Photochemical & Photobiological Sciences (2023) 22:1611–1624
https://doi.org/10.1007/s43630-023-00402-8

ORIGINAL PAPERS

A comparison of consistent UV treatment versus inconsistent UV treatment in horticultural production of lettuce

Martin Weiland[1,2] · Caspar Friedrich Weßler[3,2] · Thomas Filler[4] · Johannes Glaab[4] · Neysha Lobo Ploch[4] · Ulrike Winterwerber[4] · Melanie Wiesner-Reinhold[2] · Monika Schreiner[2] · Susanne Neugart[5]

Received: 3 November 2022 / Accepted: 28 February 2023 / Published online: 29 March 2023

Abstract
UV radiation is an underrated radiation currently missing in many horticultural production systems of vegetables in protected cultivation. It can be added e.g., in LED light sources. Using lettuce as a model plant, this study determined whether the use of UVB LEDs is suitable (1) for use in consistent systems (indoor farming) or (2) inconsistent systems (greenhouse). Blue and red LEDs were selected as additional artificial lighting to UVB LEDs. Both approaches led to a reproducible increase of desired flavonol glycosides, such as quercetin-3-*O*-(6″-*O*-malonyl)-glucoside or quercetin-3-*O*-glucuronide and the anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside in lettuce. The impact of the consistent UVB treatment is higher with up to tenfold changes than that of the inconsistent UVB treatment in the greenhouse. Varying natural light and temperature conditions in greenhouses might affect the efficiency of the artificial UVB treatment. Here, UVB LEDs have been tested and can be recommended for further development of lighting systems in indoor farming and greenhouse approaches.

Keywords UVB · Vertical farming · Anthocyanins · Flavonoid glycosides · Hydroxycinnamic acid derivatives

1 Introduction

UVB (280–315 nm) is the highest energy radiation that life on Earth is usually confronted with [1], followed by the UVA (315–400 nm) and the visible light including photosynthetically active radiation (PAR; 400–700 nm). Molecular oxygen (O_2 and O_3) leads to the strong attenuation of solar ultraviolet radiation (UV; 100–400 nm), especially in the UVC region (100–280 nm), so that effectively no UVC is present in the terrestrial solar spectrum [2]. To protect themselves from harmful radiation qualities and quantities, plants have developed the biosynthesis of a variety of pigments within the framework of secondary metabolism [3], in addition to enzymatic mechanisms. Pigments obtain their protective function through the presence of at least one carbon ring and associated functional groups or conjugated double bonds. The original delocalized π-electron-based absorption spectrum of the carbon ring (ca. 250–270 nm), can thus be bathochromically extended and functionally specified [4]. Phenols are a group of pigments that protect plants from radiation [4]. One or more phenolic rings form the basis for over 10,000 known phenolic compounds in plants. Their biosynthesis takes place in chloroplasts or in the cytosol. Incorporation then takes place in the vacuole, cell wall, cuticle, or epidermis [4]. Starting from L-phenylalanine, synthesis of numerous compounds occurs in the course of the phenylpropanoid pathway. One possible product is hydroxycinnamic acid derivatives in the form of caffeic acid esters such as chicoric acid (ester with tartaric acid) or chlorogenic acid (ester with quinic acid). It is assumed that their protective function lies in the preventive, direct

✉ Susanne Neugart
Susanne.neugart@uni-goettingen.de

1 Institute for Biology, Experimental Biophysics, Humboldt-Universität zu Berlin, Invalidenstr. 42, 10115 Berlin, Germany

2 Leibniz Institute of Vegetable and Ornamental Crops e.v., Plant Quality and Food Security, Theodor-Echtermeyer-Weg 1, 14979 Grossbeeren, Germany

3 Institute of Horticultural Production Systems, Leibniz Universität Hannover, Herrenhäuser Straße 2, 30419 Hannover, Germany

4 Ferdinand-Braun-Institut (FBH), Gustav-Kirchhoff-Str. 4, 12489 Berlin, Germany

5 Division Quality and Sensory of Plant Products, Georg-August-Universität Göttingen, Carl-Sprengel-Weg 1, 37075 Goettingen, Germany

absorption of UV radiation rather than in the reaction with reactive oxygen species (ROS), as they have their absorption maxima in the range between 310 and 325 nm and hardly react to strong UVB exposure [4, 5]. Besides hydroxycinnamic acid derivatives, flavonoids are the second products of the synthesis pathway. More than 7000 different flavonoids have been identified in plants to date [6]. The flavonol group is probably the most widespread one [7]. They are mostly present in glycosylated form [6] and, if necessary, esterification with carboxylic acids, such as quinic, malic, tartaric, or malonic acid, which have different origins in plant metabolism. Unlike hydroxycinnamic acid derivatives, flavonols absorb in the wavelength range above 335 nm and show specific induction upon UVB exposure, reducing oxidative damage, formation of ROS but also the penetration of UV into leaves [5]. In general, known biological functions of flavonoids include protection against UV, herbivores, and pathogens, attracting pollinators, communicating with insects and microorganisms, affecting auxin transport, fertility of male individuals, and quenching free radicals or chelating metal ions [7]. Furthermore, they appear to accumulate in a site-specific manner with respect to ROS [5].

Mainly descended from the prickly lettuce (*Lactuca serriola*), garden lettuce (*Lactuca sativa)* [8] is classified in the Asteraceae family and has been cultivated for more than 4000 years [9]. Furthermore, it is currently one of the most consumed leafy vegetables and is grown almost worldwide [9, 10]. In Germany, lettuce has been ranked among the most consumed vegetables and is grown in the open field, under plastic film or in greenhouses all year round. To date, at least 171 different metabolites were identified, with hydroxycinnamic acid derivatives of tartaric and quinic acid and derivatives of kaempferol dominating [9, 10]. From an empirical point of view, there is strong evidence that vegetable consumption in general, but also phenolics or sesquiterpene lactones in lettuce in particular, have beneficial health effects to reduce risks or progression of diabetes, cardiovascular disease, cancer, and inflammation [11, 12]. Other secondary plant compounds of *L. sativa*, such as carotenoids, vitamins or dietary fiber, also influence its positive health effects.

Due to its high diversity of varieties, compact growth and short cultivation period, lettuce is suitable for cultivation in controlled environments [13]. In particular, the advances in LEDs (light emitting diodes), which are superior in many respects to traditional high-pressure sodium lamps or compact fluorescent lamps [14, 15], give these growing methods more applications and, under certain circumstances, superiority over conventional growing methods [16, 17]. From a plant physiology perspective, LEDs are of particular value because they have narrowband (Full Width at Half-Maximum: FWHM between 4 and 30 nm [18]) spectra and thus can generate differential responses of plant metabolism [15]. Due to this, the cultivation of microgreens and leafy vegetables could benefit, but this does not necessarily apply to cash crops, such as rice, sugar cane, wheat, corn, or potatoes [15]. Currently, the use of UVB LEDs to produce lettuce for consumption is banned in Germany [19]. Nevertheless, this technology in particular is also developing in relation to agricultural production. UVC is already being used for decontamination in greenhouses.

Although the biosynthesis of phenolics is not induced solely as protection from radiation [4, 5] and PAR UV-specific induction of various phenolic compounds has been demonstrated in numerous experiments with *L. sativa*. In terms of spectral composition, films with selective transparency [20, 21] or compact fluorescent lamps [22–25] were used so far. Because they are broadband sources, differential stimulation of metabolism is more difficult, and interference from undesirable wavelengths can affect the results. Samuolienė et al. (2013) [26] used UVA LEDs, and Goto et al. (2016) [27] used UVB LEDs (in combination with red LEDs); the latter, however, without precise analysis of phenolics and in hydroponics. According to current knowledge, increased UVB radiation in *L. sativa* leads to an increase in total phenolic content (TPC) in all cases, although in detail luteolin, quercetin glycosides [22, 24, 28], and anthocyanins [21, 22, 25] or chicoric acid or chlorogenic acid do not show a uniform response [22, 24]. Thus, the ratio of quercetin to kaempferol glycosides may be an indicator of UVB radiation as UVB is known to especially affect the synthesis of polyhydroxylated flavonoids [5, 29]. Genetic predisposition (cultivar) is the determining factor in the induction of phenol metabolism [13, 21]. Thus, red cultivars produce more anthocyanins and flavonoid glycosides than green cultivars [25]. Blue light can also stimulate the synthesis of quercetin and luteolin glycosides, while red light can lead to a decrease in quercetin glycosides and an increase in phenolic acids [30]. While this effect has been observed several times for blue light [14, 31], there is no clear trend for red light, so synergistic effects must be considered [19, 32].

The experiments described in this paper applied currently developed UVB LED technology to achieve a reproducible, differential stimulation of phenolics in *L. sativa* by means of a narrow-banded application. A low dose was chosen so that signal transduction via UVR8-COP1-HY5 can be assumed and UVB radiation will act as eustressor. During the experiments, the following questions will be clarified: Does ecophysiologically low, narrow-band UVB irradiation cause a reproducible change in the polyphenol profile in *L. sativa*? Does consistent UV treatment (mimicking indoor farming facilities) produce better results than an inconsistent treatment in greenhouses? What are the effects of ambient PAR conditions in greenhouses? What influence does the additional blue or red irradiation have?

2 Materials and methods

For the chemical analyses of the secondary metabolites, the following chemical products were used: methanol (MeOH) and acetic acid (HAc) were purchased from Roth (Karlsruhe, Germany) while acetonitrile (ACN) was purchased from J. T. Baker (Fisher Scientific GmbH, Grießheim, Germany). For the calibration series chlorogenic acid, quercetin-3-glucoside, kaempferol-3-glucoside, and cyanidin-3-glucoside from the company Roth (Karlsruhe, Germany) were used.

