African Journal of Biotechnology Vol. 11 (82), pp. 14786-14791, 11 October, 2012
Available online at http://www.academicjournals.org/AJB
DOI: 10.5897/AJB12.2020
ISSN 1684-5315 ©2012 Academic Journals

Full Length Research Paper

Effects of day-night supplemental UV-A on growth, photosynthetic pigments and antioxidant system of pea seedlings in glasshouse

Liu Wenke[1,2]* and Yang Qichang[1,2]

[1]Institute of Environment and Sustainable Development in Agriculture, Chinese Academy of Agricultural Sciences, Beijing 100081, China.
[2]Key Laboratory of Energy Conservation and Waste Management of Agricultural Structures, Ministry of Agriculture, Beijing 100081, China.

Accepted 23 July, 2012

A glasshouse experiment was carried out to investigate the effects of day-night supplemental UV-A on growth, photosynthetic pigments and antioxidant system (contents of antioxidant substances and antioxidant enzyme activity) of pea seedlings substrate-cultivated in glasshouse. The results show that supplemental UV-A did not significantly affect shoot and root to shoot ratio of pea seedlings, but root and total biomass were modified. Day supplemental UV-A slightly increased root and total biomass of pea seedlings, while night supplemental UV-A significantly decreased the root and total biomass. Supplemental UV-A did not affect the contents of shoot chlorophyll a and carotenoid of pea seedlings. Nevertheless, day supplemental UV-A significantly decreased chlorophyll b content in shoot of pea seedlings, while night supplemental UV-A slightly decreased shoot chlorophyll b content. In addition, supplemental UV-A did not significantly affect the contents of shoot vitamin C, phenolic compounds and flavonoid of pea seedlings. However, day supplemental 6 h and night supplemental 3 h slightly decreased shoot anthocyanin content of pea seedlings, while night supplemental 6 h significantly reduced shoot anthocyanin content. Finally, supplemental UV-A did not significantly affect shoot peroxidase activity and catalase activity.

Key words: UV-A, photosynthetic pigment, antioxidant enzymatic activity, antioxidant substance, pea seedling.

INTRODUCTION

Nowadays, consumption of sprouts has become popular worldwide because they are often considered as kinds of healthy vegetables containing rich health-promoting phytochemicals with antioxidant properties, such as vitamin C, phenolic compounds, flavonoids and anthocyanin. Notably, sprouts of cruciferous plants such as radish, broccoli and those of legumes such as pea are known for their health-promoting phytochemicals (Plum et al., 1997; Takaya et al., 2003; Zielinski et al., 2007). More importantly, most sprouts are green and agrochemical free foods because they can be cultivated without applying fertilizer and pesticides. Pea seedling, as a kind of green sprout vegetable, is considered to be more health-beneficial and nutritive. Although pea seedling has only ten or dozens of days for growth, light environment is an important and adjustable factor which has potential in regulating yield and nutritional quality (Pérez-Balibrea et al., 2008).

UV-A is a kind of ultravoilet with wavelength range from 320 to 400 nm, which is near to photosynthetic active spectrum band (400 to 700 nm). Additionally, UV-A contributes more than 90% of UV that arrives on earth surface. More importantly, UV-A and UV strength inside protected facilities decrease sharply from ambient values caused by filtration of cover materials. Some morphogenesis and nutritional quality problems occurred

*Corresponding author. E-mail: liuwke@163.com.

Table 1. Treatments designed in the experiment.

