

Original Research

Long-Term Gamma Radiation Effect on Functional Traits of *Tradescantia Flumnensis* L.

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Abstract

The effect of radioactivity on the environment, especially on plants, has always been one of the vital researches of radio-ecology. However, long-term irradiated ecosystems are very rare. Occasionally, one special miniature ecological environment object consisted of thorium mineral (Th-232) and *Tradescantia flumnensis* L. over 10 years is carried out to evaluate the effect of long-term low-radioactivity gamma radiation on herbaceous plant. Several functional features of *Tradescantia flumnensis* L. are selected and measured with physical measurement (PM), HPGe portable gamma-ray spectrometer (HPGe PGS), wavelength dispersion X-ray fluorescence (WDXRF), and advantage isotope ratio mass spectrometer (AIR-MS). The results show that functional traits varied with the increasing of radiation dose rate. These results also indicate that the plants can adapt to low-intensity gamma radiation (<332.6 nGy•h-1) through adjustment of physical properties. In addition, low-intensity gamma radiation has positive influence on plant's water use efficiency, growth and nutrition acquisition of roots. Restrictions on plant phosphorus demand have also been eased. However, high-intensity gamma radiation (>528.7 nGy•h-1) has exceeded the tolerance of plants, making most functional traits unhealthy or abnormal. This rare case is of great reference significance of setting biological indicators of long-term radiation system.

Keywords: radio-biology, *Tradescantia flumnensis* L., long-term radiation, functional traits

Introduction

In the field of radio-ecology, studying plant functional traits is one of the effective ways to understand the adaptability of plants to the ecosystem environment [1]. Plant functional traits can directly reflect the response and adaptability of plants to external environmental changes. Relatively, the gradient distribution of environmental factors also affects the functional traits of plants [2-5]. Practically, researchers prefer to select morphological and physiological manifestation of plant functions that reflect plant adaptation to the environment and/or affect plant productivity, such as leaf traits (leaf size, leaf thickness, specific leaf area, concentration of N and P, etc.) and root traits (root size, root length, specific root length, etc.) [5-8]. These plant functional traits can effectively show the relationship between ecosystems and environmental factors (light, water and soil nutrients, etc.) [5-6, 9]. Besides, the co-evolutionary relationship between plants and environmental factors can be analysed by comparing and verifying plant functional traits.

The behaviour and end-result of radioactive materials in the ecological environment influence the ecosystem (mainly on the growth of plants and animals) [10-11]. In 2017, phone electromagnetic field radiations test (EM-r) have proved that EM-r caused oxidative damage to plant roots [12]. Some studies have indicated that long-term growth of plants in the irradiation area may lead to changes in genetic information, such as leaf thickness, plant development and organ growth, etc. [13-14]. As the establishment and formation of an ecosystem is a long process, the effect of radiation (especially on plants) should be considered as a long-term process. However, current studies are more focused on short-term experiment [15-16], resulting in the fact that the research of effect of long-term nuclear radiation on plants is very scarce.

In natural ecosystems, plants can adapt to changes in external environmental conditions by altering their functional traits, such as height, leaf area, leaf mass, leaf life span, seed size and transmission mode. This

peculiarity also demonstrates features in ecosystem function [17]. In the previous studies, some scholars adopted different doses of gamma radiation on early bamboo irradiation treatment to conclude that with the increase of radiation dose, early bamboo growth potential decrease and the photosynthesis become weaker [18]. However, some researches show that low-intensity gamma radiation can significantly promote the photosynthesis of plants [19]. Therefore, the effect of radiation on the growth of plants may have a greater relationship with the species and nutrients receiving radiation. In this paper, *Tradescantia flumensis* L. has been receiving low-intensity long-term gamma radiation for at least 10 years, the relatively vital factors are species and nutrients, which are considered as the main experiment objects.

