DOSIMETRIC MEASUREMENT OF THE VISIBLE AND UV
EXPOSURES ON FIELD GROWN SOYBEAN PLANTS

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Abstract

This paper describes a dosimeter system for measuring both the PAP (photosynthetically active photons) (400-700 nm) and ultraviolet-B (UVB) (280-320 nm) exposures in the supplemental UVB irradiation of field grown soybean (*Glycine max* [L.] Merr.) plants. At the V2 growth stage, the dosimeters positioned at the same position and orientation as the trifoliate leaves of the plants that were measured received 12 to 38% more PAP and 5 to 82% more UVB than the unifoliate leaves. For the crop maturity stage, the plants exposed to high levels of UV irradiance (high UV treatment) received approximately 40% more UVB on a horizontal plane at the top of the plant canopy compared to the control group of plants (control treatments). For the other measurement sites over the plants that were orientated at 45 degrees to the vertical in the north, south, east and west directions, the additional amount of UVB for the supplemental treatment compared to the corresponding sites for the plants in the control treatments varied between -39% and 37%, due predominantly to the shading provided by the other plants. Furthermore, the supplemental UVB changed the natural partitioning of UVB and PAP and the ratio of PAP to UVB over the plants. All these variations to the UVB and PAP over the plant canopy cannot be predicted by exposure measurement in the wavebands on a horizontal plane. Consequently, for the case of the complex topography of plants, the dosimeters described in this paper have the advantage of allowing the exposures to be measured simultaneously at multiple sites that are at any orientation.

**Keywords:** Ultraviolet; Dosimeters; Photosynthetically active radiation; Plants
1. Introduction

Agricultural production may be influenced by decreased stratospheric ozone and in order to develop reliable quantitative prediction of the influence on plant growth, research employing the irradiation of plants with supplemental UV is necessary. Research in greenhouses and growth cabinets provides information on the mechanisms of the effects of UV on plants. Additionally, field research with supplemental UV on plants under realistic microclimate conditions is required. For example, alfalfa (*Medicago sativa* L.) seedlings grown in laboratory conditions were found to be more sensitive to UV induced DNA damage compared to plants grown outdoors (Takayanagi et al., 1994). McLeod (1997) provides a review of outdoor supplementation systems for studies of increased UV on plants.

These studies require information on both the UV exposures and the photosynthetically active radiation (400–700 nm) of plants. Measurements of the photosynthetically active radiation are reported as photosynthetic photon flux density (PPFD) for instantaneous levels or photosynthetically active photons (PAP) for time integrated exposures (CIE, 1993). Knowledge of the PPFD in supplemental UV studies is necessary as research has shown that plants grown under high PPFD conditions are less susceptible to UV damage (Mirecki and Teramura, 1984). Previous research has measured the UV exposures on a horizontal surface as a proxy for other surfaces (Caldwell et al., 1994). However, more research is required on the irradiances on non horizontal surfaces. Models have been developed to calculate the light penetration into a plant canopy (Grant, 1997) and through tree canopies (Grant and Heisler, 1999). The difference between irradiance on a horizontal flat plane and fluence rate, or the radiation intercepted by a sphere of unit cross section per unit time, was examined by Bjorn and Vogelmann (1996) for natural light conditions who reported that it is not possible to calculate fluence rate from a single irradiance measurement without detailed information of the conditions. The UV and photosynthetically active radiation irradiances were measured over a hemispherical surface at the base of a corn (*Zea mays* L.) canopy (Grant, 1991). A system of diodes has also been used in plant canopies with each diode either mounted on leaves or fixed in the canopy (Gutschild et al., 1985; Sassenrath-Cole, 1995). Sassenrath-Cole (1995) found that a species with cupped leaves resulted in more photosynthetically active radiation to the lower leaves compared to a species with a more regular leaf structure.

Polysulphone dosimeters have been previously employed for the measurement of the UV exposures on humans (Diffey et al., 1996). These dosimeters respond only to wavelengths shorter than 330 nm (CIE, 1992). A system of UV dosimeters measured the UV exposures on the leaves of soybean (*Glycine max* [L.] Merr.) plants in supplemental UV research in a greenhouse (Parisi et al., 1998a). As the relative proportion of the diffuse to direct radiation varies across the UV and visible wavebands, it is also necessary to measure the PAP in supplemental UV research. To the authors' knowledge, no dosimetric system has been previously employed to measure both the PAP and UVB (280-320 nm) exposures in a field crop using a supplemental UV system. An innovative, reliable and passive dosimeter for measuring the PAP has been previously described (Parisi et al., 1998b). The PAP dosimeter and the polysulphone dosimeter will be
combined in this paper to provide a system to measure both the PAP and UVB exposures on crop plants to an outdoor supplemental UV system.

