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Muhammad Haroon U Rashid
Qingjun Xie
Xiangqing Ma
Zeyan Wu
Taimoor Hassan Farooq
Irfan Ahmad
Pengfei Wu
fjwupengfei@126.com

Fujian Agriculture and Forestry University https://orcid.org/0000-0001-7426-2335

Research Article

Keywords: Cunninghamia lanceolata, phosphorus deficiency, root classes, phosphorus acquisition, antioxidant defense.

Posted Date: October 29th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-11271459/v1

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Root Class-Specific Phosphorus Dynamics and Antioxidant Defense Responses in Contrasting Chinese Fir Clones Under Phosphorus Deficiency

Muhammad Haroon U Rashid[1,2,3], Qingjun Xie[2], Xiangqing Ma[2], Zeyan Wu[1], Taimoor Hassan Farooq[4], Irfan Ahmad[3], Pengfei Wu[2]*

[1]College of Life Science, Fujian Agriculture and Forestry University, Fuzhou, 350002, China
[2]Chinese Fir Engineering Technology Research Center of State Forestry Administration, College of Forestry, Fujian Agriculture and Forestry University, Fuzhou, 350002, China
[3]Department of Forestry and Range Management, University of Agriculture, Faisalabad, 38000, Pakistan
[4]Bangor College China, a Joint Unit of Bangor University and Central South University of Forestry and Technology, Changsha, Hunan, China

***Corresponding Author:** Pengfei Wu (fjwupengfei@126.com)

Abstract

Aims

Phosphorus (P) deficiency is a serious constraint on the productivity of forest plantations, especially in Chinese fir (*Cunninghamia lanceolata*). This study aims to determine how P availability affects root class-specific P acquisition, antioxidant responses, and physiological adaptations in two contrasting Chinese fir clones (020 and 061) across three harvesting periods.

Methods

Seedlings were grown in P-deficient (P_0) and P-replete (P_1) conditions, and P dynamics and antioxidant responses were determined across root orders, shoots, and needles.

Results

Generally, P concentration and accumulation increased with harvesting time and were significantly higher in P-replete conditions, with clone-061 being the better P acquirer than clone-020. P concentration and accumulation differed significantly among root classes, indicating functional specialization in P acquisition and distribution. P uptake and translocation were highest during early harvestings, while P use efficiency was higher at later growth stages. Under P deficiency, the activities of antioxidant enzymes, such as superoxide dismutase, peroxidase, and catalase, increased with harvesting time, especially in clone-061, and the malondialdehyde content decreased with harvesting time and remained comparatively lower in clone-061, indicating reduced oxidative damage. According to principal component analysis, both P acquisition and antioxidant enzyme activity were positively associated with antioxidant responses and negatively associated with malondialdehyde in a distinct, root-class-specific manner.

Conclusions

Overall, clone-061 exhibited superior P acquisition, utilization efficiency, and antioxidant capacity, highlighting its greater adaptation to phosphorus-limited environments. These findings reveal root-specific P-use strategies and support selecting P-efficient Chinese fir genotypes for sustainable plantation management.

Keywords: *Cunninghamia lanceolata*; phosphorus deficiency; root classes; phosphorus acquisition; antioxidant defense.

Introduction

Phosphorus (P) is an important macronutrient for plant growth, metabolism, and physiological functioning, but it can be limiting to productivity in terrestrial ecosystems. P deficiency is especially common in the highly weathered, acidic red soils of southern China, as soluble P can readily form insoluble complexes with aluminum (Al^{3+}), iron (Fe^{3+}), and calcium (Ca^{2+}) ions, significantly reducing its mobility and bioavailability (Wu et al. 2017). P depletion has increased over the past decades due to nutrient removal from continuous harvesting of short-rotation forests, especially Chinese fir plantations. Plants have evolved a series of adaptive strategies to deal with P-deficient environments, which include changes in root system architecture, stimulation of root proliferation and elongation, more organic acids and phosphatases released from roots, and changes in rhizosphere processes to increase the efficiency of P acquisition and use (Wu et al. 2011; Rashid et al. 2023).

Fine roots (non-woody roots < 2 mm in diameter) are a critical component of plant adaptation to changing environmental conditions and are highly sensitive to variations in forest ecosystems. These roots play a fundamental role in water and nutrient acquisition, thereby contributing significantly to plant growth, productivity, and survival (Fantozzi et al. 2024). Fine root development and functional activity are affected by genetic and environmental factors such as nutrient availability, soil physicochemical properties, temperature, and precipitation patterns. The fine-root system is highly structurally and functionally heterogeneous and is frequently divided in terms of branching order. Distal lower-order roots (e.g., first-order) are generally thinner, have higher specific root length and shorter lifespan, which is a sign of their higher metabolic activity and efficiency in nutrient acquisition. By contrast, higher-order roots tend to be larger in diameter, more structurally advanced, and are primarily associated with transport, storage, and mechanical support (Staszel et al. 2022; Rashid et al. 2023). The functional differentiation among root classes is therefore important for understanding plant strategies for resource acquisition and adaptation in a changing environment (Wang et al. 2021; Li et al. 2022).

Root order is an important functional trait that defines the physiological functions and resource acquisition strategies of fine-root systems. Each root class has specific anatomical, physiological, and morphological traits that differ in their ability to absorb nutrients and in metabolic activity. Lower-order roots tend to have a higher nutrient uptake capacity due to their higher metabolic activity, respiration rates, and surface area-to-biomass ratio (Freschet et al. 2021). Higher-order roots, by contrast, serve as transport and storage organs that move absorbed nutrients and water to above-ground tissues.

Under nutrient-limited conditions, plants can modify root-order-specific traits to optimize resource acquisition and maintain growth (Gao et al. 2025). Therefore, investigating nutrient dynamics across different root classes provides valuable insights into the mechanisms underlying plant adaptation to environmental stresses, particularly phosphorus deficiency, in which efficient root exploration and nutrient utilization are critical for maintaining productivity.

Cunninghamia lanceolata is one of the most valuable and important subtropical conifer species in China and has been cultivated for wood production for a long time (Farooq et al. 2019; Wang et al. 2024). Chinese fir is regarded as an important plantation species in China because of its good wood qualities and suitability for plantation cultivation, and its planted area accounts for nearly 30% of China's plantation forests, totaling about 11 million hectares (FAO 2015). Despite the economic importance of Chinese fir, the physiological mechanisms underlying its adaptation to P-limited

environments remain poorly explored, particularly with respect to root class-specific P acquisition, utilization, and antioxidant defense.