With the development of economy and technology, various large-scale radiation projects have been created, the surrounding ecosystems have been affected by continuous nuclear radiation. In this study, the effects of long-term nuclear radiation on plants are explored by studying the functional traits of *Tradescantia flumensis* L. The result can be important indicators to estimate the effects of long-term gamma radiation and provides a basis and reference on the impact of nuclear radiation on ecosystems.

Material and Methods

Sample Introduction

The thorium mineral (refers to radiation source Th-232 isotope) is located in the grassland of Chengdu University of Technology, Chengdu, China. It has been kept without moving for more than 10 years, resulting in long-term radiation exposure to the surrounding environment. Large number of *Tradescantia flumensis* L. in this area has experienced multiple generations of reproduction in the nuclear radiation environment.

A plane coordinate system with the thorium mineral as the origin of coordinates was set up. 90 samples in total are randomly collected and the sampling location

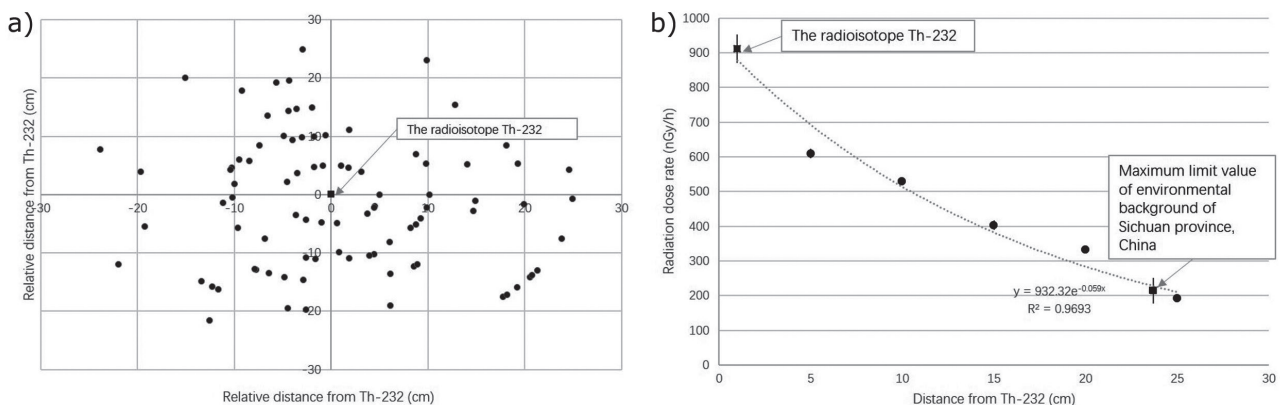


Fig. 1. a) Sampling position distribution, b) Absorption dose rate of gamma radiation gradient diagram.

are divided into six different gradients (1±0.1 cm, 5±0.5 cm, 10±0.5 cm, 15±0.5 cm, 20±0.5 cm, 25±0.5 cm). Fig. 1a) shows the sampling distribution, and the red dot refers to the thorium, which was divided into 6 different gradients.

As further quantitative analyse is necessary, gamma radiation dose rate around thorium mineral was measured by using the HPGe portable gamma ray spectrometer (trans-SPEC-DX-100T, relative measurement efficiency is 40%, energy resolution is 1.6 keV, 2.3keV at 122 keV, 2332 keV, respectively). In Fig. 1b), it shows the highest absorbed dose rate was 910.964±41.09 nGy/h while the lowest was 192.906±5.05 nGy/h. The red square refers to the maximum limit value of environmental background of Sichuan province, China.

Experimental Method

90 healthy growth plants of *Tradescantia flumensis* L. of current year in five gradients were randomly selected and uprooted as samples. The following steps are sample disposal procedures and physical trait records.

1. Samples were sorted and washed by distilled water.
2. The thickness of the leaves, the length of the roots and the height of the plants were measured with a vernier caliper.
3. The leaf area was calculated by Photoshop pixel analysis.
4. The samples were placed in a constant temperature drying oven for 8 h under 75°C.
5. Analytical balance was used to weigh the plant mass.

