



Research Article

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EFFECT OF ABIOTIC ELICITORS ON BIOCHEMICAL STATUS OF TWO CULTIVARS OF *VIGNA RADIATA* (L.) WILCZEK

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ABSTRACT

The present investigation has been conducted to analyse the effect of different concentrations of abiotic elicitors, Salicylic acid (0.01mM, 0.025mM and 0.05mM) and NaCl (50mM, 100mM and 150mM) individually and their combination on biochemical status of seeds of two cultivars, Pusa Vishal (PV) and SML 668 of *Vigna radiata*. All the biochemical analysis was conducted after 3rd and 6th day of imbibition. Highest amount of protein was observed in 100mM NaCl+0.025mM salicylic acid treated seeds of PV after 6 day as compared to all treatments as well as control set of both cultivars (PV and SML 668). Maximum proline content was recorded in 150mM NaCl treated seeds of PV after 3rd day. Highest phenolic content was recorded in 150mM NaCl+0.025mM salicylic acid-treated seeds of SML 668 cultivar after 3rd day. It was noticed that control seeds after 3 days showed higher phenolic content as compared to the 6th day. In general, treatment of any elicitor individually or in combination, led to increase in phenolic content after 3 days but to decrease after 6 days. Similar results were obtained for proteins too. Any concentration of NaCl resulted in increase in proline content in both of the cultivars. All the results indicate that treatment with NaCl on PV and SML 668 seeds always led to increase in the concentration of proteins.

Keywords: NaCl, Phenolics, Proline, Protein, Salicylic acid (SA), *Vigna radiata*

INTRODUCTION

Vigna radiata, commonly known as mung bean, is an important pulse crop plant, which is native to India. It is grown in arid and semi arid regions of the world as it has heat tolerating capacity. It has short growing season, high protein content and nitrogen fixing capacity that helps in improving soil fertility. The seeds of *Vigna radiata* contain 22%-28% proteins, 60%- 65% carbohydrates, 1%- 1.5% fat, 3.5% - 4.5% fibres, 4.5%-5.5% ash¹. Vitamins like niacin, thymine, carotene, riboflavin, folic acid, ascorbic acid are also present in mung bean. A number of essential amino acids like arginine, histidine, tryptophan, phenylalanine, cysteine, methionine, leucine is also present in *V. radiata*. It contains flavonoids, phenolic acids and organic acids as well². These contribute to the nutritional aspect of *Vigna radiata*.

Elicitors are compounds that induce a certain response in a living cell. These result in some specific kind of biosynthetic response. Elicitors that are of non-biological origin are referred to as abiotic elicitors.

Salt stress also known as salinity stress results in adverse growth and germination rates of plants. Salinity is a burning problem in many parts of the world particularly in arid and semi arid regions.

Salicylic acid is a naturally occurring plant hormone that has adverse effects on various biotic and abiotic stress tolerances among plants. It regulates many important physiological functions of plants like ion uptake, stomatal closure, inhibition of ethylene biosynthesis, stress tolerance, etc.³. It can have both inhibitory as well as stimulatory functions in plants.

MATERIALS AND METHODS

The present investigation was carried out to find out the biochemical changes occurring due to abiotic elicitors. Abiotic elicitors that had been used were NaCl and Salicylic acid. Seeds of both the cv. of *Vigna radiata* (L.) Wilczek, i.e. Pusa Vishal and SML 668 were procured from Seed Distribution Centre, IARI, New Delhi. The experimental design was completely randomised. Biochemical analysis was conducted after treatments of NaCl (50 mM, 100 mM, 150 mM), salicylic acid (0.01 mM, 0.025mM, 0.050mM) as well as their combinations (50mM NaCl-0.01mM S.A., 50mM NaCl-0.025mM S.A., 50mM NaCl-0.050mM S.A., 100mM NaCl-0.01mM S.A., 100mM NaCl-0.025mM S.A., 100mM NaCl-0.050mM S.A., 150mM NaCl-0.01mM S.A., 150mM NaCl-0.025mM S.A., 150mM NaCl-0.050mM S.A.). A separate set in distilled water as a control was also analysed.

