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Effect of Elevated Ultraviolet-B Radiation on Microbial Biomass Carbon and Nitrogen in Barley Rhizosphere Soil

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Abstract As one of the important problems in global change, elevated ultraviolet-B (UV-B) radiation induced by the depletion of stratospheric ozone layer has received more and more attentions around the world. Field experiment with barley was conducted to investigate the effects of elevated UV-B radiation on microbial biomass carbon and nitrogen in rhizosphere and nonrhizosphere soil. The experiment was designed with two UV-B radiation levels, i.e., elevated (E, simulating 25% stratospheric ozone depletion) and ambient (A, control), and performed at the Station of Agricultural Meteorology, Nanjing University of Information Science and Technology, Nanjing, China. Compared with the control, elevated UV-B radiation significantly depressed shoot biomass by 13.2–42.6% and root biomass by approximately 50% from jointing to ripening stage. Elevated UV-B radiation significantly increased microbial biomass C and N in nonrhizosphere soil in most cases, but significantly decreased microbial biomass C and N in rhizosphere soil. Further researches are needed to elucidate whether the above findings are connected with the changes in composition and amount of root exudates induced by elevated UV-B radiation, which can mainly affect the dynamics of soil microbial biomass.

Keywords UV-B radiation · Barley · Rhizosphere · Microbial biomass C and N

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1 Introduction

The stratospheric ozone layer formed a protective atmospheric filter to avoid biologically harmful solar ultraviolet (UV) radiation. Anthropogenic releases of chlorofluorocarbons and nitrogen oxides resulted in ozone depletion. Accordingly, elevated levels of UV-B radiation (280~320 nm) had been detected since early 1980s. The elevated UV-B radiation induced damage to various plant processes, including growth inhibition, lipid peroxidation, photosynthetic depression, ultrastructural change, and decrease in crop yield (Barsig and Malz, 2000; Björn, 2002; Jordan, 2002; Teramura, 1983). These reports were mainly concerned with the ecological and physiological processes in aboveground part of plant (shoot). In contrast, few reports have been

available regarding the effect of elevated UV-B radiation on belowground part of plant (root), especially ecological processes in rhizosphere soil under field conditions (Gallo et al., 2006).

The rhizosphere is first of all a unique hot spot in the soil at the viewpoint of microbial ecology as soil microorganisms are considerably stimulated in the vicinity of the roots, as a consequence of root exudates containing a range of C-compounds (Jones et al., 2004). Some researchers reported that elevated UV-B radiation significantly decreased the quantity of bacteria, actinomyces and fungi in the rhizosphere soil of spring wheat (Li et al., 1999). UV-B radiation could not penetrate into the soil, and had no direct effects on microorganisms in rhizosphere soil (Moorhead and Callaghan, 1994). Therefore, the effect of UV-B radiation on microorganisms in rhizosphere soil should be indirect likely through root exudates or root debris as UV-B radiation largely affected root growth in barley and maize (Wu et al., 2007; Wu et al., 2010). Roughly 30–60% of the net photosynthesized C was allocated below ground, and as much as 40–90% of this fraction entered the soil in the forms of root exudates, mucilage, sloughed-off cells, and decaying roots (Lynch and Whipps 1990).

As decomposers in terrestrial ecosystem, microorganisms directly influence the turnover of organic matter and nutrient mineralization. However, little attention has been paid to dynamics of microbial biomass C and N in rhizosphere soil under elevated UV-B radiation. Barley is the fourth most important worldwide cereal crop, after wheat, maize, and rice; but very few studies have been conducted concerning its response to elevated UV-B radiation. The purposes of this study were to investigate the response of barley growth (shoot and root) as well as microbial biomass C and N to elevated UV-B radiation in rhizosphere soil under field conditions.

2 Materials and Methods

2.1 Experimental Setup

The field experiment with barley (namely cultivar Dan 2) was conducted from November 2008 to May 2009 at the Station of Agricultural Meteorology (32°14′N, 118° 42′E), Nanjing University of Information Science and Technology, Nanjing, China. The tested soil was classified as a Typic Stagnic Anthrosol (Soil Taxonomic Classification Research Group of China, 1993). The soil contained total organic C of 20.1 g kg^{-1}, total N of 1.52 g kg^{-1}, pH of 6.25 (1:1, soil/water ratio), clay content of 27.5% (<1 μm). Total carbon was determined using dichromate oxidation, and total N with Kjeldahl method. Soil pH (1:1 soil–water paste) was measured with electrometry (pH electrode), and clay content with pipette method (Page et al., 1982). The treatments consisted of two UV-B radiation levels, i.e., elevated (E) and ambient (A, control). Each treatment was replicated three times.