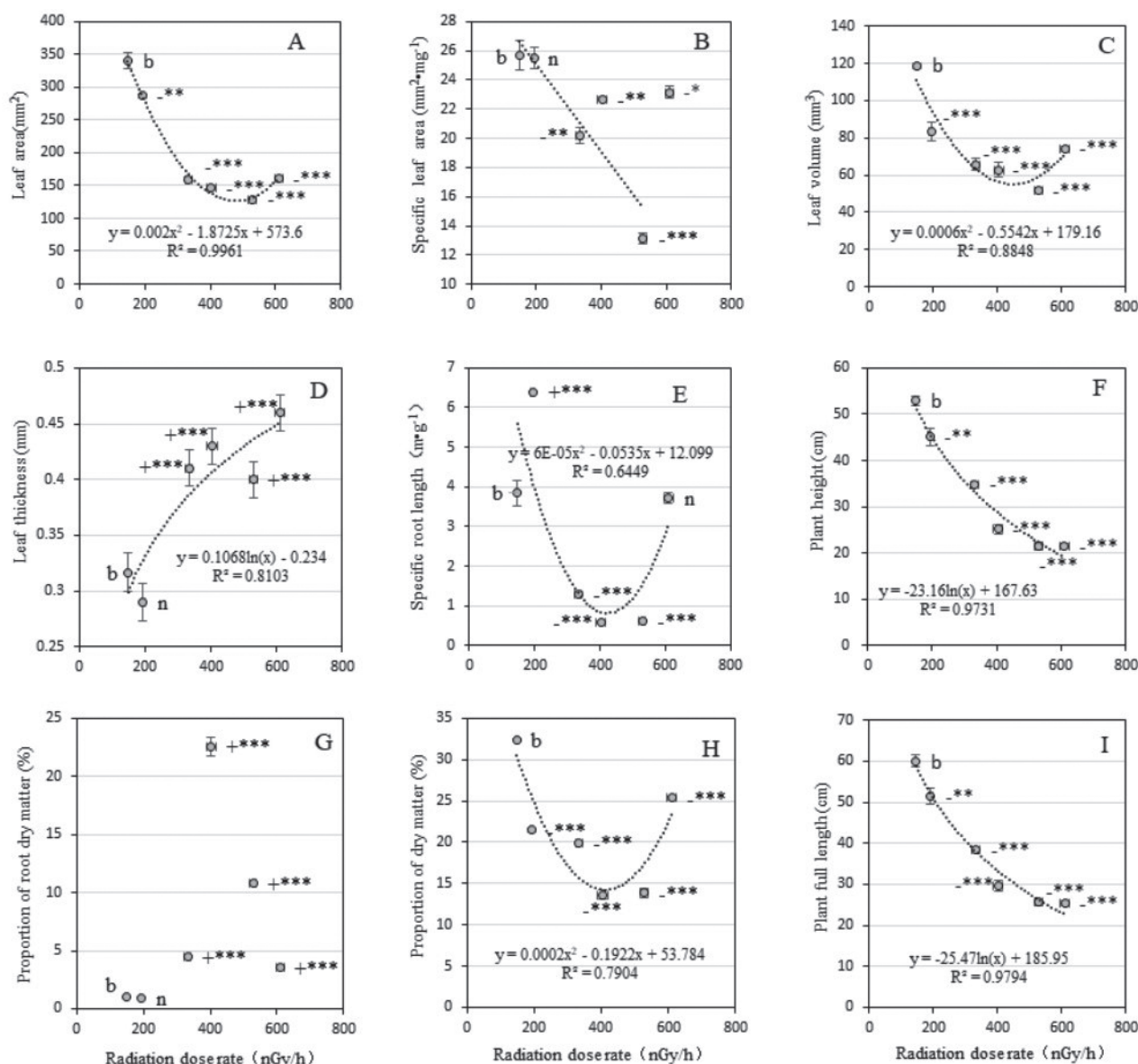


Fig. 2. Relationships between gamma radiation dose rate and a) leaf area, b) specific leaf area, c) leaf volume, d) leaf thickness, e) specific root length, f) shoot length, g) proportion of root dry matter to total dry matter, h) proportion of plant dry matter, and i) plant length.

