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Total Root and Shoot Biomass Inhibited by Paclobutrazol Application on Common Landscape Trees

By Chad M. Rigsby, E. Thomas Smiley, Sean Henry, Liza Holmes, and Andrew L. Loyd

Abstract. Background: Paclobutrazol (PBZ) is used in the arboriculture industry to reduce the growth of trees. It works by inhibiting gibberellin biosynthesis, a group of phytohormones associated with cell elongation. A substantial amount of variation exists within the literature as to the impact of PBZ on woody plant root systems. The purpose of this study was to assess the impact of PBZ on belowground growth and biomass allocation among plant species with varying levels of PBZ sensitivity in a controlled setting. Methods: We treated containerized silver maple, white oak, pecan, laurel oak, and stone pine trees with Cambistat® at the full label rate, one category lower, two categories lower, or water-only controls. After a 14-month incubation period, leaf, stem, and root tissue dry mass were quantified, root:shoot ratios were calculated, the length of the longest root quantified, and total root length of a subset of replicates was estimated. Species were statistically analyzed separately and collectively to assess trends. Results: Paclobutrazol application resulted in significantly lower root dry mass and total root length for all species analyzed, and significantly reduced longest root length of all species except for silver maple. Across species and dosage combinations, we saw few dose effects on any response variable and no major trends in root:shoot ratios. Conclusion: The impact of PBZ on trees in the landscape appears to be influenced by a number of factors, but we observed relatively consistent results on belowground biomass when growing conditions were uniform in our controlled experiment.

Keywords. Anti-Gibberellin; Growth Reduction; Paclobutrazol; Plant Growth Regulators; Root Growth.

INTRODUCTION

Synthetic compounds used to reduce the shoot length of plants without altering developmental patterns or being phytotoxic are referred to as plant growth regulators (PGRs), though they are more accurately described as plant growth retardants (Rademacher 2000). Many of these compounds function by inhibiting the synthesis of gibberellins (Rademacher 2000; Gao et al. 2017), a large family of tetracyclic diterpenoid phytohormones that regulate many different aspects of plant development and growth, including cell division and elongation, seed germination, stem and hypocotyl elongation, root elongation and root hair development, and flowering (Bottini et al. 2004; Sun and Gubler 2004; Sun 2011). Of these known functions, the involvement of gibberellins in cell elongation and general plant growth is the most understood, with an abundance of primary research and review literature having been devoted to this topic (e.g., Davière and Achard 2013). Paclobutrazol (PBZ) is a triazole-type PGR that functions by acting as a competitive inhibitor of *ent*-kaurene oxidase (Rademacher 2000), a key enzyme in the gibberellin biosynthesis pathway (Gao et al. 2017).

Paclobutrazol is commonly used in agriculture, horticulture, arboriculture (commercial, residential, and utility), and urban forestry. Within the arboriculture and urban forestry industries, PBZ can drastically reduce the frequency of pruning (Mann et al. 1995; Burch et al. 1996). The applications of PBZ to plants result in the phenotype, generally, of reduced shoot growth/internode length, leaf size, and stem diameter (Davis et al. 1985; Watson 1996; Hartman et al. 2009; Smiley et al. 2009; Bai et al. 2005; Tanis et al. 2015), effects that are uniformly reported in the literature. Interspecific variation in sensitivity to PBZ exists among woody plant species (Bai et al. 2005), but mechanistic explanations for this variation are not well understood. Titration effects of PBZ on *ent*-kaurene oxidase enzyme abundance (Swain et al. 2002), variation in enzyme susceptibility to PBZ inhibition (Miyazaki et al. 2011), and alternative (e.g., environmental) inputs driving growth gene expression (Monson et al. 2022) could all contribute to species variation in PBZ susceptibility. Aside from anatomical responses, researchers have studied a variety of other effects of PBZ treatment including defense against herbivory (e.g., Chorbadjian et al. 2011), pathogenic fungi (e.g., Watson and Jacobs 2012), drought stress (e.g., Percival and AlBalushi 2007), light and thermal stress (e.g., Mahoney et al. 1998), and the physiological mechanisms of stress tolerance (e.g., Percival and Noviss 2008) to name a few.

