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Effects of Clipping Intensity on the Physiology of *Dicranopteris pedata* and Its Interroot Soil in the Rare-Earth-Mining Area in Southern China

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Abstract: Clipping is crucial during phytoremediation. However, research into the effects of clipping intensity on the physiology of *Dicranopteris pedata* (*D. pedata*) and its interroot soil in the rare-earth-mining area in southern China is lacking. A clipping experiment was conducted to verify the phytoremediation effect of *D. pedata*. The physiology of *D. pedata*, such as biomass, antioxidant enzymes, chlorophyll, and rare-earth elements (REEs), were determined after clipping. And the microbial community diversity and soil enzyme activities in the interroot soil of *D. pedata* were investigated. The phytoremediation efficiency was determined at the end of the experiment. The results showed that the compensatory growth effect of *D. pedata* was stronger with increasing clipping intensity. There was no significant difference in the α diversity of interroot soil microorganisms of *D. pedata* at different clipping intensities, but β diversity analysis showed that the clipping treatment group deviated from the control group. Only urease activity decreased among the interroot soil enzymes in *D. pedata* after clipping, while the soil catalase and sucrase were less responsive to clipping. The REEs accumulated by *D. pedata* were dominated by light REEs in the aboveground part of the plant, while the amounts of light and heavy rare-earth elements accumulated in the underground part of the plant were similar. The phytoextraction of REEs gradually increased with increasing clipping intensity. It was concluded that 100% clipping once a year is the most appropriate when considering *D. pedata's* phytoremediation potential and soil system. The time it takes for 100% clipping of *D. pedata* to reduce the soil TREEs (total rare-earth elements), LREEs (light rare-earth elements), and HREEs (heavy rare-earth elements) to below-average soil REE concentration in China was estimated to be 25.54 years, 19.56 years, and 65.43 years, respectively, which was significantly lower than that for other clipping intensities and the control group. It is concluded that clipping *D. pedata* is an effective way to promote phytoextraction efficiency in the southern rare-earth-mining areas. The soil can still support the resumption of *D. pedata* growth after high-intensity clipping.

Keywords: clipping intensity; rare-earth elements; *Dicranopteris pedata*; phytoremediation

Citation: Lin, Y.; Chen, Z.; Li, W.; Chen, Z. Effects of Clipping Intensity on the Physiology of *Dicranopteris pedata* and Its Interroot Soil in the Rare-Earth-Mining Area in Southern China. *Sustainability* **2024**, *16*, 664. https://doi.org/10.3390/su16020664

Academic Editors: Robson Andreazza and Filipe Selau Carlos

Received: 10 December 2023
Revised: 4 January 2024
Accepted: 10 January 2024
Published: 11 January 2024

1. Introduction

REEs consist of 15 lanthanides, scandium, and yttrium, which are generally categorized into two groups: light REEs (La, Ce, Pr, Nd, Sm, and Eu) and heavy REEs (Gd, Tb, Dy, Ho, Er, Tm, Yb, and Lu) [1]. Owing to their unique and excellent properties, REEs have been widely used in a variety of fields [2]. For instance, REEs are known to have hormonal effects on organisms, which are characterized by stimulatory effects at low doses and inhibitory effects at high doses [3]. Therefore, low doses of REEs are widely used in agriculture [3]. Excessive mining activities cause the release of REEs into the environment [4,5]. After entering the human body through the food chain, REEs can accumulate in the blood, brain, bones, and hair, resulting in neurotoxic effects [6]. Therefore, appropriate restoration measures are needed for rare-earth-mining areas.

Regarding the remediation of rare-earth-mining sites, both physical and chemical methods have limitations, such as high costs, high labor intensity, irreversible changes in soil properties, and interference with the native soil microbiota. Considering the costs and long-term ecological benefits, phytoremediation is currently the most affordable option for treating rare-earth-mining sites [7,8]. The five techniques used in phytoremediation include phytoextraction (or phytoaccumulation), phytostabilization, phytofiltration, phytodegradation, and phytovolatilization [9]. Phytoextraction and phytostabilization have found wider application in the actual remediation of contaminated mine sites [10]. Phytoextraction is the uptake of pollutants from soil or water by plant roots and their transfer to and accumulation in aboveground biomass [11,12]. Phytostabilization can reduce the bioavailability of metals by immobilizing them in the rhizosphere, which prevents metal dispersal [12].

The global demand for REEs has grown rapidly in recent years. China, as the major REE raw material supplier, accounted for 60–70% of global production between 2010 and 2020 [13]. Ion-adsorption rare-earth ores are the major supplier of medium and heavy REEs in southern China [14]. The REE concentration of *D. pedata* growing in abandoned rare-earth mines in southern China can reach 7000 $mg \cdot kg^{-1}$, which is the highest known REE enrichment capacity for a plant [5,15,16]. It has strong resistance to drought, heat, and barrenness., while its well-developed root system also stabilizes water and soil [5,15,16]. Therefore, *D. pedata* has been proven to have extraordinary potential in the ecological restoration of abandoned rare-earth-mining areas [16]. Although phytoremediation has been widely accepted by society, it is time-consuming, which is detrimental to the economic development of developing countries. Currently, the greatest challenge is the low biomass of *D. pedata*, which drastically extends the time needed for phytoremediation.