In order to take further study on the chemical characteristics, the plants were grinded to 200 mesh powder with a mortar, the $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, carbon and nitrogen contents ($\text{g}\cdot\text{g}^{-1}$; %) of the sample were determined by DELTA V Advantage Isotope Ratio Mass Spectrometer. Phosphorus content ($\text{g}\cdot\text{g}^{-1}$; %) of the sample was determined by Wavelength Dispersion X-ray Fluorescence (WDXRF) [20] with AxiosmAX produced by PANalytical B.V. The spectrometer uses Rh-target X-ray tube, the set voltage is 60 kV, the current is 125 mA and the measurement time is 20 minutes. The quantitative analysis software is super Q 4.0.

30 soil samples were collected at 6 different gradients, which were located at exactly the same radiation dose of the plants. Each gradient contains 5 soil samples.

Statistics

The values presented in the figures are given as means \pm standard errors of means. Statistical analyses

are performed with IBM SPSS statistics 23.0 (IBM Inc., NY, USA).

Results and Discussion

The leaf functional traits: leaf area (LA), specific leaf area (SLA) and leaf volume (LV), are decreased with greater radiation dose rate until $528.7 \text{ nGy}\cdot\text{h}^{-1}$, and then increased under $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate (Fig. 2a-c). However, leaf thickness (LT) gets fatter while radiation dose rate goes higher significantly ($P<0.001$) (Fig. 2d). Specific root length (SRL) is getting shorter with greater radiation dose rate until $528.7 \text{ nGy}\cdot\text{h}^{-1}$, and then changes to grow longer under $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate (Fig. 2e). Shoot length goes down with increasing radiation dose rate significantly ($P<0.001$) (Fig. 2f). Proportion of root dry matter to total dry matter (PRM) reaches higher with greater radiation dose rate until $403.3 \text{ nGy}\cdot\text{h}^{-1}$, and then turns to low value with greater radiation dose rate (Fig. 2g). In contrast, proportion of plant dry matter

