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Effect of supplemental UV-A irradiation in solid-state lighting on the growth and phytochemical content of microgreens**

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A b s t r a c t. In this study, we sought to find and employ positive effects of UV-A irradiation on cultivation and quality of microgreens. Therefore, the goal of our study was to investigate the influence of 366, 390, and 402 nm UV-A LED wavelengths, supplemental for the basal solid-state lighting system at two UV-A irradiation levels on the growth and phytochemical contents of different microgreen plants. Depending on the species, supplemental UV-A irradiation can improve antioxidant properties of microgreens. In many cases, a significant increase in the investigated phytochemicals was found under 366 and 390 nm UV-A wavelengths at the photon flux density (12.4 μmol m^{-2} s^{-1}). The most pronounced effect of supplemental UV-A irradiation was detected in pak choi microgreens. Almost all supplemental UV-A irradiation treatments resulted in increased leaf area and fresh weight, in higher 2,2–diphenyl–1–picrylhydrazyl free-radical scavenging activity, total phenols, anthocyanins, ascorbic acid, and α-tocopherol.

K e y w o r d s: ultraviolet-A, microgreens, growth, antioxidants

INTRODUCTION

Microgreens are seedlings of vegetables and herbs that are grown to fully opened cotyledons or the first true leaf stage. They are harvested at 7-20 days after germination, depending on the species. Due to a variety of flavours, colours, and textures, microgreens may be used as ingredients in salads, soups, sandwiches and so on (Treadwell *et al*., 2010; Xiao *et al*., 2012). They may contain much higher levels of bioactive compounds such as vitamins, minerals, and antioxidants than mature plants or seeds (Xiao *et al*., 2012). Nowadays, this type of vegetables is assigned to the group of 'functional foods' that have health promoting or disease preventing properties. About 80-100 crops and crop varieties may be cultivated as microgreens. It is most popular to grow microgreens from cabbage, beet, kale, kohlrabi, mizuna, mustard, radish, Swiss chard, and amaranth (Treadwell *et al.,* 2010). They can be cultivated in fields or greenhouses and growth chambers under artificial light (Samuoliene *et al*., 2013; Treadwell *et al.,* 2010; Xiao *et al*., 2012). High-pressure sodium lamps – an artificial light source – are still popular for supplemental lighting in greenhouses due to their high electrical efficiencies, long operating life, and a wide spectrum of light (Wheeler, 2008). However, recent technologies, such as solid-state lighting, based on light-emitting diodes (LEDs) propose more progressive properties and capabilities. LEDs have a narrow light spectrum, low power consumption, and emit less heat. One of the main advantages of LEDs is the ability to control the spectral output of the lighting system. LEDs are available in the spectral range from near ultraviolet (UV) to near infrared (IR): thus, lighting spectra can be adapted for specific crops and optimized for maximum production without wasting energy on nonproductive wavelengths (Morrow, 2008). The small scale, integrity of LEDs, and attractive design possibilities further expand the growing opportunities of microgreens. They can be grown not only in greenhouses or growth chambers, but also in various