One of the effects of PBZ on woody plants that has received significant research attention is its impact on root system growth, development, and recovery from injury. Research using herbaceous model plants (e.g., *Arabidopsis* spp.) demonstrates clearly and mechanistically that root elongation requires gibberellins (e.g., Wittenmayer and Merbach 2005; Ubeda-Tomás et al. 2008; Ubeda-Tomás et al. 2009; Barker et al. 2021). In agreement with this work with model plants, much research using woody plants has shown PBZ to have either a neutral effect or a significant reduction in root growth/size metrics using several study species and growth stages/size classes (e.g., seedlings, saplings, mature trees), including apple (*Malus domestica*)(Wieland and Wample 1985; Wang and Faust 1986; Hodairi et al. 1988; Zeller et al. 1991), peach (*Prunus persica*)(Rieger and Scalabrelli 1990; Avidan and Erez 1995), nectarine (*P. persica* var. nucipersica)(Avidan and Erez 1995), ash (*Fraxinus* spp.) (Sterrett and Tworkoski 1987; Tanis et al. 2015), bur oak (*Quercus macrocarpa*)(Watson and Hewitt 2017), and white oak (*Q. alba*)(Watson 2006). However, other studies have reported a positive effect of PBZ on root systems also using several different study species and growth stages, including apple (Wang and Faust 1986; Ma 1990), sweet orange (*Citrus sinensis* cv. Valencia)(Vu and Yelenosky 1992), pin (*Q. palustris*) and white oak (Watson 1996; Watson and Himelick 2004), and American elm (*Ulmus americana*) (Watson 2001). Similar variation exists among studies assessing the ability of PBZ to aid in root system recovery/enhancement following root injury. Watson (2004) found a positive impact for PBZ treatment for root-injured black maple (*Acer nigrum*) but not for green ash (*F. pennsylvanica* 'Summit'). Similarly, Gilman (2004) found no impact of PBZ on root-injured live oak (*Q. virginiana*), but Martínez-Trinidad et al. (2011) reported reduced root growth of live oak following root pruning.

Paclobutrazol-mediated shifts in biomass allocation to belowground tissues have been reported in several studies (e.g., Tanis et al. 2015; Wang et al. 2019). These shifts in biomass allocation may be due to reductions in aboveground growth with little or no reductions in belowground growth (e.g., Hodairi et al. 1988) or absolute increases in root growth (Cregg and Ellison-Smith 2020). However, this is not uniformly reported in the literature, either. Ruter (1994) measured reductions in root to shoot ratios in potted *Pyracantha* x 'Mojave' regardless of dose, illustrating that PBZ treatment may have a greater impact on below- than aboveground growth. The variation in results within the literature suggest that the impact of PBZ on woody plant root systems and biomass allocation appears to be situational, and likely depends on numerous factors (Watson 2001) including species (e.g., Watson 2004), health condition of the plant (e.g., Watson 1996, 2001, 2006), PBZ dose (e.g., Wang and Faust 1986), and also other likely factors such as soil-PBZ-water interactions (e.g., Milfont et al. 2008; Wu et al. 2013).

Given the variation reported in the literature, studies incorporating additional species with varying levels of PBZ sensitivity subjected to uniform growing conditions are needed to assess general trends for the effects of PBZ on woody plant root systems and shifts in biomass allocation. Here, we present the results of a study assessing the impact of full and partial doses of PBZ (Cambistat®, Rainbow Ecoscience, Minnetonka, MN, USA) on the biomass of containerized ornamental woody plant species along a PBZ susceptibility gradient. We hypothesize that all species will roughly adhere to their PBZ-treated aboveground "phenotype" (i.e., more pronounced impact of PBZ on more susceptible than less susceptible species resulting in decreased stem and leaf mass) known within the industry and reported in the literature (e.g., Bai et al. 2005). We also hypothesize that these aboveground phenotypes will be mirrored belowground with decreased root system development in PBZ-treated plants, and that the mirroring of above- and belowground phenotypes will not result in major shifts in biomass allocation above- and belowground. Lastly, we hypothesize that reduced PBZ dosages will likewise reduce the magnitude of growth reduction (i.e., reduced PBZ dose will relieve growth suppression).