Treatment code	Day (D) or night (N)	Treatment time (h)
CK	-	0
D-UV3	D	3
D-UV6	D	6
N-UV3	N	3
N-UV6	N	6

when protected vegetables were cultivated under low UV radiation strength. Therefore, aiming to improve nutritional quality and economic value of protected vegetables, many studies were carried out to investigate the possible beneficial effects of UV-A supplementation on protected horticultural plants through artificially elevating UV radiation level (Klein, 1976, 1990; Tsormpatsidis et al., 2008). Additionally, seedling sprouts or leafy vegetables grown under artificial lighting are severely short of UV-A irradiation. Therefore, UV-A supplementation is more important for these plants. Up to date, the biochemical and biological functions of UV-A on protected horticultural plants have not been clearly investigated. Previous study showed that UV-A was involved in anthocyanin synthesis (Klein, 1976). Klein (1990) examined the effect of UV-A radiations on the concentration of floral petal anthocyanin of flowering plants. However, no tested plant under supplementary UV-A radiation was enhanced in floral anthocyanin content. Li and Kubota (2009) reported that anthocyanin concentration of lettuce under white fluorescent lamps as the main light source increased by 11% with supplemental UV-A. Voipio and Autio (1995) reported increases in anthocyanin content of lettuce plants under supplementary UV-A radiation. So far, many previous studies have showed the effects of supplemental UV-A on accumulation of health - promoting phytochemicals of vegetables in chamber with artificial light or greenhouse (Voipio and Autio, 1995; Tsormpatsidis et al., 2008; Li and Kubota, 2009). However, little information has been revealed about the responses of photosynthetic pigments and antioxidant system of vegetables to supplemental UV - A, particularly the difference of day and night supplemental lighting.

Pea seedling is one kind of important sprout vegetables that is popularly consumed worldwide. Pea seedlings are usually cultivated under protected systems with artificial light or sunlight. Generally, light environment, particularly light intensity and spectral quality, is a necessary and crucial factor that is related to growth and quality of pea seedlings during short-term cultivation. Nowadays, effects of spectral quality, such as monochromatic light from light-emitting diodes and ultravoilet radiation on growth and nutritional quality of pea seedlings are not fully investigated. The objectives of this study were to (1) investigate the effects of supplemental UV-A on growth, photosynthetic pigments and antioxidant system (contents of antioxidant substances and antioxidant enzyme activity) of pea seedlings substrate-cultivated in glasshouse; and (2) differentiate diurnal and dose effects on nutritional quality of supplemental UV-A.

MATERIALS AND METHODS

Plant material and growth conditions

Cultivar of pea used in this experiment was ‘Shenchun’. Uniform pea seeds were selected and soaked in tap water for 40 min and then sown in plug tray with six holes (filled with vermiculite). Three seeds were planted in each round hole (top diameter=5 cm, height=5 cm). Plug trays were partly (about 0.5 cm) immersed into distilled water for germination and growth.

Experimental design and UV-A treatments

Five treatments were designed (Table 1), including control without UV-A treatment, day supplemental UV-A treatments and night supplemental UV-A treatments. Day and night treatments included two treatment intensities, that is, 3 h (day 9 to 12 and night 20 to 23 o’clock) and 6 h (day 9 to 15 and night 20 to next day 2 o’clock), respectively (Table 1). Each treatment was replicated thrice. The pea seeds were planted on 14 October, 2011, and UV-A treatments started on 18 October, 2011. UV-A lamps was positioned 5 cm above pea seedlings. Every two days, the positions of various treatments were changed randomly. 30 W UV-A lamp (main wavelength is 365 nm) was used, and light intensity 5 cm below ultraviolet lamp was 208 μW/cm^2 or 31.2 μmol/m^2/s using UV ultraviolet radiometer (3414 F made by Fieldscout). After ten days’ cultivation, pea seedlings were harvested and sampled. During cultivation, the treatment positions of replicates and treatment pots under ultravoilet lamps were regularly changed each day. Pea seedlings were cultivated in a glasshouse, and day changes of photosynthetically active radiation (PAR) and UV radiation in the glasshouse are as shown in Figures 1 and 2.

