



# Engineering insect resistance in Rice

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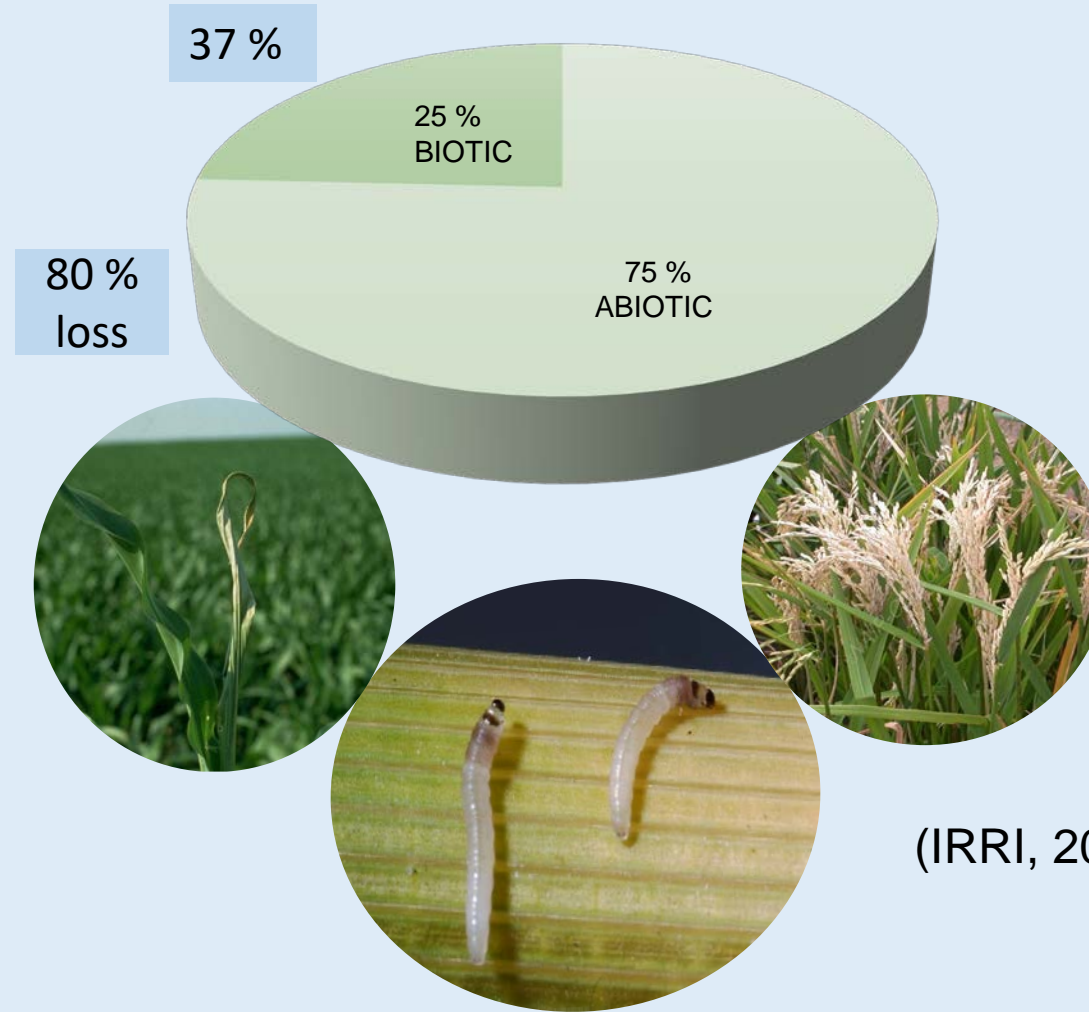
# WHY?

## RICE

Major calorie source of more than 50 % of global population (IRRI, 2013)