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indoor spaces in restaurants, hospitals, kindergartens, and residential houses. Literature data show that different plants are successfully cultivated under various lighting combinations of blue, red, and far-red LEDs (Stutte *et al*., 2009; Tarakanov *et al*., 2012). However, microgreens are generally grown from seeds of open-field vegetables that are physiologically adapted to natural sunlight. It is known that solar UV radiation can cause various responses in higher plants and it is traditionally divided into three wavelength ranges. UV-C (200-280 nm) is very harmful to living organisms, but is absorbed almost completely by the ozone layer of the atmosphere and is not important under natural solar irradiation (Hollósy, 2002). UV-B (280-320 nm) represents only 1.5% of the total solar spectrum and a greatest portion of it is absorbed by the ozone layer, but can induce a variety of damaging effects in plants. The level of such radiation is affected by changes in stratospheric ozone depletion, so most investigations have focused on UV-B (Hollósy, 2002; Zhou *et al.,* 2007). According to literature data, UV-B reduced plant height, leaf area and length, fresh and dry biomass, and such response depends on the irradiation intensity level, environmental conditions, plant species (Krizek *et al*., 1998; Tsormpatsidis *et al.,* 2008; Turcsányi and Vass, 2000). Moreover, the change in various internal plant parameters also depends on different levels of UV-B radiation. A high level of UV-B causes serious damage to DNA, membranes, and proteins, decreases the chlorophyll content, and has a negative effect on photosynthesis (Hollósy, 2002; Tsormpatsidis *et al.,* 2008). However, a low level of UV-B stimulates accumulation of UV-absorbing pigments and increases the concentration of flavonoids and anthocyanins (Tsormpatsidis *et al.,* 2008; Zhou *et al.,* 2007). UV-A (320-400 nm) represents about 6.3% of solar radiation that penetrates the atmosphere to reach the earth surface. It is a less harmful part of UV and is reported to diminish the damaging effect of UV-B (Hollósy, 2002; Zhou *et al.,* 2007). A high level of UV-A sometimes causes similar responses as a low level of UV-B irradiation: necrosis or impairment of the photosynthesis system (Turcsányi and Vaas, 2000; Zhou *et al.,* 2007). A low level of UV-A may increase the content of photosynthetic pigments as well as antioxidant compounds and stimulate plant growth (Brazaitytė *et al*., 2010; Helsper *et al*., 2003; Tsormpatsidis *et al.,* 2008). Microgreen cultivation under controlled conditions without UV radiation could impair their nutritional quality. For example, Iwai *et al*. (2010) stated that it is difficult to harvest perilla with high internal quality in the greenhouse, because it generally is cultivated in the open field and requires full sunlight for efficient growth. The positive effects of UV-A suggest a possibility to use low levels of UV-A to increase the content of various plant phytochemicals that have human health-promoting activity (Helsper *et al*., 2003). Some studies of the UV-A effect on plants were carried out using various short-wave cut-off films (Tsormpatsidis *et al.,* 2008). Only in recent years, UV-A LEDs were used for plant growing experiments as a part of different lighting systems (Brazaitytė *et al*., 2009; 2010; Li and Kubota, 2009) or as a sole source of light (Lee *et al*., 2010; Phyo and Chung, 2013). However, these studies deal with one or another UV-A wavelength effect. In this study, we sought to find and employ positive effects of UV-A irradiation on cultivation and internal quality of microgreens. Therefore, the goal of our study was to investigate the influence of 366, 390, and 402 nm UV-A LED wavelengths, supplemental for the basal solid-state lighting (LED) system at two UV-A irradiation levels on the growth and phytochemical contents of different microgreen plants.

MATERIALS AND METHODS

The experiments were carried out at the Institute of Horticulture, Lithuanian Research Centre of Agricultural and Forestry Science. Microgreens of basil (*Ocimum basilicum* L., Sweet Genovese), beet (*Beta vulgaris* L., Bulls Blood), and red pak choi (*Brassica rapa* var. chinensis, Rubi) were grown to harvest time (respectively 10, 14, and 20 days) in a growth chamber on a peat substrate (Profi 1, JSC Durpeta, Lithuania) (pH 6). The average amount of nutrients (mg l^{-1}) in the substrate was as follows: N – 110, P_2O_5 – 50, K_2O – 160, microelements – Fe, Mn, Cu, B, Mo, Zn. Electrical conductivity was 0.5-0.7 mS cm^{-1}. 1 g of basil, 3.5 g of beet, and 1 g of red pak choi seeds (CN Seeds Ltd., UK) were seeded into 0.5 l vessels (18 x 11 x 6 cm), which represented one replicate. Four vessels were used for each species. The vessels were arranged randomly and were systematically rotated every day for improving the uniformity of the light environment. The plants were watered when needed. The photoperiod was 16 h, day/night temperature 21/17°C, and the average of relative air humidity – 65%.

The microgreens were cultivated under custom–made lighting equipment containing separate modules for parallel growth runs under individually controlled illumination conditions. The light-emitting diodes were mounted on a flat aluminium heat sink with reflectors and were arranged to ensure optimal homogeneity of the flux. Each module contained four basal groups of high-power LEDs with different peak wavelengths. The main photosynthetically active photon flux was provided by deep red, red, and blue LEDs with the peak wavelength of 665, 638, and 447 nm, respectively (Luxeon Rebel LXM3-PD01-0300, Luxeon LXHL-LD3C, Luxeon LXHL-LR3C, respectively; all by Philips Lumileds Lighting Co., USA). The far-red component of the spectrum was provided by LEDs with the peak wavelength of 731 nm (L735-05-AU, Epitex Inc., Japan). Three modules were equipped with the fifth group of supplemental high-power UV LEDs emitting at 366 nm (NCSU033B, Nichia Corp., Japan), 390 nm (NCSU034B, Nichia Corp., Japan), and 402 nm (ACULED VHL ACL01–SC–UUUU–E05-C01-L-U000, PerkinElmer, Inc., USA). The emission spectra of the LEDs were measured

using a photonic multichannel analyser (Hamamatsu PMA-12, Japan) and are presented in Fig. 1. All LED groups were driven independently by custom-made high-power current regulators with digital control. Lighting regime was pre-programmed by a controller unit using a remote computer. Each module has an illuminated area of 0.28 m^2 sufficient for simultaneous growth of vegetable plants in amounts large enough for acquisition of statistically reliable data.