2. Materials and Methods

2.1 Supplemental UV System

Soybean seeds (cv. Essex) were planted 7 cm apart in east-west rows spaced at 0.48 m intervals at the start of summer (1 December 1997) in Toowoomba (27.5° S latitude). The soil was cultivated with fertiliser applications of Urea (46% N) at 100 kg/ha and CK88 (14.8% N, 4.3% P, 11.3% K and 13.4% S) applied at 200 kg/ha. A UV irradiation system was installed over the plants with six different treatments and three replicates of the six treatments in a randomised block design. The six treatments were a high and low supplemental UV treatment and a control and dummy treatment for each of the supplemental UV treatments. A buffer of 4 rows was provided between each treated row, along with 1.2 m of plants between each treatment in a row (Figure 1). Each treatment plot was 2.4 m x 2.4 m and the trial arranged in a series of three replicates side by side with each replicate consisting of 3 by 2 treatments. The total trial covered 7.2 m x 14.4 m. A frame of hardwood timber 25 cm x 50 cm was fabricated over each treatment to allow suspension of the UV lamps and reflectors.

The supplemental UV irradiance treatments, designated ‘TL’ (low) and ‘TH’ (high) were produced by 1.2 m long Phillips TL40/12 lamps (supplier, Lawrence and Hanson, Toowoomba, Australia) suspended over the soybean rows as shown in Figure 2. The low UV treatment refers to supplemental UV, but at a lower level than that of the supplemental UV provided in the high treatment. Both were provided by suspending the lamps at heights of 40 cm and 25 cm respectively above the plant tops. Two sets of dummy treatments were employed, namely lamp holders with no lamps set at a height of 40 cm and 25 cm above the average plant top (DL and DH respectively) and two sets of control treatments supplied by lamps filtered through Mylar (supplier, Cadillac Plastics, Australia) that provided zero irradiance at wavelengths shorter than 320 nm and set at a height of 40 cm and 25 cm above the average plant top (CL and CH respectively).

Each lamp was fitted with a white reflector shield over the top of the lamp. In addition to directing the UV downwards, this also provided the benefit of keeping water off the lamps. The electronic ballasts were removed from each lamp and installed in a weather-proof box at the end of each replicate. The electrical wires were run from the boxes and attached to the appropriate lamp ends. A sleeve cover approximately 20 to 30 cm was fabricated from a bicycle inner tube and employed to protect the connection of the wires to the lamp ends from the weather. For each treatment, the measurements were made in the region that was at least two soybean plants inside the ends of the lamp where the irradiance was relatively uniform. The lamps were raised each week as the plants grew to maintain a constant average height above the plants.

The supplemental UVB was provided the UV lamps with a 0.13 mm cellulose acetate filter (CA) (supplier, Artery, Hobart, Australia) around each lamp tube as previously described (Mirecki and Teramura, 1984). This provided a zero irradiance at
approximately 292 nm (Parisi et al., 1996). Both the Mylar and CA filter material were pre-solarised for eight hours prior to wrapping around the lamps. The CA filters were replaced twice weekly and the Mylar filters were replaced weekly as both materials degrade with use due to the effects of heat and UVB radiation. The supplemental UV was commenced at seedling emergence and the lamps turned on and off with an automatic timer at 9:00 and 15:00 Australian Eastern Standard Time (EST) respectively.

2.2 Dosimeter System
The development and testing of a dosimeter system for the measurement of the PAP was described by Parisi et al. (1998b). Briefly the system consists of 35 mm AGFA 25 APX photographic film with two pieces of pre-exposed film acting as neutral density filters. The system is based on a 9 cm x 9 cm frame, with a total weight of 13 g, fabricated from black plastic to prevent stray light leakage with an opening of 1 cm x 2 cm to expose the film. Following exposure, the film was processed with a standardised developing process and the change in absorbance at 800 nm, measured with a spectrophotometer (model UV-1601, Shimadzu Co., Kyoto, Japan) and compared to a piece of processed unexposed film to quantify the amount of darkening of the dosimeter due to exposure. The dosimeter was calibrated for PAP by exposing a series of dosimeters on a horizontal plane for different times while measuring the visible spectrum between 400 and 700 nm with a calibrated spectroradiometer. The photosynthetic photon flux density, PPFD was calculated as given in (McCree, 1981):

