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Full Length Research Paper

The effect of ultraviolet radiation on water-logging resistance in Tibetan peach

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The effect of ultraviolet (UV) radiation on the water-logging resistance of Tibetan peach (*Amygdalus mira* Koehne) remains unclear. In this study, Tibetan peach seedlings were subjected to 9-days of UV-B (280 - 320 nm) supplementation, water-logging or the combination of both, and the growth indicated by leaf number, net photosynthetic rate and stomatal conductance were monitored. In addition, the activities of protection enzymes SOD and POD, as well as the concentrations of hormones ABA, ZR, GAs and ZR in leaves were examined. The results show that UV-B or water-logging or the combination of both factors restrained the growth trend, net photosynthetic rate and stomatal conductance. Both UV-B and water-logging increased SOD activity, and synergistically led to a drastic increase in POD activity. Furthermore, UV-B and water-logging condition increased the concentrations of ABA, ZR and GAs, while it decreased ZR. The results suggest that the changes of hormone concentrations in Tibetan peach leaves may explain the increased activities of protection enzymes and decreased photosynthesis under water-logging condition.

Key words: Tibetan peach, water-logging, UV-B, photosynthesis, protection enzymes, hormones.

INTRODUCTION

Tibetan peach (Amygdalus mira Koehne), as one of the original wild fruit tree germplasm resources, is widely distributed in the Tibet Plateau of China（Wang et al., 1997). Many studies show that peaches are highly susceptible to water-logging and prefer perfect drainage (Chen and Liu, 2002); however, the Tibetan peach can survive in places close to a river or grow in a water-logged area in healthy condition. Undoubtedly, the long period of evolution may be the reason for this special resistance. Among all the environmental factors, higher ultraviolet-B radiation on the Tibet Plateau may be the most important one to form this special resistance.

In the past few decades, stratospheric ozone reduction caused by trace gases such as chlorofluocarbons (CFCs) and NOx has resulted in increasing UV-B (280 - 315 nm) radiation reaching Earth's surface. Previous studies indicated that a wide range of biochemical, physiological, morphological, anatomical and growth responses of plants are attributed to elevated UV-B radiation and the effects of enhanced UV-B radiation on Earth's ecosystems might well continue in the coming decades (Caldwell et al., 2007; Feng et al., 2000; Zhang et al., 2003).

In addition, under high dose of UV-B radiation, plants usually encounter several stresses simultaneously under field conditions. The stresses may cause a variety of plant responses that can be additive, synergistic or antagonistic (Alexieva et al., 2001). Owing to the commonalities among stress responses, plants may activate similar defense systems to reduce cellular damages and a phenomenon known as cross-tolerance may occur (Hideg et al., 2003; Stratmann, 2003). Researchers also found that UV-B radiation can decrease the sensitivity of plant to other stress environment and relieve the damaging effects (Krause et al., 2003; Balakumar et al., 1993; Petropoulou et al., 1995; Manetaset al., 1997).

As an original plant resource, Tibetan peach has been

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Abbreviations: ABA; abscisic acid, **GA;** gibberellic acid, **IAA;** indole-3-acetic acid, **ZR;** zeatin riboside, **SOD;** superoxide dismutase, **POD;** peroxidase.