Although numerous studies have investigated plant responses to P deficiency at whole-root and above-ground levels, the physiological mechanisms underlying functional differentiation among fine-root classes remain poorly understood. Fine roots exhibit substantial structural and functional heterogeneity, with different root orders contributing differently to nutrient acquisition, transport, and storage; however, how P availability influences root class-specific P acquisition, utilization efficiency, and stress responses remains unclear. In particular, the interactive roles of P uptake, P-use efficiency, and antioxidant defense systems among contrasting root classes under P deficiency have received limited attention. Since P deficiency restricts nutrient acquisition and induces oxidative stress, understanding root-specific physiological and biochemical responses is essential for elucidating plant adaptive strategies under nutrient-limited conditions.

Therefore, the present study aimed to investigate the effects of P availability on P dynamics, antioxidant defense mechanisms, and physiological adaptation among different root classes in contrasting Chinese fir clones. Specifically, this study was designed to: (1) evaluate P concentration, accumulation, translocation, uptake efficiency, and use efficiency in different root classes, stems, and leaves under contrasting P supply levels; (2) investigate antioxidant defense responses, including superoxide dismutase (SOD), peroxidase (POD), catalase (CAT), and malondialdehyde (MDA) accumulation across different root classes and other plant organs; (3) determine the relationships between P acquisition, P-use efficiency, antioxidant defense mechanisms, and functional differentiation among different root classes under P stress; and (4) identify the clone exhibiting superior P-use efficiency and adaptive responses to P deficiency. We hypothesized that different root classes exhibit distinct P-acquisition strategies and antioxidant defense responses, which, together, enhance the adaptation of Chinese fir clones to P-limited environments.

Materials and Methods

Plant material and growth conditions

Two contrasting Chinese fir (*Cunninghamia lanceolata*) clones (020 and 061) were selected based on differential growth performance under P deficiency reported by Wu et al. (2017). Healthy 1-year-old seedlings were prepared by tissue culture and obtained from Yangkou State-owned Forest Farm, Fujian, China. Seedlings of both clones were similar in growth characteristics prior to treatment, with average heights of 20.1 ± 0.12 cm and 22.92 ± 0.16 cm for clones 020 and 061, respectively. Similarly, the mean root collar diameter measured 3.1 ± 0.08 mm in clone-020 and 3.5 ± 0.06 mm in clone-061. Healthy, disease-free, morphologically uniform seedlings were selected to ensure experimental consistency and minimize variability among treatments.

Study design

The experiment was conducted in a greenhouse at the Forest and Environment Research Institute, Fujian Agriculture and Forestry University, China. One-year-old Chinese fir seedlings were transplanted into polyethylene pots (32 cm in height; 18 cm at the upper and 9 cm at the lower diameter) filled with thoroughly washed sand. Two contrasting Chinese fir clones (020 and 061) were subjected to two P supply levels: P-deficient (P_0, no P addition) and P-sufficient (P_1, 1.0 mmol L^{-1} KH_2PO_4). Each treatment consisted of 24 replicates, comprising 48 pots per clone and 96 pots overall. Seedlings were harvested at 40, 80, and 120 days after treatment initiation.

To supply essential nutrients other than P, a modified Hoagland nutrient solution containing one-third of the standard concentration was applied following Wu et al. (2011). The solution contained KNO_3 (5.0 mmol L^{-1}), $MgSO_4 \cdot 7H_2O$ (2.0 mmol L^{-1}), H_3BO_3 (46.3 μmol L^{-1}), $CuSO_4 \cdot 5H_2O$ (0.3 μmol L^{-1}), $ZnSO_4 \cdot 7H_2O$ (0.8 μmol L^{-1}), $MnSO_4 \cdot H_2O$ (9.1 μmol L^{-1}), and $H_2MoO_4 \cdot 4H_2O$ (0.4 μmol L^{-1}). Each pot received 110 mL of nutrient solution every 10 days and 200 mL of distilled water daily. Potassium concentration was balanced across treatments using KCl, while nutrient solution pH was maintained at 5.8 using NaOH or diluted HCl. Greenhouse environments were maintained at approximately 18-28°C under a 14 h light/10 h dark photoperiod, with relative humidity above 80%.

Determination of P concentration in plant organs

Fresh biomass of different root classes, shoots, and leaves were measured at each harvesting stage. Afterward, the plant samples were oven-dried at 80°C until constant weight, ground, and sieved through a 1 mm mesh. Dried samples were acid-digested (sulfuric acid, H_2SO_4, and perchloric acid, $HClO_4$, at 10:1 (v/v)) to determine P. About 10 mL of digestion solution was added to each sample, and digestion was carried out under controlled heating in the fume hood until the solutions were clear. The samples were digested and then placed in 50 mL volumetric flasks and filtered through filter paper into plastic containers. The levels of P in different root classes, stem, and needles were then determined by ICP-OES (Optima 8000, PerkinElmer, USA). The results were reported on a dry weight basis.

P accumulation and distribution

P accumulation in different root classes, stem, and needle tissues was calculated by multiplying P concentration (mg g^{-1}) by the dry biomass of each organ. To assess nutrient allocation and nutrient distribution patterns under contrasting P supply levels, P accumulation was quantified separately for each organ. Total plant P was calculated as the sum of all plant tissues and presented as mg $plant^{-1}$.

P uptake, translocation and use-efficiency

P uptake efficiency (PUpE), P translocation efficiency (PTE), and P use-efficiency (PUE) were calculated using the following equations:

$$P\ uptake\ efficiency\ (g \cdot kg^{-1}) = \frac{Root\ P}{Root\ dry\ mass}$$

$$P\ translocation\ efficiency\ (\%) = \left(\frac{Shoot\ P}{Total\ P}\right) \times 100$$

$$P\ use - efficiency\ (g \cdot g^{-1}) = \frac{Shoot\ dry\ mass(g)}{Shoot\ P\ (g)}$$

Determination of antioxidant enzyme activity and lipid peroxidation

Antioxidant enzyme activities, including superoxide dismutase (SOD), peroxidase (POD), and catalase (CAT), and malondialdehyde (MDA) content were determined in different root classes, shoots, and needles at each harvesting stage. Fresh tissue samples (0.2 g) were homogenized in liquid nitrogen, extracted with phosphate-buffered saline (PBS), and centrifuged at 10,000 rpm for 20 min at 4°C. The supernatant was used for enzymatic analysis. SOD, POD, CAT, and MDA were quantified spectrophotometrically at 560, 470, 240, and 450/532/600 nm, respectively, using an Infinite 200 Pro NanoQuant analyzer (Tecan, Switzerland) following standard protocols.