Clipping is a cheap and non-polluting technique that is beneficial in increasing the biomass of plants and has been applied to the phytoremediation of contaminated soils [17]. For example, a proper clipping pattern improves the phytoremediation ability of *Leersia hexandra*, *Solanum photeinocarpum*, and *Pennisetum purpureum* Schumach [18–20]. The stability of the plant–soil system is important for clipping and determines whether clipping can be repeated, which affects the sustainability of phytoremediation. It has been suggested that clipping leads to a loss of plant biomass and an imbalance in soil water and heat, thereby causing disturbances in the plant-soil system [21]. Ilmarinen [22] found no evidence of clipping creating general soil feedback during plant growth. Different plant species have different nutrient requirements, acquisition strategies, and forms of nutrient uptake, which, together with differences in geographic location and soil properties, lead to different feedbacks from the soil to plant clipping [23]. Interroot soil is the soil in a narrow area 1–3 mm below the surface of the plant root system [24]. The microbial community in and enzymatic activity of soil are recognized as sensitive indicator parameters for assessing the soil environment [25]. The interroot soil microbial community can reflect the compositional and functional diversity of an ecosystems as well as the health of vegetation, which is critical for the feedback regulation of soil microorganisms and vegetation [26]. Therefore, the effect of clipping on the stability of the plant–soil system can be evaluated by the soil's microbial community and enzyme activity.

Most of the current research on clipping focuses on its effect on forage yield, while the research on the application of clipping technology in the remediation of contaminated soil is insufficient. For the purpose of improving the efficiency of phytoremediation by *D. pedata*, it is important to be aware of the physiological characteristics of *D. pedata* and understand the variability in the soil environment to ensure that it can be clipped for a long time to develop a reasonable clipping intensity. As far as we know, our study is the first to investigate the effect of clipping intensity on the phytoremediation efficiency of *D. pedata*. Our study had the following three main objectives: (1) to understand the recovery of *D. pedata* growth at different clipping intensities; (2) to investigate the effect of different clipping intensities on the interroot soil, and (3) to evaluate the effect of different clipping intensities on the phytoremediation efficiency in rare-earth mining areas.

2. Research Methodology

2.1. Study Area

The experimental site was at Niushitang (25°36′ N, 116°26′ E), Hetian Town, Changting County, Fujian Province, China, which has abundant and scattered rare-earth mines. With an average annual temperature of 18.5 °C and an average annual rainfall of 1885 mm, it has a subtropical monsoon climate with simultaneous rain and heat. The terrain is mainly hilly. The soil in the study area is derived from quaternary red clay and classified as Argi-Udic Ferrosols (Chinese Soil System Classification) [27]. The main vegetation types are *Pinus massoniana*, *D. pedata*, *Liquidambar formosana*, *Schima superba*, *Paspalum wetsfeteini*, etc. Among them, *D. pedata* is the dominant vegetation species.

2.2. Sampling and Pretreatment

A well-grown and uniform *D. pedata* flat was selected as the experimental site, which is in an abandoned rare-earth-mining area. Subsequently, it was divided into fifteen subexperimental sites (1 m × 1 m). Clipping is the removal of the above ground portion of the *D. pedata*. The above ground portion of the *D. pedata* dries out in winter, which facilitates clipping. In mid-January 2021, *D. pedata* was clipped in proportion to area to create the clipping intensity, which was categorized as 100%, 75%, 50%, 25%, and unclipped (Consider the unclipped as the experimental control group. See Figure 1). Three replications were set for each clipping intensity. The 30 cm of *D. pedata* at the periphery of the sample square was removed to avoid disturbance, and no measurements were taken during this period. The experiment was conducted in the field for 365 days. In January 2022, the cover of *D. pedata* was measured. Plant samples were collected separately using the "five-point method". The root system of *D. pedata* was carefully dug out, and then an interroot soil sample was obtained from each plant using the "shake down method". One subsample was immediately stored at −80 °C until it was used for DNA extraction, and the other subsample was stored at 4 °C for physicochemical analysis.

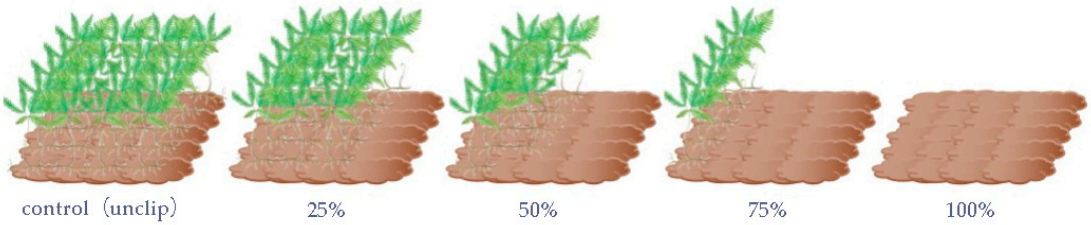


Figure 1. Schematic diagram of the clipping method for *D. pedata*.

