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Research Article

ASSESSMENT OF NODULATION IN THREE VARIETIES OF BLACK GRAM UNDER ELEVATED UV-B RADIATION

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ABSTRACT
Among symbiotic nitrogen fixing systems, nodulated legumes have been used in cropping systems for centuries. But change in environmental conditions like elevated UV-B can hinder the process of symbiotic nitrogen fixation. In this connection the impact of UV-B rays on nodulation in black gram (*Vigna mungo* (L.) Hepper), the nitrogen fixing grain legume is evaluated. The fully developed root systems were harvested on 30 and 45 DAS (days after seed germination) from three varieties of black gram *viz.* VAMBAN-3, NIRMAL-7 and T-9 after *in situ* supplementary UV-B radiation (2 hours daily @ 12.2 kJ m^{-2} d^{-1}; ambient = 10 kJ m^{-2} d^{-1}). Nodule number fell below control after UV-B exposure at all stages of plant growth. NIRMAL-7 recorded heavy reduction in nodule number on 30 DAS (63.63 %) and 45 DAS (80 %). VAMBAN-3 showed only little inhibition (28.20 %) in nodulation on 30 DAS, but the suppression was severe on 45 DAS (82.35 %). T-9 which showed only 4.65 % reduction in nodule number at early stage, recorded 41.17 % reduction on 45 DAS. Nodules weighed less than their controls in NIRMAL-7 (14.37 to 64.86 %), VAMBAN-3 (26.67 to 41.66 %) and T-9 (6.29 to 9.36 %).

Keywords: *Ultraviolet-B, Black Gram, Three Varieties, Nodulation*

INTRODUCTION
The economic and environmental costs of the heavy use of chemical nitrogen fertilizers in agriculture are a global concern. Sustainability considerations mandate that alternatives to nitrogen fertilizers must be urgently sought. Biological nitrogen fixation, a microbiological process which converts atmospheric nitrogen into a plant-usable form, offers this alternative Bohlool *et al.*, (1992). Adverse environmental conditions created by stresses like salt, drought, acidity, alkalinity, nutrient deficiency, fertilizers, heavy metals, and pesticides suppress the growth and symbiotic characteristics of most *Rhizobia*; however, several strains, distributed among various species of *Rhizobia,* are tolerant to stress effects (Zahran 1999). The present work is an effort to screen the effect of yet another abiotic stress *viz.*, ultraviolet-B rays on nodulation in three varieties of black gram.

MATERIALS AND METHODS
Black gram (*Vigna mungo* (L.) Hepper) the nitrogen fixing grain legume was chosen for the study. Viable seeds of the three varieties of black gram *viz.* VAMBAN-3, NIRMAL-7 and T-9 were procured from Saravana Farms, Villupuram, Tamil Nadu and from local farmers in Pondicherry. The seeds were selected for uniform colour, size and weight and used in the experiments. The crops were grown in pot culture in the naturally lit greenhouse (day temperature maximum 38 ± 2 ºC, night temperature minimum 18 ± 2 ºC, relative humidity 60 ± 5 %, maximum irradiance (PAR) 1400 µmol m^{-2} s^{-1}, photoperiod 12 to 14 h). Supplementary UV-B radiation was provided in UV garden by three UV-B lamps (*Philips TL20W/12 Sunlamps*, The Netherlands), which were suspended horizontally and wrapped with cellulose diacetate filters (0.076 mm) to filter UV-C radiation (< 280 nm). UV-B exposure was given for 2 h daily from 10:00 to 11:00 and 15:00 to 16:00 starting from the 5th day after sowing. Plants received a biologically effective UV-B dose ($UV\text{-}B_{BE}$) of 12.2 kJ m^{-2} d^{-1} equivalent to a simulated 20 % ozone depletion at Pondicherry (12º2'N, India).
The control plants, grown under natural solar radiation, received $UV\text{-}B_{BE}$ 10 kJ m^{-2} d^{-1}. Ten plants from each treatment were carefully uprooted from the soil at 30 and 45 DAS when the nodulation was at its peak and the number and fresh weight of nodules were recorded after removing the soil particles by

washing them repeatedly and blotting to dryness. Whole plants and plant parts were photographed in daylight using a Sony digital camera fitted with appropriate close-up accessories. At least ten replicates were maintained for all treatments and control. The experiments were repeated to confirm the trends. The result of single linkage clustering (Maskay, 1998) was displayed graphically in the form of a diagram called dendrogram (Everstt, 1985).