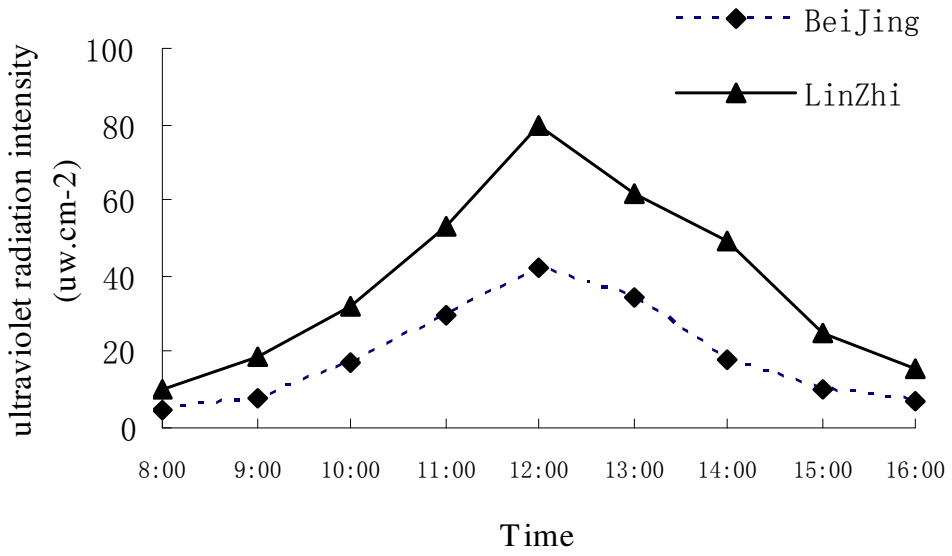

Figure 1. Diurnal changes of UV-B radiation in different area.

living in Tibet tableland for thousands of years. In such a long period, whether the high dose of UV-B radiation has any beneficial effects on the water-logging resistance is still uncertain. In this experiment, Tibetan peach seedlings were treated with water-logging, diurnal change of UV-B radiation simulating the dose of UV-B radiation in Tibet and the combination treatment of both, the changes of biological and physiological parameters under these conditions were investigated.

MATERIALS AND METHODS

Plant materials

The seeds of Tibetan peach were acquired from Linzhi (3,300 m) located in the southeast of the Tibet tableland of China. The healthy seedlings with uniform height were chosen and transferred to single flowerpots containing homogeneous natural soil grown in a greenhouse. The air temperature and relative humidity were 20-25 /10-15℃ (day/night) and 40 - 60%, respectively. 30 seedlings were selected for each group.

Experimental design

Four treatments were employed in our study: (1) No UV-B supplementation and well-watered condition (CK); (2) UV-B supplementation and well-watered condition (UV); (3) No UV-B supplementation and water-logging condition (W); (4) UV-B supplementation and water-logging condition (UV*W). Ambient UV-B radiation was set according to the diurnal change of UV-B radiation in Beijing. UV-B supplementation was set according to the diurnal change of UV-B radiation in Linzhi in Tibet. UV-B radiation in Beijing and Linzhi is illustrated in Figure 1.

UV-B radiation (280 - 320 nm) was provided by UV-B Lamps (Beijing Normal University, Beijing, China). Four 40 W fluorescence sun lamps were added to form the ambient environment; two UV-B Lamps and four 40 W fluorescence sun lamps were used for the treatment. The lamps were suspended 60 - 80 cm above the plants and the height was adjusted every hour to simulate the diurnal change of UV-B radiation above the plants. The lamps were wrapped with 0.13 mm thick cellulose diacetate film which absorbs radiation below 290 nm. The intensity was measured by a UV-B detection instrument. The seedlings of Tibetan peach were treated for 6 hours every day (09:00 - 15:00).

In the well-watered treatment, all pots were watered to 100% of field capacity by supplying an amount of water equal to the transpiration losses and the soil water content was always kept at 33.0 %. In the water-logging treatment, all pots were submerged by water. Evaporation from the soil surface was prevented by enclosing the pots in plastic bags that were tied to the stems of the plants. Transpiration water loss was measured gravimetrically by weighing all pots every four days.

Photosynthesis measurement

Stomatal conductance and net photosynthetic rate for leaves of Tibetan peach seedlings were measured by the analysis system (LI-6400, LI-COR Biosciences, Lincoln, NE). Measurements were made at 10:00 am. The fifth to the eighth leaves from the top of each seedling were collected for analysis.

Hormone measurement

The fourth leaf from the top of each seedling was collected for analysis in the UV-B treatment. Extraction and determination of hormones IAA, GAs, ZR and ABA were performed as described by Li et al. (2003).

SOD and POD measurement

SOD and POD enzyme activities were assayed in the supernatant from leaves of each treatment according to Abassi et al. (1998).

Statistical analysis

Two-way ANOVA was used in the study. Values presented are

Table 1. The effects of water-logging and UV-B radiation on growth properties.