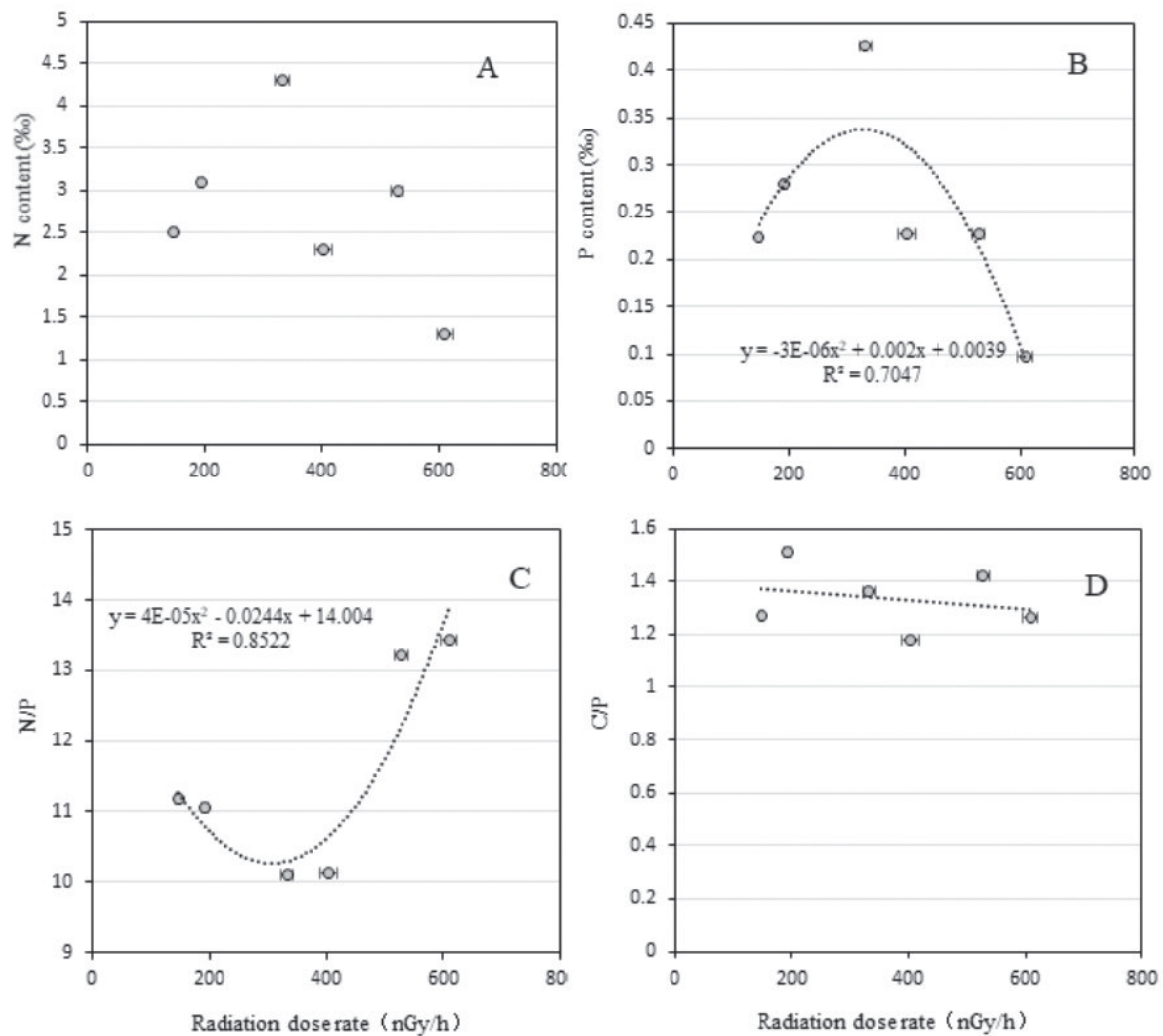


Fig. 3. Relationships between gamma radiation dose rate and a) N concentration, b) P concentration, c) N/P ratio, d) C/P ratio.

(PDM) decreases with greater radiation dose rate until $403.3 \text{ nGy}\cdot\text{h}^{-1}$, and then increases with greater radiation dose rate (Fig. 2H). Plant length and radiation dose rate show a significantly negative correlation ($P < 0.001$) (Fig. 2i).

Concentrations of N and P show positive correlation with radiation dose rate until $332.6 \text{ nGy}\cdot\text{h}^{-1}$, and then decreases while radiation dose rate goes higher (Fig. 3a-b). N/P ratio gets lower while radiation dose rate goes higher until $332.6 \text{ nGy}\cdot\text{h}^{-1}$, and then turns higher with greater radiation dose rate (Fig. 3c). Surprisingly, C/P ratio is not correlated with radiation dose rate (Fig. 3d), no linear dependence is found in the result. Concentrations of C, N and P are positive correlated with each other significantly (Fig. 4). The $\delta^{13}\text{C}$ value gets smaller with increasing radiation dose rate, except under $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate (Fig. 5a). However, $\delta^{15}\text{N}$ exhibits a positive relationship with soil depth, except under $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate significantly ($P < 0.001$) (Fig. 5b).