\[ \text{PPFD} = 8.35 \times 10^{-9} \int_{400}^{700} \lambda S(\lambda) d\lambda \]  \( \mu \text{mol m}^{-2} \text{s}^{-1} \)  (1)

where \( S(\lambda) \) is the incident spectral irradiance in \( \mu \text{W m}^{-2} \text{nm}^{-1} \) and \( \lambda \) is the wavelength in nanometres. The time integrated exposures over a period, \( t \), were calculated in terms of photosynthetically active photons, PAP (CIE, 1993) as follows:

\[ \text{PAP} = t \text{PPFD} \times 10^{-6} \text{ mol m}^{-2} \]  (2)

The dosimeter film has been measured to be independent of temperature between 25 and 42°C and the error in the use of these dosimeters due to sources such as timing of the exposure, calibration and film development is of the order of \( \pm 20\% \) (Parisi et al., 1998b).

The polysulphone dosimeters were fabricated from 40 \( \mu \text{m} \) thick polysulphone film. The sheet was cut and mounted into 25 mm x 25 mm rigid plastic holders, each with a 1 cm\(^2\) central aperture. Polysulphone is sensitive to wavelengths shorter than 330 nm and was employed in this research for the measurement of the UVB waveband. The UV induced change in optical absorbance at 330 nm, \( \Delta A \), was determined by measuring the pre- and post- exposure optical absorbance in the spectrophotometer. The change in optical absorbance of the dosimeters is related to the UVB radiant exposure, by (Diffey, 1989):

\[ K(\Delta A + \Delta A^2 + 9\Delta A^3) \]  (3)

where \( K \) in units of \( J \text{m}^{-2} \) is a constant determined in the calibration process (Diffey, 1989). The ratio of the UVB exposures to the specific orientations described in the next section over the plants compared to the exposures on a horizontal plane at the tops of the canopies was employed and the constant \( K \) cancelled in the calculation of the ratio.
2.3 Measurements in the Field
The dosimetric measurements of the UVB and PAP were undertaken at the full expansion of the first trifoliate leaves above the unifoliate node (V2 growth stage) and at the crop maturity stage. These stages corresponded to 28 and 105 days respectively from the planting date. The measurements were made between 8:30 and 9:00, 11:30 and 12:00 and 14:30 and 15:00 EST. For these times, the solar zenith angle at the V2 stage measurements were 39°, 5° and 42° respectively and 50°, 25° and 50° for the crop maturity stage measurements. These periods will be referred to as 9:00, 12:00 and 15:00 EST in the following text. The cloud cover was 4, 7 and 8 okta for each respective time at the V2 stage and 0, 0 and 1 okta for each respective time at the crop maturity stage.

The dosimeters were deployed over one plant in each treatment on a lightweight frame constructed from 2 mm diameter steel rods (Parisi et al., 1996). At the V2 growth stage, two dosimeters were positioned at the same position and orientation as the unifoliate and trifoliate leaves. At the crop maturity stage, five dosimeters were orientated over the measurement plant. The first was at the approximate top of the plant canopy on a horizontal plane. The remaining four were at an angle of 45 degrees to the horizontal and angles of 0, 90, 180 and 270 degrees relative to north. These orientations will be respectively referred to as Top, N, E, S and W. This prevented any changes in the natural angle and position of the leaves. At the V2 stage, the dosimeters were exposed for 15 minutes and 60 seconds for the UVB and PAP dosimeters respectively. These exposure times were a compromise between a sufficient time to produce a measurable change in the dosimeters and minimisation of changes in cloud cover and wind induced leaf orientation. The PAP dosimeters produced a response in a shorter time than the UVB dosimeters and required a shorter exposure time. The dosimeters were orientated in the approximate plane of the unifoliate and trifoliate leaves and held on the frame by lightweight clips. At the crop maturity stage, the exposure times were 10 minutes and 30 seconds for the UVB and PAP dosimeters respectively. These shorter exposure times compared to those for the V2 stage were employed due to the comparatively lower amount of cloud cover. For each time and dosimeter orientation the UVB exposures were averaged for the two control and two dummy treatments. The relative values refer to the exposure on a horizontal plane at 12:00 EST for the high UV treatment.