2.1 Plant material and UVB radiation treatment

To document the growing conditions and to keep the lighting situation uniform during the treatment period, regular measurements of the UV illumination were made. For the measurement of the UV irradiance, a radiometer (Gigahertz Optik GmbH, Türkenfeld, Germany) was used while a calibrated spectrometer (Ocean Optics Inc, Ostfildern, Germany) was used to determine the weighted irradiation.

2.1.1 Consistent UV treatment

A consistent UV treatment was applied mimicking the situation at indoor farming facilities. For the climate chambers, a UV radiation module was constructed from 20 UVB LEDs. The LEDs (peak wavelength = 307 nm ± 5 nm), 108 red LEDs (peak wavelength = 657 nm ± 11 nm), and 84 blue LEDs (peak wavelength = 455 nm ± 6 nm) were arranged over an area of 50 cm × 50 cm. The UV-LEDs are hermetically packaged to protect them from environmental impact, particularly humidity. The current of each UVB LED is adjustable via separate drivers. To ensure maximum efficiency and reliability of the UV-LEDs, self-heating of the devices was minimized using ceramic AlN packages, aluminum-core printed circuit boards with a high heat conductivity, and metal heat sinks actively cooled by fans. As a result, the temperature rise at the LEDs is limited to 3 K above ambient temperature. The operation of the irradiation module is managed by a microcontroller, which monitors the temperature and controls the current of all LEDs as well as the sequence of irradiation. The arrangements of the UVB LEDs as well as the shape of the surrounding aluminum reflector were optimized with respect to a maximum uniformity of the UV irradiance distribution by ray tracing simulations using the ZEMAX-EE commercial software package from ZEMAX Development Corporation. Separately adjusting the current of each LED through individual current drivers resulted in a uniformity factor of about 85% at a distance of 30 cm and more than 90% at a distance of 50 cm from the LEDs. The visible LEDs were purchased from Osram Opto Semiconductors GmbH (OSLON® SSL LED family). The blue and red LEDs are combined into twelve groups, each of which is assigned a driver to adjust the currents for each group individually. This allows homogeneities of 93% to be achieved for all three wavelengths at a distance of 45 cm from the bottom edge of the module. The irradiances can be set separately for all three colors, with maximum values of 37 mW m^{-2} for UVB, 60 W m^{-2} for blue, and 46 W m^{-2} for red at a distance of 45 cm from the bottom edge of the module. Before plant irradiation began, the spectral irradiance of the UVB radiation was measured over the entire irradiated area using a calibrated spectrometer (USB 2000 + Fiber Optic Spectrometer, Ocean Optics Inc., Ostfildern, Germany).

Seeds of *Lactuca sativa* 'Navarro' (red-leaved; Albert Treppens & Co Samen GmbH, Berlin, Germany) were sown in shallow trays on soil (Einheitserde type P, Fitz Kausek GmbH & Co. KG, Mittenwalde, Germany), grown at 18 °C in the dark until germination, and pricked out at the seedling stage into pots (Ø 8 cm for the climate chamber). The further cultivation in the climatic chamber took place under controlled conditions (day/night temperature: 22 °C/18 °C; relative humidity 70%; light: 415 µmol m^{-2} s^{-1} PAR for 10 h) for 3 weeks. Control plants remained in this chamber the whole time until harvest. Plants at the 4–5 leaf stage (baby leaf salads) were treated with an irradiance of 10 or 20 mW m^{-2} UVB and photon fluxes of 66.5 µmol m^{-2} s^{-1} (blue light) and 364 µmol m^{-2} s^{-1} (red light) (blue to red ration of 5.5). The UVB radiation treatment took place for four consecutive days for 10 h each day at a distance of 30 cm. All other parameters were identical to the growing conditions. On the fifth day, 24 h after the last treatment, the plants were harvested in three biological replicates with two plants each. The experiment was repeated twice with independent sets of plants.

2.1.2 Inconsistent UV treatment

In a second experiment, the UV treatment took place in a greenhouse. The LED modules in the greenhouse were installed on the watering module, so that an inconsistent application resulted from regular passes over the plants (Fig. 1). This is advantageous as it not only reduces shading due to the device but also lowers the costs using a smaller number of UVB LEDs for a large number of plants. The LEDs and their performance correspond to those of the aforementioned module. The 150 UVB LEDs (peak wavelength = 307 nm ± 5 nm) were arranged over an area of 200 cm × 30 cm. The light modules were mounted on the casting trolley at a height of 39 cm from the crop and were controlled via a central computer. The mounting on the casting trolley ensured a homogeneous distribution of the light,

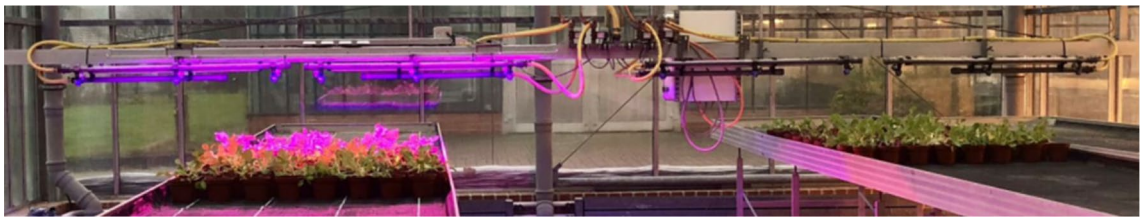


Fig. 1 inconsistent UV treatment in the greenhouse with a lighting module (left) attached to a watering module

as it was driven at a constant speed of 5.14 cm s^{-1} over the culture area.

For the second experiment, the *Lactuca sativa* 'Navarro' seeds were directly sown in Ø 10 cm pots for the greenhouse. Further cultivation took place at a relative humidity of 30–93% (Ø 65%), temperatures between 10 and 28 °C, and day length between 13 and 9 h (09 September–09 November 2018, 52°21′33.912″N 13°18′32.194″E). Plants were not fertilized and but were irrigated as needed. Plants at the 4–5 leaf stage (baby leaf salads) were treated with an irradiance of 215 mW m^{-2} (UVB) and photon fluxes of 104 µmol m^{-2} s^{-1} (blue light) and 245 µmol m^{-2} s^{-1} (red light), respectively, depending on which emission band was dominating in the treatment. The photon flux ratio of blue to red light was either 3/7 or 7/3. In contrast to the climatic chamber, only maximum values can be specified with regard to irradiance. The treatment with UVB, and blue and red radiation took place for four consecutive hours per day (approximately 100 passes of the module) and one hour after dawn taking into account the shift of twilight. All other parameters were identical to the growing conditions. The experiment was repeated two times for each light treatment (UVB combined with 3/7 and 7/3 blue/red). Control plants were in the same greenhouse and were passed by the watering module as well (Fig. 1). The harvest took place 24 h after the last light treatment in three biological replicates with two plants each.

2.2 Chemical analyses of phenolic compounds and antioxidant activity

The plant material was directly frozen in liquid nitrogen at harvest, lyophilized, and ground to a fine powder by a Retsch mill.

For analyzing the phenolic compounds, 10 mg lyophilized plant sample was extracted three times with at least 300 µl 60% acidified methanol and supernatants were combined according to Neugart et al. (2015) [33]. For the quantitative analysis of flavonoid glycosides and hydroxycinnamic acid derivatives, a HPLC series 1100 (Agilent Technologies, Waldbronn, Germany) was used [34]. Identification was performed by a mass spectrometer (Amazon SL, Bruker Daltonics GmbH & Co. KG, Bremen, Germany) according to Neugart et al. (2015) [33]. Due to the lack of the exact reference standards, a semi-quantification of phenolic compounds was done via HPLC–DAD. External calibration curves were plotted (0.1–100 µg ml^{-1}) for chlorogenic acid (hydroxycinnamic acid derivatives), quercetin-3-glucoside (quercetin-3-*O*-glucuronide and quercetin-3-*O*-(6″-*O*-malonyl)-glucoside), kaempferol-3-glucoside (kaempferol-3-*O*-glucuronide), and cyanidin-3-glucoside (cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside). The peak purity was verified by DAD (λ 190–600 nm) for the peaks. The variation coefficient of accuracy was below 5% for the investigated phenolic compounds of which the variation coefficient of precision was below 1%. The signal-to-noise ratio was above 10 for all compounds in any chromatogram. No limit of detection or limit of quantification was estimated for the phenolic compounds in lettuce.

2.3 Data analyses

Using one-factorial analysis of variance (ANOVA) and two-factorial analysis of variance (ANOVA) followed by a Tukey HSD, $p \leq 0.05$ with Apache Open Office 4.1.5, the significant differences in quantity of hydroxycinnamic acid derivatives, flavonol glycosides, and the anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside were determined as well as the interactions of PAR and artificial light for the greenhouse experiments. For each light treatment, 3 independent experiments were done.

3 Results and discussion

3.1 Light conditions during the experiments

Light conditions were monitored throughout the experiments to ensure the standardization of the experimental repetitions.

