

IMPACT OF SUPPLEMENTAL UV-B RADIATION ON CHLOROPHYLL DEVELOPMENT IN SOYBEAN (*GLYCINE MAX* L.).

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Abstract: An attempt has been made to study the impact of supplemental UV-B radiation on chlorophyll content in soybean (*Glycine max* L.). Plants were irradiated daily with supplemental UV-B radiation supplied by sun lamps, 300 watt held in frames suspended 1 meter above the plants in the fields. The total supplemental UV-B irradiance received at the top of the plants beneath the lamps was $24.23 \text{ Jm}^{-2}\text{s}^{-1}$. Control plants were not exposed to supplemental UV-B radiation. Plants of plots T₂, T₃ and T₄ were exposed to supplemental UV-B radiation for 1 hr, 2 hr and 3 hr daily respectively till maturity. Result indicated that chlorophyll content was inhibited at all supplemental UV-B exposures during lab condition. Shorter exposure (1 hr) of supplemental UV-B radiation inhibited the chlorophyll pigment however longer exposures of supplemental UV-B radiation (2 and 3 hr) promoted the chlorophyll pigment during crop growth in field condition. Chlorophyll 'a' seems to be more sensitive to supplemental UV-B radiation. There was a gradual increase in chlorophyll content as crop grows towards maturity.

Keywords: Chlorophyll content, Soybean, supplemental UV-B radiation

INTRODUCTION

Soybean is one of the most important legume crops. It is rich source of the vegetarian protein in the world. It is used in human foods and live stock feeds. It is an important crop in Central India and provides significant returns in many farm enterprises. Teramura and Murali, (1986) in *Glycine max* L. observed that chlorophyll pigments reduced under supplemental UV-B radiation. Jain and Goyal, (1985); Duysen *et al.*, (1985) and Sharma *et al.*, (1988) also observed that chlorophyll contents viz. chl 'a', chl. 'b' and protochlorophyll were greatly reduced due to the UV exposures. Bornman and Vogelmann, (1991) reported the reduced chlorophyll content and chlorophyll fluorescence induction kinetics caused by UV-B radiation in *Brassica* cultivars. Dillenburg *et al.*, (1995) observed that chlorophyll accumulation and the development of photosynthetic capacity were more rapid in plants when exposed to higher UV-B radiations. Deckymn and Impens, (1995) observed that plant growth at high UV-B radiation retained their photosynthetic capacity longer; maximum photosynthesis, chlorophyll and nitrogen content of leaves were higher under high UV-B doses in bean (*Phaseolus vulgaris* L.) plants. No changes in chlorophyll 'a' and 'b' contents were observed by exclusion of solar UV radiation but the content of carotenoids was significantly (34 to 46%) lowered in *Glycine max* L. (Kadur *et al.*, 2007). Adebooye *et al.*, (2008) studied the impact of UV-B radiation on two tropical variants of *Trichosanthes cucumerina* L. They observed that chlorophyll 'a' and total chlorophyll contents of the control plant were significantly higher than those of plants exposed to UV-B for 4 hr and 8 hr. The chlorophyll 'b' contents were not significantly affected by the UV-B exposure. Physiological activities of the plants are regulated by a number of physical and edaphic factors viz. light,

temperature, humidity, rainfall, carbon dioxide concentration, type of the soil etc. Previous studies have shown that UV-B irradiation influenced the crop growth and development in terms of plants height, leaf area, fresh weight, dry weight and mineral composition. These effects are directly or indirectly related to the physiological processes of plants. It can be said that these morphological changes caused by UV-B irradiation are the results of physiological distortion. So it was desired to investigate certain physiological parameters in relation to the UV-B radiation. So in this study, effect of supplemental UV-B radiation on chlorophyll content during the seedling growth and crop growth was taken.

MATERIAL AND METHOD

The experiment was conducted in the Department of Botany, M.M.H. College Ghaziabad. Seeds of *Glycine max* L. were sown in sandy loam soil in rows spaced 0.1m apart in 4 plots of 1 x 1 square meter each. After seedling emergence, plants were irradiated with supplemental UV-B radiation supplied by sun lamps (Philips), 300 watt held in frames suspended 1 meter above the plants in the fields. The total UV-B irradiance received at the top of the plants beneath the lamps was $24.23 \text{ Jm}^{-2}\text{s}^{-1}$. Control (T₁) plants were not exposed to UV-B radiation. Plants of plots T₂, T₃ and T₄ were exposed to supplemental UV-B radiation for 1 hr, 2 hr and 3 hr daily respectively till maturity. The samples for chlorophyll analysis during seedling growth were taken after 7 days grown under laboratory condition. The samples for chlorophyll analysis during crop growth were taken regularly at 15 days intervals after the seedling emergence till maturity of the crop. The chlorophyll content was estimated by the formulae given by Arnon (1949).