The term dendrogram is used in numerical taxonomy for any graphical drawing giving a tree-like description of a taxonomic system. The similarity indices between the three varieties of black gram under study were calculated using the formula given by Bhat and Kudesia (2011).

$$\text{Similarity index} = \frac{\text{Total number of similar characters}}{\text{Total number of characters studied}} \times 100$$

Based on the similarity indices between the three varieties of black gram, dendrogram was draw to derive the interrelationship between them and presented in tables and plates.

RESULT AND DISCUSSION

Nodule number in all the UV-B irradiated varieties of black gram were always less than the control on 30 and 45 DAS. A reduction of 63.63 % and 80 % was recorded on 30 and 45 DAS respectively in NIRMAL-7. Even though VAMBAN-3 recorded only 28.20 % reduction in nodulation on 30 DAS, the suppression was severe on 45 DAS as it reached 82.35 % reduction. T-9 which showed a sign of recovery with only 4.65 % less than control crops, on prolonged UV-B exposure recorded 41.17 % reduction on 45 DAS.

The inhibitory tendency of UV-B continued in fresh weight of nodules also. The nodules in NIRMAL-7 variety of black gram weighed less by 14.37 to 64.86 % compared to control on 30 and 45 DAS followed by VAMBAN-3 (26.67 to 41.66 %). T-9 was least affected by UV-B as the nodules weighed better than other two varieties showing only 6.29 to 9.36 % reduction (Table 1, Plate 1).

Reduction in nodulation after UV-B exposure was also reported in green gram (Rajendiran and Ramanujam, 2006), *Sesbania grandiflora* (L.) Pers. (Sudaroli and Rajendiran, 2013a), *Vigna unguiculata* (L.) Walp. c.v. BCP-25 (Sudaroli and Rajendiran, 2013b), ten varieties of cowpea (Kokilavani and Rajendiran, 2014), in *Vigna unguiculata* (L.) Walp. cv. COVU-1 (Sudaroli and Rajendiran, 2014a), *Vigna mungo* (L.) var. T-9 (Sudaroli and Rajendiran, 2014b), *Vigna unguiculata* (L.) Walp. c.v. CO-1 (Sudaroli and Rajendiran, 2014c), *Lablab purpureus* L. var. Goldy (Arulmozhi and Rajendiran, 2014a), hyacinth bean (Arulmozhi and Rajendiran, 2014b), *Vigna unguiculata* (L.) Walp. cv. COFC-8 (Arulmozhi and Rajendiran, 2014c), *Cyamopsis tetragonoloba* (L.) Taub. var. PNB (Vijayalakshmi and Rajendiran, 2014a), *Phaseolus vulgaris* L. cv. Prevail (Vijayalakshmi and Rajendiran, 2014b) and in *Vigna unguiculata* (L.) Walp. cv. CW-122 (Vijayalakshmi and Rajendiran, 2014c). According to Rajendiran (2001) UV-B stressed shoot system decreased allocation of food to root system which reacted quickly with reduced root system, thereby providing lesser surface area for *Rhizobium* inoculation and root nodule formation.