MATERIALS AND METHODS

Plants and Treatments

Experiments took place at the Bartlett Tree Research Laboratories (BTRL) in Charlotte, NC, USA. Five species across a PBZ-susceptibility gradient were acquired on 2019 March 21 as 30 to 46 cm tall bare root plants from Rennerwood Nursery (Tennessee Colony, TX, USA). These species were: silver maple (*A. saccharinum*), white oak (*Q. alba*), pecan (*Carya illinoinensis*), laurel oak (*Q. laurifolia*), and stone pine (*Pinus pinea*). Upon arrival, plants were heeled into a pile of pine fines (bulk shredded pine bark mulch; SiteOne Landscape Supply, Roswell, GA, USA) and kept moist for approximately one week until they could be potted. Plants were dormant when they were potted into 2-gallon (7.5-L) pots in a 60:40 mix of sand (bulk course sand; SiteOne Landscape Supply) and pine fines in early 2019 April. Potted plants were then transferred to the BTRL glasshouse (glasshouse conditions: 12 to 15 °C in winter and 27 to 30 °C in summer, natural day light exposure, 75% to 80% humidity year round). Plants were organized randomly on benchtops (i.e., complete randomization of all species and treatment groups) and allowed to acclimate for one month prior to treatment. Once potted and throughout the duration of the experiment, monitoring and irrigation occurred every other day with approximately 1 L water.

Species were treated in accordance with and relative to the published susceptibility rating on the Cambistat® label: category “D” (silver maple and white oak), category “E” (pecan), and category “F” (laurel oak and stone pine). This species selection represented the categories on the Cambistat® label that are considered to have moderate-low levels of PBZ sensitivity (category “A” represents the most sensitive plants and category “F” represents plants with low levels of sensitivity). Plants within a species were split into 4 treatment groups: non-treated controls (dosed with water only), full rate (dosed in accordance with the label rate), 1 category lower rate (1CL)(e.g., an F species was dosed at the E rate), and 2 category lower rate (2CL)(e.g., an F species was dosed at the D rate), each with 10 replicate plants. A linear equation was derived from the Cambistat® label where volume of product was plotted as a function of stem diameter and the equation was used to calculate volume of product required based on the plant stem caliper of our plants. This volume of product was then diluted (11-parts tap water to 1-part Cambistat®)(e.g., 5 mL product was diluted in 55 mL water) and applied by pouring the solution on the soil surface around the stem. Applications of PBZ were made on 2019 May 3, plants monitored regularly through the duration of the study, and plants that died throughout the duration of the experiment were removed as they died. Plants were allowed to grow for 14 months, after which they were destructively harvested (Table 1).

Plants were all harvested within one week which was done by carefully removing the plant from pots and extensively washing the root systems with water to remove debris and media. During the washing of root systems, we attempted to minimize the loss of fine roots during manual washing, but no attempts were made to recover any fine roots that were potentially lost during the process. Plants were then divided into leaves, stems, and roots. Leaf and stem tissues were placed in an oven to dry for at least 24 hours or until tissues were a constant mass, and then weighed to quantify dry mass. For root systems, the length of the longest root was first measured for each replicate, then subsets of species × treatment combinations were randomly selected and used to estimate the total root length prior to drying and quantifying dry mass. We were only able to estimate average total root length for subsets of samples within species × treatment combinations due to limitations in time availability and person-power, but as many replicates as possible were used (Table 1). Replicates that were not selected for the estimation of average total root length were immediately subjected to drying for the measurement of dry mass. For those replicates that were selected, total root length was estimated using the commercially available software Assess 2.0 (Lamari 2008) using a high-resolution tabletop scanner (Epson Perfection V700 Photo Duel Lens System, Suwa, Nagano, Japan). The program measured the total length of all roots in the scanned image by contrasting the roots behind a standard blue background and scale bar. After total root length was measured, these root systems were then dried as the other tissues and their root mass was measured. These data were also

Table 1. Summary of experimental set up.