RESULT AND DISCUSSION

Chlorophyll development during seedling growth

Table 1 (graph 1) showed that exposures of supplemental UV-B (1 hr, 2 hr and 3 hr) caused a marked decline in different chlorophyll pigments viz. chl. 'a', chl. 'b', total chl. and a/b ratio to these pigments at seedling stage in lab condition. The inhibition of chlorophyll development starts from 1 hr supplemental UV-B exposure and it was observed 27%, 28%, 28% 0% and 12%, 13%, 12% 0% at 2 hr and 9%, 11%, 7%, 2% at 3 hr supplemental UV-B exposures in chl. 'a', chl. 'b', total chl. and a/b ratio

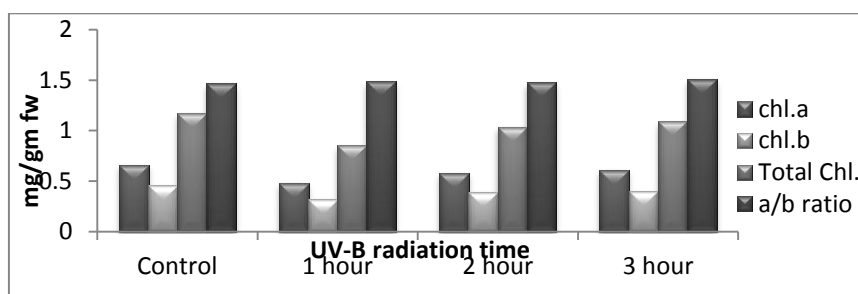
respectively. Marked inhibition in different chlorophyll contents (chl. 'a', chl. 'b' and total chl.) was observed 27%, 28% and 28% at 1 hr supplemental exposure respectively. Promotion in a/b ratio was observed 2% at 3 hr however no change was observed at 1 hr and 2 hr supplemental UV-B radiation when compared with control.

Maximum inhibition was observed at 1 hr supplemental UV-B radiation in chl. 'a', chl. 'b' and total chl. Maximum promotion in chl. a/b ratio was observed at 3 hr however no inhibition was observed at 1 hr and 2 hr supplemental UV-B exposures due to more inhibition in chl. 'b'.

Table 1: Effect of supplemental UV-B radiation on the chlorophyll development (mg/gm fw) during seedling growth in soybean (*Glycine max* L.).

Daily UV-B Irradiation	CHLOROPHYLL CONTENT (mg/gm. fw.)			
	Chlorophyll 'a'	Chlorophyll 'b'	Total chlorophyll	a/b ratio
Control	0.659 ± 0.013	0.449 ± 0.012	1.168 ± 0.025	1.467 ± 0.017
1 hour	*0.482 ± 0.014	*0.324 ± 0.014	*0.849 ± 0.030	1.487 ± 0.016
2 hour	*0.580 ± 0.023	*0.393 ± 0.023	*1.035 ± 0.013	1.475 ± 0.039
3 hour	*0.610 ± 0.024	*0.404 ± 0.019	1.087 ± 0.038	*1.509 ± 0.024

± = standard deviation, *= significant at 5% level.



Graph 1: Effect of supplemental UV-B radiation on chlorophyll development during seedling growth in soybean (*Glycine max* L.).

Results indicated that supplemental UV-B radiation was inhibitory to chlorophyll development at seedling stage in soybean (*Glycine max* L.) crop. Similar findings were also reported by Strid *et al.*, (1990) in *Pisum sativum*. Significant reduction in different chlorophyll pigments by supplemental UV-B exposures were also investigated by Jain and Goyal, (1985) in *Cucumis utillissimus*; Duysen *et al.*, (1985) in *Glycine max* L.; Sharma *et al.*, (1988) in *Pisum sativum* and Goyal *et al.*, (1991) *Linum usitatisimum* but Deckymn and Impens, (1995)

reported enhancement in chlorophyll content in *Phaseolus vulgaris* L.