There was no detectable UVB radiation during the cultivation of the plants in the climate chamber (Table 1). However, the UVA radiation in the climate chamber used for cultivation and control plants was approximately 20% of the maximum daily irradiation during the test period (580 J m^{-2} in the open field). A priming effect due to the UVA might be possible but would rather lead to a lower effect of UVB

Table 1 Daily radiation dose of the artificial light sources at the plant's highest leaf during the experiments in the climatic chamber or the greenhouse

Treatment	UVB	UVA	PAR	Blue light	Red light
Climate chamber control	< 3.6 J m^{-2}	108 J m^{-2}	21 mol m^{-2}	3.3 mol m^{-2}	17 mol m^{-2}
Climate chamber 10 mW m^{-2} UVB	360 J m^{-2}	2.1 J m^{-2}	16 mol m^{-2}	2.4 mol m^{-2}	13 mol m^{-2}
Climate chamber 20 mW m^{-2} UVB	720 J m^{-2}	2.3 J m^{-2}	16 mol m^{-2}	2.4 mol m^{-2}	13 mol m^{-2}
Greenhouse 215 mW m^{-2} UV/blue	280 J m^{-2}	260 J m^{-2}	0.6 mol m^{-2}	0.4 mol m^{-2}	0.2 mol m^{-2}
Greenhouse 215 mW m^{-2} UV/red	280 J m^{-2}	150 J m^{-2}	0.6 mol m^{-2}	0.2 mol m^{-2}	0.4 mol m^{-2}

than expected. The ratio of red to blue was 5 in the climate chamber used for cultivation and control plants. This ratio was also set in the climate chamber for the UVB treatment. However, the power of the LEDs did not allow identical PAR intensities. Nevertheless, the intensities were within normal ranges for lettuce. Due to the narrow-band UVB LEDs, the UVA during the UV treatment in the climatic chamber was lower than in the control chamber.

In the greenhouse, the artificial irradiation (Table 1) was accompanied by the natural solar radiation, which is further modified by the greenhouse glazing and was subject to strong fluctuations. Spot measurements during the experimental period revealed maximum daily irradiances of 170 J m^{-2} for UVB, 150 J m^{-2} for UVA, and 10 mol m^{-2} for PAR radiation in the greenhouse cabin while ventilation flaps are open. Although these values varied by up to 80% due to weather conditions, UVB, UVA, and PAR values were well above due to the additional treatment. The experiments took place during the equinox period. Solar PAR decreased by 60% during the experimental period (see supplemental section Figure S1). Comparable reductions were also expected for UVB and UVA. The artificial PAR was 0.32% at the beginning of the experimental series and 0.89% at the end of the experimental series.

3.2 Biomass

A treatment for only a few days at the end of a production cycle was not expected to have an influence on the biomass and growth parameters. Generally, UVB and UVA radiation in lettuce reduces biomass but increases the formation of quercetin and luteolin and anthocyanins [35, 36]. At harvest time, there was a stronger red coloration of the UVB-treated plants. This was evident in plants from the climate chambers and also, but weak, in plants from the greenhouse. The recorded fresh and dry masses did not show any differences between UV treated plants and control plants in their relation to each other. The relative dry masses of plants from the climate chamber (9.7 ± 1.9%) were higher than those of plants from the greenhouse (6.5 ± 0.6%). At lower intensities, blue and red LEDs increased rosette area and dry weight of lettuce, which was not the case for higher intensities [37]. It can be concluded that in the present experiment, the lower PAR intensities in the climate chamber conditions were beneficial for dry weight production of lettuce. A higher UVA content in light, as in the greenhouse treatments here, led to higher fresh weights in lettuce, previously, but not always to higher dry weights [35, 38]. Nevertheless, the end-of-production treatment in the greenhouse did not have an effect on fresh and dry weight, whereas the greenhouse conditions in general, did result in lower dry weight of lettuce.

3.3 Identification and quantification of phenolic compounds and antioxidant activity

One of the main responses of plants to UVB is the enhancement of phenolic compounds that are assumed to have a higher antioxidant activity [5, 39]. Different phenolic acids, flavonol glycosides, and anthocyanins were tentatively identified by their deprotonated pseudo molecular ions $[M-H]^-$ and characteristic product ions after collision-induced dissociation (CID) in red lettuce (see supplemental section Figure S2). Among them, the main phenolic compound is chicoric acid (*meso*-dicaffeoyltartaric acid) $[M-H]^-$ *m/z* 473, then [caftaric acid − H]$^-$ *m/z* 311, followed by quercetin-3-*O*-(6″-*O*-malonyl)-glucoside $[M-H]^-$ *m/z* 549, [M − H-malonyl]$^-$ *m/z* 505, [quercetin − H]$^-$ 301, and cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside$[M-H]^-$ *m/z* 533, $[M-H]^-$ *m/z* 489, [cyanidin − H]$^-$ 285, quercetin-3-*O*-glucuronide $[M-H]^-$ *m/z* 477, [quercetin − H]$^-$ *m/z* 301, chlorogenic acid (5-*O*-caffeoylquinic acid) $[M-H]^-$ *m/z* 353, [quinic acid − H]$^-$ 191, caffeoylmalic acid $[M-H]^-$ *m/z* 295, [caffeic acid − H]$^-$ *m/z* 179. These compounds were previously reported for red leaf lettuce [9, 10, 40].

The concentration of phenols in each experiment was dominated by phenolic acids (Fig. 2), approximately 80% of the phenolic compounds as previously shown for lettuce [40]. Control plants tended to have higher levels of phenolic acids, and UVB-treated plants had correspondingly higher levels of flavonol glycosides. The concentrations of flavonol glycosides increased more in the experiments in the climatic chamber than in the greenhouse after UVB treatment compared to the control, while phenolic acids decreased. Although phenolic acids are often shown to increase or are unaffected by UVB radiation [19], it is also possible that these compounds decreased due to their shared biosynthetic pathway with flavonoids but a higher activity of the enzymes 4-cumaroyl CoA-ligase, chalcone synthase,

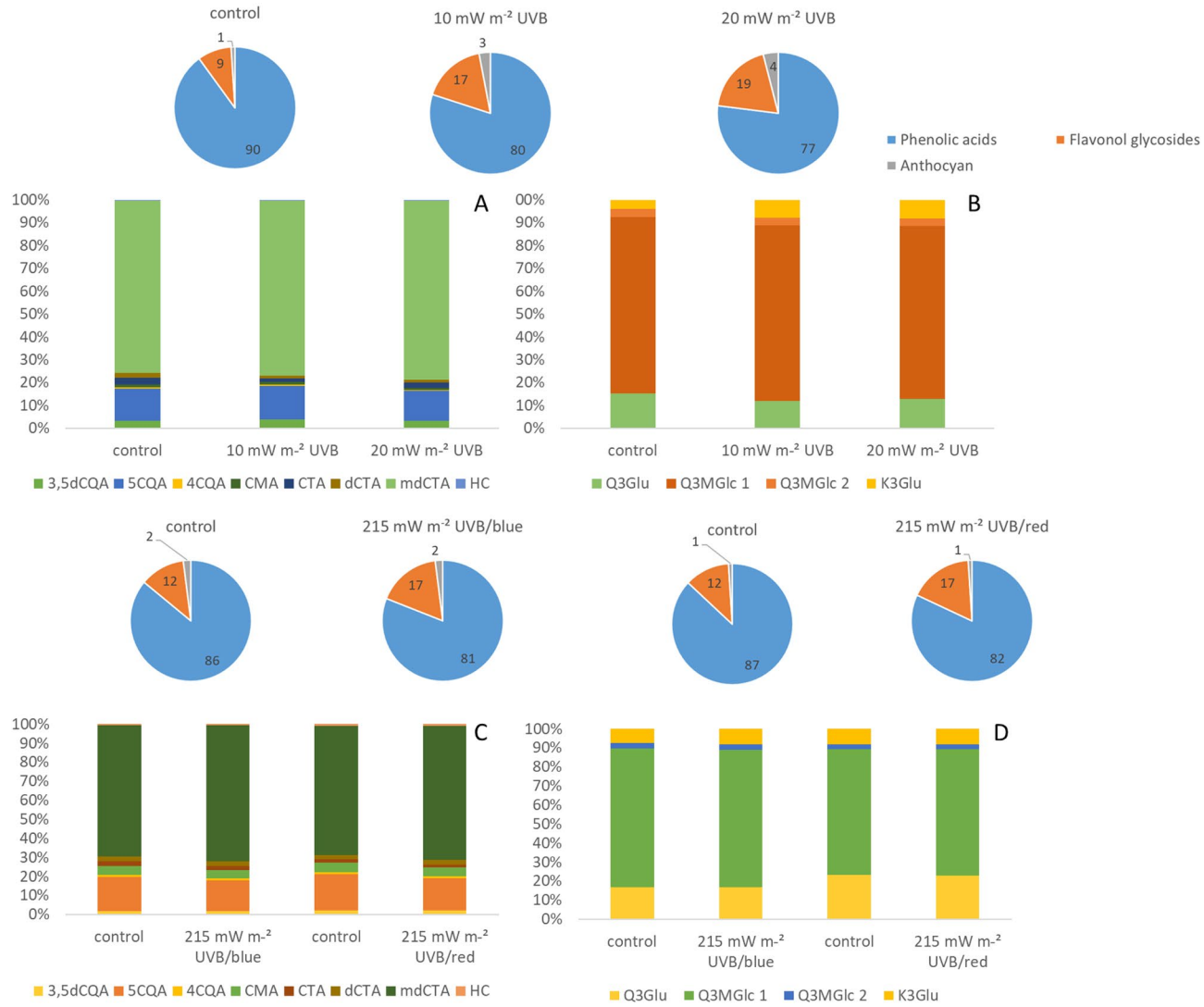


Fig. 2 Ratios (%) of phenolic compounds (phenolic acids (**A** and **C**) and flavonol glycosides (**B** and **D**)) in lettuce grown in a climate chamber (**A** and **B**) under consistent UV treatment and in the greenhouse (**C** and **D**) under inconsistent UV treatment. *dCQA* dicaffeoylquinic acid, *CQA* caffeoylquinic acid, *CMA* caffeoylmalic acid, *CTA* caffeoyltartaric acid, *dCTA* dicaffeoyltartaric acid, *mdCTA* *meso*-dicaffeoyltartaric acid, *HC* hydroxycinnamic aicd derivative, *Q* quercetin, *K* kaempferol, *C* cyanidin, *Glu* glucuronide, *MGlc* Malonylglucoside

and chalcone isomerase leading to the flavonoids. The anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside proportion increased in the climate chamber treatments as seen by the coloration and remained unchanged in the greenhouse treatments, which showed low coloration. Red coloration of lettuce with anthocyanins by UVB is associated with increased expression of relevant genes, such as flavone-3-hydroxylase and dihydroflavonol reductase [41].