Tree species (label category)	Treatment[1] (label category)	Replicates after mortality[2]	Replicates used to quantify average total root length	Cambistat® dose[3] (mL/cm)	Paclobutrazol dose[4] (g/cm)
Silver maple (D)	Full rate (D)	3	2	4.6	1.1
	1CL (C)	3	3	4.2	1.0
	2CL (B)	3	2	2.3	0.6
	Non-treated	8	8	0.0	0.0
White oak (D)	Full rate (D)	10	4	4.6	1.1
	1CL (C)	10	4	4.2	1.0
	2CL (B)	10	4	2.3	0.6
	Non-treated	10	4	0.0	0.0
Pecan (E)	Full rate (E)	5	4	5.0	1.2
	1CL (D)	5	4	4.6	1.1
	2CL (C)	5	3	4.2	1.0
	Non-treated	9	6	0.0	0.0
Laurel oak (F)	Full rate (F)	9	4	6.7	1.6
	1CL (E)	8	3	5.0	1.2
	2CL (D)	9	4	4.6	1.1
	Non-treated	10	4	0.0	0.0
Stone pine (F)	Full rate (F)	10	3	6.7	1.6
	1CL (E)	10	3	5.0	1.2
	2CL (D)	9	4	4.6	1.1
	Non-treated	9	3	0.0	0.0

[1]Treatment rate based on the species-specific category on the Cambistat® label is indicated as "full rate"; the category below full rate is indicated as "1CL"; 2 categories below full rate is indicated as "2CL"; and non-treated were controls.
[2]Replicates that survived the duration of the experiment were used to quantify all response variables except for average total root length.
[3]Rates were based on stem caliper of plant. The amount of water applied per application was 11 mL of water/mL of Cambistat® for all treatments.
[4]Dosage based on the amount of grams of paclobutrazol administered based on stem caliper of plant.

used to calculate root:shoot dry mass where aboveground dry mass was calculated by summing leaf and stem dry mass.

Statistical Analysis

An analysis of variance (ANOVA) was performed for each response variable and with PBZ treatment as the predictor variable. Models were first constructed separately for each individual species and models with a significant predictor (PBZ dose) effect were subjected to a post-hoc Tukey test to separate means by PBZ dose. Significance of predictor variables was considered at the 0.05 level. Prior to ANOVA construction, data were manually inspected for normality and transformed if required; total root length was the only response variable that required transformation (log transformed). Due to replication issues, average total root length for silver maple was not statistically analyzed, but the results are reported. Lastly, to analyze general biomass allocation trends across species, a restricted maximum likelihood mixed model was constructed using log-transformed predictor variables (to normalize variances) where treatment was a fixed effect and species was a random effect. This was done for each of the biomass response variables (root, shoot, leaf, and root:shoot dry mass). As before, means were