2.3. Determination of D. pedata and Soil Physical and Chemical Indices

D. pedata tissues were divided into aboveground (leaf) and underground parts (rhizome and root). Tissues were washed separately with deionized water and dried at 65 °C. The aboveground biomass, underground biomass, and total biomass of *D. pedata* per unit area were measured. Briefly, the crude enzyme solution was extracted by placing 1 g of fresh leaves in cold extraction buffer (0.1 mol·L^{-1} PBS, pH 7.0, 1% PVP, 0.1% mercaptoethanol) and grinding them quickly with a precooled mortar and pestle. The homogenates were centrifuged at 4000 rmin^{-1} and 4 °C for 15 min, and the supernatant was reserved for further enzyme activity measurement. SOD activity was determined based on the photochemical reduction of nitro blue tetrazolium (NBT). Guaiacol colorimetry was used for the measurement of POD activity. CAT activity was determined by monitoring the decline in 240 nm sections [28].

The *D. pedata* leaves from each site were placed in 10 mL of ethanol solution (96%, *v*/*v*) and incubated in the dark at room temperature for four days until the leaf samples turned completely white. The solution for each sample was placed in three cuvettes, and the light absorption of each cuvette was measured at three wavelengths, 665, 649, and 470 nm, using a Varian Cary 100 Bio UV—Visible Spectrophotometer (Thermo Electron Corporation, Madison, WI, USA). Chlorophyll concentrations were calculated according to Lichtenthaler [29].

A Microwave Laboratory System (Multiwave 3000, Anton Paar, North Ryde, Australia) was used to mineralize the soil and tissue samples. Soil samples were digested using an acid mixture of HF 40%: HCl 38%: HNO_3 70% (1:1:3), and the tissue samples were digested using an acid mixture of H_2O_2 30%: HNO_3 70% (1:3). Fifteen REEs from all samples were determined using an inductively coupled plasma-mass spectrometer (ICP-MS, X Series 2, Thermo Scientific, Waltham, MA, USA). With the introduction of Chinese national certified standards (e.g., soils GBW07405 and GBW07407), the recoveries of these elements were in the range of 85–115%, and the relative standard deviations of all samples were less than 5%.

Soil sucrase was determined using the 3,5-dinitrosalicylic acid colorimetric method. Soil urease was determined with the sodium phenol-sodium hypochlorite colorimetric method, and soil peroxidase was determined using the potassium permanganate titration method [30].

2.4. Soil Genomic DNA Extraction, PCR Amplification, and Sequencing

Soil genomic DNA was extracted using an E.Z.N.A. Soil DNA Kit (Omega Biotek, Inc., Norcross, GA, USA) following the manual. The concentration and quality of the genomic DNA were examined with a NanoDrop 2000 spectrophotometer (Thermo Scientific Inc., USA). DNA samples were stored at −20 °C for subsequent experiments. The V3-4 hypervariable region of the bacterial 16S rRNA gene was amplified with the universal primers 338F (5′-ACTCCTACGGGAGGCAGCAG-3′) and 806R (5′-GGACTACNNGGGTATCTAAT-3′). For each sample, an 8-digit barcode sequence was added to the 5′ end of the forward and reverse primers (provided by Allwegene Company, Beijing, China). PCR was carried out on a Mastercycler Gradient (Eppendorf, Hamburg, Germany) using 25 μL reaction volumes containing 12.5 μL of 2 × Taq PCR MasterMix (Vazyme Biotech Co., Ltd., Nanjing, China), 3 μL of BSA (2 ng/μL), 1 μL of forward primer (5 μM), 1 μL of reverse primer (5 μM), 2 μL of template DNA, and 5.5 μL of ddH_2O. The cycling parameters were 95 °C for 5 min, followed by 28 cycles of 95 °C for 45 s, 55 °C for 50 s, and 72 °C for 45 s with a final extension at 72 °C for 10 min. The PCR products were purified using an Agencourt AMPure XP Kit (Beckman Coulter, Inc., Brea, CA, USA). Sequencing libraries were generated using the NEB Next Ultra II DNA Library Prep Kit (New England Biolabs, Inc., Ipswich, MA, USA) following the manufacturer's recommendations. The library quality was assessed with a Nanodrop 2000 (Thermo Fisher Scientific, Inc., Waltham, MA, USA), an Agilent 2100 Bioanalyzer (Agilent Technologies, Inc., Santa Clara, CA, USA), and an ABI StepOnePlus Real Time PCR System (Applied Biosystems, Inc., Foster City, CA, USA).

2.5. Data Processing and Statistical Analysis

Statistical analysis software, such as Excel 2016 and R (v3.6.0), was used for data processing. The data were log transformed to satisfy the assumptions of normality and homogeneity as needed. We used one-way ANOVA to compare the differences in data; otherwise, Dunnett's T3 test was used. Significant differences were tested using least significant difference (LSD) multiple comparisons. Levels of $p < 0.05$ were considered statistically significant in the multiple comparisons analysis. Pearson correlation analysis was used to analyze the relationship between the indicators.

The raw data were separated into different samples based on the barcode sequence through the QIIME [31] (v1.8.0) software. Pear [32] (v0.9.6) software was used to filter and splice raw data. The sequences were disregarded if they were shorter than 120 bp, had a low quality score (≤20), or contained ambiguous bases. The minimum overlap was set to 10 bp

and the mismatch rate was 0.1 during the splicing process. The bacterial 16S sequences with lengths less than 230 bp were removed after splicing using Vsearch [33] (v2.7.1) software, and the chimeric sequences were removed using the UCHIME [34] method based on the Gold Database. After splicing, the fungal ITS sequences with lengths less than 230 bp were removed using Vsearch (v2.7.1) software, and chimeric sequences were removed by the UCHIME method based on the Unite Database. Eligible sequences were clustered into operational taxonomic units (OTUs) using the Uparse [35] algorithm in Vsearch (v2.7.1) software with a similarity threshold of 97%. The rarefaction curves were generated using QIIME (v1.8.0) and the richness and α diversity indices were calculated according to the OTU information. The matrix of β diversity distances between samples were calculated using the Bray-Curtis algorithms and plotted using partial least squares discriminant analysis (PLS-DA).