Table 1: Changes in nodulation of three varieties of 30 and 45 DAS *Vigna mungo* (L.) Hepper under control and supplementary UV-B exposed conditions – *In situ*

Varieties	Treatment	Nodule number plant $^{-1}$		Fresh weight of nodule plant $^{-1}$ (g)	
		30 DAS	45 DAS	30 DAS	45 DAS
VAMBAN-3	Control	39	17	0.024	0.656
	UV-B	28	3	0.014	0.481
NIRMAL-7	Control	22	25	0.037	0.668
	UV-B	8	4	0.013	0.572
T-9	Control	43	17	0.641	0.620
	UV-B	41	7	0.581	0.581

Figure 1: VAMBAN-3

Figure 2: NIRMAL-7

Figure 3: T-9

Plate 1: Comparative gross morphology of root systems showing nodulation in three varieties of *Vigna mungo* (L.) Hepper on 45 DAS. (1: Control, 2: UV-B)

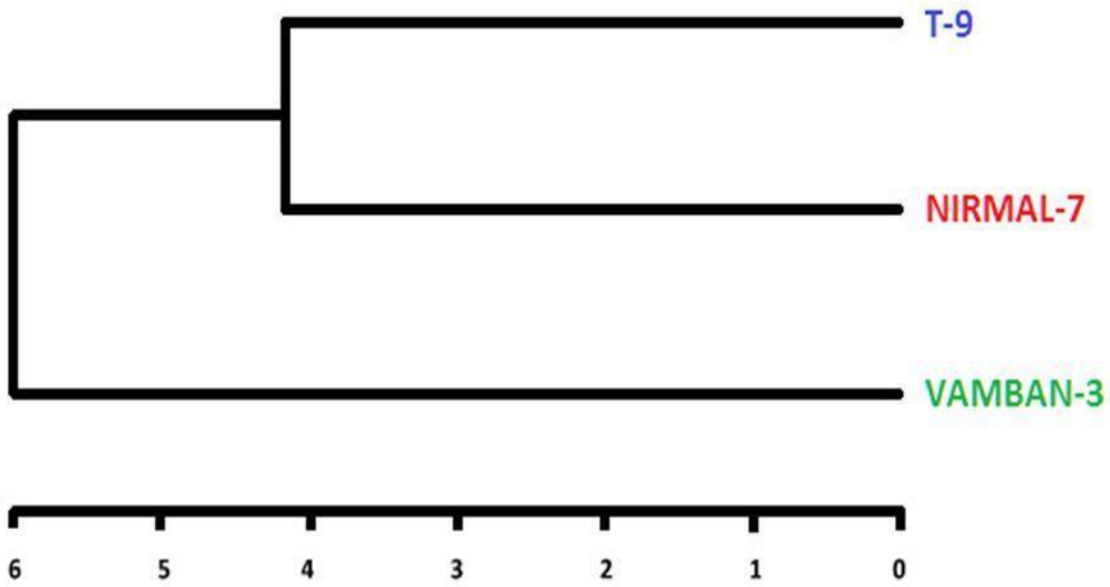

Plate 2: Dendrogram showing the interrelationship between the three varieties of *Vigna mungo* (L.) Hepper in nodulation under control and supplementary UV-B - *In situ*

Table 2: The similarity indices in nodulation of three varieties of *Vigna mungo* (L.) Hepper under supplementary UV-B exposed conditions – *In situ*

Varieties	VAMBAN-3	NIRMAL-7	T-9
VAMBAN-3	100%	62.5%	50%
NIRMAL-7	62.5%	100%	37.5%
T-9	50%	37.5%	100%

The number and fresh weight of nodules in three varieties of black gram exhibited differences on 30 and 60 DAS due to *in situ* supplemenaty UV-B radiation. NIRMAL-7 and T-9 varieties formed one group with 37.5 % similarity index between them (Table 2; Plate 2). VAMBAN-3 remaining alone in the cluster had 62.5 % and 50 % similarity indices with NIRMAL-7 and T-9 respectively.

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REFERENCES
Arulmozhi D and Rajendiran K (2014a). Effect of supplementary ultraviolet-B radiation on nodulation and nitrogen metabolism in *Lablab purpureus* L. var. Goldy. *International Journal of Advanced Biological Research* **4**(3) 343-346.
Arulmozhi D and Rajendiran K (2014b). Supplementary ultraviolet-B induced reduction in nodulation and nitrogen metabolism in hyacinth bean. *International Journal of Geology, Earth and Environmental Sciences* **4**(2) 73-77.
Arulmozhi D and Rajendiran K (2014c). Effect of elevated ultraviolet-B irradiation on the nodulation and nitrogen metabolism in *Vigna unguiculata* (L.) Walp. cv. COFC-8. *International Journal of Food, Agriculture and Veterinary Sciences* **4**(2) 184-188.
Bhat TM and Kudesia R (2011). Evaluation of Genetic Diversity in Five Different Species of Family Solanaceae using Cytological Characters and Protein Profiling. *Genetic Engineering and Biotechnology Journal* **2011** 1-8.