Two experiments were carried out to determine the UV-A effects on the microgreens. Table 1 presents the lighting regimes of both growth runs. The overall photon flux density of blue, red, deep red, and far red LEDs in each lighting module was 300 μmol m^{-2} s^{-1} at the crop level. The regimes differed in the UV LEDs photon flux density. The first treatment (EXP1) was provided with the supplemental UV-A photon flux density of 6.2 μmol m^{-2} s^{-1} (lower UV-A irradiance level) and in the second treatment (EXP2) the supplemental UV-A photon flux density was increased by a factor of 2 (higher UV-A irradiance). A module without UV LEDs was used for reference growth runs.

After the lighting experiment, microgreen cotyledons with stems were harvested just above the ground. Samples were taken from the central vessel part, leaving plants 1.5 cm from vessel edges as guard. Plant height, hypocotyl length, leaf area, and fresh weight were measured for determining the UV-A effect on microgreen growth. Leaf area was measured using a 'WinDias' leaf area meter (Delta-T Devices Ltd, UK).

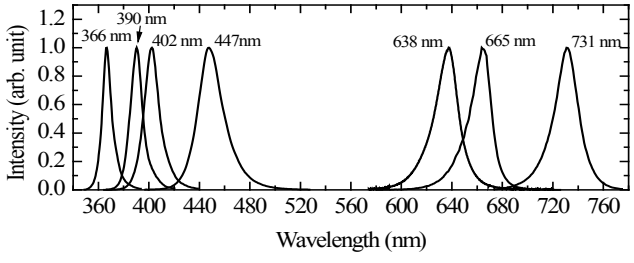

Fig. 1. Normalized emission spectra of the LEDs used in the lighting equipment.

Conjugated biological samples of the fresh matter of randomly selected plants were used for phytochemical analysis. Antioxidant activity of microgreen fresh matter (μmol g^{-1}) was evaluated spectrophotometrically as the 2,2–diphenyl–1–picrylhydrazyl (DPPH) free-radical scavenging capacity (Ragaee *et al*., 2006) of methanolic extracts (fresh tissue ground with liquid nitrogen and diluted with 80% methanol at the ratio 1:10 (m/v)). The absorbance was scanned for 16 min at 515 nm (Thermospectronic, USA).

The total content of phenolic compounds (mg g^{-1}) in microgreen fresh matter was determined by preparing methanolic extracts (fresh tissue ground with liquid nitrogen and diluted in 80% methanol at the ratio 1:10 (m/v)) and using a colorimetric Folin-Ciocalteau method (Ragaee *et al*., 2006). Absorbance was measured at 765 nm using a Genesys 6 spectrophotometer (Thermospectronic, USA) against water as a blank. Total phenolics were determined by a calibration method using gallic acid as a standard.

The total amount of anthocyanins (mg g^{-1}) was determined using the spectrophotometric pH-differential method proposed by Stanciu *et al*. (2009). Fresh tissue was ground with liquid nitrogen and diluted with 2% HCl in methanol. The absorption values of the extract in 0.025 M potassium chloride (pH 1.0) or 0.4 M sodium acetate buffer (pH 4.5) were measured at 420, 520, and 700 nm wavelengths. Anthocyanins were expressed as a cyanidin 3-glucoside equivalent, mg g^{-1} in fresh microgreen weight, using a molar extinction coefficient of 25740 M^{-1} cm^{-1} and molecular weight of 485 mg mol^{-1}.

Ascorbic acid (mg g^{-1} in fresh weight) was assessed by a spectrophotometric method (Janghel *et al*., 2007), based on methyl viologen reduction to a stable blue free radical ion. Samples were prepared by homogenising fresh plant material with an oxalic acid solution. The coloured radical ion was measured at 600 nm and ascorbic acid contents were evaluated using the calibration method.

The α-tocopherol content (mg g^{-1} in fresh weight) was evaluated according to Fernandez-Orozco *et al*. (2003) using the high performance liquid chromatography (HPLC)

T a b l e 1. Photon flux densities at the crop level in μmol·m^{-2} s^{-1} produced by LEDs with the indicated peak emission wavelengths in two plant growth experiments denoted as EXP1 and EXP2

Experiments	Illumination						
	Supplemental			Basal			
	UV			blue	red	deep red	far red
	366	390	402	447	638	665	731
	(nm)						
	Photon flux density (μmol m^{-2} s^{-1})						
EXP1	6.2	6.2	6.2	21	122	155	2.2
EXP2	12.4	12.4	12.4	21	122	155	2.2

method. Fresh microgreen tissue was ground with liquid nitrogen and hexane extracts (1:10 m/v) were prepared. An HPLC 10A system equipped with RF-10A fluorescence detector (Shimadzu, Japan) and Pinacle II silica column, 5-µm particle size, 150 x 4.6 mm (Restek, USA) was used for the analysis. The peak was detected using an excitation wavelength of 295 nm and emission wavelength of 330 nm. The mobile phase was 0.5% isopropanol in hexane, flow rate 1 ml min^{-1}.