Treatment	Plant height (cm)		Leaf number	
	0 d	9 d	0 d	9 d
CK	15.4 ± 0.9 (100)a	19.8 ± 1.8 (128.6)a	10.1 ± 2.0 (100)a	14.7 ± 1.9 (145.5)a
UV-B	15.3 ± 0.3 (100)a	18.5 ± 1.2 (121.2)b	9.9 ± 1.6 (100)a	14.5 ± 1.1 (146.5)a
W	15.3 ± 0.8 (100)a	18.6 ± 1.6 (120.8)b	10.2 ± 1.9 (100)a	14.1 ± 2.6 (140.6)a
UV*W	15.3 ± 0.5 (100)a	18.3 ± 1.3 (119.8)b	9.7 ± 1.8 (100)a	14.2±1.9 (146.4)a

Values presented are means of 6 replicates ± SD. Values marked with different letters are significantly different according to Students t- test ($p < 0.05$).

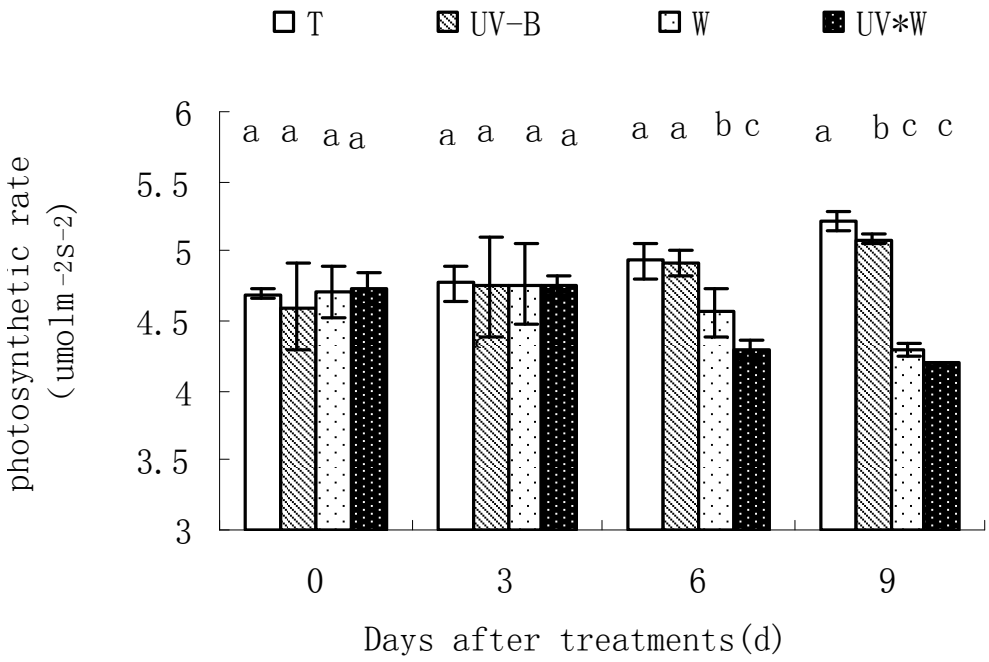

Figure 2. The effects of water-logging and UV-B radiation on net photosynthetic rate. The bars with different letters are significantly different from each treatment ($p < 0.05$). Values are means of six replicates.

means of 6 replicates ± SD. Values marked with different letters are significantly different according to Students t- test ($p < 0.05$).

RESULTS

Water-logging and UV-B radiation retard growth

As shown in Table 1, after 9 days of treatment under UV-B radiation, water-logging or in combination, the plant height of Tibetan peach seedlings was clearly constrained in comparison with CK. However, combination treatment of UV-B radiation and water-logging was not significantly different from the separate treatments. In contrast with plant height, the leaf number of Tibetan peach seedlings did not vary significantly under all conditions.