Statistical analysis

Three-factor factorial ANOVA was performed to assess the effects of clone, stress, and treatment time on the traits under study. Results were statistically analyzed using Tukey's test at a $P < 0.05$ level of significance. All statistical analyses were conducted using SPSS Statistical Package (SPSS 19.0, SPSS Inc., IL, USA). Principal component analysis was conducted to examine the relationships among P acquisition, P-use efficiency, and antioxidant defense across different root classes.

Results

Phosphorus (P) concentration

The interaction among clone, P treatment, root class, and harvesting time significantly affected P concentration in roots, shoots, and needles ($P < 0.05$). As shown in Figure 1, root P concentration varied significantly among root classes and generally increased with harvesting time under P-replete (P_1) conditions. The highest P concentration was recorded in 2C roots of clone 061-P_1 at the 2nd harvest (3.24 ± 0.58 mg plant^{-1}), followed by 1C (2.36 ± 0.07 mg plant^{-1}) and 3C roots (2.08 ± 0.25 mg plant^{-1}) at the 3rd harvest. The lowest values were observed under P deficiency (P_0), particularly in 3C roots of clone 061 at the 1st harvest (0.18 ± 0.05 mg plant^{-1}). Overall, 2C roots exhibited the highest P concentration, followed by 1C and 3C roots, with P supplementation consistently enhancing P concentration across all root classes.

Shoot P concentration increased with harvesting time and was higher under P_1 than P_0, reaching a maximum in 061-P_1 at the 2nd harvest (5.16 ± 0.36 mg plant^{-1}) and a minimum in 061-P_0 at the 1st harvest (2.96 ± 0.21 mg plant^{-1}). Needle P concentration was highest in 020-P_1 at the 1st harvest (6.80 ± 0.73 mg plant^{-1}) and lowest in 061-P_0 at the 2nd harvest (3.23 ± 0.61 mg plant^{-1}), indicating that P supplementation consistently increased P concentration in above-ground tissues.

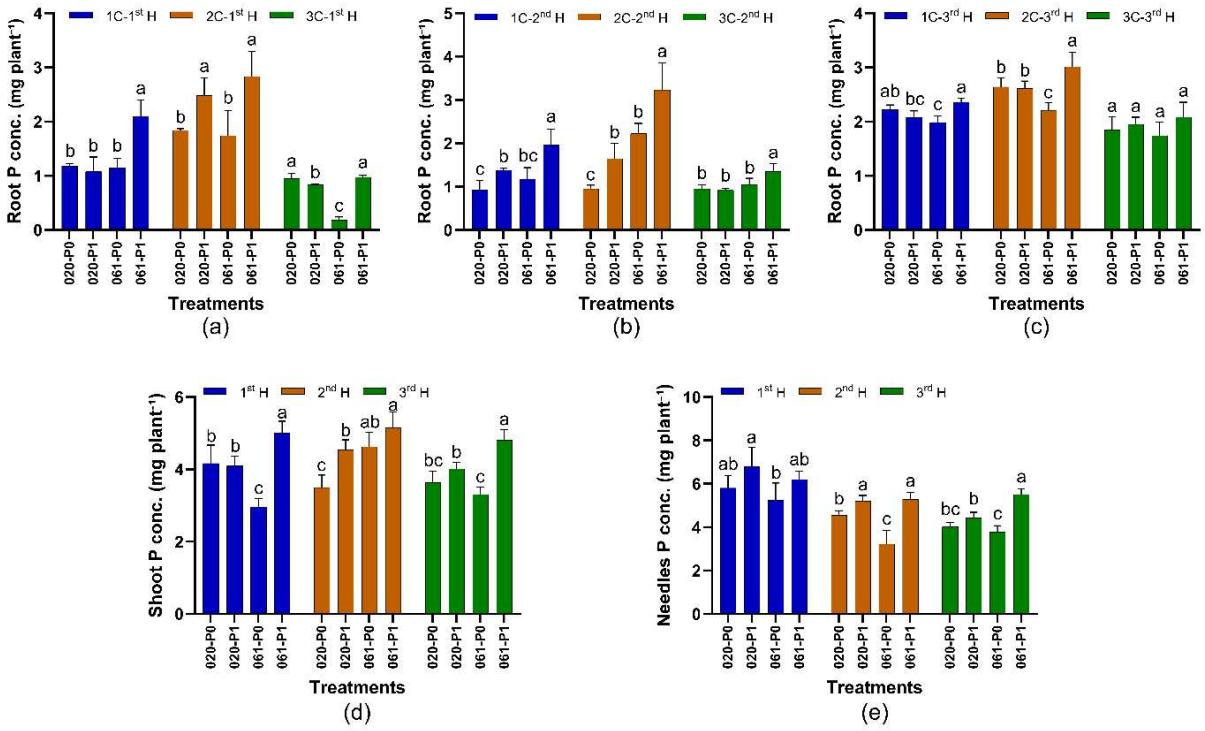


Figure 1. 1C, 2C, 3C roots, shoot, and needles P concentration of two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Phosphorus (P) accumulation

Figure 2 shows that P accumulation in roots, shoots, and needles generally increased with harvesting time and was enhanced under P-replete conditions. Among root classes, 2C roots exhibited the highest P accumulation, reaching a maximum in 061-P_1 at the 3rd harvest (1.40 ± 0.13 mg plant^{-1}), followed by 3C roots (1.39 ± 0.11 mg plant^{-1}) and 1C roots (0.53 ± 0.07 mg plant^{-1}). The lowest root P accumulation was observed under P deficiency during the early harvesting period, particularly in 061-P_0. Similarly, shoot and needle P accumulation increased over time, with maximum values recorded in 061-P_1 at the 3rd harvest (6.59 ± 0.93 and 13.80 ± 1.51 mg plant^{-1}, respectively). Overall, clone-061 showed greater P accumulation capacity than clone-020, especially under P-replete conditions, indicating superior P acquisition and utilization.

