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Research paper

The Effect of UV Radiation on the Growth and Peroxidase Activity in Soybean (*Glycine max*)

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ABSTRACT

In this study, the soybean variety NRC-7 was cultivated under vinyl filters designed to exclude specific portions of the ultraviolet (UV) spectrum, namely UV-B (<320 nm) and UV-A (380 nm), from natural solar radiation. The results demonstrate that the exclusion of UV-B and UV-A+B significantly enhanced growth parameters, including leaf area, internodal length, and plant height. Notably, the activity of guaiacol peroxidase, an important antioxidant enzyme, was significantly reduced with the removal of both UV-B and UV-A+B. These findings indicate that under normal solar radiation conditions, the guaiacol peroxidase activity is heightened in control plants, potentially as a defense mechanism against oxidative stress induced by the presence of UV-A and UV-B radiation. Specifically, the study recorded an 86% reduction in peroxidase activity with the exclusion of UV-B and a 91% reduction with the exclusion of UV-A+B. This suggests that UV radiation plays a critical role in regulating the antioxidant defense system in soybean plants.

1. Introduction

In recent years, studies on the biological effects of UV-B radiation have garnered considerable attention due to the depletion of the ozone layer caused by pollutants such as chlorofluorocarbons (CFCs), primarily used as refrigerants and aerosol propellants. Research highlights that oxides of nitrogen can lead to spectral changes in the UV-B radiation range (280-320 nm), further exace-rbating the issue (Molina and Rowland, 1974; Kataria et al., 2014). Data collected over the past two decades indicate that nearly 50% of crop plants are negatively affected by elevated levels of solar UV-B radiation. Numerous studies have demonstrated that increased UV-B exposure can have detrimental effects on plant growth and development, photosynthesis, and biomass pro-duction across various cultivated and native plant species (Caldwell, 1971; Teramura, 1983; Tevini and Teramura, 1989; Baroniya et al., 2023; Tevini et al., 2023).

The supplementation of natural solar radiation with UV-B, either through ambient sunlight (Ambler et al., 1975; Semenuik and Stewart, 1979; Kanungo et al., 2023) or artificial light sources (Krizek, 1975; Sisson and Caldwell, 1976; Basiouny et al., 1978; Stefanello et al., 2023), has frequently resulted in suppressed growth of roots, cotyledons, and leaves. The observed effects encompass inhibition of photosynthesis, increased pigmentation, decoloration, phytotoxic damage such as bronzing and glazing of cotyledons, leaf distortion, and ultimately abscission, as well as abnormal shoot curvature across several plant species. Recent attenuation studies further reveal that solar UV-B exposure can decrease biomass production in some species by an estimated 10% to 35% (Mazza et al., 1999; Xiong and Day, 2001; Jorge Alberto Zavalla and Javier Francisco Botto, 2002; Baroniya et al., 2011, 2022; Tevini et al., 2023).

The adverse effects of UV-B on plant growth and physiology are frequently linked to increased oxidative stress (Rao et al., 1995; Baroniya et al., 2013), which is characterized by the generation of reactive species such as singlet oxygen, superoxide radicals, hydrogen peroxide, and hydroxyl radicals. In response to this oxidative stress, plants activate their antioxidant defense systems, which include low molecular weight antioxidants such as ascorbate, glutathione, α-tocopherol, and β-carotenoids (Alscher and Hess, 1993; Saleem et al., 2024), in addition to antioxidant enzymes like superoxide dismutase (SOD), guaiacol peroxidase (POD), ascorbic acid peroxidase (APX), and glutathione reductase (GR) (Bowler et al., 1992; Creissen et al., 1994; Baroniya et al., 2013; Hasanuzzaman et al., 2020).