The profile of phenolic acids across experiments (Fig. 2) had a very high proportion of *meso*-dicaffeoyltartaric acid (68–78%) and a much lower proportion of 5-*O*-caffeoylquinic acid (13–19%). Both compounds are known as main phenolic acids in lettuce [40]. Nevertheless, there was in total a number of eight compounds that were quantified and qualified in lettuce here. Nearly all of them were caffeic acid derivatives. It is known that caffeic acid is a potent antioxidant while quinic acid is not [42] and other organic acid might not be as well. The profile of flavonol glycosides across experiments had a comparably high proportion of quercetin-3-*O*-(6″-*O*-malonyl)-glucoside (66–77%) and a much lower proportion of quercetin-3-*O*-glucuronide (13–23%). Quercetin glycosides in general can have a high antioxidant activity [43]. The anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside was identified as the only anthocyanin and therefore is 100% of the anthocyanin profile. These compounds were previously identified as main flavonoids

in lettuce [40]. The ratios of the investigated substances to each other was only slightly changed by UVB radiation. There are indications in the literature that polyhydroxylated compounds are more strongly induced in response to UVB radiation [5, 29]. In lettuce, a large number of compounds appeared to respond very similarly to UVB treatments, changing in absolute concentrations but not in ratios to each other. It should be emphasized that caffeic acid, quercetin, and cyanidin have a catechin structure that is considered polyhydroxylated, so lettuce already has a large number of polyhydroxylated compounds.

3.3.1 Effect of a consistent UV treatment in a climate chamber on phenols and antioxidant activity

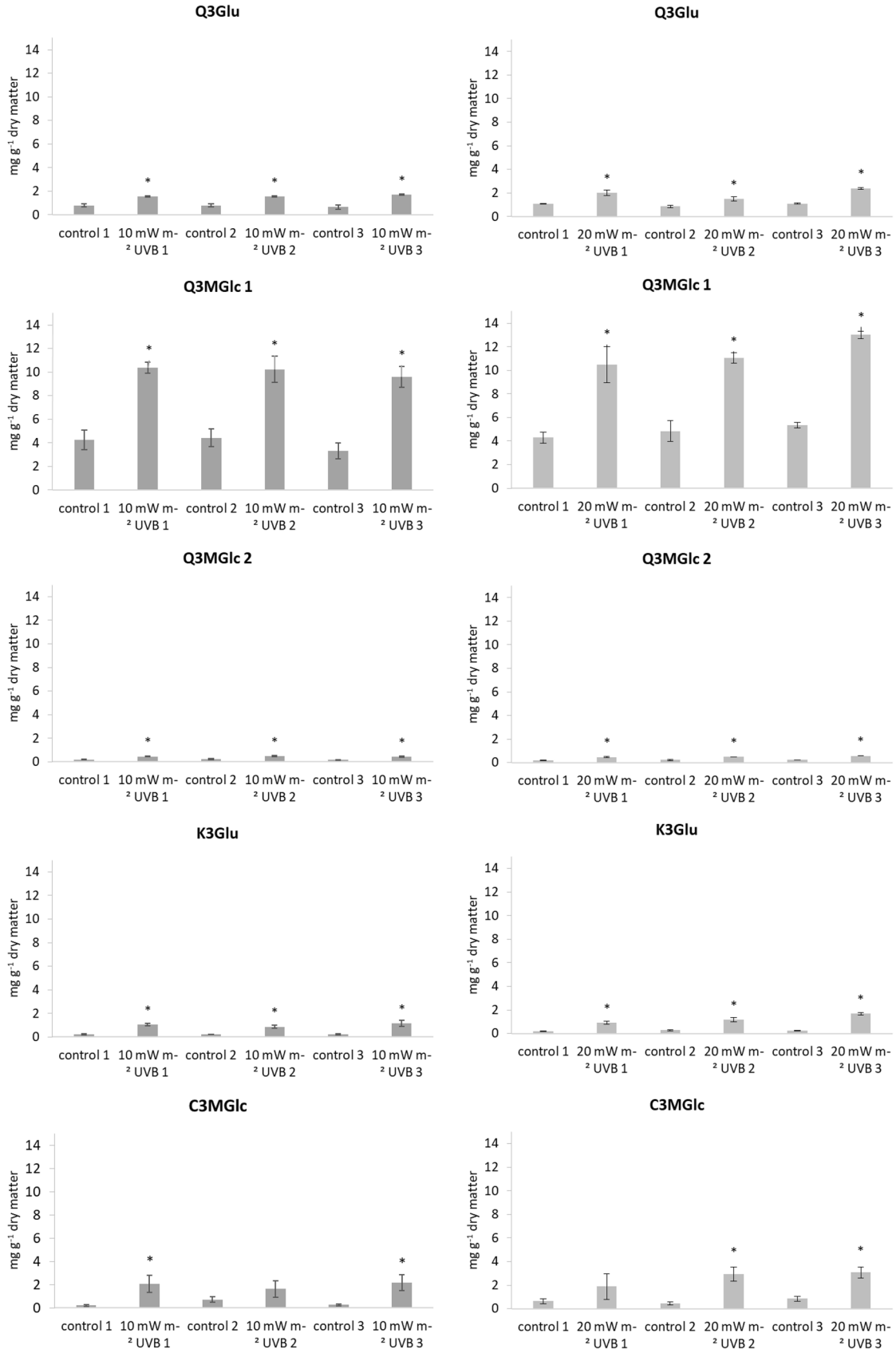
In the consistent UV treatment, dicaffeoyltartaric acid showed a reduction (0.5–0.6-fold) in all three replicates of the experiment (Table 2). Caffeoyltartaric acid also frequently showed a tendency to decrease. In contrast, *meso*-dicaffeoyltartaric acid often tended to increase slightly suggesting a structural relocation. 4-*O*-caffeoylquinic acid tended to be decreased at 20 mW or significantly decreased by 0.5–0.6-fold at 10 mW, whereas 5-*O*-caffeoylquinic acid tended to be increased by UV treatment. Even if there was no significance, the tendency showed a structural relocation favoring 5-*O*-caffeoylquinic acid, a well-known antioxidant [44]. For all flavonol glycosides (Fig. 3), a reproducible increase in concentration was found (quercetin glycosides up to 2.3-fold; kaempferol-3-*O*-glucuronide up to 6.7-fold) as a result of UV treatment. This is a well-known effect of UV treatment in plants [19, 39]. In this experiment, the ratios of the individual quercetin glycosides to the kaempferol glycoside kaempferol-3-*O*-glucuronides ranged from 0.35 to 0.45. The increase of the quercetin to kaempferol ratio has been found previously as an indicator for UV treatment and underlines the assumption that polyhydroxylated compounds such as quercetin derivatives are favored over monohydroxylated compounds such as kaempferol derivatives in UV response [5, 29]. However, in the present study, UV treatment, the quercetin to kaempferol ratio decreased by a factor of 2, although all compounds increased. However, the treatment resulted in a greater increase in kaempferol-3-*O*-glucuronide relative to the quercetin glycosides which leads to the assumption that the amount of polyhydroxylated compounds in lettuce was already high and a second mechanism of shielding might be important as well [45]. An increase of anthocyanins in *L. sativa* due to UVB radiation was previously found [21, 22, 25]. The anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside increased up to tenfold in some cases, but in others increases were only a trend. It seems that the biosynthesis of anthocyanins, late in the biosynthetic pathway, is only used when necessary. During the treatments, the leaves were not exactly at the same height but rather overlaid and in layers with different distance to the UV-LEDs which might affect the outcome here. Nevertheless, the red coloration of lettuce treated with UV radiation could be an optical marker for the successful treatment. Stronger increases in cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside and quercetin glycosides were found in the 10 mW m^{-2} treatment compared to the 20 mW m^{-2}, suggesting a potential oxidation while being used as an antioxidant at higher radiation intensities [46]. In another study, the increase of anthocyanins was found after UVA irradiation, although it must be mentioned that UVB produces similar efficacy in lettuce and UVC even

Table 2 Concentration of phenolic acids in mg g^{-1} dry matter in lettuce grown in a climate chamber under consistent UV treatment either at 10 mW m^{-2} or at 20 mW m^{-2} in three experimental replicates

