A novel resistance mechanism to *Bacillus thuringiensis* Cry1Ac toxin in diamondback moth

With the support by the National Natural Science Foundation of China, the Ministry of Science and Technology of China, the Agricultural Science and Technology Innovation Program and the Beijing Key Laboratory for Pest Control and Sustainable Cultivation of Vegetables, Prof. Zhang Youjun's laboratory at the Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences, reported a novel resistance mechanism to *Bacillus thuringiensis* Cry1Ac toxin in diamondback moth, which was published in *PLoS Genetics* (2015, 11(4): e1005124).

Biopesticide and transgenic crops based on *Bacillus thuringiensis* (Bt) Cry toxins are widely used worldwide, yet the development of field resistance seriously threatens their sustainability. Unraveling these resistance mechanisms is of great importance for delaying insect field resistance evolution. The diamondback moth, *Plutella xylostella* (L.), was the first insect to evolve field resistance to Bt biopesticides and it is an excellent model to study Bt resistance mechanisms. In this work, we present strong empirical evidence supporting that (1) field-evolved resistance to Bt in *P. xylostella* is tightly associated with differential expression of a membrane-bound alkaline phosphatase (*ALP*) and a suite of ATP-binding cassette transporter subfamily C genes (*ABCC2* and *ABCC3*), and (2) a constitutively transcriptionally-activated upstream gene (*MAP4K4*) in the MAPK signaling pathway is responsible for this *trans*-regulatory signaling mechanism. These findings identify key resistance genes and provide the first comprehensive mechanistic description responsible for the field-evolved Bt resistance in *P. xylostella*, which greatly advances our comprehensive understanding of insect resistance mechanisms to Bt Cry1Ac toxin and provides new insights into how insects evolve resistance to Bt entomopathogen.

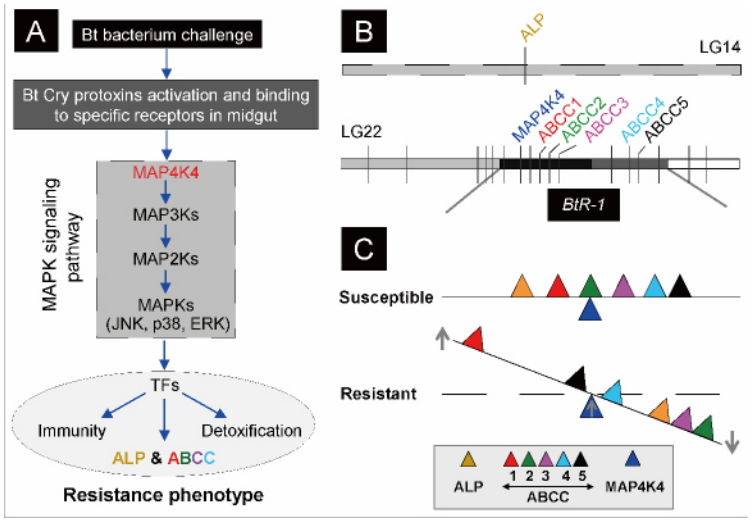

Figure Schematic drawing depicting a novel Cry1Ac resistance mechanism in *P. xylostella*. (A) Cry protoxins produced by Bt bacterium are activated by midgut proteases and then bind to specific midgut receptors to form ionic pores leading to insect death. In resistant insects, over-activation of the MAPK signaling pathway (highlighted in the dashed box, *MAP4K4* is an important upstream regulator) serves to alter the expression of different genes associated with resistance phenotype, including receptor genes involved in Bt resistance probably through the regulation of diverse transcription factors (TFs). Blue arrows indicate recruitment, activation, or production. (B) Diagram of the physical location of the genes involved in Cry1Ac resistance in *P. xylostella*. *ALP* is located in the linkage group 14 and outside the *BtR-1* resistance locus, whereas other genes are located within the *BtR-1* locus in the linkage group 22. (C) *MAP4K4*-mediated interplay between *ALP* and *ABCC* among *P. xylostella* strains with different susceptibility to Cry1Ac toxins. Based on this study, we propose that *MAP4K4* serves as a general switch modulating the differential expression of *ALP* and ABCC1-5 in *P. xylostella*. The data do not strongly support the conclusion that changes in expression of ABCC4 and ABCC5 affect the development of resistance. Up-and downward arrows represent the up-and down-regulation of genes, respectively, in Cry1Ac-resistant and susceptible *P. xylostella* strains. The distance remove from the fulcrum point reflects the qualitative magnitude of gene regulations.