Historically, early research investigating the effects of UV-B illumination on terrestrial plants was largely conducted indoors using growth chambers or greenhouses. Consequently, there has been a substantial gap in the literature concerning the attenuation effects of UV-A and UV-B components in natural solar radiation environments. This study aims to address this gap by assessing the impact of excluding ambient levels of UV-A and UV-B from solar radiation on the growth parameters and peroxidase activity in soybean (Glycine max, var. NRC-7). This particular soybean variety is increasingly recognized as an important crop both in India and globally (Soy Stats, 2022), noted for its rich nutritional profile, comprising approximately 40% protein, 23% carbohydrates, and 20% cholesterol-free oil.

By investigating these critical interactions between UV radiation and soybean growth, this study aims to improve our understanding of plant responses to changing environmental conditions and foster the development of agricultural practices that enhance crop resilience.

2. Background

2.1 Overview of UV Radiation Types

UV radiation is categorized into three primary types based on wavelength:

UVA (320-400 nm): Constitutes the majority of UV radiation that reaches the Earth's surface. UVA can penetrate deeper into plant tissues and may induce various physiological changes (Zhang et al., 2020).

UVB (280-320 nm): Represents a smaller fraction of UV radiation that has a more pronounced impact on plants. Despite its shorter wavelength, UVB is highly energetic and can cause direct DNA damage and oxidative stress in plants (Tevini et al., 2023).

UVC (<280 nm): Mostly absorbed by the ozone layer and does not generally reach the Earth's surface under natural conditions. Its effects on plant biology are therefore less relevant in the context of environmental exposure (Khalil et. al., 2024).

2.2 Role of Peroxidase in Plants

Peroxidase is an enzyme that plays a crucial role in various physiological processes, particularly in response to environmental stressors. Its functions include:

Stress Responses: Peroxidase is involved in the detoxification of reactive oxygen species (ROS), which are generated during stress conditions such as UV exposure. This enzyme helps mitigate oxidative damage by catalyzing the reduction of hydrogen peroxide (H_2O_2) and other harmful compounds (Sharma et al., 2021).

Cell Wall Metabolism: Peroxidase contributes to the formation and remodeling of plant cell walls, facilitating growth and development (Francoz et al., 2015).

Defense Mechanisms: The peroxidase system is an integral component of plant defense mechanisms, providing protection against pathogens and abiotic stresses, thus enhancing overall plant resilience (Baroniya et al., 2014).

A research by Baroniya et al. (2014) highlighted the effects of UV radiation on various physiological aspects of plant growth, emphasizing the role of antioxidant enzymes, including peroxidase, in alleviating oxidative stress induced by UV exposure. Their findings underscore the importance of further investigating how UV radiation influences growth dynamics and biochemical responses in crops like soybean.

3. Objectives

This study aims to:

- Assess the effects of different UV radiation intensities on soybean growth, specifically measuring parameters such as leaf area, plant height, and internodal length.
- Measure the activity of guaiacol peroxidase in soybean plants in response to varying levels of UV exposure, elucidating its role in oxidative stress management and growth regulation.

4. Materials and Methods

Seeds of soybean (Glycine max) cultivar NRC-7 were obtained from the ICAR-Indian Institute of Soybean Research (IISR), Indore, Madhya Pradesh. The cultivation of these plants was conducted in our botanical garden. The seeds were sown in the field inside an iron cage measuring 120 cm x 90 cm, arranged in three rows lengthwise, and exposed to natural sunlight.

To exclude UV-A and UV-B radiation from sunlight, vinyl filters (Garware Polyester Limited, Mumbai) were used. These filters were wrapped around iron cages measuring 4 feet in length, 3 feet in width, and 3.5 feet in height, where the plants were cultivated. The plants grown under these exclusion filters, which were designated as -UV-B or UV-A+B treatments, were compared to control plants that were grown in cages covered with clear polythene. The clear polythene transmitted all UV radiation present in sunlight.

The emission and absorption characteristics of the filters were measured using a Shimadzu UV-160 spectrophotometer (Fig. 1). The plants were maintained in these filters until they reached full maturity. The levels of UV-A and UV-B attenuation at the center of the treatment cages were quantified using a Radiometer IL 1350 (International Light Inc., USA).

Morphological characters, such as internodal length and plant height, were measured upon completion of the experiment. Leaf area was measured at different growth stages of fully expanded leaves, specifically from the 2nd to the 7th node.