Chlorophyll development during crop growth

Table 2 (graph 2-5) showed that shorter exposure of supplemental UV-B (1 hr) inhibited chlorophyll development, however longer exposures of 2 hr and 3 hr supplemental UV-B radiation promoted the various chlorophyll pigments (viz. chl. 'a', chl. 'b', total chl. and chl. a/b ratio) respectively.

Table 2: Effect of supplemental UV-B radiation on chlorophyll content (mg/gm fw) in field grown soybean (*Glycine max* L.) crop.

Daily UV-B Irradiance		CROP AGE IN DAYS					
Chl. Pigments	15	30	45	60	75	90	
Control							
Chl 'a'	0.417 ± 0.008	0.375 ± 0.006	0.394 ± 0.006	0.395 ± 0.006	0.291 ± 0.020	0.114 ± 0.013	
Chl 'b'	0.479 ± 0.016	0.505 ± 0.008	0.477 ± 0.012	0.484 ± 0.015	0.216 ± 0.007	0.182 ± 0.013	
Total Chl	1.010 ± 0.020	1.015 ± 0.014	0.906 ± 0.008	0.981 ± 0.029	0.530 ± 0.008	0.304 ± 0.009	
a/b ratio	0.870 ± 0.018	0.742 ± 0.011	0.825 ± 0.014	0.816 ± 0.015	1.347 ± 0.010	0.626 ± 0.008	
1 hour							
Chl 'a'	*0.248 ± 0.004	*0.176 ± 0.009	*0.285 ± 0.007	*0.323 ± 0.005	0.286 ± 0.006	*0.389 ± 0.019	
Chl 'b'	0.453 ± 0.021	*0.390 ± 0.011	*0.361 ± 0.010	*0.303 ± 0.009	*0.265 ± 0.010	*0.468 ± 0.014	

Total chl	*0.713 ± 0.016	*0.568 ± 0.030	*0.692 ± 0.009	*0.612 ± 0.014	*0.697 ± 0.013	*1.005 ± 0.011
a/b ratio	*0.547 ± 0.005	*0.451 ± 0.007	0.789 ± 0.015	*1.066 ± 0.012	*1.079 ± 0.012	*0.831 ± 0.008
2 hour						
Chl 'a'	*0.459 ± 0.018	*0.479 ± 0.020	*0.434 ± 0.014	*0.429 ± 0.010	*0.394 ± 0.015	*0.411 ± 0.012
Chl 'b'	0.494 ± 0.019	*0.649 ± 0.017	*0.521 ± 0.007	0.501 ± 0.015	*0.511 ± 0.024	*0.315 ± 0.015
Total chl	1.032 ± 0.007	*1.152 ± 0.010	*1.045 ± 0.040	*0.937 ± 0.011	*0.983 ± 0.013	*0.780 ± 0.012
a/b ratio	0.929 ± 0.026	0.738 ± 0.011	*0.833 ± 0.011	0.856 ± 0.01	*0.771 ± 0.009	*1.304 ± 0.016
3 hour						
Chl 'a'	*0.457 ± 0.006	*0.346 ± 0.009	*0.357 ± 0.009	*0.602 ± 0.008	*0.454 ± 0.010	*0.348 ± 0.007
Chl 'b'	*0.392 ± 0.007	*0.573 ± 0.015	0.489 ± 0.016	*0.754 ± 0.028	*0.538 ± 0.012	*0.353 ± 0.008
Total chl	*0.945 ± 0.006	*1.035 ± 0.012	*1.011 ± 0.020	*1.386 ± 0.015	*1.042 ± 0.016	*0.784 ± 0.014
a/b ratio	*1.165 ± 0.009	*0.603 ± 0.007	*0.730 ± 0.011	*0.798 ± 0.009	*0.843 ± 0.014	*0.985 ± 0.010

Chl = Chlorophyll, ± = standard deviation, * = significant at 5% level.

At 15 day stage, chlorophyll contents inhibited 41% however promoted 6% and 30% at 1 hr supplemental UV-B radiation in terms of chl. 'a', chl. 'b', total chl. respectively. However, these pigments were promoted 10%, 3% and 2% at 2 hr; 9%, 18% and 7% at 3 hr supplemental UV-B exposures in chl. 'a', chl. 'b' and total chl. respectively. Chl. a/b ratio was inhibited 37% at 1 hr however promoted 7% at 2 hr and 34% at 3 hr supplemental UV-B radiation.

At 30 day stage, inhibition in chl. 'a' was observed 54%, 28% and 8% at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively. Inhibition in chl. 'b' and total chl. was observed 23% and 44% at 1 hr however promotion was observed 28%, 13% and 13%, 2% at 2 hr and 3 hr supplemental UV-B radiation respectively. Chl. a/b ratio was inhibited 39%, 1% and 19% at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively.