UVB treatment	3,5dCQA	5CQA	4CQA	CMA	CTA	dCTA	mdCTA	HC
Control 1	1.89 ± 0.23	8.58 ± 0.62	**0.43 ± 0.05**	1.00 ± 0.16	0.83 ± 0.06	**0.93 ± 0.07**	**35.93 ± 1.90**	0.29 ± 0.07
10 mW m^{-2} UVB 1	2.35 ± 0.22	9.19 ± 0.29	**0.27 ± 0.02**	0.78 ± 0.08	0.97 ± 0.19	**0.64 ± 0.09**	**42.58 ± 0.99**	0.26 ± 0.02
Control 2	2.06 ± 0.37	8.21 ± 0.77	**0.36 ± 0.08**	**0.57 ± 0.04**	**1.44 ± 0.07**	**1.44 ± 0.06**	48.20 ± 1.25	0.18 ± 0.06
10 mW m^{-2} UVB 2	2.12 ± 0.35	8.84 ± 0.37	**0.20 ± 0.03**	**0.46 ± 0.08**	**1.05 ± 0.03**	**0.67 ± 0.08**	48.09 ± 0.73	0.13 ± 0.06
Control 3	1.59 ± 0.41	8.29 ± 0.48	**0.36 ± 0.06**	0.65 ± 0.03	1.18 ± 0.21	**1.08 ± 0.11**	37.05 ± 4.17	0.26 ± 0.04
10 Mw m^{-2} UVB 3	2.29 ± 0.43	9.06 ± 0.94	**0.24 ± 0.01**	0.92 ± 0.19	1.01 ± 0.45	**0.61 ± 0.12**	47.73 ± 4.77	0.36 ± 0.11
Control 1	1.06 ± 0.52	6.25 ± 0.32	0.23 ± 0.03	0.51 ± 0.06	3.07 ± 0.73	**1.31 ± 0.10**	**46.68 ± 0.23**	0.12 ± 0.03
20 mW m^{-2} UVB 1	1.64 ± 0.19	7.24 ± 0.39	0.20 ± 0.03	0.46 ± 0.06	1.85 ± 0.09	**0.93 ± 0.11**	**51.58 ± 0.89**	0.14 ± 0.04
Control 2	1.28 ± 0.17	7.16 ± 0.22	0.30 ± 0.03	0.80 ± 0.17	1.41 ± 0.27	**1.07 ± 0.11**	**40.92 ± 2.41**	0.22 ± 0.04
20 mW m^{-2} UVB 2	1.74 ± 0.39	7.56 ± 0.99	0.28 ± 0.02	0.87 ± 0.24	1.33 ± 0.27	**0.80 ± 0.07**	**48.53 ± 0.91**	0.22 ± 0.04
Control 3	2.20 ± 0.48	8.77 ± 0.18	**0.46 ± 0.03**	0.40 ± 0.03	1.40 ± 0.23	**1.03 ± 0.15**	39.35 ± 4.26	0.21 ± 0.02
20 mW m^{-2} UVB 3	2.50 ± 0.21	9.00 ± 0.30	**0.33 ± 0.02**	0.38 ± 0.03	1.10 ± 0.24	**0.74 ± 0.02**	43.06 ± 0.44	0.24 ± 0.05

Bold values indicate significant difference between the control and the treatment ($p \leq 0.05$ by Tukey's HSD test ($n = 3$; subsample = 3))

dCQA dicaffeoylquinic acid, *CQA* caffeoylquinic acid, *CMA* caffeoylmalic acid, *CTA* caffeoyltartaric acid, *dCTA* dicaffeoyltartaric acid, *mdCTA* *meso*-dicaffeoyltartaric acid, *HC* hydroxycinnamic aicd derivative


Q3Glu
Q3Glu
Q3MGlc 1
Q3MGlc 1
Q3MGlc 2
Q3MGlc 2
K3Glu
K3Glu
C3MGlc
C3MGlc
mg g-1 dry matter
0
2
4
6
8
10
12
14
control 1
10 mW m-² UVB 1
control 2
10 mW m-² UVB 2
control 3
10 mW m-² UVB 3
20 mW m-² UVB 1
20 mW m-² UVB 2
20 mW m-² UVB 3

◂**Fig. 3** Concentration of flavonol glycosides and anthocyanin in mg g^{-1} dry matter in lettuce grown in a climate chamber under consistent UV treatment either at 10 mW m^{-2} (left graphs) or at 20 mW m^{-2} (right graphs) in three experimental replicates. Asterisks indicate significant difference between the control and the treatment ($p \leq 0.05$ by Tukey's HSD test ($n = 3$; subsample = 3)). *Q* quercetin, *K* kaempferol, *C* cyanidin, *Glu* glucuronide, *MGlc* Malonylglucoside

higher concentrations of anthocyanins [23]. At 20 mW m^{-2}, however, the efficacy for kaempferol-3-*O*-glucuronide was higher, suggesting that compounds may be affected by UV treatments depending on their chemical structure [19].

3.3.2 Effect of an inconsistent UV treatment in a greenhouse on phenols and antioxidant activity

A two-factorial analysis of variance (ANOVA) revealed interactions between the factors solar PAR and UV treatment on all compounds except caffeoyltartaric acid (Data not shown). In addition, asymmetry existed between the blue-dominated and red-dominated series of experiments, as these experiments could not be run in parallel for technical reasons (see supplemental section Figure S1). Therefore, for each treatment, there was a concurrent control to which the results can be normalized to exclude the influence of solar PAR.

The phenolic acids (Table 3) showed only occasional responses to UV treatment. In particular, dicaffeoyltartaric acid or *meso*-dicaffeoyltartaric acid increased up to 1.1–1.5-fold, while selected 5-*O*-caffeoylquinic acid also increased in the same order of magnitude. Previously, in lettuce, the 5-*O*-caffeoylquinic acid was also increased by UV light [47]. Since caffeic acid is a major antioxidant [48], it seems to be increased due to UV stress, but lettuce has a variety of caffeic acid derivatives and structural relocation rather seems to take place. The additional application of blue or red light was expected to be more promising to increase the phenolic compounds in lettuce in autumn, when the experiments took place, than in spring. However, in previous experiments, only a few substances were affected and quercetin can be increased, whereas rutin decreased [49]. Here, flavonol glycosides (Fig. 4) showed an increase (1.3–2.7-fold) as a result of UV treatment in the blue-dominated and red-dominated treatments despite different solar PAR ratios due to environmental conditions in the greenhouse. Compared to red light, blue light showed higher expressions of flavonoid biosynthesis genes, such as phenylalanine ammonium lyase, flavone-3-hydroxylase, and dihydroflavonol reductase, and an increase in the corresponding metabolites such as quercetin-3-malonyl-glucoside or dicaffeoyl quinic acid [30]. In the present experiment, it was found that under blue-dominated treatment, quercetin-3-*O*-(6″-*O*-malonyl)-glucoside 1, quercetin-3-*O*-glucuronide, and kaempferol-3-*O*-glucuronide increased more than under red-dominated treatment which lines up with the results found in the literature. While solar UVA light, that was present in the greenhouse during the experiment, does not seem to increase anthocyanins, antioxidant activity and TPC in lettuce, blue light may do so [50, 51]. Other studies showed that supplemental UVA light and blue light can increase anthocyanins and antioxidant activity in lettuce while red light does not show this effect and there is also a dose–response effect [51–53]. Furthermore, combinations of blue and red light with UVA showed higher concentrations of anthocyanins in lettuce [54]. Here, combinations of blue and red light with UVB were successful to increase flavonol glycosides and the anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside of lettuce. Nevertheless, red light may act as a primer for subsequent response

Table 3 Concentration of phenolic acids in mg g^{-1} dry matter in lettuce grown in the greenhouse under inconsistent UV treatment either at blue-dominated additional light or at red-dominated additional light in three experimental replicates

UVB treatment	35dCQA	5CQA	4CQA	CMA	CTA	dCTA	mdCTA	HC
Control 1	0.52 ± 0.18	5.46 ± 0.92	0.25 ± 0.06	1.19 ± 0.25	1.27 ± 017	**0.96 ± 0.12**	**26.22 ± 2.10**	0.18 ± 0.05
215 mW m^{-2} UVB/blue 1	0.63 ± 0.12	6.22 ± 0.74	0.33 ± 0.07	1.40 ± 0.10	1.29 ± 0.27	**1.42 ± 0.16**	**35.57 ± 3.19**	0.22 ± 0.04
Control 2	1.01 ± 0.41	7.04 ± 0.59	0.39 ± 0.103	1.50 ± 0.45	1.06 ± 0.19	0.91 ± 0.17	26.03 ± 4.82	0.22 ± 0.09
215 mW m^{-2} UVB/blue 2	0.79 ± 0.31	6.62 ± 1.07	0.34 ± 0.10	1.12 ± 0.19	1.03 ± 0.25	1.03 ± 0.05	29.86 ± 1.83	0.25 ± 0.09
Control 3	0.57 ± 0.07	**7.17 ± 0.20**	**0.36 ± 0.03**	**2.50 ± 0.38**	0.58 ± 0.06	**0.72 ± 0.05**	**23.95 ± 1.01**	0.21 ± 0.05
215 mW m^{-2} UVB/blue 3	0.67 ± 0.05	**7.92 ± 0.09**	**0.45 ± 0.02**	**3.09 ± 0.33**	0.56 ± 0.05	**0.94 ± 0.04**	**29.33 ± 1.29**	0.22 ± 0.04
Control 1	**0.94 ± 0.20**	7.60 ± 0.57	**0.40 ± 0.08**	**1.45 ± 0.15**	0.83 ± 0.21	**0.90 ± 0.06**	**27.73 ± 2.23**	0.28 ± 0.08
215 mW m^{-2} UVB/red 1	**1.32 ± 0.20**	8.33 ± 0.51	**0.54 ± 0.08**	**1.15 ± 0.20**	0.87 ± 0.13	**1.38 ± 0.21**	**36.68 ± 4.11**	0.40 ± 0.08
Control 2	0.42 ± 0.11	**6.93 ± 0.31**	0.31 ± 0.08	2.04 ± 0.26	0.82 ± 0.11	0.78 ± 0.20	**27.56 ± 1.93**	0.21 ± 0.08
215 mW m^{-2} UVB/red 2	0.48 ± 0.08	**7.60 ± 0.37**	0.35 ± 0.03	2.27 ± 0.42	0.76 ± 0.21	0.88 ± 0.09	**31.78 ± 1.42**	0.23 ± 0.04
Control 3	1.13 ± 0.11	7.38 ± 0.24	0.62 ± 0.03	2.37 ± 0.23	0.50 ± 0.09	0.96 ± 0.05	**24.66 ± 0.89**	0.53 ± 0.04
215 mW m^{-2} UVB/red 3	0.91 ± 0.26	7.72 ± 0.27	0.64 ± 0.08	2.46 ± 0.09	0.60 ± 0.14	1.05 ± 0.08	**27.43 ± 1.67**	0.51 ± 0.07