The compensatory growth pattern of *D. pedata* is expressed by the compensatory growth index G/C proposed by Belsky [36]. In this paper, G is the biomass of the aboveground parts of *D. pedata* after different clipping intensities, and C is the biomass of the aboveground parts in the untreated control. If G/C > 1, this indicates overcompensation; if G/C = 1, this indicates equivalent compensation; and if G/C < 1, this indicates low compensation.

The phytoremediation efficiency of *D. pedata* was evaluated using the following equation:

$$\text{Phytoextraction: } T = C_{ab} \times B_{ab} \times VC/1000 \quad (1)$$

Here, C_{ab} indicates the REEs concentration in the aboveground parts of *D. pedata*. B_{ab} indicates the biomass of *D. pedata*. VC indicates the coverage of *D. pedata*.

The phytoremediation efficiency was calculated using the equation proposed by Chen [37].

$$PT = (TREEs - ATREEs) \times BD \times TH \times YE/(T \times VC) \quad (2)$$

In the equation, ATREEs (186.76 $mg \cdot kg^{-1}$) represents the average TREE concentration in the soil in Fujian Province, China. BD ($g \cdot cm^{-3}$) represents the soil bulk weight; PT (year) represents the time taken for the extraction of total soil REEs concentration by *D. pedata* to reach ATREEs; TH (m) represents the soil thickness; YE (year) represents the year that *D. pedata* was clipped once; and VC represents the coverage of *D. pedata*. The same method was used to calculate the phytoextraction time for soil LREEs and HREEs to reach the average soil ALREE (average light rare-earth element concentration, 80.83 $mg \cdot kg^{-1}$) and AHREE (average heavy rare-earth element concentration, 56.04 $mg \cdot kg^{-1}$) in Fujian Province, China.

3. Results

3.1. Effect of Clipping Intensity on the Growth and Physiology of D. pedata

3.1.1. Growth and Physiology Indices of *D. pedata* after Clipping

As seen in Table 1, *D. pedata* grew well after one year, and its coverage gradually returned to normal levels. All *D. pedata* exhibited overcompensated growth after clipping, and the CI of *D. pedata* gradually increased with increasing clipping intensity. The CI of the 100% clipped *D. pedata* treatment was significantly greater than that of the other treatments ($p < 0.05$). In antioxidant enzymes, CAT activity increased with increasing clipping intensity ($p < 0.05$), while SOD and POD decreased with increasing clipping intensity ($p < 0.05$). The chlorophyll content in the leaves after all clipping treatments was significantly higher than that of the control group ($p < 0.05$). The content of REEs in the leaves of *D. pedata* under all clipping treatments was significantly higher than that of the control group ($p < 0.05$).

Table 1. Physiological indices of *D. pedata* under different clipping intensities.

Treatment		Clipping Intensity				Control
Sorts		**100**	**75**	**50**	**25**	**0**
VC in 2022 (%)		89	82	88	92	100
Compensation index		1.82 ± 0.13 a	1.58 ± 0.11 b	1.53 ± 0.10 b	1.53 ± 0.07 b	0 c
Physiological	CAT(U·mL^{-1})	4.51 ± 0.83 a	3.08 ± 0.67 b	3.04 ± 0.33 b	2.79 ± 0.23 b	3.11 ± 0.57 b
	SOD(U·L^{-1})	1395.48 ± 106.82 b	1445.36 ± 217.17 b	1705.46 ± 345.88 ab	2043.57 ± 517.62 a	2151.00 ± 183.74 a
	POD(U·L^{-1})	6.72 ± 0.99 b	8.66 ± 0.69 b	12.05 ± 2.71 a	12.41 ± 1.68 a	12.81 ± 1.42 a
	Chl(U·L^{-1})	1.99 ± 0.32 a	1.83 ± 0.13 a	1.83 ± 0.06 a	1.91 ± 0.09 a	1.40 ± 0.25 b
REEs (mg·kg^{-1})	Light	1759.07 ± 126.64 a	1617.97 ± 85.48 a	1433.36 ± 339.09 a	1542.07 ± 285.58 a	935.65 ± 253.93 b
	Heavy	263.89 ± 23.17 ab	228.22 ± 76.62 b	328.60 ± 73.38 a	307.42 ± 12.85 ab	217.97 ± 33.53 b
	Total	2022.96 ± 149.74 a	1846.19 ± 122.16 a	1761.96 ± 330.61 a	1849.50 ± 272.75 a	1153.63 ± 286.26 b

Note: values followed by the same or no letters are not significantly different, and those followed by different letters are significantly different at $p < 0.05$.