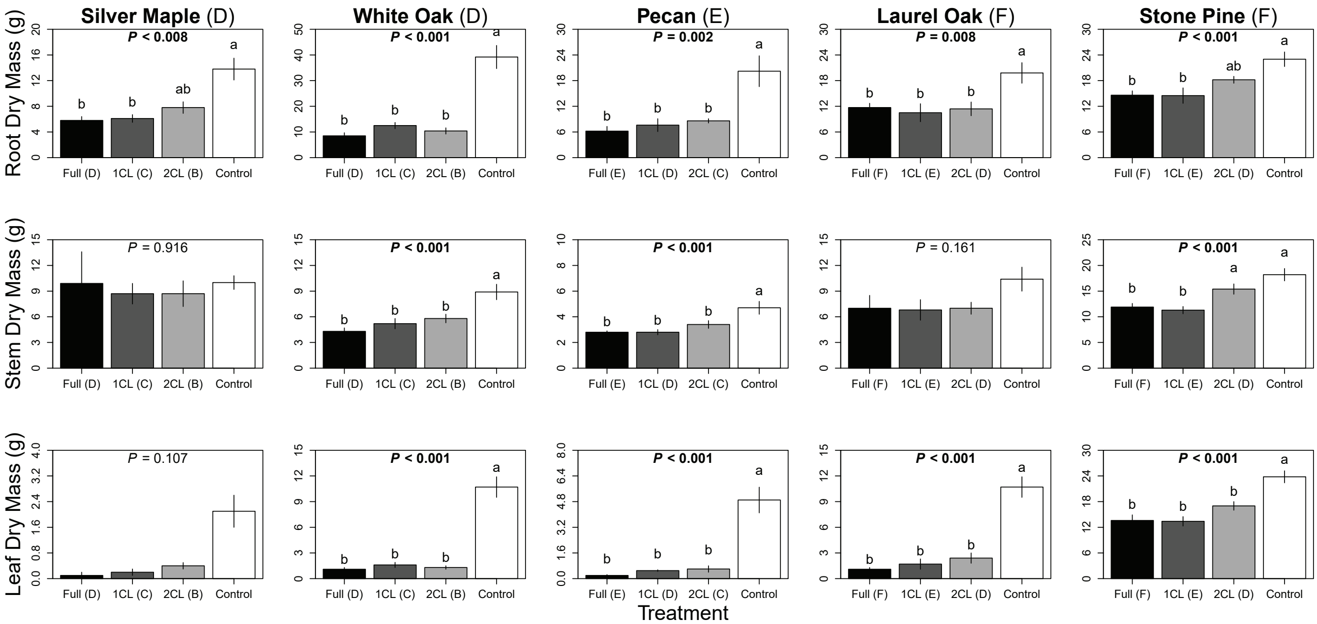

Figure 1. Root, stem, and leaf dray mass (g) associated with different application rates of paclobutrazol (Cambistat®) across 5 species. *P*-values below 0.05 are in bold font and considered significant; different letters indicate significant differences between dose rates. Columns are species and rows are the response variable. Treatment rate based on the species-specific category on the Cambistat® label is indicated as "Full"; the category below full rate is indicated as "1CL"; 2 categories below full rate is indicated as "2CL"; and non-treated were controls. The species-specific category on the Cambistat® label is listed beside the species name as well as beside the "Full" treatment. The dose category is also listed beside 1CL and 2CL.

separated via a Tukey HSD post-hoc calculation when PBZ dose had a significant effect on the response variable. All statistics were performed in JMP16 (SAS Institute Inc. 2021).

RESULTS

There was a significant decrease in leaf dry mass of PBZ-treated white oak, pecan, laurel oak, and stone pine, but there was no dose effect of PBZ for these species (i.e., significant differences between untreated controls and plants treated with PBZ, but no significant differences between PBZ-treated plants)(Figure 1). Stem dry mass was significantly decreased by PBZ treatment in white oak, pecan, and stone pine. There was a dose effect observed for stone pine stem dry mass, where the full and 1CL doses were significantly smaller than the 2CL dose and controls. Stem dry mass was not affected by PBZ treatment in silver maple and laurel oak (Figure 1). All species included in the experiment had significantly reduced root dry mass as a result of PBZ treatment, and a dose effect was observed with silver maple and stone pine where the full and 1CL doses were significantly smaller than the 2CL and controls (Figure 1). Treatment effects were observed on the root:shoot mass for silver maple and laurel oak (Figure 2). There was a significant effect of PBZ treatment for silver maple root:shoot, but only the 2CL treatment was statistically differentiable from untreated controls. Ratios of silver maple treated at the full and 1CL rates were not statistically differentiable from controls or the full and 1CL rates. For laurel oak, PBZ treatment had a significant impact on root:shoot mass. Untreated controls had the lowest ratios while plants treated at the full rate had the highest, and these two treatments were significantly different from one another, but the 1CL and 2CL treatments were not statistically differentiable from untreated controls or plants treated at the full rate (Figure 2). It is also worth mentioning that while only marginally significant ($P > 0.05$), root:shoot mass decreased for white oak.

Treatment with PBZ significantly reduced the length of the longest root for white oak, pecan, laurel oak, and stone pine with a dose effect detected only for stone pine. Though a significant impact of PBZ was detected for pecan, treatment means could not be separated with post-hoc analysis. The length of the longest root for silver maple was not affected by PBZ.