Determination methods

Pea seedlings were randomly sampled for determining fresh shoot and root biomass, contents of chlorophyll and carotenoid, vitamin C, phenolic compounds, flavonoid and anthocyanin as well as peroxidase and catalase activities of shoot. Shoot contents of chlorophyll a, chlorophyll b and carotenoid were determined by colorimetric method after being extracted with 80% acetone solution (Li et al., 2000). Vitamin C content in shoots was 2,6-dichlorophenolindophenol titration (Cao et al., 2007). Phenolic compounds, flavonoid and anthocyanin contents of shoot were measured by colorimetric method after undergoing methanol-HCl extraction (Cao et al., 2007). Phenolic compounds, flavonoid and anthocyanin contents of pea seedling shoot were expressed by absorbance values per gram fresh shoot weight, which was, OD280, OD325, and OD600 to OD530 g^{-1}, respectively. In addition, peroxidase and catalase activities of shoot were determined by colorimetric method as described by Cao et al. (2007). Activities of peroxidase and catalase activities were expressed as u/(g.min) and mg/(g·min), respectively.

Statistical analysis

Data analysis was processed by ANOVA software.

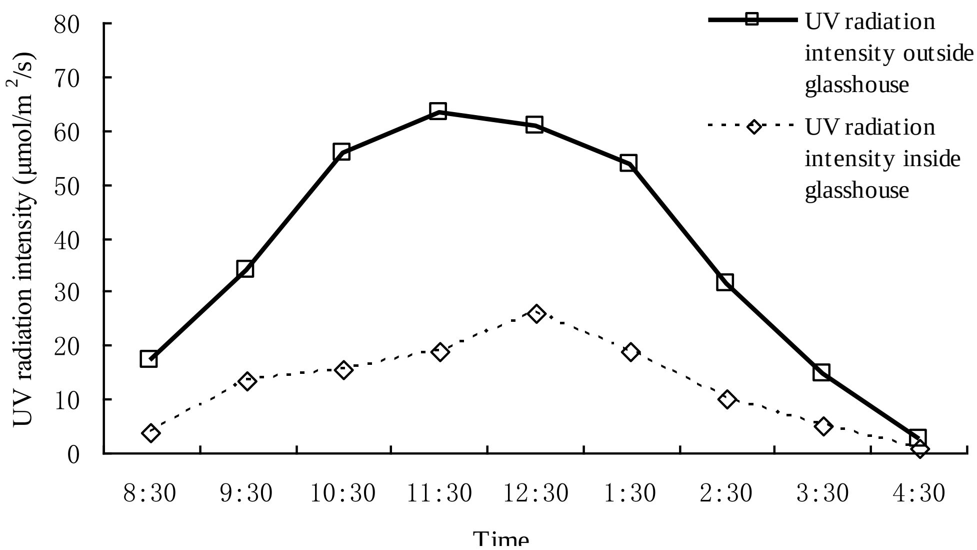

Figure 1. UV radiation intensities inside and outside the experimental glasshouse.