Water-logging and UV-B radiation inhibit photosynthesis and decrease stomatal conductance

As shown in Figures 2 and 3, the net photosynthetic rate and stomatal conductance decreased under all three different stress conditions. The water-logging and combination treatment had a stronger inhibitory effect on the net photosynthetic rate and the stomatal conductance than the UV-B radiation. Nevertheless, there was no significant difference between the water-logging and combination treatment, suggesting that UV-B radiation had no relief effect on the net photosynthetic rate and the stomatal conductance under the water-logging condition.

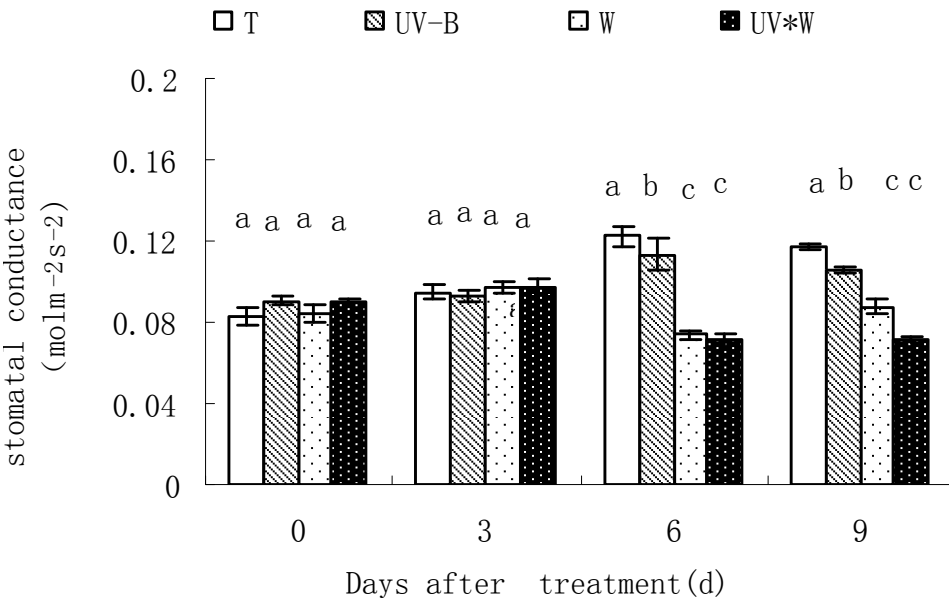

Figure 3. The effects of water-logging and UV-B radiation on stomatal conductance. The bars with different letters are significantly different from each treatment ($p < 0.05$). Values are means of six replicates.

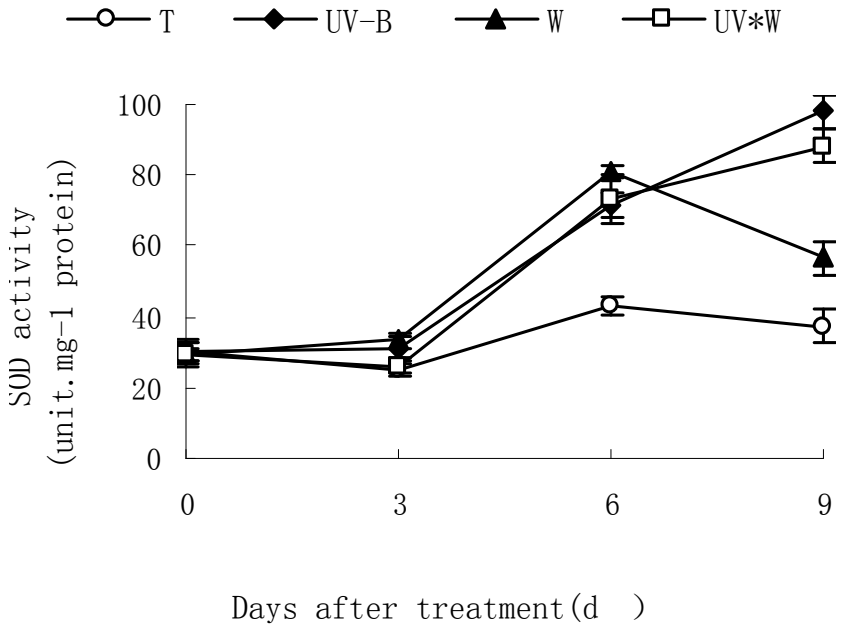

Figure 4. The effects of water-logging and UV-B radiation on SOD activity. Error bars represent standard deviation of the data. Values are means of six replicates.