The seeds were disinfected, and 20 seeds of each cv. of *Vigna radiata* (Pusa Vishal and SML 668) were put in the petri plates with different concentrations of each treatment. This was done in the Plant Physiology and Tissue Culture Laboratory, Department of Botany, C.C.S. University, Meerut. Biochemical analysis of both the varieties was done after 3 days and 6 days after imbibition. Biochemical analysis includes the estimation of total proteins⁴, total phenolics⁵ and total proline content⁶.

RESULTS AND DISCUSSION

After biochemical analysis of all the treated as well as control seeds of both cultivars following results were obtained.

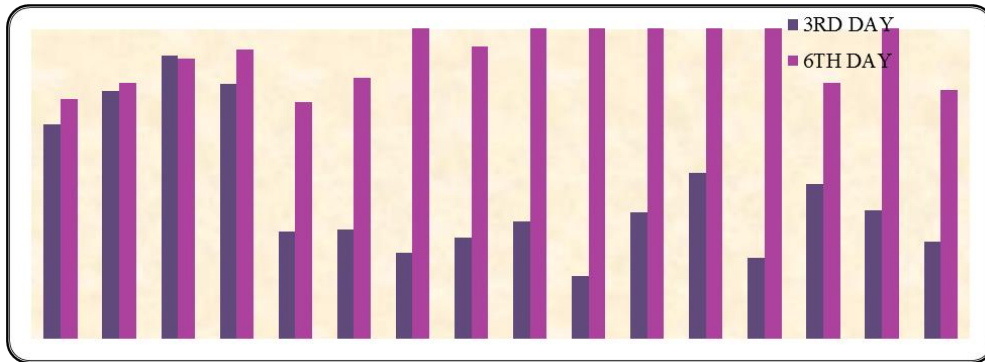


Figure 1: mg Casein eq./g F.W. Protein in *V. radiata* cv. PUSA VISHAL on 3rd and 6th Day after imbibition in different treatments

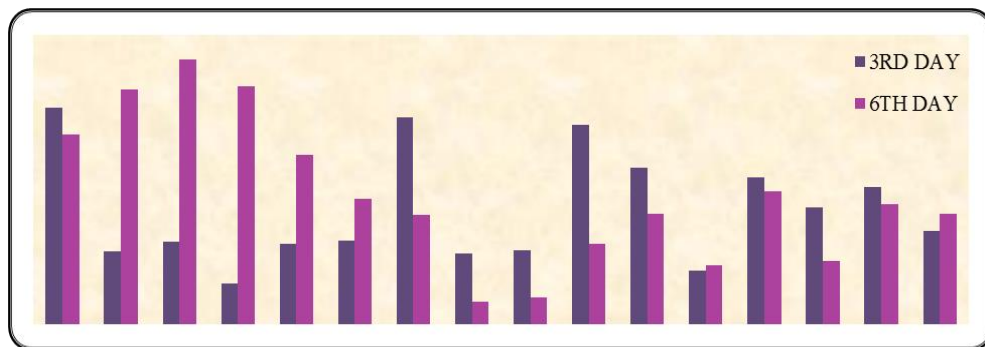


Figure 2: mg Casein eq./g F.W. Protein in *V. radiata* cv. SML-668 on 3rd and 6th Day after imbibition in different treatments

It was noted that proteins increased from 3 to 6 days in all the treatments as well as in control sets of seeds of *V. radiata* cv. PusaVishal. However, maximum amount and maximum increment (9 times over 3 days) was noted under 100mM NaCl+ 0.025 mM SA, as against individual 0 and 2.5 times increment by 100mM NaCl and 0.025 mM SA, respectively (Figure 1). Interestingly, till 3 days after treatment with SA, or with any

concentration of NaCl with any concentration of SA, the seeds were lower than control in proline content, lowest being in 0.05 mM SA treatment. This indicates antagonistic activity of SA up to 3 days but synergistic stress response of both 100mM NaCl and 0.025 mM SA (elicitors) on seeds of *V. radiata* cv. Pusa Vishal after 6 days of treatment (Figure 3).