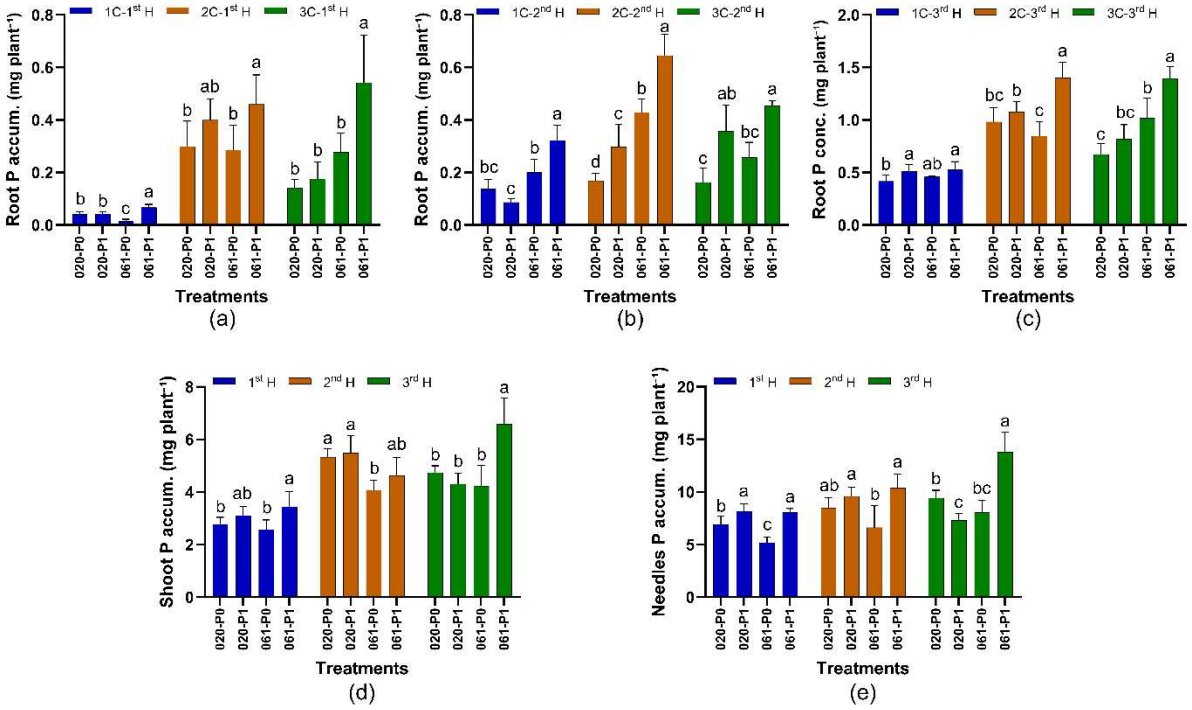


Figure 2. 1C, 2C, 3C roots, shoot, and needles P accumulation of two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Phosphorus (P) uptake, translocation, and use efficiency

The interactions among clone, P treatment, and harvesting time significantly affected P uptake, translocation, and use efficiency ($P < 0.05$). P uptake and translocation were generally higher during the 1st and 2nd harvestings and declined thereafter, whereas P use-efficiency increased with harvesting time. The highest P uptake (22.63 ± 3.40 g kg^{-1}) was recorded in clone-061 under P-replete conditions at the 1st harvesting, while maximum P translocation (38.38 ± 2.32%) occurred in clone-020 under P-deficient conditions at the 2nd harvesting. In contrast, the greatest P use-efficiency was

exhibited in clone-061 under P-deficiency at the 3rd harvesting (1.29 ± 0.06 g kg^{-1}). Overall, clone-061 exhibited greater P acquisition and utilization efficiency than clone-020 (Figure 3).

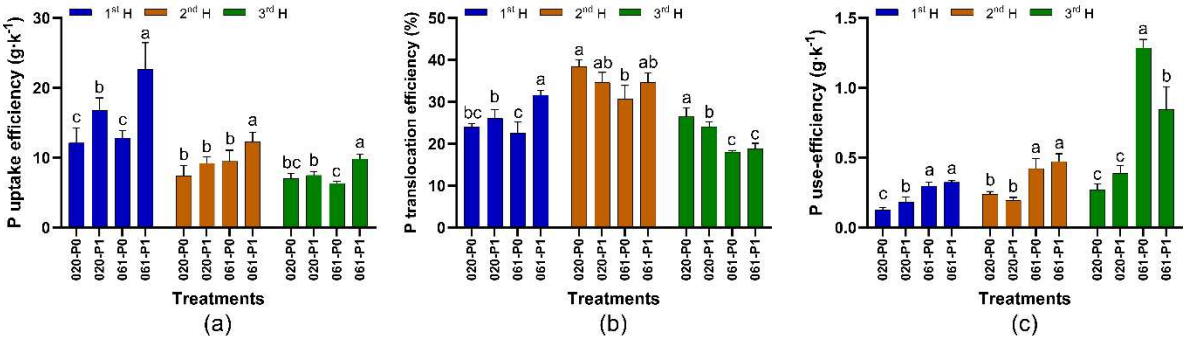


Figure 3. P uptake efficiency, P translocation efficiency, and P use efficiency of two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Antioxidant enzyme activities and lipid peroxidation

Superoxide dismutase (SOD) activity

As shown in Figure 4, SOD activity in different root classes generally increased during later harvesting periods and was enhanced under P-deficient conditions, particularly in clone-061. Among root classes, the highest SOD activity was observed in 1C roots of 061-P_0 at the 3rd harvest (479.4 ± 21.1 mg g^{-1} FW), followed by 2C roots (370.9 ± 28.5 mg g^{-1} FW) and 3C roots (359.2 ± 36.2 mg g^{-1} FW). In contrast, the lowest SOD activity was observed under P-replete conditions in clone-020. Similar trends were observed in shoots and needles, where maximum SOD activities were recorded in clone-061 at the 3rd harvest in shoots (376.8 ± 30.5 mg g^{-1} FW) and needles (426.0 ± 18.0 mg g^{-1} FW). Overall, clone-061 exhibited stronger SOD-mediated antioxidant defense than clone-020, especially under P deficiency and prolonged stress conditions.