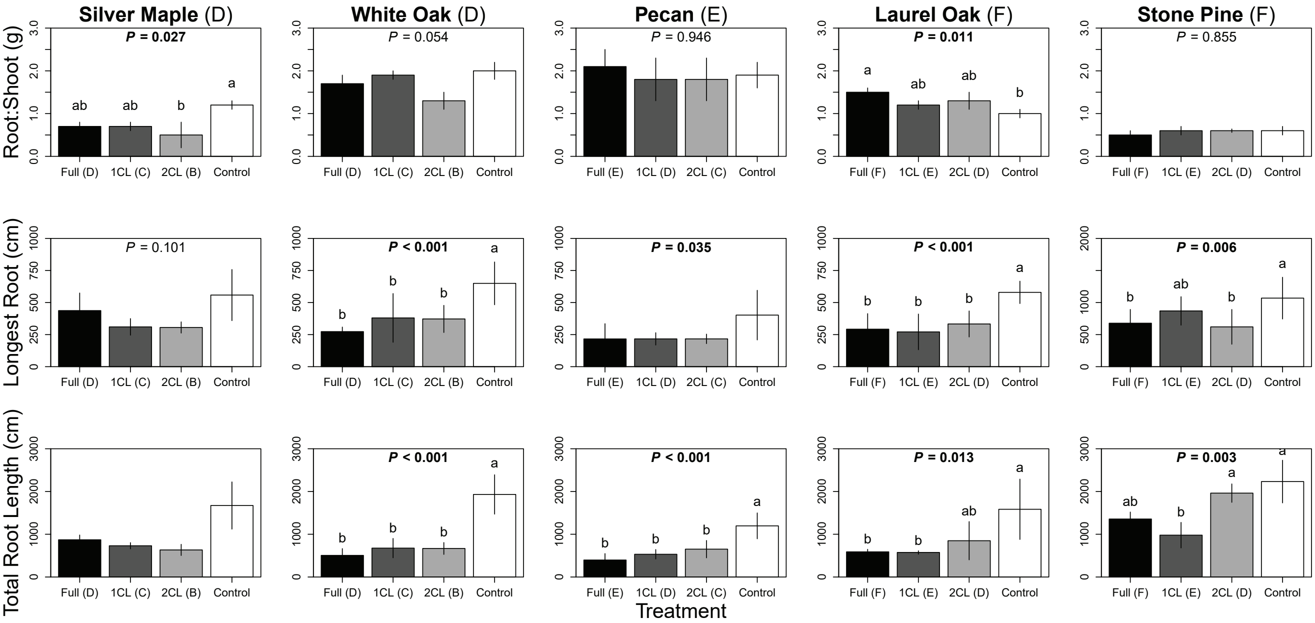

Figure 2. Root:shoot ratios (g), length of longest root (cm), and estimated total root length (cm) associated with different application rates of paclobutrazol (Cambistat®) across 5 species. *P*-values below 0.05 are in bold font and considered significant and different letters indicate significant differences between dose rates. Columns are species and rows are the response variable. Treatment rate based on the species-specific category on the Cambistat® label is indicated as "Full"; the category below full rate is indicated as "1CL"; 2 categories below full rate is indicated as "2CL"; and non-treated were controls. The species-specific category on the Cambistat® label is listed beside the species name as well as beside the "Full" treatment. The dose category is also listed beside 1CL and 2CL. The total root length for silver maple was not statistically analyzed due to low replication.

For each species statistically analyzed, total root length was significantly reduced by PBZ treatment. It is important to note that the average total root length of laurel oak treated at the 2CL rate and stone pine treated at the full and 2CL rates were undifferentiable from their respective controls (Figure 2). Despite our inability to statistically analyze silver maple total root length, we do note that the total root length of PBZ-treated silver maple tended to be reduced (Figure 2).

For general trends across species, PBZ treatment was a significant predictor for each response variable with the exception of root:shoot mass. PBZ was found to reduce root, shoot, and leaf dry mass significantly across all species. Dose effects were only observed for stem and leaf dry mass where increasing doses of PBZ led to decreases in biomass. There was no dose effect observed for root dry mass (Table 2). Treatment was found to have no statistical effect on root:shoot dry mass across species (Table 2).

