ^{32}P, while Fedorovskii (7) has reported as much as 4 to 25% of corn (Zea mays L.) plant P may be effluxed.

In the present investigation, 'Helleri' holly plants in Experiment 2 were fertilized daily while plants in Experiment 3 were fertilized every other day. This increase in the volume of solution percolating through the medium in Experiment 2 could have caused the greater loss of P from roots, compared to Experiment 3. The net increase in P content for Experiment 3 indicates that indigenous P of pine bark is absorbed by 'Helleri' holly. The fact that a net decrease in P content was observed in Experiment 2 does not mean that P was not first absorbed from the pine bark and then effluxed.

Mucks and soils that contain a large percentage of organic matter often do not contain the quantity of Fe and Al (8) found in mineral soils. Thus organic colloids possess a low P adsorption capacity (4, 8). Applied P in such soils is leached (12) and adsorbed P is water soluble (8). A similar situation exists with pine bark in that the amount of P adsorbed as determined by equilibrium solution analysis, was greater than that determined by analysis of ashed pine bark. This

indicates that some adsorbed P was lost when the pine bark was leached prior to ashing. The amount of P lost by leaching at each treatment is proportional to that adsorbed at each treatment as determined by analysis of ashed bark. Thus, P precipitation is unlikely.

These data show that when applying 10 ppm P solutions, 7.5 µg of P are bound per g of pine bark. Previous experiments have shown that maintaining 10 ppm P in the pine bark medium results in maximum shoot dry weight of 'Helleri' holly (17). On the basis of a 1 liter container, 1.2 mg of P are bound and if available for plant uptake, this would be a negligible part of the 24 mg of P used for 'Helleri' holly growth. Thus the amount of P bound by the pine bark is incidental compared to that absorbed by the plant. Furthermore, these data support previous experiments (17) which indicate that P supplied as a pine bark amendment is leached.

Acknowledgement. The author wishes to thank the Virginia Nurserymen's Association for their support of this research.

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General Conclusion

Studies were conducted to characterize the P nutrition of 'Helleri' holly and pine bark P relationships. These studies indicate that nurserymen should maintain 10 ppm P in the pine bark medium for maximum shoot growth of 'Helleri' holly. P in superphosphate-amended pine bark leaches rapidly and a stable pine bark P level is not maintained. Amending the pine bark medium with superphosphate, did not increase the growth of 'Helleri' holly when subsequently fertilized with a complete slow-release granular or water soluble fertilizer. Based on these findings superphosphate is not recommended as a P source for plants grown in pine bark.

Pine bark-P relationships revealed that indigenous P in pine bark leaches rapidly limiting its uptake by plants. 'Helleri' holly absorbed 0.08 mg of indigenous P during a 12 week experiment, a small fraction of the P utilized by the plant during a growing season. Pine bark adsorption studies revealed that 7.5 μg of P were bound per g of pine bark when 10 ppm P solutions are applied. On the basis of a 1 liter container, 1.2 mg of P would be bound by pine bark, a negligible part of the 24 mg of P used by a 'Helleri' holly plant during a growing season.

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Phosphorus Nutrition of *Ilex crenata* 'Helleri'
Grown in a Pine Bark Medium

by

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(ABSTRACT)

The purpose of this research was to characterize the phosphorus nutrition of *Ilex crenata* 'Helleri' and pine bark P relationships. Branched liners of 'Helleri' holly were grown in a pine bark medium in which 0, 5, 10, 15, 20, 25, or 30 ppm P were maintained. Shoot dry weight increased as the pine bark P level increased to 10 ppm P, while root dry weight decreased with increasing pine bark P levels. Total mg of P in shoot tissues continued to increase with P treatments higher than 10 ppm, indicating luxury consumption of P. Total mg of P in root tissues increased to the 10 ppm P treatment. Total µg of Fe, Cu, and Zn in shoot tissues followed the dry weight response, increasing to the 5-10 ppm P treatment then tending to decrease as pine bark P levels increased. Root tissue Fe was erratic while Cu did not vary with treatment and Zn decreased with increasing pine bark P levels. Total µg of Mn in shoot tissues increased with P treatments while total µg of Mn in root tissues decreased with increasing pine bark P levels.

Dry shoot weights of 'Helleri' holly grown in a pine bark medium amended with either 270, 540, or 810 g/m^3 of P or fertilized with 10 ppm P were not different while root dry weights decreased with increasing pine bark P levels. Water extractable P for the 810 g/m^3 treatment decreased 245 ppm during the experiment and by week 5 was below 10 ppm.

Amending the pine bark medium with 270 g/m^3 of P did not increase the dry shoot weight of 'Helleri' holly when subsequently fertilized with a complete slow-release granular or water soluble fertilizer.

The pine bark medium contained indigenous P which leached rapidly. When 'Helleri' holly were fertilized with a nutrient solution without P, 0.08 mg of indigenous P were absorbed. The pine bark adsorbed 7.5 µg of P per g of pine bark when equilibrated with a 10 ppm P nutrient solution.

These studies indicate that maintaining 10 ppm P in the pine bark medium results in the greatest dry weight of 'Helleri' holly. A stable pine bark P level was not attained with superphosphate-amended pine bark, thus superphosphate is not recommended as a P source. Pine bark P relationships revealed that 'Helleri' holly absorbed indigenous P while a negligible amount of P was bound by the pine bark compared to the amount of P used by a 'Helleri' holly grown in a 1 liter container.