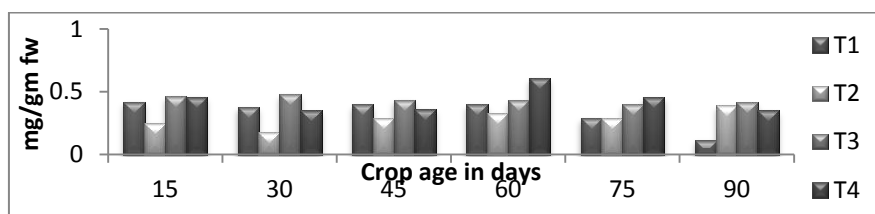
At 45 day stage, inhibition was observed 28%, 25% and 24% at 1 hr however promotion was observed 10%, 9% and 15% at 2 hr supplemental UV-B radiation in terms of chl. 'a', chl. 'b' and total chl. respectively. At 3 hr supplemental UV-B radiation, chl. 'b' and total chl. was promoted 2% and 11% however chl. 'a' was inhibited 11%. Promotion in chl. a/b ratio was observed 1% at 2 hr supplemental UV-B radiation however inhibited 5% at 1 hr and 12% at 3 hr supplemental UV-B radiation.

At 60 day stage, chl. 'a' was inhibited 18% at 1 hr however promoted 8% at 2 hr and 52% at 3 hr supplemental UV-B radiation. Chl. 'b' was also inhibited 38% at 1 hr however promoted 3% at 2 hr and 56% and 3 hr supplemental UV-B exposure.

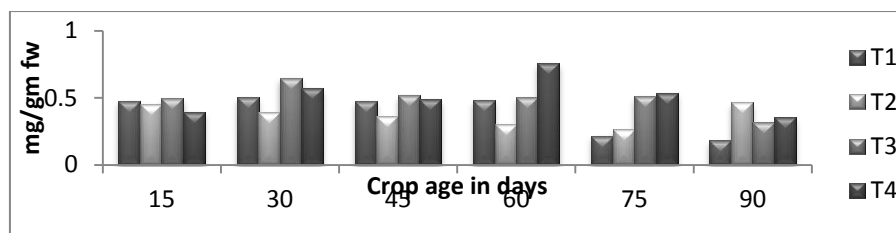
Total chl. was inhibited 38% at 1 hr and 5% at 2 hr however promoted 41% at 3 hr supplemental UV-B radiation. Promotion in chl. a/b ratios was observed 30% at 1 hr and 5% at 2 hr however inhibition was observed 2% at 3 hr supplemental UV-B radiation.

At 75 day stage, chl. 'a' was inhibited 2% at 1 hr however promoted 35% at 2 hr and 56% at 3 hr supplemental UV-B radiation. Chl. 'b' showed promotory effect over control in all treatments and it was promoted 22%, 136% and 149% at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively. Total chl. was also promoted 31%, 85% and 96% at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively. Inhibition in chl. a/b ratio was observed 20%, 43% and 33% at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively.

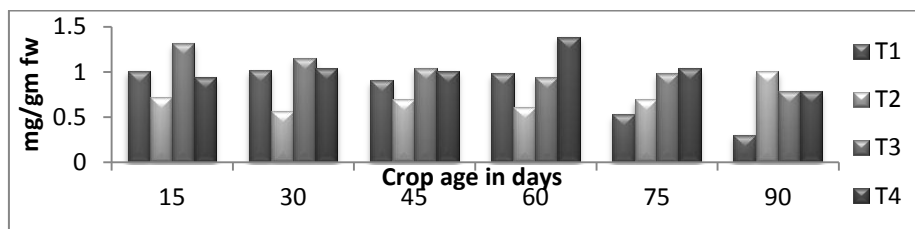
At 90 day stage, promotion was observed at all supplemental UV-B radiations and it was promoted 241%, 260% and 205% in chl. 'a'; 153%, 73% and 94% in chl. 'b' and 230%, 156% and 158% in total chl. at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively. Chl. a/b ratio was reached at its maximum promotion and it was observed 32%, 108% and 57% at 1 hr, 2 hr and 3 hr supplemental UV-B radiation respectively when compared with control. Thus above results indicated that supplemental UV-B radiation was promotory to chlorophyll development especially in winter season at 1 hr, 2 hr and 3 hr UV-B irradiance. Chl. 'a' seems to be more sensitive to supplemental UV-B radiation. There was a gradual increase in chlorophyll content as crop grows towards the maturity.