DISCUSSION

The purpose of this study was to assess general trends of PBZ treatment on plant tissues by including several woody plant species across a known gradient of sensitivity to PBZ, and under uniform conditions expose plants to varying doses of PBZ. Though some variation in aboveground responses to PBZ were detected, a significant reduction in root dry mass was measured for all species and this mostly adhered to our hypotheses. Additionally, most species had reduced longest root lengths and all species that were statistically analyzed showed significantly decreased total root lengths. When considering species individually, root:shoot ratio responses to both PBZ treatment and PBZ dose were variable, and this did not conform to our hypothesis that growth reductions both above- and belowground would result in consistent ratios among treatments within a species. However, when considering all species together, PBZ significantly reduced root, shoot, and leaf dry mass but did not affect root:shoot dry mass, and these results did adhere to our hypotheses. Though dose effects were observed for certain metrics and species, few clear trends were apparent when species were analyzed separately. However, when analyzed together, no dose effect was observed for root dry mass, but a dose effect

Table 2. Growth effects associated with different application rates of paclobutrazol (Cambistat®) with all 5 species included in analysis. Biomass parameters were log transformed to normalize variance prior to running a restricted maximum likelihood mixed model, where treatment was a fixed effect and species was considered a random effect. *P*-values in bold are significant, and means separation letters not connected by the same letter are significantly different according to the Tukey's honestly significant difference (HSD) post-hoc calculation.

Treatment[1]	Root dry mass (g)(± SE)	Stem dry mass (g)(± SE)	Leaf dry mass (g)(± SE)	Root:shoot (g)(± SE)
Full rate	10.3 (0.8)[b]	7.3 (0.7)[c]	4.5 (1.1)[c]	1.3 (0.1)
1 CL	11.5 (0.9)[b]	7.2 (0.6)[bc]	4.7 (1.0)[bc]	1.3 (0.1)
2 CL	12.4 (0.9)[b]	8.4 (0.8)[b]	5.5 (1.2)[b]	1.2 (0.1)
Non-treated	23.6 (1.8)[a]	10.4 (0.8)[a]	10.5 (1.2)[a]	1.4 (0.1)
F-statistic$_{df}$	$44.0_{3,148}$	$17.8_{3, 148}$	$89.8_{3, 148}$	$0.8_{3, 148}$
P-value	***< 0.0001***	***< 0.0001***	***< 0.0001***	*0.7502*

[1]Treatment rate based on the species-specific category on the Cambistat® label is indicated as "full rate"; the category below full rate is indicated as "1CL"; 2 categories below full rate is indicated as "2CL"; and non-treated were controls.

was observed for leaf and shoot dry mass, which mostly abided by our hypotheses.

Literature on the role of gibberellins in root system growth and development, primarily using herbaceous model plants such as *Arabidopsis* spp., is relatively extensive. Gibberellins are synthesized in root meristem (Shani et al. 2013) and bioactive forms of gibberellins, synthesized by processes downstream of *ent*-kaurene oxidase, are synthesized locally along the root tip (Barker et al. 2021). In the proliferation zone of root tips, high gibberellin levels elicit cell division and low levels promote cell differentiation, but in the elongation zone of the root tip gibberellins promote cell expansion (Fonouni-Farde et al. 2016; Barker et al. 2021). Fine adventitious root formation (Davis et al. 1985; Liu et al. 2018) is a commonly reported result of PBZ treatment. Adventitious root formation is a result of decreased bioactive gibberellin levels during the initial stage of adventitious root development followed by gradual increases thereafter (Lei et al. 2018). Reports of the PBZ impacts on fine roots various by species (Watson 1996; Tanis 2015). Though we were careful to avoid the loss of fine roots while harvesting our samples, some loss of fine roots is unavoidable and unpredictable. We did not make any attempt to separate fine roots from the rest of the root system or quantify fine root-specific responses such as turnover or density. However fine roots certainly contributed, to various extents, to the metrics that we quantified. Future research should continue to assess the impacts of PBZ on fine root dynamics (Tanis et al. 2015), as fine roots are critical for resource acquisition.