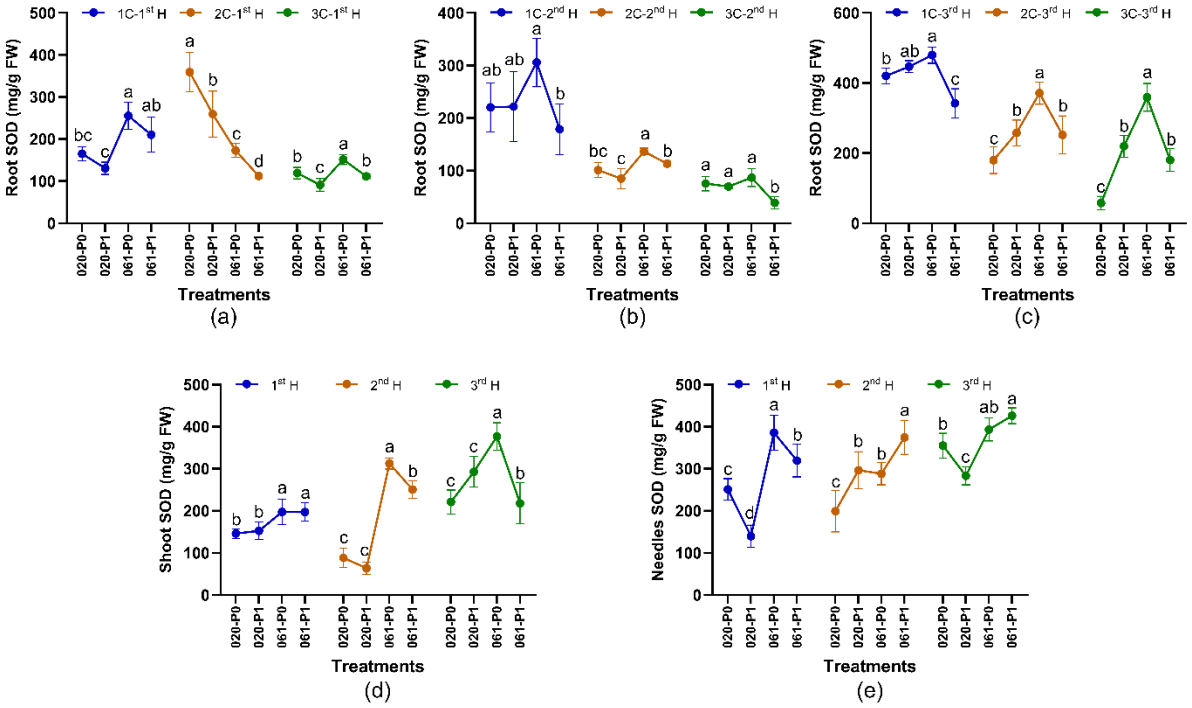


Figure 4. 1C, 2C, 3C roots, shoot and needles SOD activity in two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Peroxidase (POD) activity

POD activity in different root classes generally increased during later harvesting periods, with distinct responses among root classes. The highest POD activity was recorded in 1C roots of clone 061-P_0 at the 3rd harvest (21.80 ± 1.15 mg g^{-1} FW), whereas 2C roots showed maximum POD activity in 061-P_1 at the 3rd harvest (8.88 ± 1.08 mg g^{-1} FW). In contrast, the highest POD activity in 3C roots was observed in 020-P_0 at the 3rd harvest (16.33 ± 1.11 mg g^{-1} FW). Shoot and needles POD activities also increased at later harvests, reaching maximum values in 061-P_1 shoots (26.42 ± 1.22 mg g^{-1} FW) and 061-P_0 needles (244.16 ± 7.34 mg g^{-1} FW) at the 3rd harvest. Overall, POD activity showed root class-dependent variation, with clone-061 generally exhibiting stronger antioxidant responses under P stress (Figure 5).

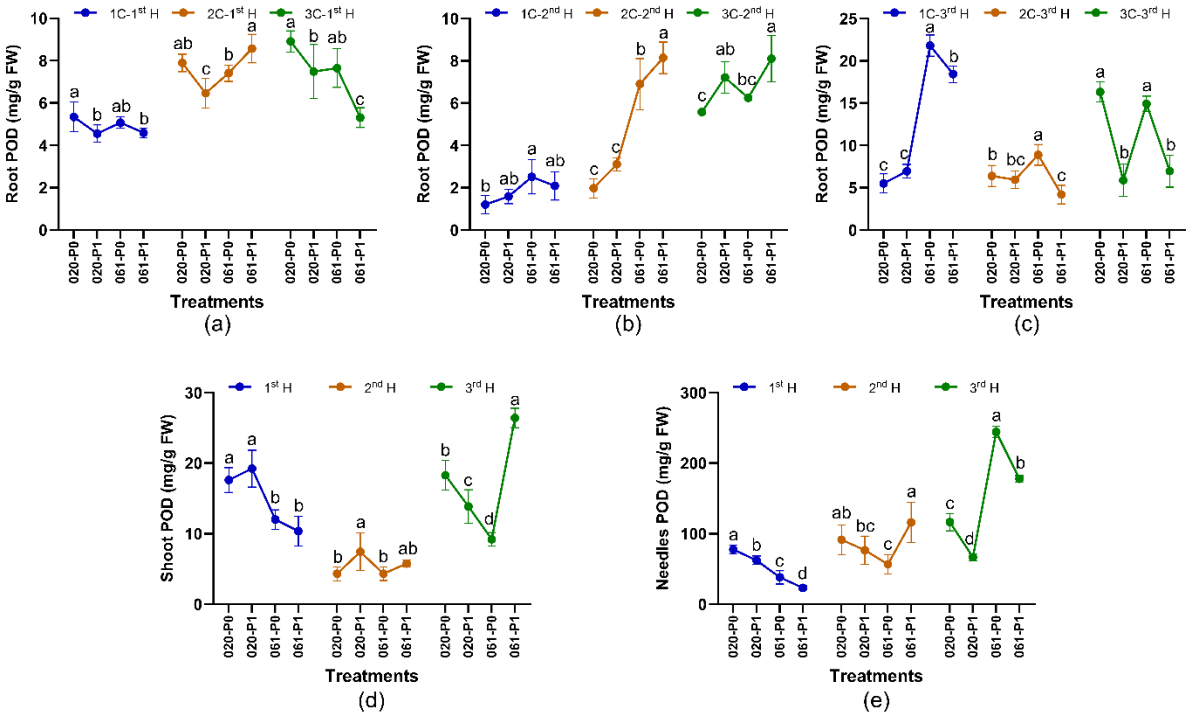


Figure 5. 1C, 2C, 3C roots, shoots, and needles POD activity in two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Catalase (CAT) activity