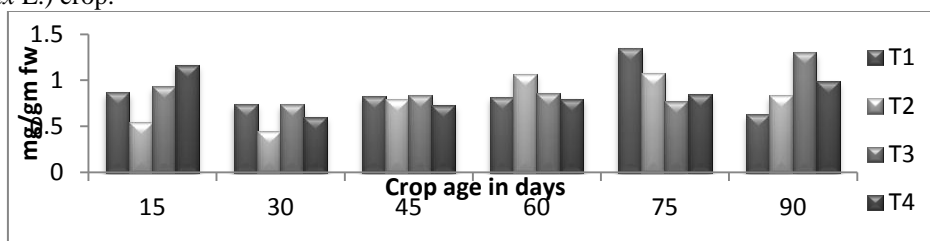
Graph 2: Effect of supplemental UV-B radiation on chl. 'a' development in field grown soybean (*Glycine max* L.) crop.



Graph 3: Effect of supplemental UV-B radiation on chl. 'b' development in field grown soybean (*Glycine max* L.) crop.



Graph 4: Effect of supplemental UV-B radiation on total chlorophyll development in field grown soybean (*Glycine max* L.) crop.



Graph 5: Effect of supplemental UV-B radiation on chl. a/b ratio development in field grown soybean (*Glycine max* L.) crop.

Chl. 'a', chl. 'b' and total chl. was maximum promoted at longer exposures however inhibited at shorter exposure of supplemental UV-B radiation. Chl. 'a' was maximum promoted at 2 hr at 90 day stage however maximum inhibited at 1 hr supplemental at 30 day stage of crop growth. Chl. 'b' was promoted maximum at 1 hr at 90 day stage however inhibited maximum at 1 hr at 60 day stage of crop growth. According to Kitajima and Hogan, (2003) the increase of chl. 'b' would be associated with certain level of acclimatization of UV-B radiation since the pigment is closely related to the pigment protein complex (LCH II) which mostly has chl. 'b' in its structure and is present in the photo system IInd. While Kadur *et al.*, (2007) observed no change in chl. 'a' and chl. 'b' content in *Glycine max* L. under exclusion of solar UV radiation. However, Musil *et al.*, (2002) observed that concentration of chl. 'a' and chl. 'b' has been significantly decreased in *Barleria obtusa* and *Vigna unguiculata* plants when exposed to supplemental UV-B radiation. Total chl. was also promoted at longer exposure of supplemental UV-B radiation however inhibited at shorter exposure of supplemental UV-B. Maximum promotion in total chl. was observed at 1 hr at 90 day stage however maximum inhibition was observed at 1 hr supplemental UV-B radiation at 30 day stage of the crop growth. No significant difference have been detected in total chl. concentration by Antonelli *et al.*, (1997) in bean plants and Santos *et al.*, (2004) in *Solanum tuberosum* when these plants were exposed

to supplemental UV-B radiation while reduction in total chl. concentration was observed by Jenkins *et al.*, (1997) in *Arabidopsis* and Singh *et al.*, (2011) in *Dolichos lab lab*.

Fluctuation in a/b ratio was observed at all supplemental UV-B radiation. Maximum promotion in chl. a/b ratio was observed at maturity of crop at 2 hr supplemental UV-B radiation however maximum inhibition was observed at 2 hr supplemental UV-B radiation at 75 day stage of crop growth. The same findings were also observed by Vu *et al.*, (1983) in *Pisum sativum* L. but Smith *et al.*, (2001) in *Phaseolus vulgaris* and Singh *et al.*, (2011) in *Dolichos lab lab* observed that a/b ratio decreased with increasing UV-B irradiances.

Our findings indicated that longer exposure (i.e. 2 hr and 3 hr) of supplemental UV-B radiation increased the chlorophyll content however shorter exposure of supplemental UV-B inhibited the chlorophyll content in soybean crop (*Glycine max* L.) when compared with control. In the present study, chlorophyll content enhanced inconsistently to the application of supplemental UV-B radiation. These results were in confirmation with the results of Caldwell and Britz, (2006) they observed that increase in chlorophylls can also explain how the indirect effect of protection against damage provoked by UV-B radiation. Synthesis of phenolic compounds is generated under high intensities which act as a barrier reducing UV-B radiation penetration.

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