Bold values indicate significant difference between the control and the treatment ($p \leq 0.05$ by Tukey's HSD test ($n = 3$; subsamples = 6))

dCQA dicaffeoylquinic acid, *CQA* caffeoylquinic acid, *CMA* caffeoylmalic acid, *CTA* caffeoyltartaric acid, *dCTA* dicaffeoyltartaric acid, *mdCTA* *meso*-dicaffeoyltartaric acid, *HC* hydroxycinnamic aicd derivative

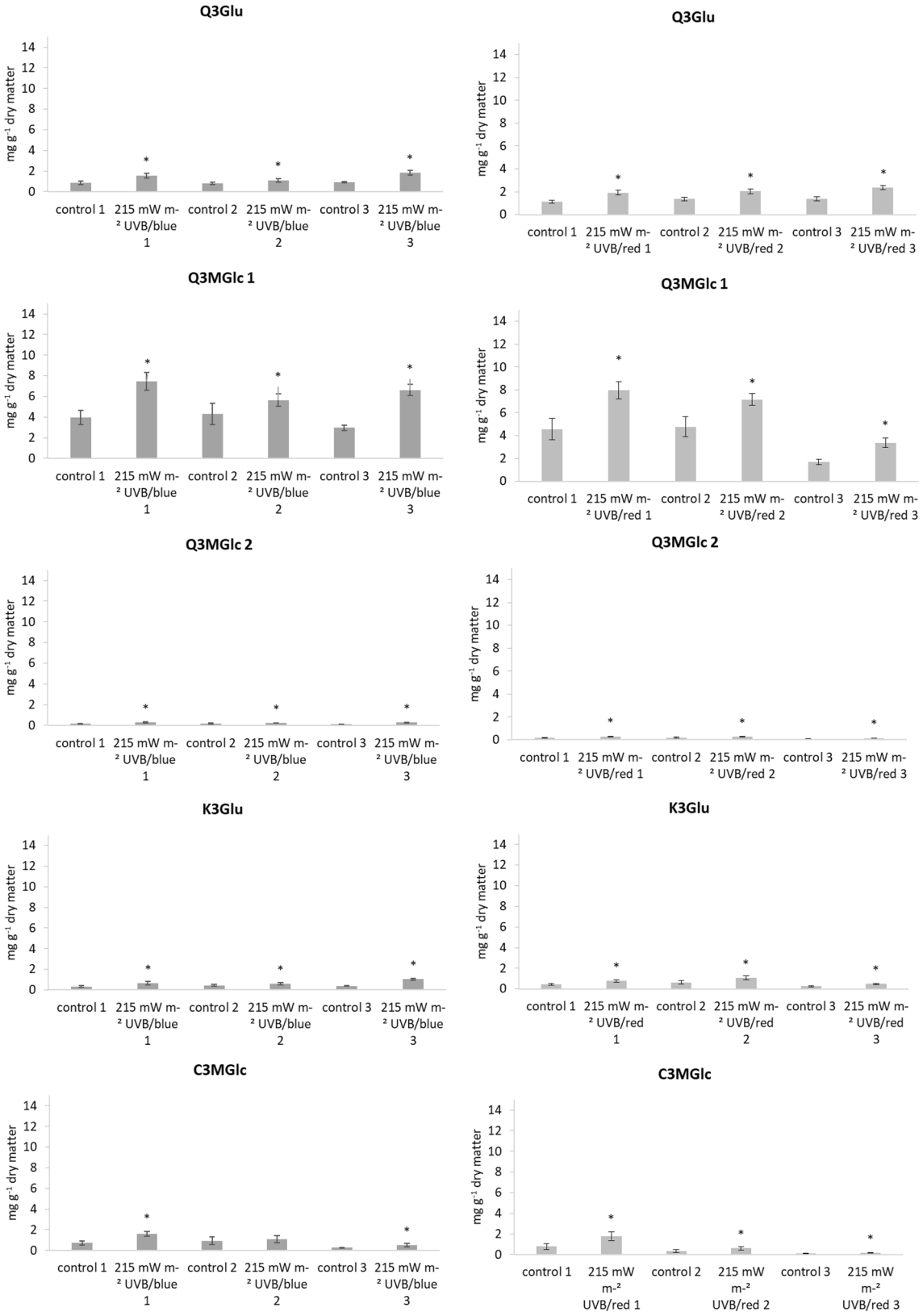


Fig. 4 Concentration of flavonol glycosides and an anthocyanin in mg g^{-1} dry matter in lettuce grown in in the greenhouse under inconsistent UV treatment either at blue-dominated additional light or at red-dominated additional light in three experimental replicates. Asterisks indicate significant difference between the control and the treatment ($p \leq 0.05$ by Tukey's HSD test ($n = 3$; subsample = 6)). *Q* quercetin, *K* kaempferol, *C* cyanidin, *Glu* glucuronide, *MGlc* Malonylglucoside

to UVA light by increasing UV-absorbing pigments [55]. It is likely that it can have a priming effect for UVB as well. In this experiment, the ratios of the individual quercetin glycosides to the kaempferol glycoside kaempferol-3-*O*-glucuronide ranged from 0.9 to 1.0. The increase of the quercetin-to-kaempferol ratio has been found previously as an indicator for UV treatment and underlines the assumption that polyhydroxylated compounds such as quercetin derivatives are favored over monohydroxylated compounds such as kaempferol derivatives in UV response [5, 29]. The treated plants had no altered quercetin to kaempferol ratio compared to the respective control. It can be concluded that kaempferol glycosides are also increased by UV treatment, as was the case here with kaempferol-3-*O*-glucuronide, and contribute to plant protection [19]. However, plants of the blue-dominated treatment showed a slightly lower quercetin-to-kaempferol ratio compared to plants of the red-dominated treatment. Similarly, the anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside increased 1.5–2.3-fold in most cases. UV-absorbing substances and anthocyanins were formed in lettuce in higher concentrations when UV was present, with the combination of UVA and UVB being more effective than UVB alone [21, 56, 57]. Higher doses of UVB and UVA also showed increased concentrations of quercetin and cyanidin in lettuce [58], and color intensity increases while the aroma of lettuce was only slightly altered and is strongly characterized by green-grassy notes [59]. Here, the anthocyanin increase resulted in a slight color change toward red leaves. Nevertheless, it must be mentioned that especially red varieties respond strongly to UV with the increase of total phenols and anthocyanins as well as quercetin, while green varieties do not always do so [60]. Also compared to red light, blue light can increase anthocyanins and total phenols [61]. However, a combination of blue and red light shows the highest levels during lettuce growth [62]. In the present experiment, both the blue-dominated and red-dominated artificial PAR resulted in an increase of the anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside in combination with the solar PAR in the greenhouse.

4 Conclusion

It has been shown that both a consistent UV treatment as it can be used in indoor farming approaches as well as an inconsistent UV treatment in greenhouses can lead to an increase of phenols especially flavonol glycosides and anthocyanins in lettuce. The effects were reproducible for flavonoid glycosides and the anthocyanin cyanidin-3-*O*-(6″-*O*-malonyl)-glucoside in both approaches. The impact of the consistent UVB treatment is higher with up to tenfold changes than that of the inconsistent UVB treatment in the greenhouse. In greenhouses, however, naturally very different light and temperature conditions are expected. Hence, a different impact of the UVB treatment might be observed in greenhouses as compared to indoor farming. The experiments were done in a period associated with decreasing global radiation and temperatures (autumn in temperate latitudes). Even though the PAR light did not have a huge effect on the outcome in this study, it needs to be considered in light experiments. Further testing is needed to determine if success can be achieved at much lower or much higher light and temperature conditions. Here, the ratio of blue to red light, given by LEDs during the UV treatment, did not seem to have an effect on the results of the UVB treatment. Nevertheless, when working with LEDs, it should be mentioned that the ratio of blue to red light as well as the presence of green light is important. Here, newly developed UVB LEDs have been tested and can be recommended for further development of lighting systems for plant growth. Lettuce was chosen as a relatively sensitive model plant for which a corresponding amount of comparison data is available and should to be used for this purpose continuously. However, light recipes for different plants have to be established.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s43630-023-00402-8.

Acknowledgements The authors thank Sarah Farrherr for the excellent technical support regarding the chemical analyses of the phenolics. The authors thank Secondary UV research project partners and subcontractors Roschwege GmbH (Greifenstein, Germany), CiS Forschungsinstitut für Mikrotechnik GmbH (Erfurt, Germany).

Author contributions Conceptualization, performing the experiments, analyzing, and interpreting these data, and writing—original draft preparation, MW, CFW, and SN; writing—review and editing, MW, CFW, TF, JG, NLP, UW, MWR, MS, and SN; supervision, SN. All authors have read and agreed to the published version of the manuscript.