Guaiacol peroxidase activity was assayed at six different stages in the leaves, nodes, and internodes utilizing the method described by George (1953) and Maehly (1955). Protein concentration was estimated by the method of Lowry et al. (1951), using bovine serum albumin (BSA) as a standard.

5. Results and Discussion

5.1 Leaf Area

The exclusion of UV-B and UV-A+B radiation significantly enhanced the leaf area at all growth stages in soybean variety NRC-7. The maximum increase in leaf area was observed at the 6th and 7th nodes, with enhancements of approximately 79% and 81%, respectively, following the exclusion of UV-B. In contrast, the exclusion of UV-A+B resulted in a maximum enhancement of about 52% at the 7th node compared to the control plants (Fig. 2). These findings align with the results reported by Baroniya et al. (2014) and complement previous studies that indicated the negative impact of ambient UV radiation on leaf development (Tevini et al., 2023). Our results suggest that ambient UV-B has an inhibitory effect on leaf expansion, while UV-A appears to play a supportive role in leaf growth (Fig. 2) (Hassan et al., 2020).

The increase in leaf area under UV exclusion could be attributed to reduced stress-induced factors that typically inhibit growth, such as the accumulation of reactive oxygen species (ROS) which are known to impair leaf development (Jansen et al., 1998).

5.2 Plant Height and Internodal Length

Both plant height and internodal length showed significant promotion due to the exclusion of UV-B radiation. The exclusion of UV-A+B further enhanced these parameters, with plant height increasing by 36.7% and internodal length by 21.4% after excluding UV-B. Following the removal of UV-A+B, plant height and internodal length increased by an additional 49.3% and 49.8%, respectively (Fig. 3a, 3b). These results suggest that both UV-A and UV-B have inhibitory effects on stem elongation in soybean variety NRC-7.

Prior studies have documented similar reductions in growth parameters due to ambient UV radiation. For instance, Krizek et al. (1997), Baroniya et al. (2011, 2014, 2023) highlight the detrimental effects of ambient levels of UV-A and UV-B on the growth of crops like cucumber, soybean, and lettuce. The findings are also consistent with those by Ballare et al. (1995) and Mazza et al. (1999), who found ambient UV-B levels inhibit stem elongation and reduce leaf expansion in Datura ferox and barley. In addition, research by Rizzini et al. (2011) indicates that UV radiation can disrupt hormonal balance, affecting growth regulators such as auxins, which are crucial for internodal elongation.

It is worth noting that the variability in responses between different plant species to UV radiation may be due to genetic and phenotypic differences, as observed in the studies performed by Chen et al. (2006), highlighting the importance of further exploring species-specific responses to UV radiation.

5.3 Peroxidase Activity

Peroxidases are heme-containing proteins that catalyze substrate oxidation and hydrogen peroxide reduction, participating in various essential metabolic processes, including cell elongation regulation, lignification, phenolic oxidation, pathogen defense, and stress response. In the control plants, peroxidase activity was notably high; however, it significantly decreased with the exclusion of UV-B alone and also with the exclusion of UV-A+B. At the cotyledonary stage, peroxidase activity was reduced by 85.7%, 50.8%, and 76.6% in the leaves, nodes, and internodes, respectively, upon exclusion of UV-B. When both UV-A and UV-B were excluded, peroxidase activity further decreased by 91%, 46%, and 89.1% in the same respective plant parts (Fig. 4).