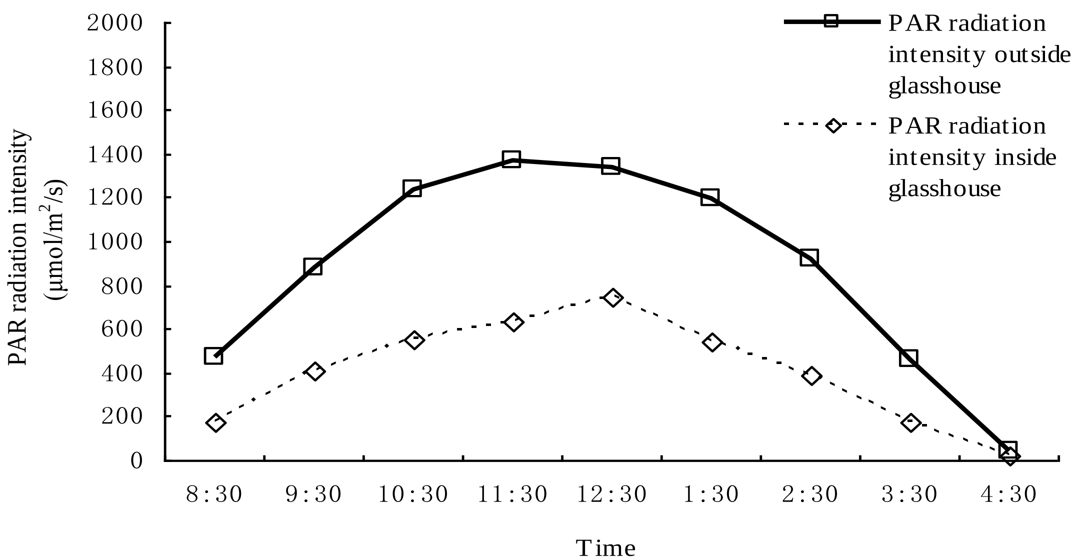

Figure 2. PAR radiation intensity inside and outside experimental glasshouse.

RESULTS

Shoot and root biomass of pea seedlings

Supplemental UV-A did not significantly affect shoot, root to shoot ratio of pea seedlings, whatever the treatment time (day or night treatments) (Table 2). However, root and total biomass were affected. Compared with the control, D-UV3 and D-UV6 treatments slightly increased root and total biomass of pea seedlings, while N-UV3 and N-UV6 treatments significantly decreased the root and total biomass of pea seedlings.

Photosynthetic pigment contents in shoot of pea seedlings

Supplemental UV-A did not significantly affect the contents of shoot chlorophyll a and carotenoid of pea seedlings, whatever the treatment time was (day or night treatments) (Table 3). Nevertheless, supplemental UV-A decreased chlorophyll b content in shoot of pea seedlings. More importantly, day supplemental UV-A significantly decreased chlorophyll b content in shoot of pea seedlings, while night supplemental UV-A slightly decreased shoot chlorophyll b content.

Table 2. Shoot, root and total biomass of pea seedlings of different UV-A supplemental treatments (fresh weight).

Treatment	Shoot biomass (g/pot)	Root biomass (g/pot)	Total biomass (g/pot)	Root to shoot ratio
CK	5.37[a]	8.48[abc]	13.85[ab]	1.58[a]
D-UV3	5.66[a]	8.87[ab]	14.53[a]	1.58[a]
D-UV6	5.50[a]	9.08[a]	14.58[a]	1.66[a]
N-UV3	5.31[a]	7.78[c]	13.09[b]	1.47[a]
N-UV6	5.15[a]	7.89[bc]	13.05[b]	1.54[a]

Table 3. Photosynthetic pigments in shoot of pea seedlings of different UV-A supplemental treatments (fresh weight).

Treatment	Chlorophyll a content (mg/g)	Chlorophyll b content (mg/g)	Carotenoid content (mg/g)
CK	0.91[a]	0.31[a]	0.22[a]
D-UV3	0.85[a]	0.25[b]	0.22[a]
D-UV6	0.85[a]	0.25[b]	0.21[a]
N-UV3	0.88[a]	0.26[ab]	0.21[a]
N-UV6	0.90[a]	0.27[ab]	0.22[a]

Table 4. Contents of antioxidant substances in shoot of pea seedlings of different UV-A supplemental treatments (fresh weight).

Treatment	Vitamin C content (mg/g)	Content of phenolic compound (OD280/g)	Flavonoid content (OD325/g)	Anthocyanin content (OD600-OD530/g)
CK	1.26[a]	2.54[a]	2.53[a]	0.040[a]
D-UV3	1.17[a]	2.48[a]	2.45[a]	0.040[a]
D-UV6	1.09[a]	2.46[a]	2.47[a]	0.037[ab]
N-UV3	1.26[a]	2.53[a]	2.51[a]	0.032[ab]
N-UV6	1.29[a]	2.52[a]	2.49[a]	0.028[b]

Table 5. Shoot antioxidant enzymatic activity of pea seedling of different UV-A supplemental treatments (fresh weight).