Funding Open Access funding enabled and organized by Projekt DEAL. This work is funded by the Federal Ministry of Education and Research in the consortium "Advanced UV for Life" (project number: 03ZZ0128). We acknowledge support by the Open Access Publication Funds of Göttingen University.

Data availability The data presented in this study are available on request from the corresponding author.

Declarations

Conflict of interest The authors declare no conflict of interest.

Institutional review board statement Not applicable.

Informed consent Not applicable.

References

1. Peñate, L., Martín, O., Cárdenas, R., & Agustí, S. (2010). Short-term effects of gamma ray bursts on oceanic photosynthesis. *Astrophysics and Space Science, 330*(2), 211–217.
2. Seckmeyer, G., Pissulla, D., Glandorf, M., Henriques, D., Johnsen, B., Webb, A., Siani, A.-M., Bais, A., Kjeldstad, B., Brogniez, C., Lenoble, J., Gardiner, B., Kirsch, P., Koskela, T., Kaurola, J., Uhlmann, B., Slaper, H., den Outer, P., Janouch, M., … Carvalho, F. (2008). Variability of UV irradiance in Europe. *Photochemistry and Photobiology, 84*(1), 172–179.
3. Solovchenko, A. E., & Merzlyak, M. N. (2008). Screening of visible and UV radiation as a photoprotective mechanism in plants. *Russian Journal of Plant Physiology, 55*(6), 719.
4. Solovchenko, A. (2010). Screening pigments: general questions, in photoprotection in plants. In A. Solovchenko & A. Solovčenko (Eds.), *Optical screening-based mechanisms* (Vol. 14, pp. 9–31). Heidelberg: Springer.
5. Agati, G., & Tattini, M. (2010). Multiple functional roles of flavonoids in photoprotection. *The New Phytologist, 186*(4), 786–793.
6. Thilakarathna, S. H., & Rupasinghe, H. P. V. (2013). Flavonoid bioavailability and attempts for bioavailability enhancement. *Nutrients, 5*(9), 3367–3387.
7. Falcone Ferreyra, M. L., Rius, S. P., & Casati, P. (2012). Flavonoids: Biosynthesis, biological functions, and biotechnological applications. *Frontiers in Plant Science*. https://doi.org/10.3389/fpls.2012.00222
8. Křístková, E., Doležalová, I., Lebeda, A., Vinter, V., & Novotná, A. (2018). Description of morphological characters of lettuce (*Lactuca sativa* L.) genetic resources. A review. *Horticultural Science, 35*, 113–129.
9. Yang, X., Wei, S., Liu, B., Guo, D., Zheng, B., Feng, L., Liu, Y., Tomás-Barberán, F. A., Luo, L., & Huang, D. (2018). A novel integrated non-targeted metabolomic analysis reveals significant metabolite variations between different lettuce (*Lactuca sativa* L.) varieties. *Horticulture Research, 5*, 33.
10. Assefa, A. D., Choi, S., Lee, J.-E., Sung, J.-S., Hur, O.-S., Ro, N.-Y., Lee, H.-S., Jang, S.-W., & Rhee, J.-H. (2019). Identification and quantification of selected metabolites in differently pigmented leaves of lettuce (*Lactuca sativa* L) cultivars harvested at mature and bolting stages. *BMC Chemistry, 13*(1), 56.
11. Ismail, H., Gillespie, A. L., Calderwood, D., Iqbal, H., Gallagher, C., Chevallier, O. P., Elliott, C. T., Pan, X., Mirza, B., & Green, B. D. (2019). The health promoting bioactivities of *Lactuca sativa* can be enhanced by genetic modulation of plant secondary metabolites. *Metabolites, 9*(5), 97.
12. Vauzour, D., Rodriguez-Mateos, A., Corona, G., Oruna-Concha, M. J., & Spencer, J. P. E. (2010). Polyphenols and human health: prevention of disease and mechanisms of action. *Nutrients, 2*(11), 1106–1131.
13. Mayorga, C., Tsukagoshi, S., & Sasaki, A. (2019). Effect on the growth and nutritional components in two red lettuces (*Lactuca sativa* L.) cultivated under UV light in a mini plant factory. *Journal of Agriculture and Crops, 56*, 71–77.
14. Taulavuori, E., Taulavuori, K., Holopainen, J. K., Julkunen-Tiitto, R., Acar, C., & Dincer, I. (2017). Targeted use of LEDs in improvement of production efficiency through phytochemical enrichment. *Journal of the Science of Food and Agriculture, 97*(15), 5059–5064.
15. Pattison, P. M., Tsao, J. Y., Brainard, G. C., & Bugbee, B. (2018). LEDs for photons, physiology and food. *Nature, 563*(7732), 493–500.
16. Pennisi, G., Orsini, F., Blasioli, S., Cellini, A., Crepaldi, A., Braschi, I., Spinelli, F., Nicola, S., Fernandez, J. A., Stanghellini, C., Gianquinto, G., & Marcelis, L. F. M. (2019). Resource use efficiency of indoor lettuce (*Lactuca sativa* L.) cultivation as affected by red:blue ratio provided by LED lighting. *Scientific Reports, 9*(1), 14127.
17. Benke, K., & Tomkins, B. (2017). Future food-production systems: vertical farming and controlled-environment agriculture. *Sustainability, 13*(1), 13–26.
18. Ajmal Khan, M., Matsuura, E., Kashima, Y., & Hirayama, H. (2019). Overcoming the current injection issue in the 310 nm band AlGaN UVB light-emitting diode. *Japanese Journal of Applied Physics, 59*(SA), SAAD01.
19. Neugart, S., & Schreiner, M. (2018). UVB and UVA as eustressors in horticultural and agricultural crops. *Scientia Horticulturae, 234*, 370–381.
20. Mormile, P., Rippa, M., Graziani, G., & Ritieni, A. (2019). Use of greenhouse-covering films with tailored UV-B transmission dose for growing 'medicines' through plants: rocket salad case. *Journal of the Science of Food and Agriculture, 99*(15), 6931–6936.
21. Garcia-Macias, P., Ordidge, M., Vysini, E., Waroonphan, S., Battey, N. H., Gordon, M. H., Hadley, P., John, P., Lovegrove, J. A., & Wagstaffe, A. (2007). Changes in the flavonoid and phenolic acid contents and antioxidant activity of red leaf lettuce (*Lollo Rosso*) due to cultivation under plastic films varying in ultraviolet transparency. *Journal of Agricultural and Food Chemistry, 55*(25), 10168–10172.
22. Assumpção, C. F., Assis, R. Q., Hermes Poletto, V. S., Castagna, A., Ranieri, A., Neugart, S., Flôres, S. H., & de Oliveira Rios, A. (2019). Application of supplemental UV-B radiation in preharvest to enhance health-promoting compounds accumulation in green and red lettuce. *Journal of Food Processing and Preservation, 43*(11), e14213.
23. Lee, M. J., Son, J. E., & Oh, M. M. (2014). Growth and phenolic compounds of *Lactuca sativa* L. grown in a closed-type plant production system with UV-A, -B, or -C lamp. *Journal of the Science of Food and Agriculture, 94*(2), 197–204.
24. Wargent, J. J., Nelson, B. C. W., McGhie, T. K., & Barnes, P. W. (2015). Acclimation to UV-B radiation and visible light in *Lactuca sativa* involves up-regulation of photosynthetic performance and orchestration of metabolome-wide responses. *Plant, Cell & Environment, 38*(5), 929–940.
25. Rodriguez, C., Torre, S., & Solhaug, K. A. (2014). Low levels of ultraviolet-B radiation from fluorescent tubes induce an efficient flavonoid synthesis in Lollo Rosso lettuce without negative impact on growth. *Acta Agriculturae Scandinavica Section B-Soil and Plant Science, 64*(2), 178–184.
26. Samuolienė, G., Brazaitytė, A., Sirtautas, R., Viršilė, A., Sakalauskaitė, J., Sakalauskienė, S., & Duchovskis, P. (2013). LED illumination affects bioactive compounds in romaine baby leaf lettuce. *Journal of the Science of Food and Agriculture, 93*(13), 3286–3291.
27. Goto, E., Hayashi, K., Furuyama, S., Hikosaka, S., & Ishigami, Y. (2016). Effect of UV light on phytochemical accumulation and