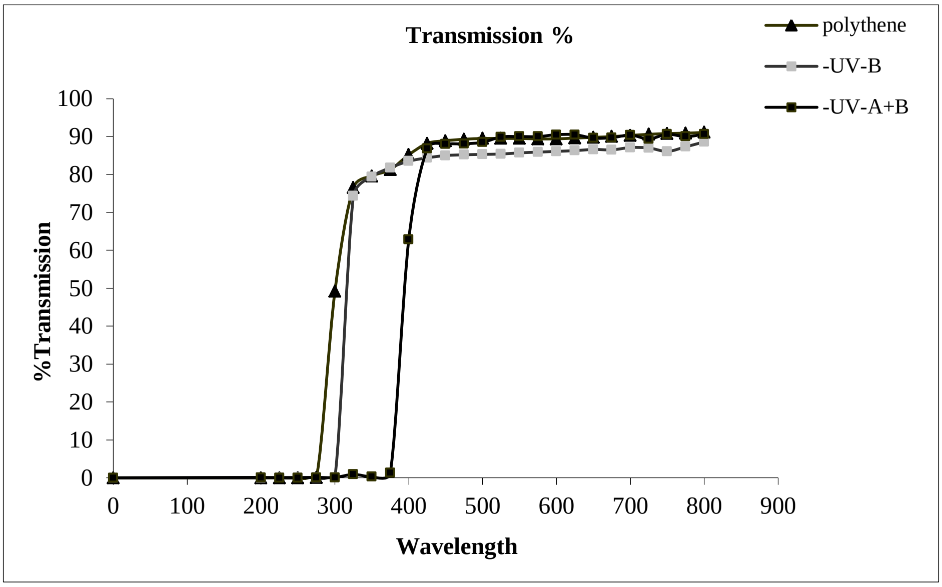

Fig. 1 % Transmission between 200 to 800 nm of the filters used in experiment

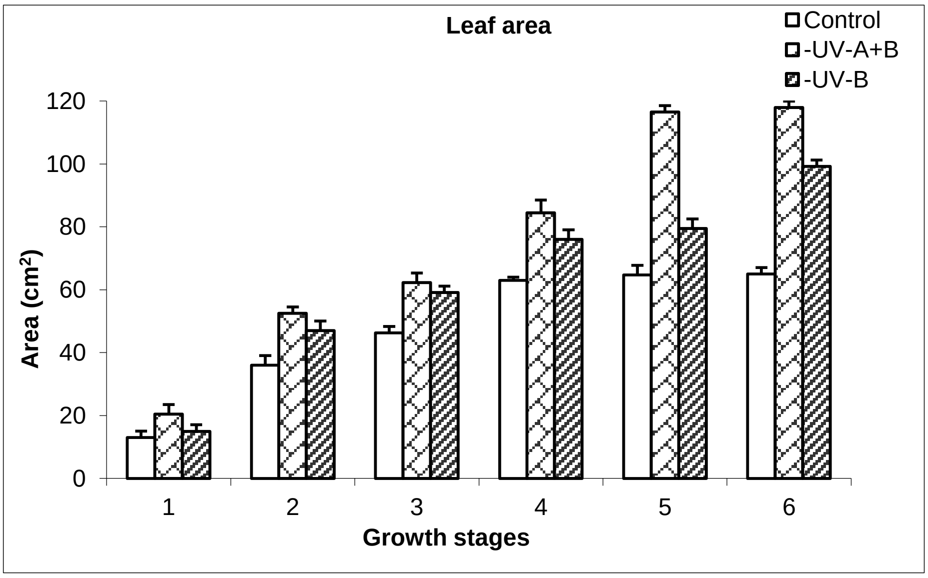

Fig. 2 Effect of exclusion of UV-B and UV-A+B on the area of leaves (1 to 6 numbers indicate the 2nd to 7th node)