3.1.2. Correlation Analysis of the Physiological Indices of *D. pedata*

As seen in Figure 2, correlation analysis showed that the REEs had a strong correlation with chlorophyll and a moderate negative correlation with SOD activity and POD activity. There was a strong correlation between biomass and CAT, while there was a strong negative correlation between biomass and SOD and POD.

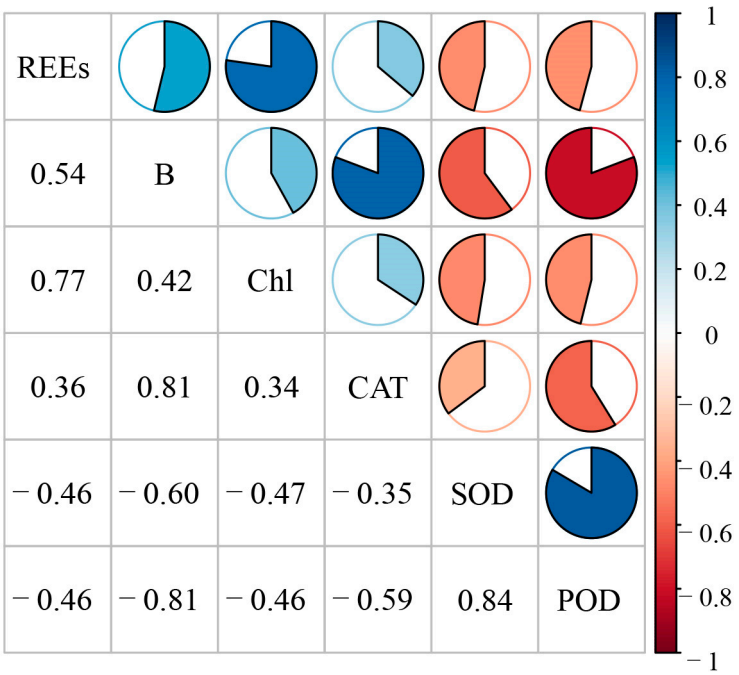


Figure 2. Correlation between REEs and physiological indicators of *D. pedata*.

3.2. Effect of Clipping Intensity on Interroot Soil

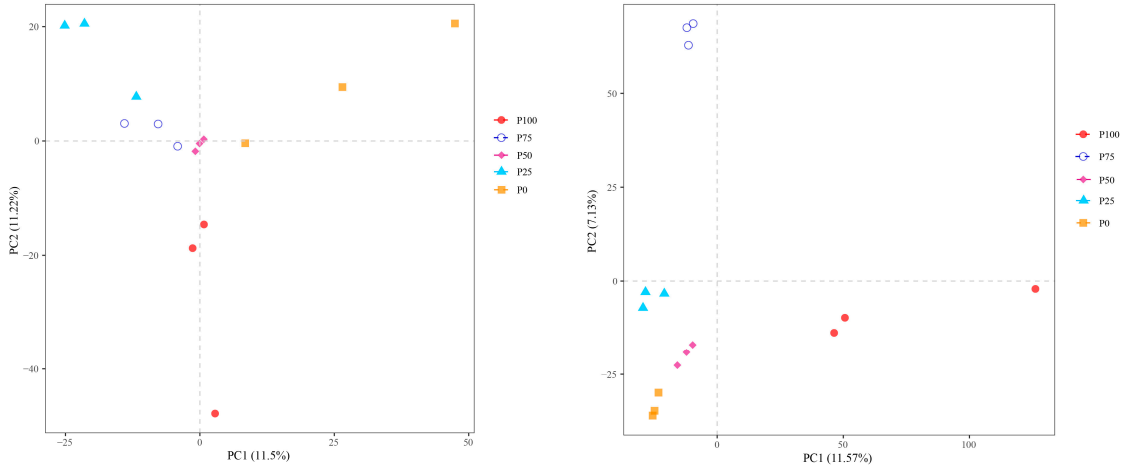
3.2.1. Interroot Soil Microbial Diversity Analysis of Different Clipping Intensities

As seen in Table 2, there was no significant difference in the α diversity index of interroot soil fungi and bacteria among different clipping intensities ($p > 0.05$).

Table 2. Interroot soil indicators under different clipping intensities.

Treatment		Clipping Intensity				Control
Sorts		100	75	50	25	0
Bacteria α diversity	Chao1	7721.46 ± 467.10	7814.18 ± 294.07	7315.51 ± 819.40	7504.08 ± 452.93	7152.80 ± 378.29
	Shannon	9.00 ± 0.27	9.20 ± 0.26	8.81 ± 0.51	8.90 ± 0.33	8.42 ± 1.17
Fungi α diversity	Chao1	1027.43 ± 131.91	1067.73 ± 184.03	1062.44 ± 222.99	1092.75 ± 130.60	1060.84 ± 276.06
	Shannon	4.36 ± 2.20	4.20 ± 1.02	3.75 ± 0.65	4.61 ± 0.71	3.47 ± 2.11

As seen in Figure 3, for the 100% clipped samples, two interroot soil fungi samples were close to each other, but one other sample deviated from the others on the PC2 axis. Individual samples in the 25% and control groups were clustered together with samples from the 75% and 50% groups.



(**a**) (**b**)

Figure 3. PLS-DA analysis of fungal and bacterial β diversity in *D. pedata* interroot soil treated with different clipping intensities. (**a**) Fungi, (**b**) bacteria.

