

## Effect of RiD on Aphid Tolerance in *Brassica juncea*

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## DESCRIPTION

Brassica juncea (Indian mustard) is one of the most important oilseed crop cultivated in India. But there is a huge loss in its productivity due to many limiting factors. One of the most devastating pests of B. juncea is Lipaphis erysimi (mustard aphid) which causes a huge loss in its yield of about 96% and reduction in oil content by 15%-32%. This mustard aphid causes nutrition deficiency leading to wilting of the plant and hence causes great economic loss even after good agricultural practices. Aphids also transmit viruses (e.g., Turnip mosaic virus and Cauliflower mosaic virus) during sap sucking which therefore causes various diseases such as mosaics. To eliminate this pest, the most conventional agriculture practice has been the use of chemical pesticides which significantly reduces aphid colonization, but causes hazardous effects on the environment as well as on human health. Therefore several attempts have been taken to develop or identify aphid tolerant varieties from the gene pool since ages and have not met with any success. This eventually led to the attempts in developing transgenic Brassica carrying genes for aphid resistance/tolerance. There are various insecticidal agents discovered till today which have been used to develop transgenic B. juncea lines expressing those insecticidal agents such as proteases and lectins which help in the reduction in aphid population to some extent. But there is always a need of identifying or discovering new insecticidal agents due to induced resistance and behavioural reorientation of the insects in due course of time. A wild crucifer, Rorippa indica has been previously reported to be tolerant towards this aphid. It survives the pest attack even when there is heavy colonization. A transcriptomic analysis on R. indica using cDNA AFLP revealed a novel defensin peptide-Rorippa indica Defensin (RiD) which was noted to be significantly upregulated during this mustard aphid-L. erysimi infestation. Plant defensins are ubiquitous cysteine-rich plant peptides, generally known as antimicrobial peptides which are a part of innate immune response against pests/ pathogens. They are responsible in host defense and are also known as host defense peptides. Some plant defensins also show potent Perspective

insecticidal activities besides antifungal and bactericidal properties as characterized till today. One of the plants defensin BrD1, a defensin gene from Brassica rapa, confers resistance towards brown plant hopper (Nilaparvata lugens) in transgenic rice. R. indica Defensin (RiD), a peptide from the wild crucifer has been reported to have high insecticidal activity against L. erysimi with an LC50 value of 9.099  $\pm$  0.621 µg/ml. RiD is also seen to be secreted into the apoplastic region indicating its role in inhibiting nutrient uptake by aphids which occurs mainly via the extracellular route to pierce through the cells. Sap sucking by aphids also induces various signalling mechanisms in plants mediated by hormonal crosstalk between different phytohormones such as JA, SA, ABA or ET. Feeding and piercing damage of plants by aphids or any phloem feeders triggers the synthesis of jasmonates which in turn regulates the expression of defense related proteins and the biosynthesis of secondary metabolites. As a result, constitutive as well as transient expression of jasmonate regulated genes has been seen to regulate plant defence. Expression of plant defensins are also induced by jasmonates as a part of defence mechanism. Thus, induction and overexpression of jasmonate related genes such as defensins which have insecticidal properties might defend aphid colonization through the development of transgenics. The suitability of RiD as an effective insecticidal agent against L. erysimi indicates that it can be a potent candidate for integrated pest management. But there has always been a myriad of concerns about the impact of transgenes for food derived from genetically modified crops. It is therefore necessary to assess the transgene for allergenicity before introducing it into a crop plant.

## CONCLUSION

According to FAO/WHO guidelines (2001) and Codex Alimentarius guidelines (2009), prediction of an allergen requires extensive sequence homology search across known databases of allergens and the identification of motifs responsible for a

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specific allergen activity. The next important strategy is to characterize the stability of the protein, which includes thermal stability assays, pepsin digestibility and allergenecity assessment of the recombinant proteins using animal models. In the aforesaid background, we assessed the risk factors and allergenecity of RiD for transgenic application. As RiD was found to be biologically safe and effective against *L. erysimi*, we tried to develop transgenic *B. juncea* plants expressing RiD to control aphid population on the said plant.