Another commonly reported phenomenon of PBZ treatment is the alterations to root:shoot biomass ratios (Rieger and Scalabrelli 1990; Tanis et al. 2015; Wang et al. 2019). Those studies that do report a significant impact of PBZ on root:shoot ratios find that PBZ tends to increase this ratio. Our results varied by species and there was no significant effect of PBZ treatment when considering all species together (i.e., construction of a maximum likelihood mixed model). For laurel oak, both the root and shoot dry mass had significantly decreased due to the application of PBZ but root:shoot ratios had increased, and there was an apparent dose effect on this ratio shift. Alternatively, silver maple root:shoot ratios significantly decreased as a result of treatment (Figure 2), and this appeared to be driven by decreases in root mass with no change to shoot mass (Figure 1). Additionally, there appeared to be no relationship between PBZ sensitivity and ratio shifts, though there is precedent for significant dose responses within the literature (Wang et al. 2019). We did not investigate any potential physiological, biochemical, or molecular mechanisms of biomass ratio shifts and therefore are unable to speak to this. However, it is interesting to note that root growth has been shown to require lower tissue levels of gibberellins while shoot growth appears to require high levels (Tanimoto 1994). Perhaps observations of increases in root:shoot ratios could be explained by the inhibition of giberellin production by PBZ that occurs at levels that inhibit aboveground, but not belowground growth.

We detected only a few true dose responses when considering species individually. The root dry mass of silver maple (Figure 1), the root:shoot ratio and average total root length of laurel oak (Figure 2), and the root and shoot dry mass of stone pine (Figure 1) all showed responses that could be somewhat described as dose dependent. For the most part, metrics with significant treatment effects were separated strictly by PBZ treatment, itself. Interestingly however, when considering all species together the aboveground metrics (leaf and shoot dry mass) showed clear dose responses, but root dry mass was only separated strictly by PBZ treatment. This might be explained by the relative difficulty in quantifying above- and belowground metrics such as biomass where detailed measurements that could show dose responses are simply difficult, leading to substantial levels of variation (Vanninen and Mäkelä 1999; Tanis et al. 2015). This issue is likely also to be the case even with potted specimens. Our experimental setup is certainly limiting with regards to the extrapolation of our results to established and/or mature trees growing in landscapes with substantial variation in soil and other environmental conditions.

CONCLUSION

Paclobutrazol is a product used in the arboriculture industry to reduce the aboveground growth of woody plants, but research has shown a substantial amount of variation of the impact of PBZ on belowground tissue growth. In order to assess general trends, we subjected 5 species of tree across a gradient of PBZ susceptibility (silver maple, white oak, pecan, laurel oak, and stone pine) to various doses of PBZ under controlled conditions and measured leaf, shoot, and root system dry mass, root:shoot ratios, the length of the longest root, and estimated total root area. Our results are largely consistent with research that has found negative impacts of PBZ on woody plant root growth and consistent with the literature addressing gibberellin and root growth dynamics in model herbaceous plants. We found generally that PBZ treatment, regardless of dose, reduced root size metrics (dry mass, length of longest root, total root length). Root:shoot ratios did not change as a result of PBZ treatment for 3 out of the 5 species, was reduced for silver maple, and increased for laurel oak. When species were all analyzed together, root, leaf, and shoot dry mass all significantly decreased with PBZ treatment while root:shoot ratio was not affected. This study offers uniform and controlled conditions for assessing trends in the effects of PBZ on root systems among several species with variation in PBZ sensitivity. Future research should focus on the plant (e.g., genetic, biochemical, physiological, life stage) and environmental factors (e.g., soil qualities, sun exposure) that contribute to the variation in root responses to PBZ observed in the literature.

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Chad M. Rigsby (corresponding author)
Bartlett Tree Research Laboratories
13768 Hamilton Road
Charlotte, NC, USA
Center for Tree Science
The Morton Arboretum
4100 IL-53
Lisle, IL, USA
crigsby@bartlett.com

E. Thomas Smiley
Bartlett Tree Research Laboratories
13768 Hamilton Road
Charlotte, NC, USA

Sean Henry
Bartlett Tree Research Laboratories
13768 Hamilton Road
Charlotte, NC, USA

Liza Holmes
Bartlett Tree Research Laboratories
13768 Hamilton Road
Charlotte, NC, USA

Andrew L. Loyd
Bartlett Tree Research Laboratories
13768 Hamilton Road
Charlotte, NC, USA
Botanic Research Institute of Texas
1700 University Drive
Fort Worth, TX, USA

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