Treatment	Peroxidase activity [u/(g·min)]	Catalase activity [mg/(g·min)]
CK	317.54[a]	10.30[a]
D-UV3	328.73[a]	10.75[a]
D-UV6	298.70[a]	13.04[a]
N-UV3	359.50[a]	11.77[a]
N-UV6	457.58[a]	11.33[a]

Contents of antioxidant substances in shoot of pea seedlings

Supplemental UV-A did not significantly affect contents of shoot vitamin C, phenolic coumpounds and flavonoid of pea seedlings, whatever the treatment time was (day or night treatments) (Table 4). However, D-UV6 and N-UV3 treatments slightly decreased shoot anthocyanin content of pea seedlings, while N-UV6 treatment significantly reduced shoot anthocyanin content of pea seedlings.

Shoot antioxidant enzymatic activity of pea seedlings

Supplemental UV-A did not significantly affect shoot peroxidase activity and catalase activity of pea seedlings, whatever the treatment time was (day or night treatments) (Table 5). However, N-UV6 treatment presented the highest shoot peroxidase activity of pea seedlings, while D-UV6 tended to improve shoot catalase activity of pea seedlings compared with control (CK).

DISCUSSION

Green leafy vegetables are a major source of dietary carotenoids which can act as lipophilic antioxidants and reduce the incidence of cataracts and macular degeneration (Moeller et al., 2000). To improve biomass and nutritional quality of protected vegetables, particularly pea seedlings, using light regulation is an efficient method revealed by previous reports (Wu et al., 2007). Nowadays, UV-A, UV-B and LED light qualities have been extensively investigated about their influence on growth and nutritional quality of protected vegetables (Li and Kubota, 2009; Wu et al., 2007). The present data showed that root biomass response to day-night UV-A supplementation is different. Compared with the control without UV-A treatment, D - UV3 and D - UV6 treatments slightly increased root and total biomass of pea seedlings, while N-UV3 and N-UV6 treatments decreased significantly the root and total biomass of pea seedlings. There are two possible reasons that should be attributed to water content variation in root biomass of different treatments caused by UV - A addition; and photosynthetic variation arisen by UV - A supplementation. Without background PAR and UV radiation presence, night UV - A supplementation may affect its beneficial role in promoting root growth.

Chlorophyll a and b are two major photosynthetic pigments in higher plants. Our experimental results suggested that day supplemental UV-A significantly decreased chlorophyll b content in shoot of pea seedlings, while night supplemental UV-A slightly decreased shoot chlorophyll b content. Chlorophyll a and carotenoid content were not affected. The carotenoid and chlorophyll pigments of vegetable leaf tissues are sensitive to plant growth conditions, and may have cultivar-specific variations (Kimura and Rodriguez - Amaya, 2003; Kobayashi et al., 1989). Fluorescence measurements indicate that photosynthetic performance index was 15% higher under the presence of UV-B and UV-A than under the presence of UV-A and 53% higher than in the absence of UV radiation, suggesting protection of the photosynthetic apparatus possibly by phenolic compounds.

Our data indicate that anthocyanin content did respond to UV-A supplementation. D-UV6 and N-UV3 treatments slightly decreased shoot anthocyanin content of pea seedlings, while N-UV6 treatment significantly reduced shoot anthocyanin content of pea seedlings. Li and Kubota (2009) found that anthocyanins concentration of baby leaf lettuce grown under white fluorescent lamps as the main light source was increased when supplemented with LED UV-A. Furthermore, there was a curvilinear relationship between the anthocyanin content and UV wavelength cutoff such that above 370 nm, there was no further reduction in anthocyanin content (Tsormpatsidis et al., 2008).

Responses of peroxidase and catalase activities of plants were considered as sensitive indicators to environmental stress. In this study, UV-A supplementation did not affect peroxidase and catalase activities of pea seedling shoot. This indicate that supplemental UV-A did not alter these antioxidant enzymatic activities, which means that UV-A is not a harmful light quality for pea seedlings. Pea seedling is a kind of vegetable popularly consumed in Asian countries, especially in China. Generally, light and nutrient are necessary factors that relates to growth and quality of pea seedlings in cultivation. High light intensity can gain high yield with the aid of nutrient addition, and high light intensity may inhibit pea seedling growth without nutrient addition.