There is no significant regulation was found between radiation dose rate and carbon and nitrogen contents (Table S1). The mean nitrogen content of No.2 (the radiation at this point is $609.745 \pm 13.45 \text{ nGy}\cdot\text{h}^{-1}$) is the highest, and the No.3 (the radiation at this point is $528 \pm 11.27 \text{ nGy}\cdot\text{h}^{-1}$) is the lowest. The mean carbon content of No. 2 (the radiation at this point is $609.745 \pm 13.45 \text{ nGy}\cdot\text{h}^{-1}$) is the highest, and the No. 4 (the radiation at this point is $403.292 \pm 14.55 \text{ nGy}\cdot\text{h}^{-1}$) is the lowest.

Influence of Gamma Radiation on Morphological and Isotopic Traits

With the increase of radiation, plants tend to reduce individual size in order to reduce unit radiation, which is consistent with natural radiation damage reduction. Therefore, the physical traits like root length, LA and LV are getting smaller while radiation dose rate getting higher significantly, but LT has to get fatter to ensure survival by enriching cells in the leaves and increasing the amount of water storage. Furthermore, SLA is getting smaller, means the utilization mass of leaves is increasing, with greater radiation dose rate. In this way, the plants could reduce exposure without impairing the function of leaves.

The $\delta^{13}\text{C}$ is an important trait correlated with the water use efficiency [21], and the decreased $\delta^{13}\text{C}$ correlated with increased water use efficiency until $528.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate. Mycorrhizal fungi have higher $\delta^{15}\text{N}$ signatures than their host plants, because they supply relatively ^{15}N -depleted N to their hosts [22]. Thus, the positive response of $\delta^{15}\text{N}$ until $528.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate, could be interpreted as evidence of increasing function of root to absorb nutrients directly, and reducing the dependence on rhizomes [21]. In order to increasing the water use efficiency and the function of roots, the plants increase the utilization mass of leaves and roots, making SLA

and SRL decrease until $528.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate. However, under 528.7 to $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate, the $\delta^{13}\text{C}$ increased but $\delta^{15}\text{N}$ decreased while radiation dose rate gets higher. These phenomenon shows the water use efficiency and the function of root-microbial system have been severely damaged under $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate [21, 23]. Therefore, such intense radiation has exceeded the tolerance limit of the plants, making most of the functional traits