The nitrate concentration was measured by a potentiometric method (Geniatakis *et al*., 2003) using an ion meter (Oakton, USA) with a nitrate-selective electrode (Cole-Parmer, USA). Samples were prepared from ~40 g fresh microgreen tissue per sample that was dried at 105°C for 24 h and ground. The ionic strength adjustor (ISA) contained 0.1 M $Al_2(SO_4)_3$. The weighed dry sample (0.2 g) was diluted with 20 ml water-ISA solution (50/50% v/v). All measurements were performed after the sensor signal had been stabilized for 3 min.

The flavonol and chlorophyll content was determined by a non-destructive method using a flavonol and chlorophyll meter Dualex4 (Dynamax Inc., USA).

Three analytical replications of DPPH, phenols, anthocyanins, ascorbic acid, a-tocopherol, nitrate, and ten biological replications of chlorophylls, flavonols, and growth parameters were performed for each treatment. Statistical analysis was performed using STATISTICA 7.0 for Windows, and differences from the reference were compared using Student t-test. Mean differences were considered significant when $p \leq 0.05$.

RESULTS

The microgreens showed distinct growth responses to different treatments of supplemental UV-A in basal illumination (Table 2). A trend was noticed that, with increasing supplemental UV-A wavelength (from 366 to 402 nm) in the basal lighting, the microgreen height and hypocotyl length increased at both UV-A LEDs intensity levels. However, at the higher (EXP2 –12.4 µmol m^{-2} s^{-1}) UV-A irradiance intensity level, basil grew significantly higher under supplemental 402 nm wavelength light and pak choi – under supplemental 390 nm wavelength light. Meanwhile, at this irradiance level, supplemental UV-A LEDs had no significant effect on the length of the microgreen hypocotyls. At lower (EXP1 6.2 µmol m^{-2} s^{-1}) UV-A LEDs irradiance, only supplemental 402 nm wavelength light caused a significant increase in the microgreen height and hypocotyl length.

Leaf area of basil and pak choi significantly increased under all supplemental UV-A LEDs lighting at the higher intensity level, although basil under supplemental 390 nm light (Table 2). Meanwhile, at a lower intensity level (EXP1), the leaf area of basil and beet significantly increased under supplemental 366 nm wavelength light and that of pak choi under supplemental 402 nm light. Similar trends were found for fresh weight of these microgreens, although there were no significant differences between the treatments at the higher UV-A intensity level (EXP2). However, at the lower intensity level, supplemental UV-A LEDs light caused a significant or slight increase in fresh weight of basil, a decrease in this index in beet, and no effect in pak choi. The same trends were found for the dry weight of the microgreens (data not shown).

Neither the different UV-A wavelengths nor the UV-A LEDs intensity level had a significant effect on basil and beet chlorophyll index (Table 2). Meanwhile, supplemental UV-A increased this index in pak choi. Supplemental 390 nm irradiation at both illumination levels had the most significant effect on the increase in the chlorophyll index.

Phytochemical concentrations and antioxidant activity in the microgreens were also significantly affected by the different supplemental UV-A treatment. Our data revealed that almost all supplemental UV-A resulted in increased DPPH free-radical scavenging activity of the microgreens (Table 3). Depending on the microgreen species and level of exposure, most notable effects were induced by supplemental 366 nm irradiance. Total phenolic compounds were found to be species depended. All supplemental UV-A significantly increased their content in basil at the higher intensity level, but had no effect at lower irradiance. The lower UV-A LEDs irradiance level (EXP1) had no effect on the phenol content in beet, but at higher intensity (EXP2) their content significantly increased with the supplemental 402 nm treatment. Supplemental 366 nm irradiance significantly increased total phenolic compounds in pak choi at both applied UV-A irradiance levels (Table 3). The flavonol index determined by a non-destructive method showed a similar trend to that in phenols, although a significant increase in beet and pak choi flavonol index was found under supplemental 366 nm irradiance at the higher intensity level (Fig. 1). The total anthocyanin concentration at the higher UV-A intensity (EXP2) was the greatest under supplemental 366 nm and 390 nm in beet and pak choi. Meanwhile, their content decreased in basil (Table 3). At the lower irradiance level (EXP1), such supplementation had no effect on the total anthocyanin concentration in all microgreens. Supplemental 366 nm irradiance caused a significant decrease in ascorbic acid in basil at both illumination levels. Its concentration in beet significantly decreased with supplemental 366 nm at the higher intensity, but significantly increased at the lower. All supplemental UV-A wavelengths increased the ascorbic acid concentration in pak choi at the higher intensity level. Meanwhile, at the lower one, their concentration significantly increased with supplemental 366 nm and significantly decreased with supplemental 390 nm and 402 nm. At the higher irradiance level (EXP2), all supplemental UV-A had a positive effect on the α-tocopherol content in the microgreens, except basil, where supplemental 390 or 402 nm irradiance resulted in a significantly decreased content thereof. Supplemental 366 nm irradiance had the greatest effect on the α-tocopherol content in all microgreens. Meanwhile, the lower intensity