Conclusion

Supplemental UV-A did not significantly affect shoot and root to shoot ratio of pea seedlings, but root and total biomass were modified. Supplemental UV-A did not affect the contents of shoot chlorophyll a and carotenoid of pea seedlings. Nevertheless, day supplemental UV-A significantly decreased chlorophyll b content in shoot of pea seedlings, while night supplemental UV-A slightly decreased shoot chlorophyll b content. In addition, supplemental UV-A did not significantly affect the contents of shoot vitamin C, phenolic coumpounds and flavonoid of pea seedlings. However, day supplemental 6 h and night supplemental 3 h slightly decreased shoot anthocyanin content of pea seedlings, while night supplemental 6 h significantly reduced shoot anthocyanin content. Finally, supplemental UV-A did not significantly affect shoot peroxidase and catalase activities.

ACKNOWLEDGEMENTS

This study was supported by the National High Technology Research and Development Plan of China (863 Project, grant No. 2011AA03A114), State Science and Technology Support Program (2011BAE01B00) and the Basic Scientific Research Fund of National Nonprofit Institutes (BSRF201004 and BSRF2012-2013).

REFERENCES

Cao JK, Jiang WB, Zhao YM (2007). Experimental Guidance of Postharvest Physiology and Biochemistry of Fruits and Vegetables. China Light Industry Press. Beijing.

Kimura M, Rodriguez-Amaya DB (2003). Carotenoid composition of hydroponic leafy vegetables. J. Agric. Food Chem. 51:2603-2607.

Klein RM (1976). Plants and near-ultraviolet radiation. Bot. Rev. 4:1-127.

Klein RM (1990). Failure of supplementary ultraviolet radiation to

enhance flower color under greenhouse conditions. HortScience 25(3):307-308.
Kobayashi K, Tsurumizu A, Toyoda M, Saito Y (1989). Contents of chlorophyll, b-carotene and pesticide residues in butter head lettuce produced by various cultivation methods. Nippon Shokuhin Kogyo Gakkaishi 36:676-681.
Li HS (2000). Principles and techniques of plant physiological biochemical experiment. Beijing: Higher Education Press.
Li Q, Kubota C (2009) Effects of supplemental light quality on growth and phytochemicals of baby leaf lettuce. Environ. Exp. Bot. 67:59-64.
Moeller SM, Jacques PF, Blumberg JB (2000). The potential role of dietary xanthophylls in cataract and age-related macular degeneration. J. Am. College Nutr. 19:522S-527S.
Pérez-Balibrea S, Moreno D, Garcia-Viguera C (2008). Influence of light on health-promoting phytochemicals of broccoli sprouts. J. Sci. Food Agric. 88:904-910.
Plum GW, Price KR, Rhodes MJ, Williamson G (1997). Antioxidant properties of the major polyphenolic compounds in broccoli. Free Radic. Res. 27:429-435.
Takaya Y, Kondo Y, Furukawa T, Niwa M (2003). Antioxidant constituents of radish sprout (Kaiware-daikon), Raphanussativus L. J. Agric. Food Chem. 51:8061-8066.
Tsormpatsidis E, Henbest RGC, Davis FJ, Battey NH, Hadley P, Wagstaffe A (2008). UV irradiance as a major influence on growth, development and secondary products of commercial importance in Lollo Rosso lettuce ‘Revolution’ grown under polyethylene films. Environ. Exp. Bot. 63:232-239.
Voipio I, Autio J (1995). Responses of red-leaved lettuce to light intensity, UV-A radiation and root zone temperature. Acta Horticulturae, 399:183-187.
Wu MC, Hou CY, Jiang CM, Wang YT, Wang CY, Chen HH, Chang HM (2007). A novel approach of LED light radiation improves the antioxidant activity of pea seedlings. Food Chem. 101(4):1753-1758.
Zielinski H, Piskuta MK, Michalska A, Koz3owska H (2007). Antioxidant capacity and its components of cruciferous sprouts. Pol. J. Food Nutr. Sci. 57:315-322