The effects of water-logging and UV-B radiation on protection enzyme system

As shown in Figures 4 and 5, compared with CK, the protection enzyme activities were strongly influenced by UV-B radiation, water-logging and the combination treatment. SOD activity was significantly stimulated by UV-B radiation and water-logging. However, at the later stage of the experiment, water-logging led to a decrease of SOD activity while UV-B radiation sustained the SOD activity at a higher level. ANOVA analysis of water-logging and the combination showed significant interactive effects of UV-B radiation and water-logging on SOD activities, indicating that the increase in SOD activity was

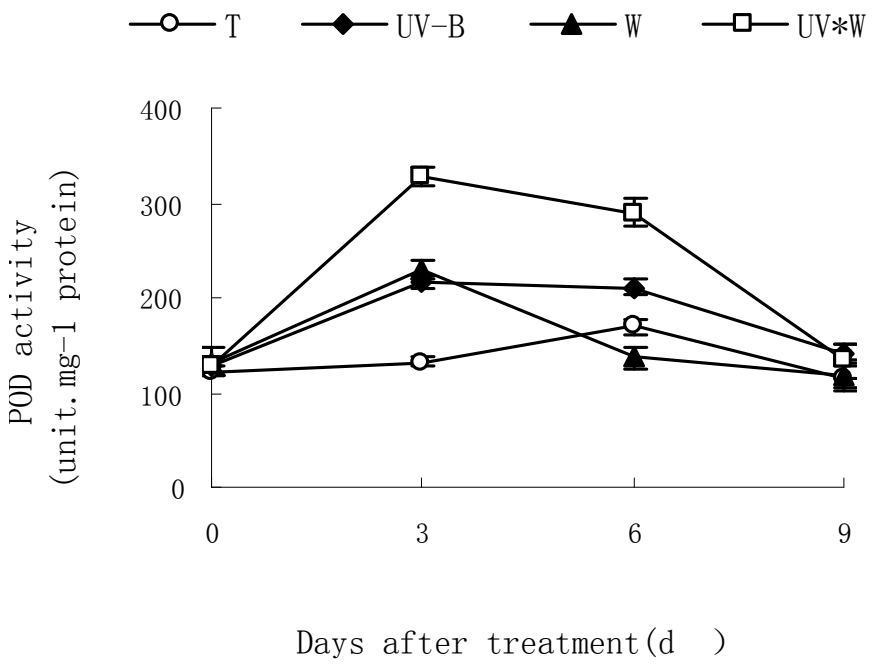

Figure 5. The effects of water-logging and UV-B radiation on POD activity. Error bars represent standard deviation of the data. Values are means of six replicates.

Table 2. The effects of water-logging and UV-B radiation on hormones.

Treatment	IAA (pmol g-1 FW)		ZR (pmol g-1 FW)		GA (pmol g-1 FW)		ABA (pmol g-1 FW)	
	0 d	9 d	0 d	9 d	0 d	9 d	0 d	9 d
CK	31.9 ± 3.1 a	34.7 ± 2.9a	7.5 ± 0.9 a	8.5 ± 2.7 b	18.0 ± 1.2 a	18.5 ± 1.5a	139 ± 10 a	138 ± 9c
UV-B	32.4 ± 1.9 a	27.2 ± 2.1b	7.9 ± 0.9 a	16.6 ± 3.6a	17.9 ± 1.6 a	20.1 ± 2.6a	134 ± 11 a	207 ± 21b
W	31.0 ± 2.2 a	26.2 ± 1.8b	7.5 ± 2.5 a	6.9 ± 1.3 b	17.7 ± 1.2 a	14.1 ± 1.2b	139 ± 16 a	182 ± 23b
UV*W	31.6 ± 1.0 a	21.4 ± 2.4c	7.6 ± 1.6 a	15.9 ± 2.9 a	18.1 ± 1.0 a	20.7 ± 2.0a	141 ± 10 a	236 ± 44a

Values presented are means of 6 replicates ± SD. Values marked with different letters are significantly different according to Students t- test ($p < 0.05$).

heavily due to UV-B radiation.