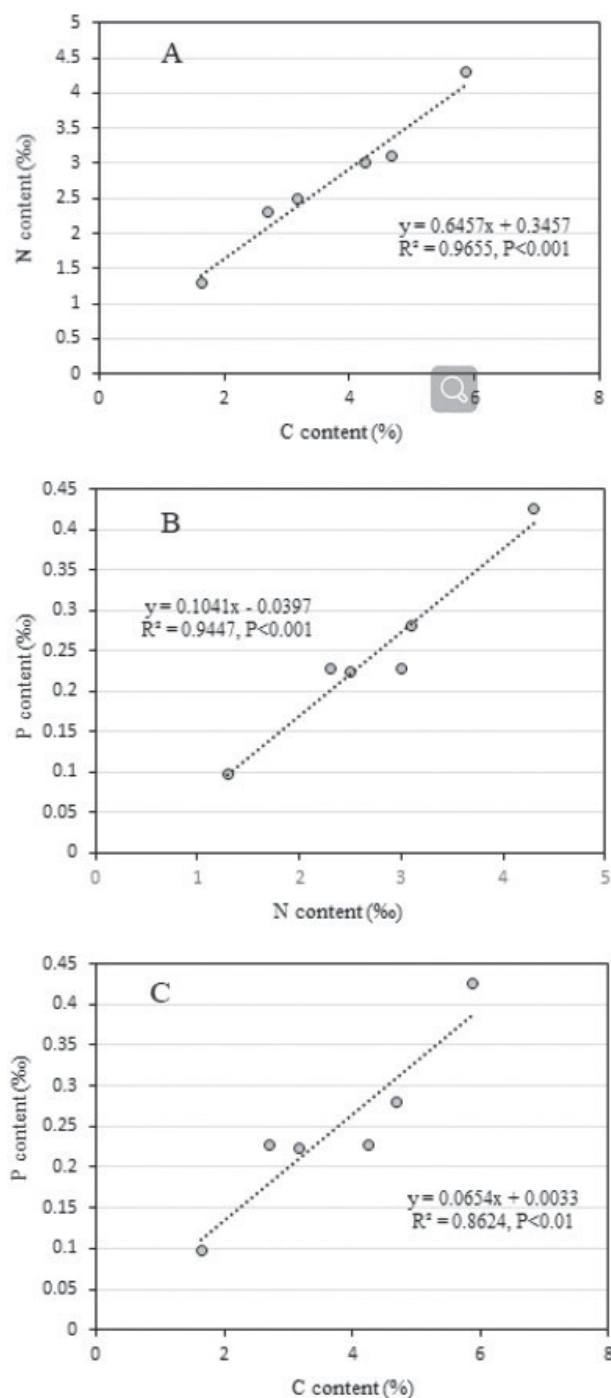


Fig. 4. Relationships between a) N concentration and C concentration; b) N concentration and P concentration; c) C concentration and P concentration.

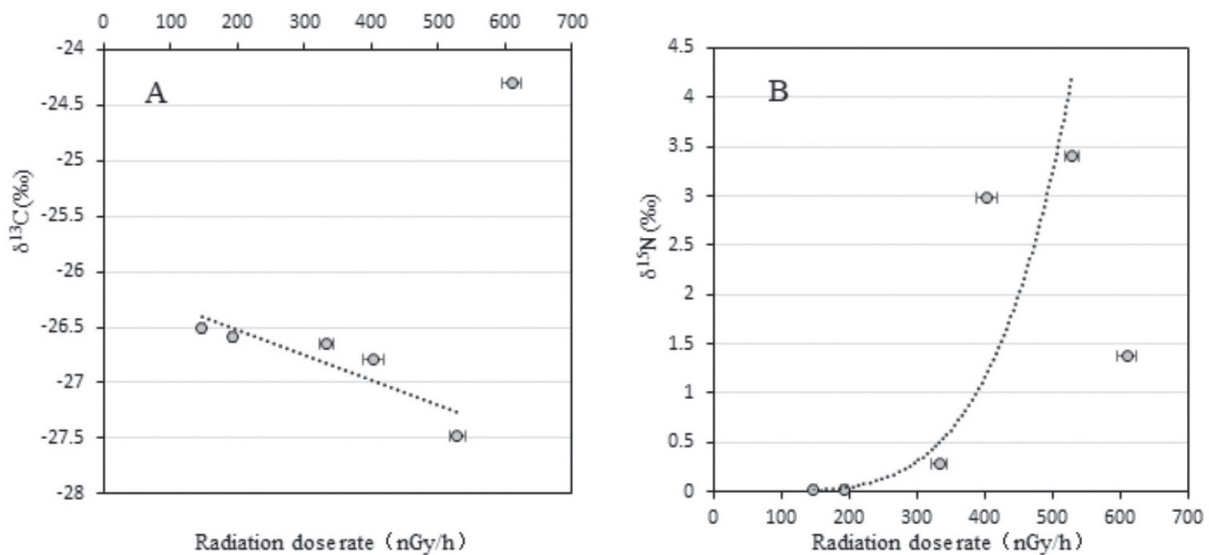


Fig. 5. Relationships between gamma radiation dose rate and (A) $\delta^{13}\text{C}$, and (B) $\delta^{15}\text{N}$.

unhealthy or abnormal under $609.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate.