T a b l e 2. The effect of UV-A on growth parameters and the chlorophyll index of the microgreens

Effect		Height (cm)	Hypocotyls length (cm)	Leaf area (cm^2)	Fresh weight (mg)	Chlorophyll index
			Basil			
EXP1	Basal	2.48±0.11	1.36±0.13	1.00±0.06	27.3±1.6	25.8±1.1
	+366	2.63±0.08	1.51±0.09	1.05±0.04A	39.0±2.3A	26.8±0.9
	+390	2.70±0.07	1.71±0.09A	0.80±0.05	27.9±0.6	24.6±1.4
	+402	2.93±0.08A	1.87±0.08A	0.89±0.03	35.4±1.9A	28.7±0.9
EXP2	Basal	3.25±0.06	2.01±0.11	1.60±0.13	48.8±2.9	30.4±1.1
	+366	3.12±0.12	1.81±0.07	2.18±0.14A	55.8±4.6	29.2±0.8
	+390	3.27±0.10	1.98±0.12	1.43±0.07	52.3±1.5	30.5±1.7
	+402	3.73±0.10A	2.32±0.10	2.35±0.08A	55.5±4.1	29.3±1.2
			Beet			
EXP1	Basal	4.70±0.13	1.94±0.16	2.46±0.15	94.1±5.3	19.5±1.1
	+366	5.38±0.19A	2.04±0.18	3.72±0.23A	89.9±9.6	16.8±1.3
	+390	4.94±0.13	1.95±0.14	2.73±0.08	75.7±5.0B	17.7±0.8
	+402	5.49±0.16A	2.56±0.16A	2.75±0.14	80.3±1.3B	18.4±0.8
EXP2	Basal	6.92±0.20	3.47±0.12	3.45±0.22	152.0±7.4	25.2±0.8
	+366	6.74±0.30	3.54±0.16	3.14±0.23	131.1±7.3	25.0±0.8
	+390	7.16±0.22	3.85±0.16	3.95±0.28	174.9±12.2	25.1±1.0
	+402	6.77±0.16	3.76±0.12	3.59±0.21	155.7±13.7	22.9±0.8
			Pak choi			
EXP1	Basal	5.95±0.09	3.79±0.07	3.02±0.13	95.6±3.8	23.9±0.84
	+366	5.92±0.14	3.73±0.12	2.98±0.08	93.2±5.9	28.8±0.97A
	+390	5.96±0.12	3.91±0.13	2.71±0.12	88.6±3.0	30.5±1.28A
	+402	6.29±0.06A	4.03±0.06A	3.77±0.15A	98.9±6.0	29.4±1.26A
EXP2	Basal	6.00±0.13	3.99±0.08	2.67±0.07	98.6±2.7	29.1±1.7
	+366	6.34±0.14	4.10±0.12	4.25±0.18A	109.0±9.5	29.2±0.9
	+390	6.57±0.11A	4.37±0.18	4.21±0.15A	106.5±6.7	34.4±1.5A
	+402	6.28±0.09	4.10±0.09	3.93±0.13A	98.7±3.6	29.6±1.5

A – significantly higher than the control (basal illumination), B – significantly lower than the control. Significance level: $p<0.05$ (t-test).