In comparison with water-logging, the combination treatment resulted in significantly higher POD activity, suggesting that UV-B radiation was the key factor to increase POD activity.

The effects of water-logging and UV-B radiation on plant hormones

Hormone concentrations in the leaves of the Tibet peach seedlings are shown in Table 2. The combination treatment influenced the IAA concentration more than UV-B radiation or water-logging alone, demonstrated by a 32.3% decrease in IAA concentration in contrast of 17.1% decrease and 16.5% decrease, respectively. UV-B radiation and the combination treatment increased the GA concentration by 11.2% and 11.4%, respectively; in contrast, water-logging reduced the GA concentration by 20.4%. The ABA concentration was increased by 50.9% and 31.9%, respectively, in response to UV-B radiation and water-logging treatment. It was also found that the 67.3% increase in the ABA concentration caused by the combination treatment was significantly higher than the single treatment. The ZR concentration increased under UV-B radiation and combination, while there was slight change on the ZR concentration shown by water-logging .The results indicated that the UV-B radiation increased ABA, GA, ZR concentration and decreased IAA concentration in Tibetan peach leaves under water-logging condition.

DISCUSSION

In Tibet tableland, peaches can often be peculiarly found

growing near the river side and water-logging area; whether this kind of special resistance has a correlation with the higher UV-B radiation in Tibet tableland was investigated in this study.

We found that UV-B radiation, water-logging and the combination treatment all inhibited the growth of Tibetan peach seedlings demonstrated by lower plant height. Numerous studies have reported that GA as the antagonist of ABA is important for stimulation and regulation of plant growth (Hoffmann and Kende, 1992; Rademacher and Growth, 2000; Rijnders et al., 1997). In our experiment, GA concentration was decreased only in the water-logging group while there was no significant change in the UV-B radiation and combination treatment groups. However, ABA concentration was increased in all three treatments. As a result, the GA/ABA ratio was decreased in all treatments, which resulted in the inhibitory effects on growth.

Pilet and Saugy, (1987) have suggested that the inhibitory effects on stomatal conductance could be influenced by the ABA/IAA ratio. In our experiment, the ABA/IAA ratio was increased due to the decrease in IAA concentration and increase in ABA concentration, which might explain the result of a lower photosynthetic rate in Tibetan peach leaves (Ellison and Farnsworth, 1997; Pezeshki et al., 1997； Mary et al., 2006).

Over the past 20 years, it has been clearly demonstrated that hormone can exert rapid and specific effects on genes at the molecular level (Karen and John, 1990; Gretchen and Tom, 2002; Esther et al., 2000; Klages et al., 1994). Under the physiological condition, the effective regulation by plant hormone on proteins, especially the protection enzyme system, was thought to be an important strategy on stress resistance. It has been reported that ABA concentration increases under stress condition and consequently induces a variety of the stress proteins including the protection enzymes (Thomashow, 1999; Ingram and Bartels, 1996; Shinozaki and Yamaguchi-Shinozaki, 1997). Similar result was found in our experiment. Additionally, the combination treatment also increased the SOD activity and sustained at a higher level than water-logging. Furthermore, POD activity was also increased by UV-B radiation under water-logging condition, likely due to the decrease in IAA concentration. Numerous experiments have shown that there is a strong negative correlation between the IAA concentration and POD activities (Ros and Tevn, 1995; Huang et al., 1997; Marcel et al., 2001).

In conclusion, the present study suggests that UV-B radiation significantly impacts on the growth and physiological response of Tibetan peach seedlings under water-logging condition. Changes in physiological response are more likely to be the result of changes in hormone concentration or the ratios of different hormones induced by UV-B radiation. Increase in protection enzyme activities is a result of the increase of ABA concentration in leaves, whereas the decrease in photosynthetic rate is due to lower GA concentration and GA/ABA ratio.

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