expression of anthocyanin biosynthesis genes in red leaf lettuce. *Acta Horticulturae, 1134*, 179–186.
28. Wiesner-Reinhold, M., Dutra Gomes, J. V., Herz, C., Tran, H. T. T., Baldermann, S., Neugart, S., Filler, T., Glaab, J., Einfeldt, S., Schreiner, M., & Lamy, E. (2021). Subsequent treatment of leafy vegetables with low doses of UVB-radiation does not provoke cytotoxicity, genotoxicity, or oxidative stress in a human liver cell model. *Food Bioscience, 43*, 101327.
29. Bilger, W., Rolland, M., & Nybakken, L. (2007). UV screening in higher plants induced by low temperature in the absence of UV-B radiation. *Photochemical & Photobiological Sciences, 6*(2), 190–195.
30. Kitazaki, K., Fukushima, A., Nakabayashi, R., Okazaki, Y., Kobayashi, M., Mori, T., Nishizawa, T., Reyes-Chin-Wo, S., Michelmore, R. W., Saito, K., Shoji, K., & Kusano, M. (2018). Metabolic reprogramming in leaf lettuce grown under different light quality and intensity conditions using narrow-band LEDs. *Scientific Reports, 8*(1), 7914.
31. Son, K.-H., & Oh, M.-M. (2013). Leaf shape, growth, and antioxidant phenolic compounds of two lettuce cultivars grown under various combinations of blue and red light-emitting diodes. *Horticultural Science, 48*(8), 988–995.
32. Bian, Z., Cheng, R., Wang, Y., Yang, Q., & Lu, C. (2018). Effect of green light on nitrate reduction and edible quality of hydroponically grown lettuce (*Lactuca sativa* L.) under short-term continuous light from red and blue light-emitting diodes. *Environmental and Experimental Botany, 153*, 63–71.
33. Neugart, S., Rohn, S., & Schreiner, M. (2015). Identification of complex, naturally occurring flavonoid glycosides in *Vicia faba* and *Pisum sativum* leaves by HPLC-DAD-ESI-MS^n and the genotypic effect on their flavonoid profile. *Food Research International, 76*, 114–121.
34. Heinze, M., Hanschen, F. S., Wiesner-Reinhold, M., Baldermann, S., Grafe, J., Schreiner, M., & Neugart, S. (2018). Effects of developmental stages and reduced UVB and low UV conditions on plant secondary metabolite profiles in pak choi (*Brassica rapa* subsp. c*hinensis). Journal of Agricultural and Food Chemistry, 66*(7), 1678–1692.
35. Hooks, T., Masabni, J., Sun, L., & Niu, G. (2021). Effect of preharvest supplemental UV-A/blue and red/blue LED lighting on lettuce growth and nutritional quality. *Horticulturae, 7*(4), 80.
36. Rajabbeigi, E., Eichholz, I., Beesk, N., Ulrichs, C., Kroh, L. W., Rohn, S., & Huyskens-Keil, S. (2013). Interaction of drought stress and UV-B radiation—impact on biomass production and flavonoid metabolism in lettuce (*Lactuca sativa* L.). *Journal of Applied Botany and Food Quality, 86*, 190–197.
37. Cammarisano, L., Donnison, I. S., & Robson, P. R. H. (2021). The effect of red and blue rich LEDs vs fluorescent light on 'Lollo rosso' lettuce morphology and physiology. *Frontiers in Plant Science, 12*, 603411.
38. He, R., Zhang, Y., Song, S., Su, W., Hao, Y., & Liu, H. (2021). UV-A and FR irradiation improves growth and nutritional properties of lettuce grown in an artificial light plant factory. *Food Chemistry, 345*, 128727.
39. Jansen, M. A. K. (2002). Ultraviolet-B radiation effects on plants: induction of morphogenic responses. *Physiologia Plantarum, 116*(3), 423–429.
40. Becker, C., Klaring, H. P., Kroh, L. W., & Krumbein, A. (2013). Temporary reduction of radiation does not permanently reduce flavonoid glycosides and phenolic acids in red lettuce. *Plant Physiology and Biochemistry, 72*, 154–160.
41. Park, J.-S., Choung, M.-G., Kim, J.-B., Hahn, B.-S., Kim, J.-B., Bae, S.-C., Roh, K.-H., Kim, Y.-H., Cheon, C.-I., Sung, M.-K., & Cho, K.-J. (2007). Genes up-regulated during red coloration in UV-B irradiated lettuce leaves. *Plant Cell Reports, 26*(4), 507–516.
42. Uranga, J. G., Podio, N. S., Wunderlin, D. A., & Santiago, A. N. (2016). Theoretical and experimental study of the antioxidant behaviors of 5-O-Caffeoylquinic quinic and caffeic acids based on electronic and structural properties. *ChemistrySelect, 1*(13), 4113–4120.
43. Fiol, M., Adermann, S., Neugart, S., Rohn, S., Mügge, C., Schreiner, M., Krumbein, A., & Kroh, L. W. (2012). Highly glycosylated and acylated flavonols isolated from kale (*Brassica oleracea* var. sabellica)—structure–antioxidant activity relationship. *Food Research International, 47*(1), 80–89.
44. Csepregi, K., Neugart, S., Schreiner, M., & Hideg, E. (2016). Comparative evaluation of total antioxidant capacities of plant polyphenols. *Molecules, 21*(2), 208.
45. Majer, P., Neugart, S., Krumbein, A., Schreiner, M., & Hideg, É. (2014). Singlet oxygen scavenging by leaf flavonoids contributes to sunlight acclimation in *Tilia platyphyllos*. *Environmental and Experimental Botany, 100*, 1–9.
46. Neugart, S., Tobler, M. A., & Barnes, P. W. (2019). Different irradiances of UV and PAR in the same ratios alter the flavonoid profiles of *Arabidopsis thaliana* wild types and UV-signalling pathway mutants. *Photochemical & Photobiological Sciences, 18*(7), 1685–1699.
47. Sytar, O., Zivcak, M., Bruckova, K., Brestic, M., Hemmerich, I., Rauh, C., & Simko, I. (2018). Shift in accumulation of flavonoids and phenolic acids in lettuce attributable to changes in ultraviolet radiation and temperature. *Scientia Horticulturae, 239*, 193–204.
48. Schröter, D., Neugart, S., Schreiner, M., Grune, T., Rohn, S., & Ott, C. (2019). Amaranth's 2-caffeoylisocitric acid—an anti-inflammatory caffeic acid derivative that impairs NF-κB signaling in LPS-challenged RAW 264.7 macrophages. *Nutrients, 11*(3), 571.
49. Brazaitytė, A., Vaštakaitė-Kairienė, V., Sutulienė, R., Rasiukevičiūtė, N., Viršilė, A., Miliauskienė, J., Laužikė, K., Valiuškaitė, A., Dėnė, L., Chrapačienė, S., Kupčinskienė, A., & Samuolienė, G. (2022). Phenolic compounds content evaluation of lettuce grown under short-term preharvest daytime or nighttime supplemental LEDs. *Plants, 11*(9), 1123.
50. Gómez, C., & Jiménez, J. (2020). Effect of end-of-production high-energy radiation on nutritional quality of indoor-grown red-leaf lettuce. *Horticultural Science, 55*(7), 1055–1060.
51. Yan, Z., He, D., Niu, G., Zhou, Q., & Qu, Y. (2020). Growth, nutritional quality, and energy use efficiency in two lettuce cultivars as influenced by white plus red versus red plus blue LEDs. *International Journal of Agricultural and Biological Engineering, 13*(2), 33–40.
52. Li, Q., & Kubota, C. (2009). Effects of supplemental light quality on growth and phytochemicals of baby leaf lettuce. *Environmental and Experimental Botany, 67*(1), 59–64.
53. Naznin, M. T., Lefsrud, M., Gravel, V., & Azad, M. O. K. (2019). Blue light added with red LEDs enhance growth characteristics, pigments content, and antioxidant capacity in lettuce, spinach, kale, basil, and sweet pepper in a controlled environment. *Plants, 8*(4), 93.
54. Kim, Y. H., & Lee, J. S. (2016). Growth and contents of anthocyanins and ascorbic acid in lettuce as affected by supplemental UV-A LED irradiation with different light quality and photoperiod. *Korean Journal of Horticultural Science & Technology*, 34(4), 596-606. https://doi.org/10.12972/kjhst.20160061
55. Kreslavski, V. D., Lyubimov, V. Y., Shirshikova, G. N., Shmarev, A. N., Kosobryukhov, A. A., Schmitt, F.-J., Friedrich, T., & Allakhverdiev, S. I. (2013). Preillumination of lettuce seedlings with red light enhances the resistance of photosynthetic apparatus to UV-A. *Journal of Photochemistry and Photobiology B, 122*(Supplement C), 1–6.
56. Krizek, D. T., Britz, S. J., & Mirecki, R. M. (1998). Inhibitory effects of ambient levels of solar UV-A and UV-B radiation on

growth of cv new red fire lettuce. *Physiologia Plantarum, 103*(1), 1–7.
57. Quintero-Arias, D. G., Acuña-Caita, J. F., Asensio, C., & Valenzuela, J. L. (2021). Ultraviolet transparency of plastic films determines the quality of lettuce (*Lactuca sativa* L.) grown in a greenhouse. *Agronomy, 11*(2), 358.
58. Behn, H., Schurr, U., Ulbrich, A., & Noga, G. (2011). Development-dependent UV-B responses in red oak leaf lettuce (*Lactuca sativa* L.): physiological mechanisms and significance for hardening. *European Journal of Horticultural Science, 76*, 33–40.
59. Gude, K., Talavera, M., Sasse, A. M., Rivard, C. L., & Pliakoni, E. (2021). Effect of light characteristics on the sensory properties of red lettuce (*Lactuca sativa* L.). *Foods, 10*(11), 2660.
60. Ordidge, M., Garcia-Macias, P., Battey, N. H., Gordon, M. H., Hadley, P., John, P., Lovegrove, J. A., Vysini, E., & Wagstaffe, A. (2010). Phenolic contents of lettuce, strawberry, raspberry, and blueberry crops cultivated under plastic films varying in ultraviolet transparency. *Food Chemistry, 119*(3), 1224–1227.
61. Chung, H.-Y., Chang, M.-Y., Wu, C.-C., & Fang, W. (2018). Quantitative evaluation of electric light recipes for red leaf lettuce cultivation in plant factories. *Hortte, 28*(6), 755–763.
62. Johkan, M., Shoji, K., Goto, F., Hashida, S., & Yoshihara, T. (2010). Blue light-emitting diode light irradiation of seedlings improves seedling quality and growth after transplanting in red leaf lettuce. *Horticultrual Science, 45*(12), 1809–1814.