Among the 100% clipped samples, there was one sample of soil interroot bacteria that had a large dispersion from the other samples on the PC1 axis. Even though the other two samples are clustered together, they are separated from the other treatments on the PC1 axis. The samples from the 50% group, 25% group, and control group are clustered together.

3.2.2. Interroot Soil Enzyme Activities at Different Clipping Intensities

As seen in Table 3, there was no significant difference in interroot soil catalase and sucrase activities under all treatments ($p > 0.05$). Urease in rhizosphere soil showed a decreasing trend ($p < 0.05$) with increasing clipping intensity.

Table 3. Interroot soil indicators at different clipping intensities.

Treatment		Clipping Intensity				Control
Sorts		100	75	50	25	0
Soil enzyme activity	CAT(U·L^{-1})	7193.64 ± 1094.37	6707.03 ± 839.43	8242.65 ± 1943.55	6867.15 ± 1317.66	8140.87 ± 440.15
	Urease(U·L^{-1})	456.27 ± 102.34 b	556.07 ± 55.62 ab	563.08 ± 25.02 a	633.36 ± 23.31 a	648.42 ± 37.66 a
	Surase(U·L^{-1})	921.25 ± 121.64	983.28 ± 33.60	966.40 ± 131.66	986.14 ± 126.04	1094.39 ± 134.64

Note: values followed by the same or no letters are not significantly different, and those followed by different letters are significantly different at $p < 0.05$.

3.3. Phytoremediation of D. pedata after Clipping

As seen in Figure 4, with increasing clipping intensity, the accumulation of light and heavy REEs in the aboveground parts of *D. pedata* gradually increased. The accumulation of REEs in the aboveground parts of *D. pedata* in the 100% clipping quadrat was significantly greater than that in the other treatments ($p < 0.05$). The accumulation of light and heavy REEs in the underground part of *D. pedata* was similar, and the accumulation of REEs in the underground part of *D. pedata* after clipping treatment was significantly greater than that in the control group ($p < 0.05$).

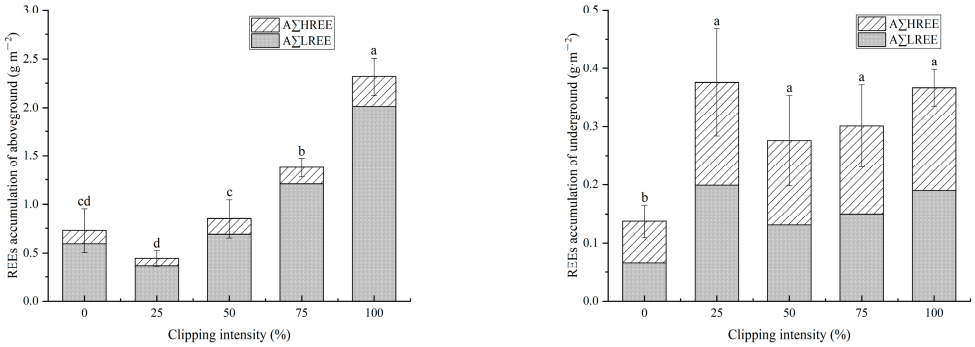


Figure 4. REE accumulation in *D. pedata* treated with different clipping intensities. Note: values followed by the same or no letters are not significantly different, and those followed by different letters are significantly different at $p < 0.05$.

As seen in Table 4, the REEs extracted by *D. pedata* are dominated by LREEs. As the clipping intensity increased, the amount of REEs extracted by *D. pedata* increased, and the phytoextraction of the 100% clipped *D. pedata* samples was significantly greater than that of the other treatment samples ($p < 0.05$). The phytoremediation time for *D. pedata* which is clipped by 100% once a year is 19.56 years for LREEs, 65.43 years for HREEs, and 25.54 years for total REEs, all of which are significantly less than that for the 25% and 50% clipping intensities ($p < 0.05$).

Table 4. Phytoremediation efficiency of *D. pedata* treated with different clipping intensities.

Treatment		Clipping Intensity				Control
Sorts		**100**	**75**	**50**	**25**	**0**
Phytoextraction ($g \cdot m^{-2}$)	Light	1.80 ± 0.21 a	0.98 ± 0.08 b	0.60 ± 0.15 c	0.34 ± 0.08 d	0 e
	Heavy	0.27 ± 0.03 a	0.14 ± 0.04 b	0.14 ± 0.04 b	0.07 ± 0.01 c	0 d
	Total	2.07 ± 0.24 a	1.12 ± 0.06 b	0.74 ± 0.15 c	0.41 ± 0.08 d	0 e
Phytoremediation time (years)	Light	19.56 ± 1.69 c	32.69 ± 3.19 bc	59.52 ± 15.06 b	108.74 ± 25.18 a	–
	Heavy	65.43 ± 5.95 c	124.48 ± 38.65 b	131.03 ± 41.25 b	265.87 ± 8.75 a	–
	Total	25.54 ± 2.22 c	42.96 ± 3.16 bc	71.62 ± 14.73 b	134.79 ± 25.80 a	–

Note: values followed by the same or no letters are not significantly different, and those followed by different letters are significantly different at $p < 0.05$.