Influence of Gamma Radiation on Plant Nutrition Distribution and Acquisition

Plants tend to decrease PDM to avoid the harmful effect caused by gamma radiation until $403.3 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate. However, the PRM increases under $<403.3 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate. This means that the plants use more nutrients for roots, meanwhile, the soil is much more resistant to radiation than air, which provides higher chance of survival. Furthermore, compared with the background value, SRL is increasing significantly but PRM has no significant change under $<192.9 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate. These results show that the plants increase the root length but do not use more nutrients for root growth under such low radiation dose rate. While the radiation dose rate is from 192.9 to $403.3 \text{ nGy}\cdot\text{h}^{-1}$, the plants distribute more nutrients to roots, SRL decreases and PRM increases significantly. However, under high-intensity gamma radiation, these adaptive strategies could no longer be maintained, the trends of SRL, PDM and PRM are just the opposite to weak radiation when exposed to $>528.7 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate.

Low-intensity gamma radiation has been found to increase plant growth and nutrition acquisition rates [24]. Accordingly, concentrations of both N and P are richer while radiation dose rate is higher until $332.6 \text{ nGy}\cdot\text{h}^{-1}$. In fact, plant growth rate and functional properties are affected by phosphorus limitation in ecosystems [25]. Besides, the concentration of C, N and P are positive correlated with each other significantly, and shows the basic nutrients required by plants have not changed with varied gamma radiation. Therefore, the N/P ratio is lower with greater radiation dose rate, which indicates the phosphorus limitation of plants has

been alleviated until $332.6 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate, and it is caused by the increasing of plant nutrition acquisition. However, under $>332.6 \text{ nGy}\cdot\text{h}^{-1}$ radiation dose rate, the concentration of N and P are smaller and N/P ratio is getting higher with greater radiation dose rate. These may indicate that the high-intensity gamma radiation is harmful to plant

Conclusions

Long-term gamma radiation could affect the functional traits significantly. Gamma radiation is disadvantageous to plant size. The plants could adapt low-intensity gamma radiation ($<332.6 \text{ nGy}\cdot\text{h}^{-1}$), and low-intensity gamma radiation is benefit for plant water use efficiency, growth and nutrition acquisition of roots. Furthermore, low-intensity gamma radiation could alleviate phosphorus limitation of plants. However, high-intensity gamma radiation ($>528.7 \text{ nGy}\cdot\text{h}^{-1}$) is harmful for the functions of plants greatly, making the plant in an unhealthy state. The functional traits could be important indicators to estimate the affections of long-term gamma radiation.

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Conflict of Interest

The authors declare no conflict of interest.

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Supplementary Material

Table S1. Carbon and nitrogen contents of the soil samples.

Radiation dose rate (nGy•h ⁻¹)	Sample	Nitrogen content (%)	Carbon content (%)
910.964±41.09	1	0.31576	5.45736
	2	0.31556	5.43598
	3	0.31732	5.45629
	4	0.31798	5.46954
	5	0.31764	5.45628
	Average value	0.31685	5.45509
609.745±13.45	1	0.32657	6.67459
	2	0.32635	6.67236
	3	0.32774	6.67605
	4	0.32869	6.67695
	5	0.32744	6.66532
	Average value	0.32736	6.67305
528.692±11.27	1	0.30251	5.96006
	2	0.29287	5.93259
	3	0.29268	5.95512
	4	0.29146	5.95349
	5	0.29376	5.93495
	Average value	0.29466	5.94724
403.292±14.55	1	0.30587	5.41004
	2	0.30665	5.40365
	3	0.30746	5.40314
	4	0.30699	5.40329
	5	0.30726	5.40264
	Average value	0.30685	5.40455
332.596±10.47	1	0.31256	5.89656
	2	0.31458	5.86595
	3	0.31963	5.90213
	4	0.31794	5.90691
	5	0.30967	5.87566
	Average value	0.31488	5.88944
192.906±5.05	1	0.32015	5.63421
	2	0.31967	5.59671
	3	0.31645	5.62546
	4	0.32158	5.61598
	5	0.31879	5.62148
	Average value	0.31933	5.61877