Bohlool BB, Ladha JK, Garrity DP and George T (1992). Biological nitrogen fixation for sustainable agriculture: A perspective. *Plant and Soil* **141** 1-11.
Everstt B (1985). *Clustering Analysis* (John Wiley and Sons, New York).
Kokilavani V and Rajendiran K (2014). Ultraviolet-B induced reduction in nodulation in ten varieties of cowpea. *International Journal of Innovative Research and Review* **2**(4) 77-82.
Maskay N (1998). Single linkage clustering. In: *Encyclopedia of Biostatistics*, edited by Armintage P and Cotton T (Wiley, New York) **5** 4121-4122.
Rajendiran K (2001). Amelioration of Ultraviolet-B radiation impacts in green gram by Triadimefon. PhD. Thesis, Pondicherry University.
Rajendiran K and Ramanujam MP (2006). Interactive effects of UV-B irradiation and triadimefon on nodulation and nitrogen metabolism in *Vigna radiata* plants. *Biologia Plantarum* **50**(4) 709-712.
Sudaroli Sudha J and Rajendiran K (2013a). Effect of elevated UV-B irradiation on the nodulation and nitrogen metabolism in *Sesbania grandiflora* (L.) Pers. *International Journal of Science and Nature* **4**(4) 664-667.
Sudaroli Sudha J and Rajendiran K (2013b). Effect of elevated UV-B irradiation on the nodulation and nitrogen metabolism in *Vigna unguiculata* (L.) Walp. c.v. BCP-25. *International Journal of Food, Agriculture and Veterinary Sciences* **3**(3) 77-81.
Sudaroli Sudha J and Rajendiran K (2014a). Impact of ultraviolet-B radiation on nodulation and nitrogen metabolism in *Vigna unguiculata* (L.) Walp. cv. COVU-1. *International Journal of Geology, Earth and Environmental Sciences* **4**(2) 224-230.
Sudaroli Sudha J and Rajendiran K (2014b). Ultraviolet-B induced reduction in nodulation and nitrogen metabolism in *Vigna mungo* (L.) Hepper var. T-9. *Global Journal of Bioscience and Biotechnology* **3**(4) 370-373.
Sudaroli Sudha J and Rajendiran K (2014c). Ultraviolet-B induced reduction in nodulation and nitrogen metabolism in *Vigna unguiculata* (L.) Walp. cv. CO-1. *Global Journal of Bioscience and Biotechnology* **4**(3) 10-14.
Vijayalakshmi R and Rajendiran K (2014a). Impact of ultraviolet-B radiation on nodulation and nitrogen metabolism in *Cyamopsis tetragonoloba* (L.) Taub. var. PNB. *International Journal of Geology, Earth and Environmental Sciences* **4**(2) 78-82.
Vijayalakshmi R and Rajendiran K (2014b). Impact of ultraviolet-B radiation on nodulation and nitrogen metabolism in *Phaseolus vulgaris* L. cv. Prevail. *International Journal of Advanced Biological Research* **4**(3) 339-342.
Vijayalakshmi R and Rajendiran K (2014c). Effect of elevated ultraviolet-B irradiation on the nodulation and nitrogen metabolism in *Vigna unguiculata* (L.) Walp. cv. CW-122. *International Journal of Food, Agriculture and Veterinary Sciences* **4**(2) 189-193.
Zahran HH *(*1999). *Rhizobium*-legume symbiosis and nitrogen fixation under severe conditions and in an arid climate. *Microbiology and Molecular Biology Reviews* **63**(4) 968-989.