As shown in Figure 6, CAT activity generally increased with harvesting time and showed distinct variation among root classes. The highest CAT activity was observed in 3C roots of clone-020 under P-deficient conditions at the 3rd harvest (90.24 ± 3.27 mg g^{-1} FW), followed by 2C roots (70.12 ± 2.76 mg g^{-1} FW) and 1C roots (58.86 ± 3.74 mg g^{-1} FW). In above-ground tissues, maximum CAT activities were recorded in shoots (85.63 ± 4.25 mg g^{-1} FW) and needles (9.46 ± 1.10 mg g^{-1} FW) of clone-061 at the 3rd harvest under P-replete conditions. Overall, CAT activity increased during later growth stages, with root and above-ground tissues exhibiting clone- and P treatment-dependent antioxidant responses.

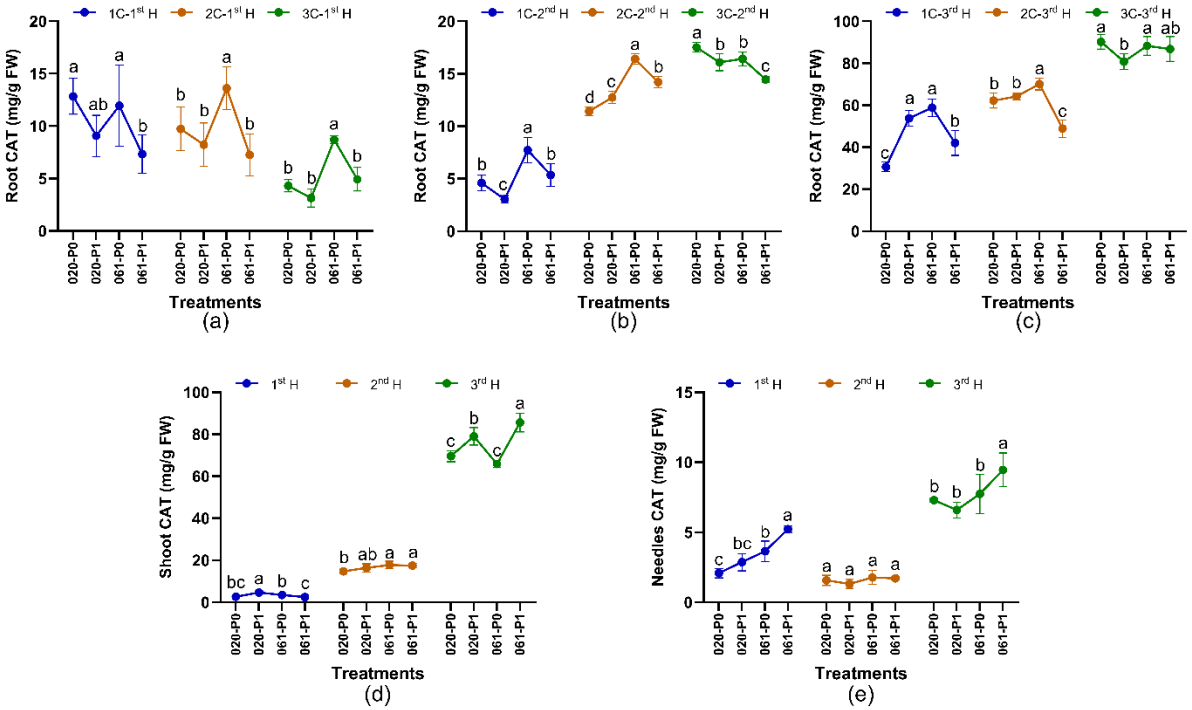


Figure 6. 1C, 2C, 3C roots, shoot, and needles CAT activity in two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Malondialdehyde (MDA) content

MDA levels across different root classes generally declined with harvesting time, indicating reduced lipid peroxidation during plant development. The highest MDA accumulation was recorded in 1C roots (2.79 ± 0.23 mg g^{-1} FW), followed by 3C roots (2.71 ± 0.14 mg g^{-1} FW) of clone-020-P_0 at the 1st harvest, whereas 2C roots showed maximum under P deficiency at the 2nd harvest (1.94 ± 0.24 mg g^{-1} FW). In contrast, clone 061 exhibited lower MDA accumulation, particularly during later harvest periods. Similar declining patterns were observed in shoots and needles, with the lowest MDA levels detected at later harvests. Overall, clone-061 maintained lower MDA accumulation than clone-020, suggesting reduced oxidative damage and enhanced membrane stability under P stress (Figure 7).