Elevated UV-B level was supplied with UV tubes made of quartz glass (HD 40 W, Hude Light Ltd., Shanghai, China). The radiation emitted by the UV tubes was in the spectrum of UV-B from 280 to 320 nm with a peak value of 300 nm. The tubes were fixed on the top of adjustable frames (2×2×2 m) made of steel pipe. The height of the UV tubes was adjusted weekly to maintain a distance of 80 cm above barley canopy. Elevated UV-B level was treated for 8 h daily (8:00–16:00) excluding cloudy and rainy days. The UV-B radiation from the tubes was determined with a radiometer (BNU297, Beijing, China) and weighted with the generalized plant action spectrum (Caldwell, 1971) normalized to 300 nm to obtain the biologically effective UV-B radiation. The biologically weighted UV-B dosage was 14.4 kJ m^{-2} d^{-1}. The dosage corresponded to approximately 25% stratospheric ozone depletion in Nanjing, China (32° 3′ N, 118° 46′ E, 8.9 m) using the model of Green (1983).

In the treatment of ambient UV-B radiation, non-burning UV-B tubes were placed on the top of the frames to make shade, as in the elevated UV-B treatment. Thus, the visible light environment was similar under ambient and elevated UV-B radiations. Shading from the tubes, tube supports, and frames was measured using a ceptometer (LP-80, Decagon, USA). On a clear day, with maximum shading (i.e., with low zenith angle), the canopy received about 90% of the PAR (photosynthetically active radiation) detected above the frames.

2.2 Field Management, Sampling, and Analysis

Barley seeds were sterilized with 10% H_2O_2 solution (*v*/*v*) for 5 min, washed with deionized water, and then germinated at 25°C in the dark. After germination, the seeds were sowed into field at a row spacing of 0.20 m.

The fertilizers of nitrogen (N, as urea), phosphorus (P, as potassium dihydrogen-phosphate) and potassium (K, as potassium chloride) were applied at a rate of 150 kg N ha^{-1}, 150 kg P_2O_5 ha^{-1}, and 150 kg K_2O ha^{-1}, respectively. Seventy percent of total N fertilizer, all P and K fertilizers were applied as basal fertilizers before sowing; the residual N was added as topdressing at tillering and booting stages. During barley-growing period (month/day: 4/1, 4/17, 4/29, 5/5, 5/10 corresponding to tillering, jointing, booting, heading, ripening, respectively), plants were sampled and separated into shoot and root, oven dried for 4 days at 70°C, and weighed to obtain dry matter biomass.

Simultaneously, soil samples in rhizosphere and nonrhizosphere were collected to measure microbial biomass C and N using the fumigation–extraction method (Brookes et al., 1985; Vance et al., 1987). The soil samples in nonrhizosphere were collected from 0 to 15 cm depth; and five cores per plot were used to make a composite sample. Four hills per plot (with five plants per hill) were used for a composite sample of the rhizosphere soil. To collect rhizosphere soil, the plants were shaken to remove the excess soil. The soil tightly adhering to the roots was taken by hand and regarded as rhizosphere soil. Soil samples were homogenized and sieved to remove possible root particles and stones.

2.3 Statistical Analysis

Statistical analysis was performed using SPSS software (SPSS Inc., Chicago, IL, USA). The significant differences in all parameters were estimated by independent samples *t* test at $P<0.05$.

3 Results and Discussion

3.1 Shoot and Root Biomass under Elevated UV-B Radiation

Compared with control, elevated UV-B radiation significantly depressed the biomass of shoot and root in barley, especially more reduction in root biomass at $P<0.05$ level. Under elevated UV-B, root biomass was reduced by approximately 50%. In contrast, shoot biomass was decreased by 13.2–42.6% from jointing to ripening stage (Fig. 1). Some researchers reported similar results, indicating that elevated UV-B significantly reduced leaf, stem, and root biomass in soybean (Feng et al., 2003). Our findings implied that root growth was more sensitive to elevated UV-B than shoot growth in barley. Extensive studies confirmed that elevated UV-B lowered plant height, leaf area, chlorophyll a/b content, and photosynthetic rate, which accounted for decreases in the biomass of shoot and root under elevated UV-B (Kakani et al., 2003). More decrease in root biomass was likely related to less supply of photosynthate or lower root activity induced by elevated UV-B radiation (Wu et al., 2007). However, further researches are needed to elucidate the mechanism for more decrease in root biomass under elevated UV-B radiation.