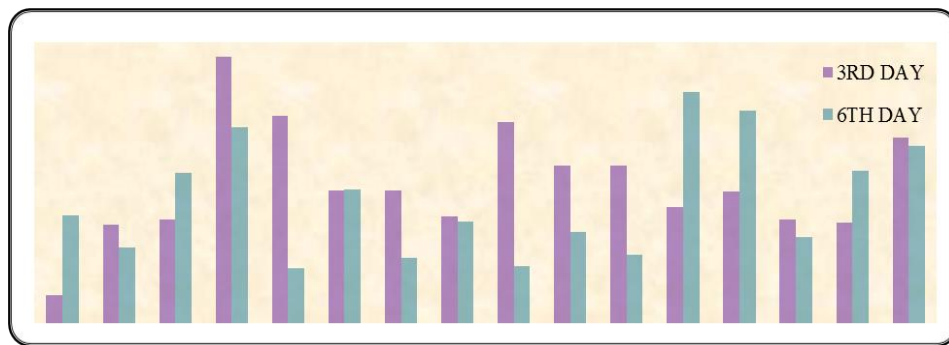


Figure 3: mg Proline eq./g F.W. Proline in *V. radiata* cv. PUSA VISHAL on 3rd and 6th Day after imbibition in different treatments

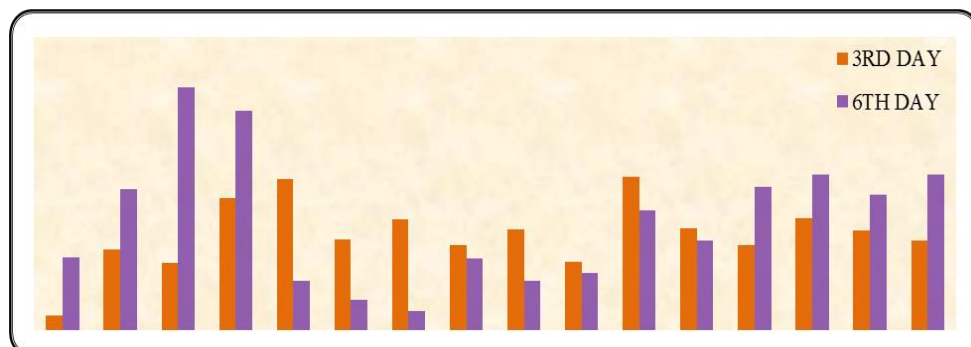


Figure 4: mg Proline eq./g F.W. Proline in *V. radiata* cv. SML 668 on 3rd and 6th Day after imbibition in different treatments

Proline content was noted to be minimum after 3 days of imbibition in control sets, but it increased almost 4 times (maximum) after 6 days. Both phenolics and proline content, in lower and higher concentrations of NaCl (50 mM, 150 mM) and Salicylic acid (0.01mM, 0.05 mM) used, declined from 3rd to 6th day after imbibition, but increased moderate to negligible in the

medium concentrations of NaCl (100mM), and SA (0.025mM) individually (Figure 5). However, maximum increment and maximum amount amongst treatments was noted in a combination of 100mM NaCl+0.025mM SA, indicating the synergistic effect of the two elicitors on proline content, too.

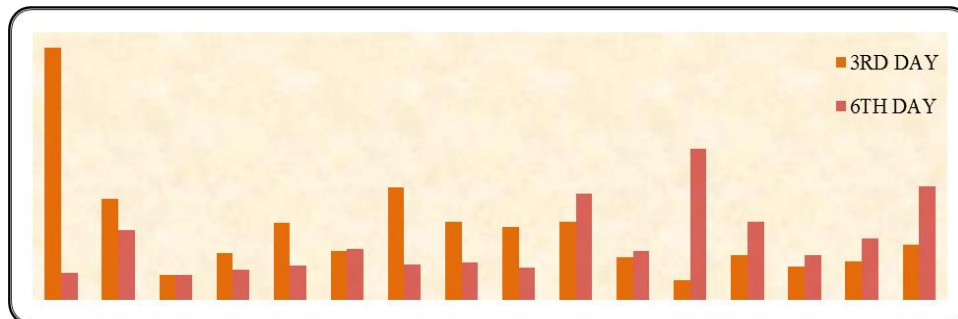


Figure 5: mg Gallic acid eq./g F.W. Phenolics in *V. radiata* cv. PUSA VISHAL on 3rd and 6th Day after imbibition in different treatments