4. Discussion

4.1. Effect of Clipping Intensity on the Growth and Physiology of D. pedata

In the present study, it was found that the supercompensatory growth of *D. pedata* was more significant with increasing clipping intensity. *D. pedata* has both sporulation and clonal reproduction. After the aboveground parts of *D. pedata* were clipped, the remaining root system did not die. *D. pedata* can deliver resources to the residual root system via the underground connected clonal organs, which is an act called clonal physiological integration [38,39]. This strategy allows the clonal organs of *D. pedata* to share resources and information and then reduce resource shortages and mitigate the effects of stress in

microscale heterogeneous habitats [38–40]. In addition, the growth habit of *D. pedata* is light-tending [41]. Clipping will improve light conditions for short *D. pedata* and reduce competition from tall species, allowing *D. pedata* to grow and coexist. Thus, clipping can promote *D. pedata*'s overcompensation. Chen [37] used the cellular automata model to simulate the growth of *D. pedata* and found that, two years after clipping, the coverage of *D. pedata* increased to 89.78–94.92% in the simulated plots. This study found that approximately 89% coverage could be achieved after one year of 100% clipping, which is sufficient to show that *D. pedata* has a stronger reproductive capacity in practice.

Scholars have argued that plant resistance plays a crucial role in the efficiency of phytoremediation [42]. Protective enzymes in plants protect both the structure and function of cell membranes and are the first to respond to stresses [43]. Jiang [44] and Song [45] found that the activities of three antioxidant enzymes of *D. pedata* from nonrare-earth-mining areas increased after heat stress. In this study, the activity of CAT increased significantly with increasing clipping intensity, thereby suggesting that the efficiency of disproportionated H_2O_2 increased in response to clipping stress. However, the activity of SOD and POD decreased. The strong correlation between antioxidant enzymes and the biomass of *D. pedata* implies that they are important for the growth of *D. pedata*. The concentration of REEs in the leaves of *D. pedata* increased after clipping. Data analysis showed a moderate negative correlation between REEs and SOD after clipping, while SOD was strongly correlated with POD. Many papers report yield improvement and resilience enhancement with REE application in plants [46]. Wang et al. [47] found that Ce acts similarly to SOD by inhibiting NBT reduction and hydroxylamine oxidation and catalyzing O_2^- disproportionation. In addition, Zhang [48] and Pang [49] et al. reported the role of lanthanides in regulating the antioxidant system of plants. Therefore, we hypothesize that REEs are involved in the physiological activities of *D. pedata*, which grow in rare-earth-mining areas, thereby affecting the SOD and POD activities of *D. pedata*. Although the SOD and POD activities of *D. pedata* were lower under high-intensity clipping treatments, REEs may replace the functions of SOD and POD and protect the cellular stability of *D. pedata*.

One of the key mechanisms of compensatory growth in plants is the increase in the leaf photosynthetic rate [50]. As the intensity of clipping increases, the photosynthetic organ of *D. pedata* decreases, which disturbs the energy balance. Therefore, to sustain its own growth and development, the chlorophyll content in *D. pedata* increases to enhance its increased light capture capacity. There was a strong correlation between the REEs' concentration and chlorophyll content in *D. pedata* leaves. REEs can be attached to the proteins in the chloroplast membrane and photosystem II after being absorbed by *D. pedata* [51,52]. Therefore, it has been hypothesized that chlorophyll–protein complexes in photosystem reaction centers are increased in the presence of REEs, resulting in enhanced photosynthesis [52]. In conclusion, clipping promotes the absorption of REEs by *D. pedata*, regulates the physiological and ecological mechanisms of *D. pedata*, enhances the photosynthetic efficiency of *D. pedata*, and promotes the occurrence of supercompensatory *D. pedata* growth. Eventually, clipping enhanced the phytoremediation efficiency of *D. pedata*, which is similar to the results of most phytoremediation studies on clipping [18–20].

4.2. Effects of Clipping on the Interroot Soil of D. pedata

Microorganisms can dissolve minerals to release nutrients that can support the growth of plants during the phytoremediation process. Therefore, the presence of soil microorganisms maintains the sustainability of soil utilization. In addition, microorganisms can also produce organic acids to improve the solubility of heavy metals and enhance their bioavailability by changing the soil environment, including pH and redox potential [53–55]. The α diversity analysis demonstrated the richness of the soil's microbial community. β diversity was used to assess structural differences in species communities between samples. In this study, there was no significant difference in the α diversity analysis of interroot soil microorganisms under different clipping treatments, indicating that the richness of interroot soil microorganisms' community was less affected by clipping. β diversity analysis showed

that the interroot soil fungi in the clipping treatment group differed from the control group, and the bacterial communities in the 100% and 75% clipping treatments differed from the control. These results imply that clipping caused a change in the characteristics of the interroot soil microbial community.

The richness of the interroot soil microbial community was not significantly affected by clipping, which could be attributed to the fact that the physical structure of the soil was not disrupted by clipping, and the below-ground root system of *D. pedata* remained biologically active after clipping. However, differences in the structure of the interroot soil microbial community may be due to the increase in surface temperature after clipping *D. pedata*. This is different from the studies by Bai [56] and Chen [57], who concluded that the fungal community composition was not sensitive to clipping. Previous studies have concluded that fungi display unique stability in phytoremediation owing to their high tolerance for extreme pH, climate, nutrients, and heavy metals [53]. Francioli [58] proposed that the richness and community structure of fungi are related to plant root traits. Fungi are major decomposers of root turnover in soil ecosystems [59], and the soil microbial community is regulated by plant root exudates [56]. Therefore, clipping has the potential to affect the interroot soil microbial community by changing the growth of *D. pedata* roots and root exudate secretion.