3.2 Microbial Biomass C Under Elevated UV-B Radiation

Microbial biomass C was significantly increased in the nonrhizosphere soil under the elevated UV-B treatment, especially at tillering, booting, and ripening

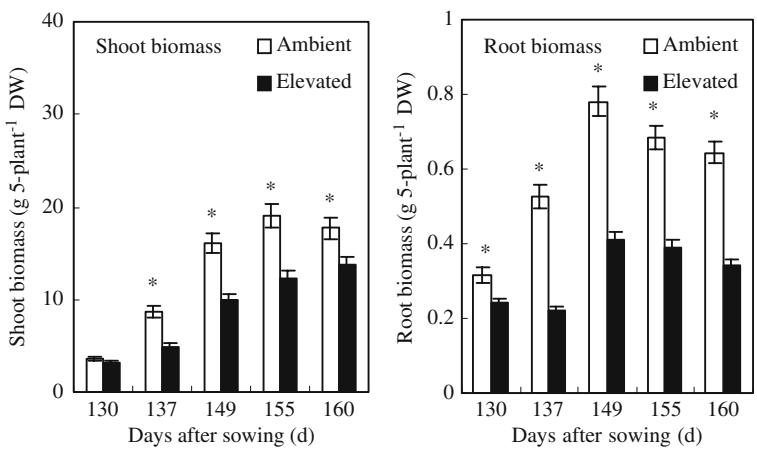

Fig. 1 Effects of elevated UV-B radiation on shoot and root biomass in barley. Values are the mean of triplicates. "Ambient" and "Elevated" represent ambient UV-B and elevated UV-B radiation treatments, respectively. *Vertical bars* indicate the standard errors of the means. *Asterisks* indicate significant differences between ambient and elevated UV-B radiation by independent samples *t* test at $P<0.05$

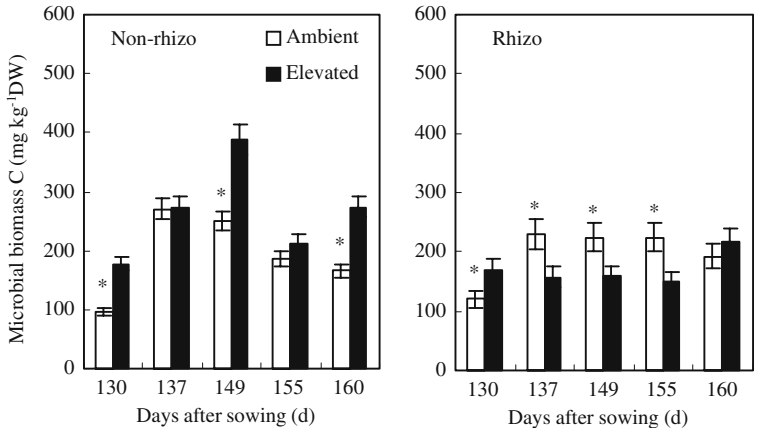

Fig. 2 Effects of elevated UV-B radiation on microbial biomass C in nonrhizospheric and rhizospheric soil with barley. Values are the mean of triplicates. "Ambient" and "Elevated" represent ambient UV-B and elevated UV-B radiation treatments, respectively. *Vertical bars* indicate the standard errors of the means. *Asterisks* indicate significant differences between ambient and elevated UV-B radiation by independent samples *t* test at $P<0.05$

stages ($P<0.05$). Due to low penetration into soil, elevated UV-B had no direct effect on microbes in soil. However, elevated UV-B significantly depressed barley root biomass (Fig. 1), root length, root volume, and root activity, and further lowered root ability to uptake nutrients and thus resulted in nutrient accumulation in nonrhizosphere, which was beneficial for microbial activity (Li et al., 1999; Yue et al., 1998; Wu et al., 2007). Under elevated UV-B, microbial biomass C in nonrhizosphere soil increased from tillering stage, reached the highest value at booting stage, and followed by an obvious decrease at heading stage, and then an increase at ripening stage. Under ambient UV-B, microbial biomass C in nonrhizosphere soil evidently increased from tillering to jointing stage, and then decreased until ripening stage (Fig. 2).