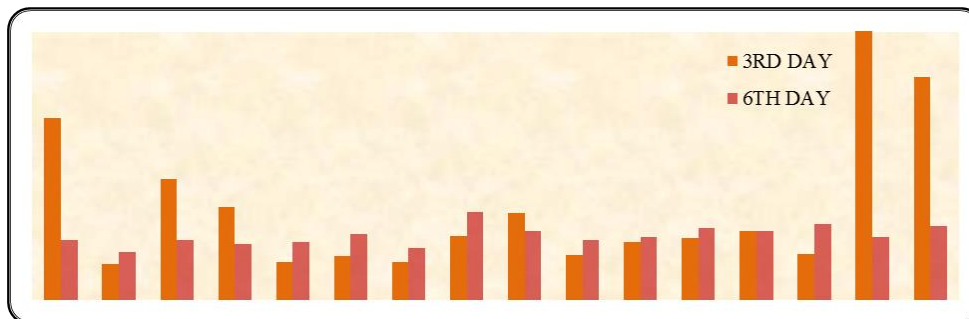


Figure 6: mg Gallic acid eq./g F.W. Phenolics in *V. radiata* cv. SML 668 on 3rd and 6th Day after imbibition in different treatments

In case of phenolic content, in control sets it was the highest of all treatments after 3 days of imbibition, but the same dropped to almost 15 times lower level by 6 days. Amongst all other treatments, interestingly, the same 100mM NaCl+0.025 mM SA exhibited the minimum (after 3 days) and maximum (after 6 days) phenolic content, though individually in 100mM NaCl or in 0.025mMSA, there was barely any change in amounts of phenolics from 3rd to 6th day, indicating a synergistic response of the two elicitors, again (Figure 5).

The second cultivar of *V. radiata* cv. SML-668, studied in the present investigation exhibited contrasting performance of control sets in terms of protein and proline, which declined from 3rd to 6th day after imbibitions (Figure 2 and Figure 4). Phenolic content of the seeds of this cultivar was moderate which though dropped by 6th day but the decline was much lesser than the previous cultivar Pusa Vishal (3 times decline in SML-668, as against 15 times in Pusa Vishal) (Figure 6). Whereas NaCl concentrations led to increase in protein, proline and phenolics, Salicylic acid concentrations led to increase in protein and phenolics upto 0.025 mM and 0.05 mM concentration, respectively during 3 to 6 days, but led to decline in proline content in all concentrations, maximum being in 0.05 mM SA. In this cultivar, 150mM NaCl combination with SA (0.025mM) led to maximum phenolic content and high protein content after 3 days of imbibition which declined by 6th day. However, proline content increased in combination of 150 mMNaCl with all concentrations of SA. The synergistic effect of NaCl with SA could be noted in this cultivar also, though at differing concentrations from Pusa Vishal.

The results indicate and reiterate the previous finding of involvement of Salicylic acid in modifying abiotic stress response⁷⁻¹⁰. Cultivar specific modification of salt stress response by salicylic acid is very starkly evident. In the cultivar Pusa Vishal, whereas single combination of 0.025 mM SA effectively modified 100mM NaCl-mediated decline or negligible increase in protein, proline and phenolics to increase in protein, proline and phenolics, whereas, in SML-668, 0.025 mM SA reverted 100mM NaCl induced increase in protein, proline and decline in phenolics to increase in phenolics and decline in proteins and proline.

Increasing NaCl concentration led to increased leakage of electrolytes and Malondialdehyde content in *Brassica napus* seedlings¹¹ and although SA is not essential for germination under normal growth conditions, it plays a promotive role in seed germination under high salinity by reducing oxidative damage. In the present investigation, seeds of both cultivars of *V. radiata* could not germinate at as low as 0.01 mM SA, indicating genus/species specific response of SA. Pre-treatment with 0.5mM S.A., there is an elevation of substantial water loss from wheat seedlings¹². It also increases drought tolerance by affecting the ascorbate-glutathione cycle. There are at least 37 proteins reported by proteomic studies that are up regulated by S.A. pre-treatment under drought stress. Several defence proteins including glutathione S-transferases, APX and 2-cysteine peroxiredoxin, are included¹³, which suggests that pre-treatment with S.A. provides protection against oxidative damage by enhancing the antioxidant defense system. Pre-treatment of tomato with SA in hydroponic culture triggered the accumulation of ABA, leading to an improved acclimation to salt stress¹⁴.

CONCLUSION

In the present investigation, SA has played a pivotal role in increasing the phenolic content under NaCl stressed seeds, protecting them from salt stress induced oxidants, by its antioxidant response in both the cultivars of *V. radiata*. This generates a future hope for finding common transcription factors for abiotic/biotic stress tolerance engineering in required crop plants.

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