3. Results

3.1 Dosimeter Measurements
The PAP exposures to the unifoliate and trifoliate leaves for the V2 growth stage for each of the three time periods are shown in Figure 3. These are the average for the six measured plants and the error bars are ±20%. These PAP exposures have been averaged over the six plants as the UV lamps have a negligible emission in the visible wavelengths and the PAP exposures should be relatively uniform across the six treatments. The PAP exposure on the trifoliate leaves at 15:00 EST is almost the same as that on the unifoliate leaves due to the high amount of cloud cover experienced at that time resulting in a highly diffuse distribution of the visible radiation. The UVB exposures to the approximate planes of the unifoliate and trifoliate leaves for the V2 stage at 12:00 EST
relative to those for the trifoliate leaf orientation of the high UV treatment at the same time are provided in Table 1. The errors are ±20% as the error in the individual polysulphone measurements is of the order of 10% (Diffey, 1989) and the value of 20% is due to the propagation of the error in the calculation of the relative UVB exposures. The values on the control/dummy treatments have been averaged as none of these treatments are exposed to supplemental UVB. The relative UV exposures to the control plants were lower (approximately 50% and 62%) for the unifoliate and trifoliate leaves respectively when compared to the trifoliate leaves of the high UV treatment. The relative UVB exposure was higher for the dosimeter orientated on the plane of the unifoliate of the low UV treatment compared to that of the high UV treatment. This is due to possible differences in the orientation of the plane of the respective unifoliate and also differences in the shading to the unifoliate leaves in each treatment resulting from the trifoliate leaves and surrounding plants. Also shown in Table 1 are the PAP values for this stage at 12:00 EST. This difference between the two sets of leaves is greater in the UV compared to the PAP due to the supplemental UV provided to the treated plants. This is due to the UV lamps providing only minimal output in the visible waveband.

For the crop maturity stage, the relative UVB exposures on the control plants for each orientation of the dosimeters at the three times are provided in Figure 4a. These are all relative to those on a horizontal plane at 12:00 EST of the high UV treatment. The range over the whole day of the relative UVB exposures for the control/dummy plants is 0.17 to 0.68. This shows that exposures larger than on a horizontal plane may occur at other orientations; in this case, the N orientation had a relative exposure of 0.68 compared with 0.61 on a horizontal plane. At the same time, the relative exposure was 0.21 for the S orientation. In comparison for the 9:00 EST measurements, the S and W orientations had the lowest exposures with values of 0.17 and 0.18 respectively. At 15:00 EST, the E and S orientations had the lowest exposures with 0.19 and 0.23 respectively and the W orientation had the highest with 0.35.

The relative UVB exposures for the high UV treatment are provided in Figure 4(b). The supplemental UV changes the exposure distribution over the plant. At 12:00 EST, the orientation with the highest relative exposure is now the top dosimeter position on a horizontal plane whereas, for the control plants, the highest exposure orientation was to the north. The corresponding PAP exposures averaged over the six plants are in Figure 5. Again, the highest exposures are not those on the horizontal plane, but the N orientation with exposures of 1.4 mol m⁻².

4. Discussion

The trifoliate leaves as measured by the dosimeters received 38%, 12% and 12% more PAP dose per unit area than the unifoliate leaves at 9:00 EST, 12:00 EST and 15:00 EST respectively for the V2 stage (Figure 3) due to possible shading by the other leaves and plants. The difference at 9:00 EST is higher than that at the other two times due to less cloud cover in the morning. Similarly, for the UVB, the dosimeters at the orientation of the trifoliate leaves also received more UVB. At 12:00 EST, the trifoliate leaves dosimeters received 82%, 5% and 24% more than the unifoliate leaves dosimeters for the
high UV, low UV and control/dummy treatments respectively (Table 1). The difference between the two sets of leaves was largest for the high UV treatment due both to shading and the trifoliate leaves being closer to the lamps. The UVB and PAP exposures on the different leaves are influenced by the position of the leaves on the plant, orientation of the leaves, shading of the leaves, time of day and the amount of cloud cover. However, from the results in this research, there does not seem to be a definite relationship between the UVB and PAP exposures on the unifoliate and trifoliate leaves due to the variations in orientations and shading of the leaves on the different plants.