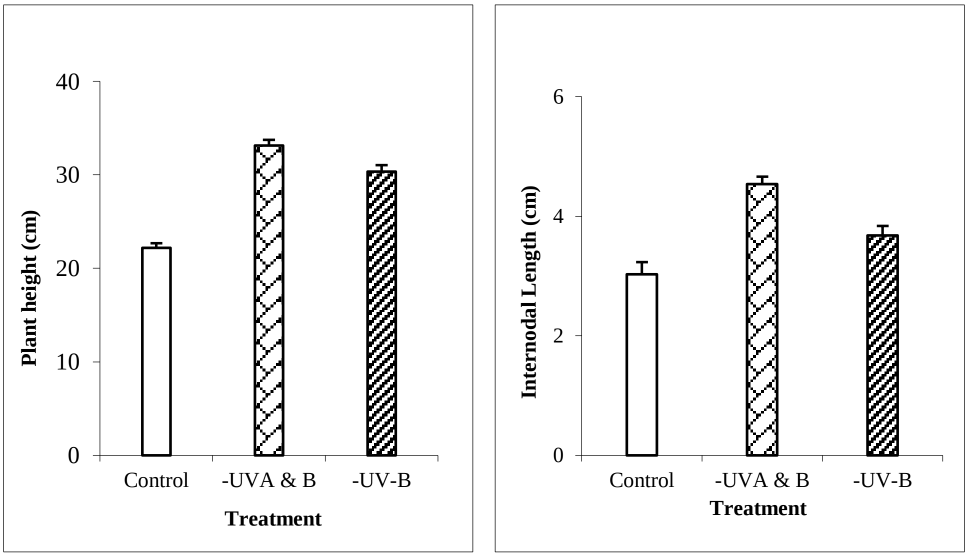

Fig. 3a & 3b Effect of exclusion of UV-B and UV-A+B on plant height & internodal length