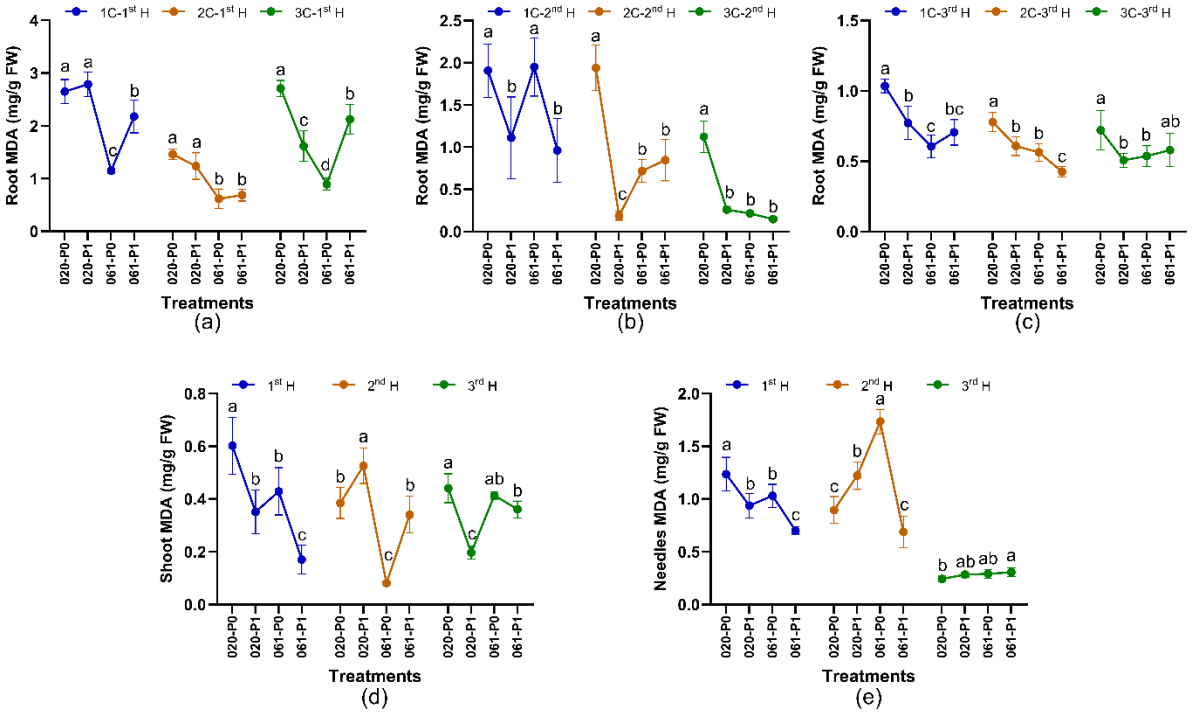


Figure 7. 1C, 2C, 3C roots, shoots, and needles MDA activity in two different Chinese fir seedlings under P-deficient (P_0) and P-replete (P_1) supply conditions.

Principal component analysis (PCA)

Principal component analysis (PCA) was performed to evaluate the relationships among root class-specific phosphorus (P) traits and antioxidant responses (Figure 8). The PCA of P concentration and accumulation revealed distinct clustering of 1C, 2C, and 3C roots, suggesting that each root class contributed differently to P acquisition and allocation across harvesting periods. Similarly, the PCA of P acquisition and use-efficiency traits demonstrated close relationships among P uptake, P translocation, and P use efficiency, reflecting coordinated P utilization during plant development. The PCA of antioxidant enzymes further showed root class-dependent variation, with SOD, POD, and CAT displaying positive associations, while MDA was negatively associated with these antioxidant traits. Overall, the PCA indicates that different root classes contribute differently to P acquisition and antioxidant defense, highlighting complementary adaptive strategies under P-deficient conditions.

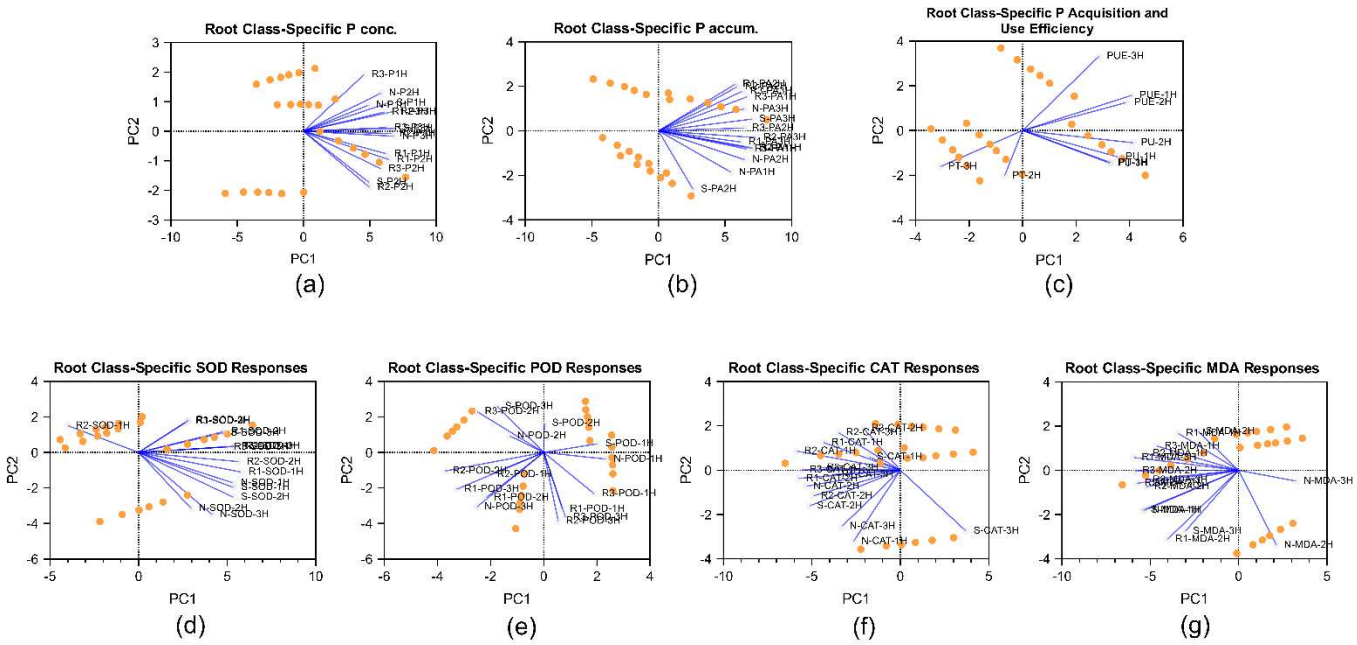


Figure 8. Principal component analysis (PCA) showing the relationships among root class-specific (a) phosphorus concentration, (b) phosphorus accumulation, (c) phosphorus acquisition and use-efficiency traits, (d) superoxide dismutase (SOD), (e) peroxidase (POD), (f) catalase (CAT), and (g) malondialdehyde (MDA) responses across different harvesting periods.