Soil enzymes mainly come from soil microorganisms. Soil enzyme activity can be used as a reference for the nutrient cycling situation in the soil system [60]. We observed that only the interroot soil's urease activity showed a significant decreasing trend with increasing clipping intensity. Urease can be used to measure nitrogen availability to plants [24]. The only factor that limits the urease activity is the availability of urea in soil [24]. As the 100% clipping treatment reduces the substrate for urease reaction in the soil, it ultimately leads to a decrease in urease activity. Moreover, the spatial structure of the *D. pedata* community changed, the ground temperature increased, the water evaporated, and the organic matter input decreased due to the removal of aboveground *D. pedata*. All of these changes led to a slight decrease in enzyme activity. In conclusion, clipping has little impact on the soil. Clipping every other year is reasonable in terms of phytoremediation, and the soil still supports the restoration of *D. pedata* growth after high-intensity clipping, which also ensures that the stability of the *D. pedata*-soil system is maintained.

4.3. Efficiency of Phytoremediation after Clipping D. pedata

Although Chen [37] has proposed clipping *D. pedata* to assist in enhancing the efficiency of phytoremediation in mining areas, the research has been limited to simulations of *D. pedata's* extraction of pollutants and have not yet delved into the practical effects of clipping techniques. The accumulation in both the above- and belowground parts of *D. pedata* increased after clipping. This was because clipping promoted the supercompensatory growth of *D. pedata*, increased the uptake of REEs by the root system, and then transferred the REEs to the aboveground parts of *D. pedata*. The phytoextraction of 100% clipped *D. pedata* was significantly higher than that of the other measures. It was found that the aboveground part of *D. pedata* mainly accumulated light REEs, while the underground part accumulated similar amounts of LREEs and HREEs. Using the simulation formula, it was found that, if 100% clipping was performed once a year, it would take approximately 25.54 years to reduce the TREEs concentration at 0–20 cm depth in the study area to the national average level. Additionally, the purification time for LREEs is expected to be 19.56 years, and the purification time for HREEs is expected to be 65.4 years, which was significantly lower than other clipping intensities and the control group. Phytoextraction of *D. pedata* can be assisted by using clipping techniques in REE-contaminated mines, which is a solution that can balance soil remediation and economic benefits.

Fertilizing combined with clipping can also be used to try to further enhance the phytoremediation efficiency of *D. pedata*. Moreover, the potential relationship between the significant changes in physio-chemical characteristics and the accumulation of REEs by *D. pedata* needs to be strengthened in the future. The aboveground parts of *D. pedata*

are clipped and contain large amounts of REEs, but these REEs are still present in the plant. If mishandled, *D. pedata* can be degraded and returned to the soil, which can result in secondary contamination of REEs. However, the demand for REEs is increasing. REE hyperaccumulators are regarded as a rich 'bio-ore'. The recovery of REEs from *D. pedata* not only improves the utilization of REEs but also reduces mining activities and reduces the environmental damage caused by mining. Therefore, we are exploring how to solve the problem of *D. pedata* harvesting products. Scholars have attempted to extract REEs from *D. pedata* based on biomass incineration using chemical processes with leaching and precipitation [61]. For example, Qin [62] used vacuum-pyrolysis condensation to convert biomass from *D. pedata* into the chemical resource ethylene oxide. Chour [63] used enhanced ion exchange leaching to achieve 78% recovery of high-purity REEs from *D. pedata*. After clipping the aboveground parts of *D. pedata*, this type of recycling technology should be applied promptly.

5. Conclusions

After clipping with different intensities, we observed that supercompensatory growth occurred in *D. pedata*. The compensatory growth effect of 100% clipping was the most pronounced, and the accumulation of REEs was the highest, thereby indicating that clipping can promote *D. pedata* purification of REEs. One year after clipping, the interroot soil of *D. pedata* was less changed and still supported the growth and development of *D. pedata*. Above all, this suggests that annual clipping does not destabilize the *D. pedata*–soil system. The time taken for 100% clipping of *D. pedata* to reduce the soil TREEs, LREEs, and HREEs to below-average soil REEs concentration in China is estimated to be 25.54 years, 19.56 years, and 65.43 years, respectively.

Author Contributions: Conceptualization, Y.L. and Z.C. (Zhiqiang Chen); data curation, Y.L.; formal analysis, Y.L. and W.L.; investigation, Y.L. and W.L.; methodology, Y.L. and Z.C. (Zhiqiang Chen); project administration, Z.C. (Zhiqiang Chen); supervision, Z.C. (Zhiqiang Chen); visualization, Y.L.; writing—original draft, Y.L.; writing—review and editing, Y.L., Z.C. (Zhiqiang Chen) and Z.C. (Zhibiao Chen). All authors have read and agreed to the published version of the manuscript.

Funding: Zhibiao Chen, The National Natural Science Foundation of China (No. 42277013), Zhiqiang Chen, Natural Science Foundation of Fujian Province (No. 2020J01143).

Data Availability Statement: No new data were created.

Conflicts of Interest: The authors declare no conflict of interest.

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