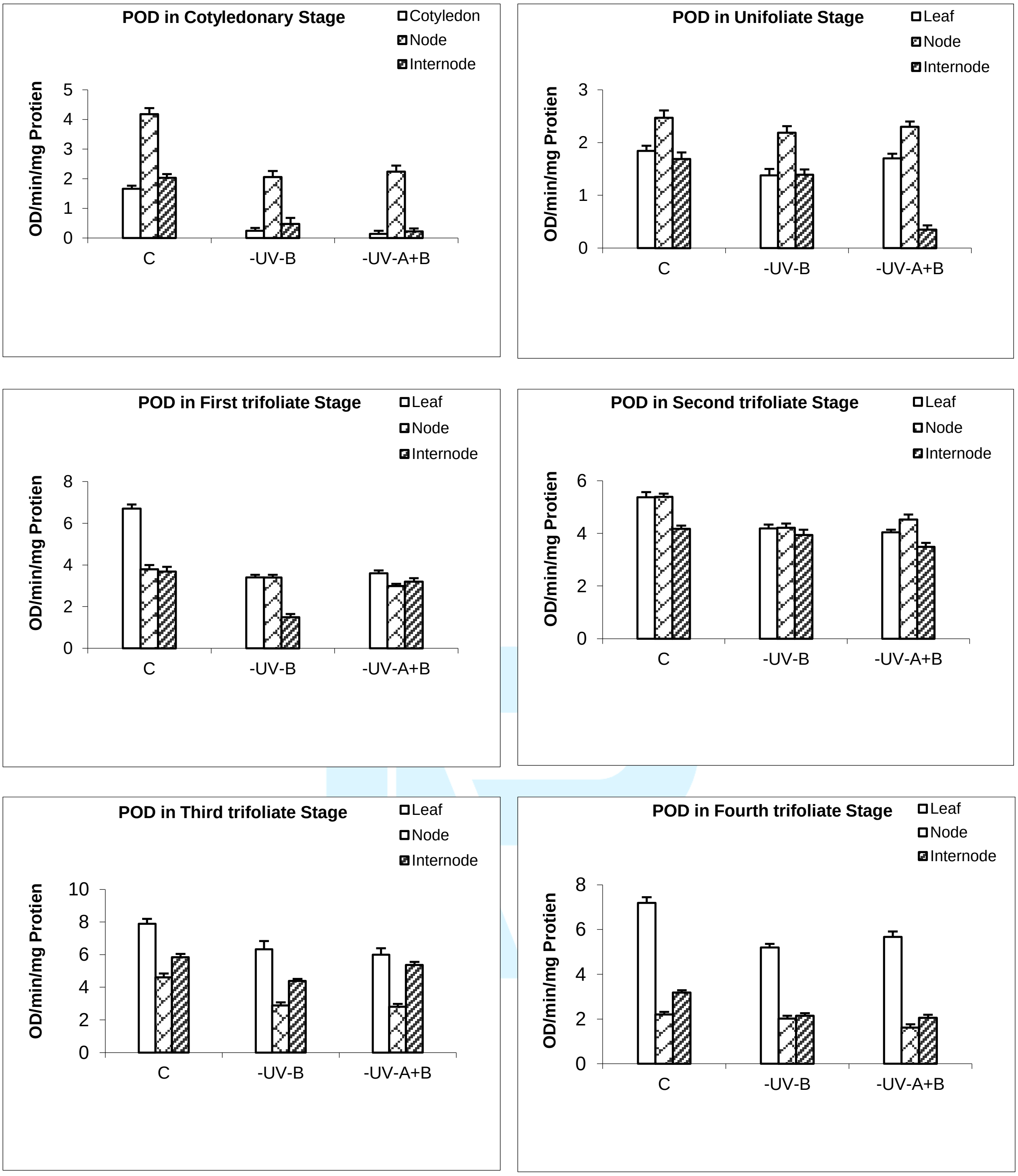

Fig. 4 Effect of exclusion of UV-B and UV-A+B on peroxidase activity

This reduction in peroxidase activity indicates that peroxidases may be activated by the stresses associated with UV exposure. High activity of peroxidase in control plants suggests a protective mechanism against oxidative stress induced by ambient UV radiation (Foyer et al., 1994). Increased peroxidase levels can help in detoxifying ROS, supporting plant resilience (Zhang et al., 2020).

The observed changes in peroxidase activity may result from de novo enzyme synthesis or a suppression of existing enzyme inhibitors. Notably, a UV-B-sensitive protein inhibitor of guaiacol peroxidase has been reported in cucumber (Cucumis sativus) cotyledons (Sunita T. and K. N. Guruprasad, 1998). Research by Birben, et al. (2012) highlighted that environmental stress factors, including UV radiation, can upregulate antioxidant defense mechanisms, further supporting the role of peroxidases during stress responses.

In summary, our study demonstrates that the exclusion of UV-A and UV-B radiation from solar exposure results in significant physiological and biochemical alterations in soybean cultivar NRC-7. Specifically, while UV-B exclusion promotes leaf area, the removal of both UV-A and UV-B enhances plant height and internodal length and reduces peroxidase activity. This research contributes to our understanding of how ambient UV radiation affects plant growth and metabolism, emphasizing the need for further studies to explore the underlying mechanisms of UV responses in different plant species.

6. Conclusion

The exclusion of UV-A and UV-B radiation from solar exposure leads to significant physiological and biochemical alterations in soybean cultivar NRC-7. The findings from this study demonstrate that the exclusion of UV-B radiation results in a marked increase in leaf area, indicating that ambient UV-B has an inhibitory effect on leaf expansion. Furthermore, the simultaneous exclusion of both UV-A and UV-B promotes plant height and internodal length, suggesting that both types of ultraviolet radiation play a crucial role in limiting vertical growth in soybean plants.

Additionally, the reduction in peroxidase activity observed under the exclusion of UV-A and UV-B radiation suggests that these stress responses are tightly linked to the presence of ambient UV radiation. The high levels of peroxidase in control plants indicate a defensive mechanism activated by oxidative stress associated with UV exposure. This research provides insights into the complex interactions between ultraviolet radiation and plant growth, emphasizing the necessity to consider UV light as a significant environmental factor influencing plant morphology and metabolic processes.

Finally, it suggests that antioxidant enzymes provide protection during oxidative injury caused by UV-B and UV-A stress. Exclusion of UV removes this stress indicated by lowering of peroxidase levels after exclusion and alters the metabolism of plants to favour primary metabolism resulting in enhanced plant height, leaf area and internodal length in Soybean var. NRC-7. These conclusions underscore the potential for optimizing growing conditions by managing UV exposure in agricultural practices, particularly for crops like soybean that may be sensitive to UV radiation. Future studies should explore the genetic mechanisms underlying these responses, investigate the effects on yield, and assess the ecological significance of UV radiation management in cultivated plants.

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