T a b l e 3. Effect of UV-A on antioxidant contents of microgreens

Effect		DPPH (μmol g^{-1})	Total phenols (mg g^{-1})	Total anthocianins (mg g^{-1})	Ascorbic acid (mg g^{-1})	α-tocopherols (μg g^{-1})
			Basil			
EXP1	Basal	9.60±0.10	1.30±0.06	0.40±0.06	2.04±0.15	148.99±3.22
	+366	10.02±0.50	1.33±0.08	0.38±0.09	1.31±0.05B	86.31±3.59B
	+390	9.56±0.46	1.31±0.03	0.43±0.08	2.20±0.03	52.10±1.39B
	+402	9.98±0.21A	1.32±0.06	0.31±0.06	2.27±0.08	68.23±1.27B
EXP2	Basal	9.49±0.21	1.80±0.02	0.97±0.04	2.52±0.28	75.57±2.76
	+366	10.04±0.03A	1.89±0.03A	0.93±0.04	1.86±0.07B	91.18±1.24A
	+390	9.81±0.08	1.93±0.01A	0.86±0.03B	2.52±0.04	64.89±1.79B
	+402	9.62±0.13	1.86±0.02A	0.88±0.02B	2.50±0.11	46.39±2.59B
			Beet			
EXP1	Basal	7.91±0.56	1.14±0.03	1.02±0.15	0.73±0.08	105.62±0.18
	+366	8.03±0.34	1.05±0.02B	0.88±0.11	1.12±0.21A	88.23±0.12B
	+390	8.41±0.37	1.09±0.05	0.81±0.07	0.69±0.02	85.86±0.19B
	+402	7.00±1.01	1.10±0.06	0.81±0.06	1.11±0.17A	76.58±0.37B
EXP2	Basal	10.43±0.05	1.08±0.03	0.28±0.03	4.24±0.12	42.17±1.82
	+366	10.61±0.03A	1.12±0.02	0.31±0.04	3.20±0.13B	80.49±1.34A
	+390	10.90±0.04A	0.91±0.01B	0.40±0.05A	5.46±0.20A	57.98±0.52A
	+402	10.53±0.03A	1.28±0.01A	0.27±0.03	7.49±0.19A	69.38±0.54A
			Pak choi			
EXP1	Basal	7.65±0.43	0.62±0.02	0.37±0.03	0.42±0.03	129.39±5.13
	+366	9.52±0.26A	0.75±0.02A	0.40±0.04	0.50±0.04A	84.33±1.14B
	+390	9.43±0.31A	0.62±0.03	0.43±0.04	0.33±0.03B	180.34±2.93A
	+402	9.26±0.50A	0.64±0.01	0.43±0.05	0.30±0.06B	208.92±0.52A
EXP2	Basal	9.13±0.34	0.73±0.02	0.76±0.03	0.83±0.05	42.46±6.24
	+366	10.07±0.24A	0.86±0.03A	0.99±0.06A	1.25±0.05A	87.25±3.96A
	+390	9.40±0.07	0.72±0.01	0.87±0.05A	1.05±0.28	71.37±3.57A
	+402	10.20±0.07A	0.75±0.02	0.75±0.02	1.18±0.09A	62.54±0.24A

Explanations as in Table 2.

supplemental UV-A irradiation (EXP1) resulted in a significantly decreased α-tocopherol content in the microgreens, except supplemental 390 or 402 nm irradiance, which caused its increase in pak choi (Table 3).

Supplemental UV-A irradiance increased the nitrate content in the microgreens at both UV-A LEDs intensities used (Fig. 3). The only exception was pak choi, where supplemental UV-A irradiance at the higher intensity level (EXP2) decreased the nitrate content. At lower intensity (EXP2), only supplemental 366 nm UV-A decreased nitrate content in basil.

DISCUSSION

Our study revealed that the UV-A irradiation effect on the microgreens was species and intensity dependent. It was determined overall trend that the supplemental UV-A irradiance increased plant height and hypocotyl length, or had no effect on it, but no decrease in elongation of the microgreens was detected in any lighting treatment. Generally, UV-A related plant responses depend on the blue-UVA light photoreceptor, *ie* the cryptochrome (Lin, 2000), and activation thereof by UV-A generally determines stem growth inhibition (Silva *et al*., 2005). However, no inhibitory effects of UV-A on the height and hypocotyls have been found in our study. Generally, supplemental UV-A had no damaging influence on leaf area and biomass parameters of the microgreens. According to literature data, supplemental UV-A did not significantly affect biomass of pea seedlings in greenhouse conditions (Wenke and Qichang, 2012) and biomass of baby lettuce cultivated in growth chambers under white fluorescence lamps (Li and Cubota, 2009). Tsormpatsidis *et al*. (2008) reported that total aboveground dry weight was positively correlated with the degree of UV radiation cutoff transmitted by films. Our investigations revealed a different effect of UV-A irradiation on the different microgreen species. A most remarkable effect of the supplemental UV-A was found on pak choi growth at the higher intensity level. Irradiation with all supplemental UV-A wavelengths increased leaf area and plant biomass, but a greater positive effect was observed under 366 and 390 nm. Meanwhile, the effect of supplemental UV-A irradiation on leaf area and biomass of basil and beet depended on the wavelengths and intensity level. The reaction of different species to supplemental UV-A was also determined in our earlier investigations with cucumber and tomato. Supplementation of the basal high-power solid-state lighting with the UV-A LEDs also resulted in increased leaf area and fresh and dry mass of tomato, but it decreased the growth and development of cucumber transplants (Brazaitytė *et al*., 2009, 2010). Various studies showed a UV-A inhibition effect on plant growth in different plant species and suggested that the decrease under UV irradiation was induced by damage to the photosynthetic apparatus by damaging photosystem II (Krizek *et al*., 1998; Turcsányi and Vass, 2000). Our investigations revealed that supplemental UV-A irradiation had no negative effect on the chlorophyll content. Meanwhile, all supplemental UV-A even increased the chlorophyll index of pak choi at the lower intensity level. Supplemental 390 nm irradiation at both intensity levels had the most significant effect on the increase in this index. Our earlier investigations revealed no UV-A effects on photosynthesis pigments of cucumber and tomato, but decreased the chlorophyll a and b ratio in cucumber (Brazaitytė *et al*., 2009, 2010).