In contrast, elevated UV-B radiation significantly decreased the content of microbial biomass C in rhizosphere soil in most cases (jointing to heading), excluding tillering and ripening stages. One reason is that elevated UV-B depressed photosynthesis and lowered plant growth (Fig. 1); thus, reduced the supply of organic substances to rhizosphere through root exudation, and finally decreased active C in rhizosphere. Another reason is that UV-B radiation promoted root to exude more momilactone B, which was able to depress microbial growth (Noguchi et al., 2007). Microbes in rhizosphere were mainly controlled by root exudation. The above explanations can be the reasons for decrease in microbial biomass C in rhizosphere under elevated UV-B. Higher content of microbial biomass C at tillering stage under elevated UV-B was related to that with temperature increasing after winter, root growth, and activity was promoted, thus dead microbial C caused by low temperature in winter began to release for microbial utilization (Liu et al., 2003). Under elevated UV-B, microbial

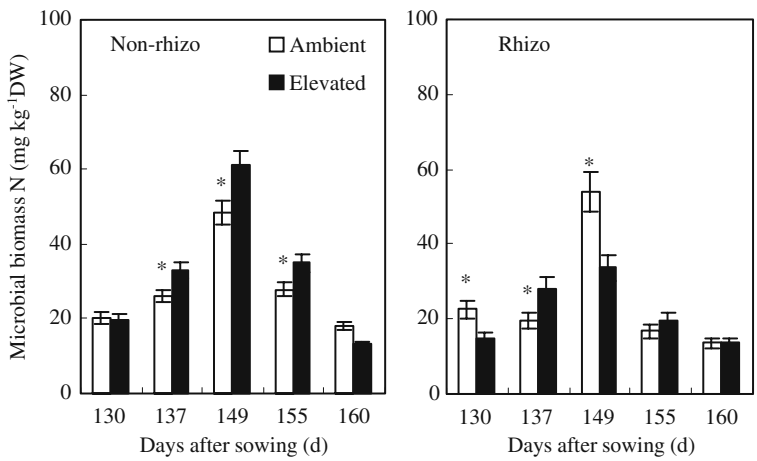

Fig. 3 Effects of elevated UV-B radiation on microbial biomass N in nonrhizospheric and rhizospheric soil with barley. Values are the mean of triplicates. "Ambient" and "Elevated" represent ambient UV-B and elevated UV-B radiation treatments, respectively. *Vertical bars* indicate the standard errors of the means. *Asterisks* indicate significant differences between ambient and elevated UV-B radiation by independent samples *t* test at $P<0.05$

biomass C in rhizosphere soil slightly decreased from tillering to heading stages, and then increased at ripening stage. Under ambient UV-B, the content of microbial biomass C in rhizosphere soil clearly increased from tillering stage, and almost maintained the top value until heading stage, and then slightly lowered at ripening stage (Fig. 2). With barley vigorous growth from tillering stage, more active C needed to build microbial body. Accordingly, microbial biomass C decreased from tillering to heading stages. With barley growing old at ripening stage, active organic C accumulated in soil by root exudation and debris was refixed into microbial body, which resulted in increase in microbial biomass C (Fig. 2).

3.3 Microbial Biomass N under Elevated UV-B Radiation

Microbial biomass N in nonrhizosphere soil enhanced from tillering stage and reached the summit at booting stage, and then decreased until ripening stage. N supply as topdressing accounted for highest content of microbial biomass N through stimulating microbial N fixation at booting stage. At jointing, booting, and heading stages, the content of microbial biomass N in nonrhizosphere soil was significantly higher under elevated UV-B than that under ambient UV-B (Fig. 3), which was related to that as described in explaining microbial biomass C in nonrhizosphere soil (Fig. 2).

Under elevated UV-B, microbial biomass N in rhizosphere soil enhanced from tillering stage and arrived at the peak value at booting stage, and then reduced until ripening stage. In contrast, under ambient UV-B, microbial biomass N in rhizosphere soil slightly decreased from tillering to jointing stage, followed by an obvious increase and reached the highest value at booting stage, and then dropped until ripening stage. Compared with control, elevated UV-B significantly depressed the content of microbial biomass N in rhizosphere soil at tillering and booting stages, but significantly stimulated the content at jointing stage at $P<0.05$ level. In general, elevated UV-B decreased microbial biomass N in rhizosphere soil by 14.6% during the entire growth period (Fig. 3). The reasons were attributed to the competition for N between soil microbes and plant as well as root exudation as affected by elevated UV-B radiation, which was similar to that as described above (Fig. 2).

4 Conclusion

In this study, elevated UV-B radiation generally depressed the contents of microbial biomass C and N in rhizosphere soil, but stimulated the contents in nonrhizosphere in most cases, which was related to decrease in shoot and root biomass induced by elevated UV-B. The change in soil nutrients induced by soil microbial biomass perhaps directly affects the growth of aboveground and underground parts of crops. However, further researches are needed to elucidate whether the above findings are connected with the changes in composition and amount of root exudates, which is dominant factor in affecting microbial dynamics, as affected by elevated UV-B radiation. The change in microbial communities of the rhizospheric soil due to elevated UV-B radiation needs to be taken into consideration in the future.