Discussion

The availability of phosphorus (P) had a significant effect on P concentration, accumulation, uptake, translocation, and P use efficiency in both Chinese fir clones, and the responses differed between the three root classes. Higher P concentrations and accumulation were consistently found in the 2C roots, followed by the 1C and 3C roots, indicating that functional differences in P acquisition and allocation occurred in the fine-root system. However, lower-order roots are generally regarded as the main functional roots for nutrient uptake due to their high metabolic activity and absorptive surface area, while the higher-order roots are primarily involved in nutrient transport and storage (Beidler and Pritchard 2017; El Amrani 2023). The higher P concentration and accumulation in 2C roots found in the current study, however, suggest that intermediate root classes are also important for P uptake and redistribution, especially during active growth. Furthermore, the constantly elevated P concentration and accumulation in clone-061 under P-replete conditions indicated a higher capacity for P acquisition and internal allocation than clone-020, consistent with previous results that efficient P acquisition is associated with increased nutrient transport and biomass production in woody species.

P accumulation tended to increase with harvesting time, suggesting continual acquisition and redistribution of P during plant growth. The increasing accumulation of P in the shoots and needles of later harvests also indicates efficient translocation of the absorbed P from the root to the above-ground tissues for photosynthesis and growth. A similar trend has been observed in forest trees, where higher P allocation to the photosynthetically active organs results in higher NUE and biomass production under different P levels (Shen et al. 2011; Wu et al. 2020). In contrast, P uptake was maximized in the early harvests, while P use efficiency was improved at the later harvests. The temporal change

may be due to the switch from P uptake from the outside to the more efficient use of the P already present in the plant and recycled by the plant during prolonged P-limiting growth. Plants under long-term P deficiency have been reported to show similar adaptive responses, allowing them to sustain growth in the face of limited P availability due to improved efficiency of P use (Veneklaas et al. 2012; Han et al. 2022). Moreover, the superior P use efficiency noted for clone-061 is further evidence of the greater physiological plasticity and adaptation to P-deficient conditions.

It has been reported that P deficiency disturbs cellular metabolism by causing excessive production of Reactive Oxygen Species (ROS), which leads to oxidative stress and damage to membranes (Tewari et al. 2021). Also, in the present study, the antioxidant responses were clearly different among root classes, suggesting a functional differentiation in oxidative stress tolerance. SOD, POD, and CAT activities were generally enhanced in later harvesting periods, and especially in P-deficient plants, which could be related to the induction of enzymatic defense responses to the extended stress situation. These antioxidant activities are higher for various root classes, indicating that fine roots are actively involved in cellular redox balance under P deficiency. Superoxide radicals are converted to hydrogen peroxide by SOD, the first line of defense against oxidative stress, which is then further detoxified by extra POD and CAT. Enhanced antioxidant enzyme activities with P deficiency have been reported in numerous woody and agricultural species and are thought to be critical in the protection of cellular structures and in preserving physiological functioning under nutrient stress (Gill and Tuteja 2010; Bano et al. 2021).

CAT activity had significant increases in the later harvesting period, suggesting that the plant had a greater ability to remove the hydrogen peroxide produced under long-term P deficiency. Similarly, increased POD activity helps maintain the integrity of the membrane by detoxifying the ROS and inhibiting oxidative damage (Sharma et al. 2022). The SOD, POD, and CAT activities across the different root classes showed variability; however, the activities of these enzymes were consistently higher in clone-061 than in clone-020, indicating that clone-061 had a more efficient antioxidant defense system to protect against oxidative stress induced by phosphorus deficiency.

On the other hand, the MDA levels slowly decreased as harvesting time increased and were always lower in clone-061 than in clone-020. MDA is a well-known marker of membrane lipid peroxidation, and its decrease reflects less oxidative damage and membrane stability under P stress (Gill and Tuteja 2010; Hasanuzzaman et al. 2020). MDA levels in the later harvesting periods were lower, suggesting that the increased antioxidant defense was able to counter oxidative damage during plant growth. In plants under nutrient deficiency, antioxidant enzyme activity and MDA accumulation have also shown an inverse correlation, suggesting that high antioxidant enzyme activity could reduce MDA levels, thereby decreasing membrane damage and enhancing stress resistance (Sharma et al. 2021). All these results together indicate that clone-061 has a higher antioxidant potential and physiological resilience than clone-020 under P-limiting conditions.

The physiological differentiation among the root classes was further confirmed by distinct associations of traits related to phosphorus and antioxidant responses across harvest dates in principal component analysis. The antioxidant enzyme activities showed a close association with root class-specific P concentration, accumulation, uptake, and P use-efficiency, while MDA showed an opposite relationship. The results indicated that functional segregation of root classes is involved in efficient P acquisition and utilization and, concurrently, helps increase antioxidant defense and

decrease oxidative damage in response to P deficiency. Coordinated physiological responses may be responsible for the greater adaptation of clone-061 in a P-limited environment.

Conclusions

The availability of phosphorus (P) had a strong effect on both root class-specific P acquisition and antioxidant defense and physiological adaptation in different Chinese fir clones. The 2C roots had the highest P concentration and accumulation amongst the three root classes, indicating their significance in acquisition and redistribution of P. Clone-061 also exhibited higher accumulation of P, use efficiency, and activities of antioxidant enzymes (SOD, POD and CAT) while accumulating a lower amount of MDA under P deficiency, thus proving to be better in terms of nutrient utilization and tolerance to oxidative stress. The coordinated relationship between root class-specific P traits and antioxidant responses, further indicated by principal component analysis, suggested complementary roles of different root classes in P acquisition and stress adaptation. In general, clone 061 showed greater physiological adaptability to P stress and is a potential genotype for improving P-use efficiency in Chinese fir plantations.

Acknowledgement

The authors gratefully acknowledge the support and facilities provided by the College of Forestry and the College of Life Sciences, Fujian Agriculture and Forestry University, China, which contributed to the successful completion of this research.

Author Contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Muhammad Haroon U Rashid, Pengfei Wu and Qingjun Xie. The first draft of the manuscript was written by Muhammad Haroon U Rashid and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Funding

This work was supported by the National Natural Science Foundation of China (grant no. 32572043) and the Key Program of Natural Science of Fujian Province, China (grant no. 2024J02010). Author P.W. has received research support for this work.

Data availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Competing interests

The authors declare that they have no conflicts of interest.

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