Plants may produce secondary products to protect them against UV light damage. Such metabolites as phenolics, flavonoids, and anthocyanins also play an important role in promoting human health and preventing diseases (Iwai *et al*., 2010; Tsormpatsidis *et al*., 2008). Literature data show different plant responses to UV-A to accumulation of these phytochemicals. Li and Cubota (2009) reported an increase in the anthocyanin concentration in baby lettuces under

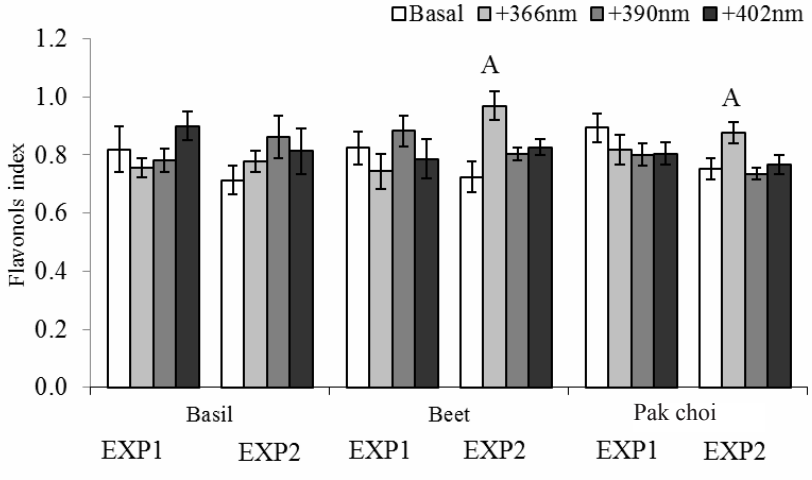

Fig. 2. The effect of UV-A on the flavonol index of the microgreens: A – significantly higher than the control (basal illumination), Significance level: $p<0.05$ (t-test).

UV-A LEDs at a 373-nm peak, but no effect on phenolic compounds. Tsormpatsidis *et al*. (2008) investigated UV transparent and blocking films and determined that radiation at 370-400 nm had no significant effects on the content of anthocyanin, flavonoid, and phenolic compounds in lettuce, and suggested that shorter wavelength UV might be more effective in increasing the concentration of these phytochemicals. Meanwhile, shorter wavelength UV caused a decrease in lettuce biomass. Supplemental UV-A with the main wavelength of 365 nm did not significantly affect the contents of phenolic compounds and flavonoids of pea seedlings, but decreased the shoot anthocyanin content (Wenke and Qichang, 2012). According to the data of Iwai *et al*. (2010), artificial illumination with UV-A enhanced the content of polyphenols in perilla, compared to greenhouse-grown plants. Total phenolic compounds and DPPH radical-scavenging activity of barley leaves cultivated under UV-A only slightly increased than of those grown under fluorescent lamps (Lee *et al*., 2010). Our data revealed that almost all investigated supplemental UV-A, especially 366 nm, increased the DPPH free-radical scavenging activity of the microgreens. Meanwhile, total phenolic compounds were found to be species dependent and the most notable effect was observed at the higher UV-A irradiance intensity level. The flavonol content measured by the non-destructive method as an index showed a similar trend as the total content of phenols. Therefore, this method could be used as quick assessment of phenolic compounds in microgreen leaves under various lighting conditions. Supplemental 366 and 390 nm at the higher intensity level showed an upward trend in the total anthocyanin concentration in beet and pak choi, but a decrease in basil. Supplemental UV-A irradiance at the lower intensity level had no effect on the total anthocyanin concentration in the microgreens. Our results agree with Kataoka *et al*. (2003), who showed that the anthocyanin content in berry skins was increased with increasing intensity of UV-A irradiation. Zhou *et al*. (2007) reported that only UV-A alone induced anthocyanin biosynthesis and application of red or far-red with UV-A did not affect the rate of UV-A induced anthocyanin accumulation in turnip.