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References

Barsig, M., & Malz, R. (2000). Fine structure, carbohydrates and photosynthetic pigments of sugar maize leaves under UV-B radiation. *Environmental and Experimental Botany, 43*, 121–130.

Björn, L. O. (2002). Effects of ultraviolet-B radiation on terrestrial organisms and ecosystems with special reference to the Arctic. In D. Hessen (Ed.), *UV radiation and arctic ecosystems*. Berlin: Springer.

Brookes, P. C., Landman, A., & Pruden, G. (1985). Chloroform fumigation and the release of soil nitrogen: a rapid direct extration method to measure microbial biomass nitrogen in soil. *Soil Biology and Biochemistry, 17*, 837–842.

Caldwell, M. M. (1971). Solar ultraviolet radiation and the growth and development of higher plant. In A. C. Giese (Ed.), *Photophysiology* (pp. 131–137). New York: Academic.

Feng, H., An, L., Chen, T., & Qiang, W. (2003). The effect of enhanced ultraviolet-B radiation on growth, photosynthesis and stable carbon isotope composition of two soybean cultivars under field conditions. *Environmental and Experimental Botany, 49*, 1–8.

Gallo, M. E., Sinsabaugh, R. L., & Cabaniss, S. E. (2006). The role of ultraviolet radiation in litter decomposition in arid ecosystems. *Applied Soil Ecology, 34*, 82–91.

Green, A. E. S. (1983). The penetration of ultraviolet to the ground. *Physiologia Plantarum, 58*, 351–359.

Jones, D. L., Hodge, A., & Kuzyakov, Y. (2004). Plant and mycorrhizal regulation of rhizodeposition. *The New Phytologist, 163*, 459–480.

Jordan, B. R. (2002). Molecular response of plant cells to UV-B stress. *Functional Plant Biology, 29*, 909–916.

Kakani, V. G., Reddy, K. R., Zhao, D., & Sailaja, K. (2003). Field crop responses to ultraviolet-B radiation: a review. *Agricultural and Forest Meteorology, 120*, 191–218.

Li, Y., Yang, J., Wang, X., & Hu, Z. (1999). The effect of UV-B radiation on the population quantity of microorganisms in rhizosphere soil of spring wheat. *China Environmental Science, 19*(2), 157–160.

Liu, M., Hu, F., & He, Y. (2003). Seasonal dynamics of soil microbial biomass and its significance to indicate soil quality under different vegetations restored on degraded red soils. *Acta Pedol Sin, 40*(6), 937–944.

Lynch, J. M., & Whipps, J. M. (1990). Substrate flow in the rhizosphere. *Plant and Soil, 129*, 1–10.

Moorhead, D. L., & Callaghan, T. V. (1994). Effects of increasing UV-B radiation on decomposition and soil organic matter dynamics: a synthesis and modeling study. *Biology and Fertility of Soils, 18*, 19–26.

Noguchi, K., Kujimeh, H., & Takeshi, I. (2007). UV-induced momilactone B accumulation in rice rhizosphere. *Journal of Plant Physiology, 164*(11), 1548–1551.

Page, A. L., Miller, R. H., & Keeney, D. R. (1982). *Methods of soil analysis, part 2—chemical and microbiological properties* (2nd ed., pp. 643–655). Madison, WI, USA: American Society of Agronomy, Inc.

Soil Taxonomic Classification Research Group of China. (1993). *Chinese soil taxonomic classification* (1st ed., p. 112). Beijing: Science Press.

Teramura, A. H. (1983). Effects of ultraviolet-B radiation on the growth and yield of crop plants. *Physiologia Plantarum, 58*, 415–427.

Vance, E. D., Brookes, P. C., & Jenkinson, D. S. (1987). An extration method for measuring soil microbial biomass C. *Soil Biology and Biochemistry, 19*, 703–707.

Wu, R., Zheng, Y., Wang, C., & Hu, Z. (2007). Effects of enhanced UV-B radiation on the growth of aerial parts and root of maize. *Ecology and Environment, 16*(2), 323–326.

Wu, J., Lou, Y., Li, Y., & Cheng, H. (2010). Effect of enhanced ultraviolet-B radiation on physiological and ecological parameters in barley. *Journal of Agro-Environmental Science, 29*(6), 1033–1039.

Yue, M., Li, Y., & Wang, X. (1998). Effects of enhanced ultraviolet-B radiation on plant nutrients and decomposition of spring wheat under field conditions. *Environmental and Experimental Botany, 40*, 187–196.