For the crop maturity stage at 12:00 EST, the PAP exposures were 0.83, 1.4, 0.36, 0.54 and 0.89 mol m\(^{-2}\) to the horizontal plane, N, E, S and W orientations respectively (Figure 5) compared to a mean of 0.81. For the same respective orientations, the relative UVB exposures for the high UV treatment were 1.0, 0.57, 0.68, 0.60 and 0.58 giving a mean of 0.69 (Figure 4b) and 0.61, 0.68, 0.53, 0.21 and 0.54 giving a mean of 0.51 (Figure 4b) for the low UV treatment. The N orientation received an additional 72% PAP compared to the horizontal plane. In comparison, the UVB exposures for the control/dummy plants at the N orientation were 11% higher than on a horizontal plane (Figure 4a). A possible reason for this is the greater amount of atmospheric scattering of the shorter wavelengths producing a greater relative proportion of diffuse radiation. In comparison, the high UV treatment had approximately 40% higher UV on a horizontal plane compared to the N and E orientations with the next highest exposures (Figure 4b). This is due to the supplemental UVB provided by the lamps. However, it is necessary to point out that the value for the N orientation is lower than the E orientation and approximately the same as the S and W orientations. This may have been due to shading of the dosimeters by the leaves of the other plants.

Comparison of the UVB exposures relative to that of the high UV treatment at noon on a horizontal plane shows that the high UV treatment received approximately 40% more UVB on a horizontal plane at the top of the plant canopy. However, for the other azimuthal orientations over the plants, the additional amount of UVB for the supplemental treatment compared to the corresponding orientations for the plants in the control and dummy treatments varied between -39% and 37%. This variation is due predominantly to the shading provided by the other plants. Furthermore, the supplemental UVB changes the natural distribution of UVB and PAP over the plant. For example, at 12:00 EST the horizontal plane orientation received the highest UVB exposures for the supplemental UVB treatment compared to the N orientation receiving the highest UVB for the plants that do not receive supplemental UVB. Similarly, previous UVB measurements over a soybean canopy in the northern hemisphere (Grant, 1999) showed a higher irradiance at noon to a south facing surface inclined at 45° compared to a horizontal surface.

Additionally, the ratio of PAP to UVB irradiances to the plant changes. For example, the ratio of PAP/UV normalised to the same ratio for a horizontal plane at 12:00 EST of the high UV treatment ranges from 1.9 to 3.0 for the different orientations for the high UV treatment at 12:00 EST compared to a range of 0.8 to 3.1 for the control/dummy treatments at the same time. All these variations to the UVB and PAP distribution over
the plant canopy cannot be predicted by exposure measurements in the respective wavebands on a horizontal plane due to the variables that need to be taken into account. Consequently, for the case of the complex topography of plants, the dosimeters described in this paper are small and lightweight and have the advantage of simultaneously measuring the exposures at different positions and orientations around a soybean plant. This can be done in the field without using expensive electronic equipment. Most plant surfaces are not horizontal and consequently, the dosimeters help provide a more realistic assessment of the PAP and UVB exposures to the plant. This paper describes a dosimeter system for measuring both the PAP and UVB exposures and the application of the system in the supplemental UVB irradiation of field grown plants. To the authors’ knowledge, this is the first application of passive dosimeters for measuring UV and PAP on field grown plants. The dosimeters described in this paper provide an important tool in research on supplemental UV effects on plants.

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References


Table 1 – The PAP doses and the relative UVB exposures for the different treatments for the V2 stage at 12:00 EST. These exposures are for the approximate planes of the unifoliate and trifoliate leaves.

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<th>Trifoliate</th>
<th>Unifoliate</th>
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<td>Relative UVB - High UV</td>
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<td>Relative UVB - Low UV</td>
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<td>0.61</td>
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<td>Relative UVB - Control/Dummy</td>
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<td>0.50</td>
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<td>PAP (mol m(^{-2}))</td>
<td>0.83</td>
<td>0.74</td>
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**Figure Captions**

Figure 1 – Plan of the irradiation setup showing the position of each treatment and the lamp holders ( ).

Figure 2 - Field supplemental UV system in place over the trial site.

Figure 3 – The PAP dose on the unifoliate and trifoliate leaves for plants at the V2 stage. The error bars are ±20%. Data are for the three time periods as given in the legend.

Figure 4 – The relative UVB at each orientation at the crop maturity stage for (a) the control/dummy plants and (b) the high UV treatments. The error bars are ±20%. These are all relative to the high UV measurements on a horizontal plane at 12:00 EST.

Figure 5 – The PAP exposure for the N, S, E, W and Top dosimeter orientations for plants at the crop maturity stage. The error bars are ±20%.
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