Ascorbic acid in association with other components of the antioxidant system protects plants against oxidative damage, resulting from various stress factors, including UV-A (Wenke and Qichang, 2012). Our results showed that supplemental 366 nm at the higher irradiance level had a harmful effect on the ascorbic acid content in basil and beet, but induced an opposite effect in pak choi. Meanwhile, the longer wavelength of UV-A in many cases had a positive effect on the ascorbic acid content. Other authors reported that UV-A irradiation had no effect on its content (Li and Kubota, 2009; Wenke and Qichang, 2012).

α-Tocopherol is the major vitamin E compound found in leaf chloroplasts and its level depends on the level of stress and species sensitivity to stress. Under high intensity light stress, tocopherol levels increase dramatically (Munne-Bosh, 2005). Our studies revealed that at the higher intensity level practically all supplemental UV-A wavelengths increased the α-tocopherol content in the microgreens. Supplemental 366 nm irradiance had the greatest effect on the α-tocopherol content. Meanwhile, at lower intensity, supplemental UV-A irradiation mainly decreased the α-tocopherol content in the microgreens. It can be stated that shorter supplemental UV-A wavelengths at the higher intensity level were stressful for the microgreens, and the α-tocopherol content increased as protection against stress.

The nitrate content is one of the quality characteristics of vegetables. Light quantity and quality have been known as one the major factors affecting nitrate contents in vegetables due to regulation of nitrate reductase activity (Lin *et al*., 2013; Santamaria, 2006). In our experiments, it was found that supplemental UV-A irradiance increased the nitrate content in the microgreens, except for pak choi under the higher UV-A irradiance level. UV-A and blue light-related plant responses depend on the same light photoreceptor. Literature data show that blue light increased the nitrogen content and was less efficient in stimulation of nitrate reduction in plants than red light (Lin *et al*., 2013). On the other hand, UV-A exposure increased the nitrate content in the microgreens, but generally their maximum level was about 400 mg kg^{-1} and such an increase should not have a negative effect on human health. According to various accepted nitrate limits in different countries, their content in fresh leafy vegetables varies from 2000 to 4500 mg kg^{-1} depending on species (Santamaria, 2006).

Summarizing, our findings and literature data indicate that it is worth to use supplemental UV-A LEDs in developing LED lighting systems for microgreen cultivation. Although the effect was species dependent, the data showed that it is better to choose a shorter wavelength and higher intensity of UV-A. Such illumination had no damaging effect on the growth parameters of the microgreens, but showed a trend to increase DPPH free-radical scavenging activity, total phenols, anthocyanins, and α-tocopherol.

CONCLUSIONS

1. Depending on species, UV-A irradiation supplemental for basal LED illumination can improve the antioxidant properties of microgreens. In many cases, a significant increase was found under the 366 and 390 nm UV-A wavelengths and the higher intensity photon flux density 12.4 μmol m^{-2} s^{-1}.

2. The most positive effect of supplemental UV-A irradiation in basal illumination was observed on the pak choi microgreens. Almost all supplemental UV-A wavelengths, especially at the photon flux density 12.4 μmol m^{-2} s^{-1} increased leaf area and fresh weight, DPPH free-radical scavenging activity, total phenols, anthocyanins, ascorbic acid, and α-tocopherol.

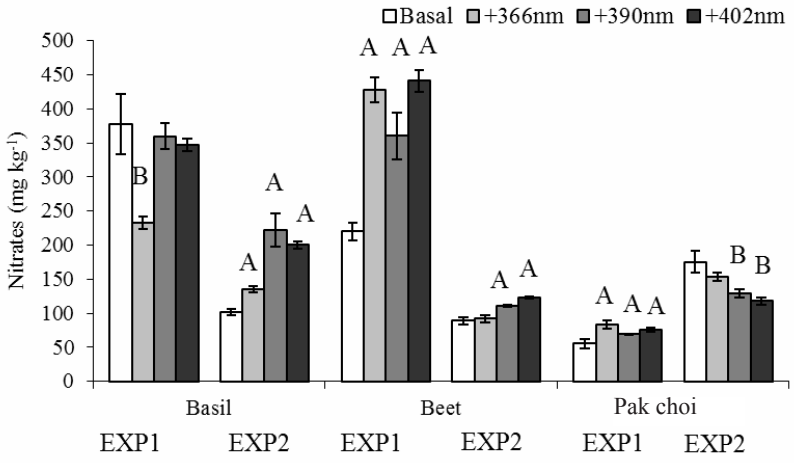

Fig. 3. Effect of UV-A on the nitrate content in the microgreens: A – significantly higher than the control (basal illumination), B – significantly lower than the control. Significance level: $p<0.05$ (t-test).

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