## INSECT RESISTANCE



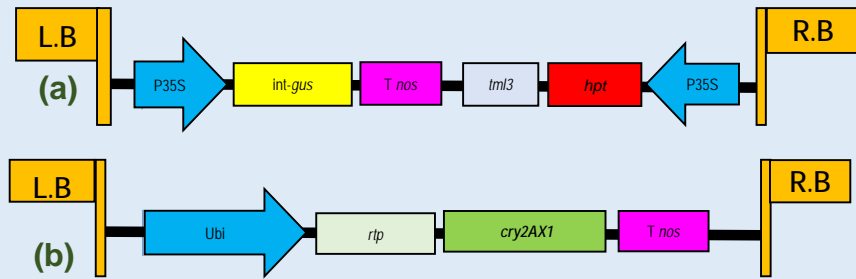
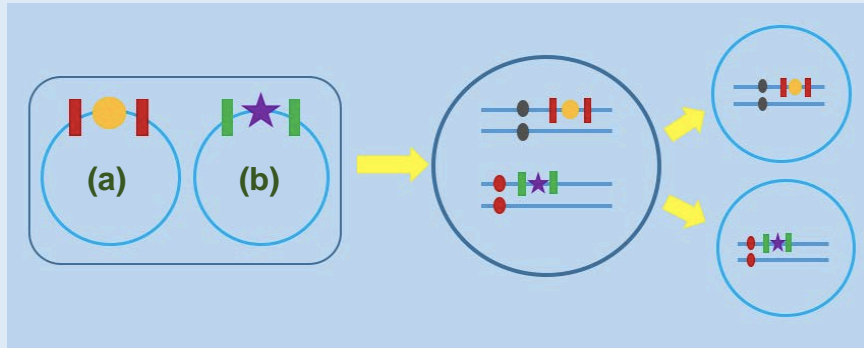
(IRRI, 2018)

## MARKER-FREE

To hasten public acceptance of transgenic crops (Qiu *et al.*, 2010)

# HOW?

## Co-transformation strategy



## Agrobacterium-mediated transformation



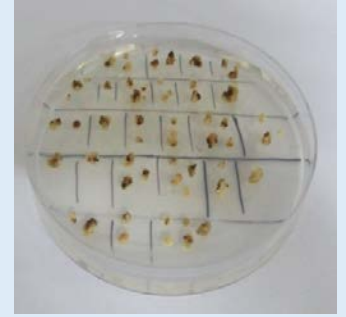
a. Rice immature embryos infected with *Agrobacterium*



b. Immature embryos 7 days after co-cultivation



c. Calli sub-cultured on resting medium



d. Calli on selection medium



e. Regeneration of embryogenic calli



f. Rooting of regenerated calli



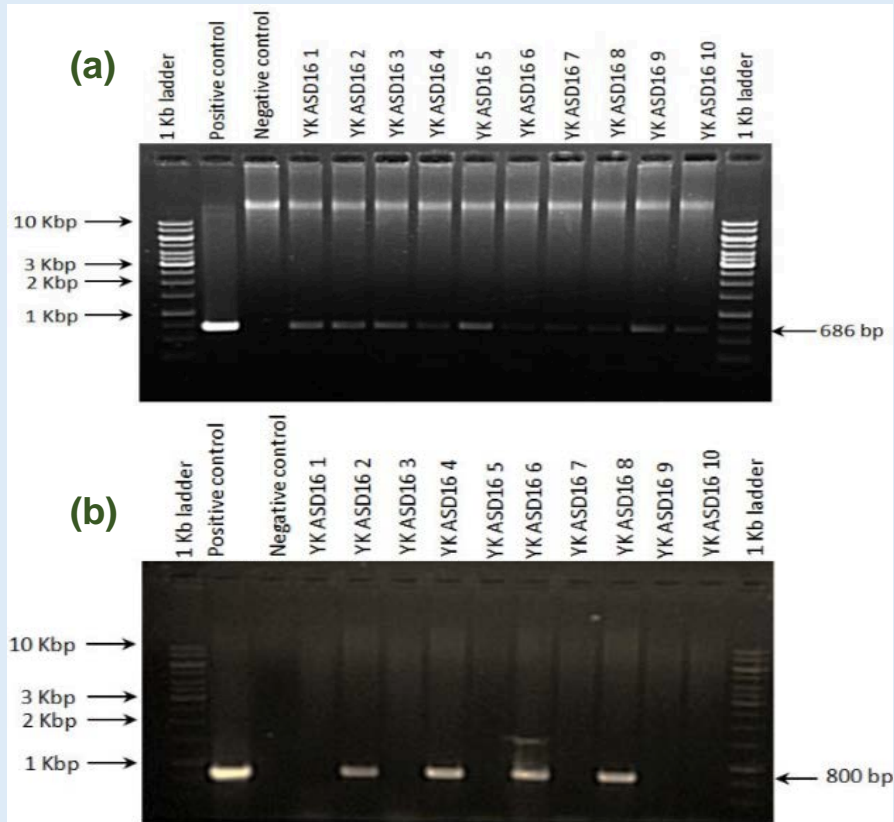
g. Putative transgenic plants on 1/2 strength MS media



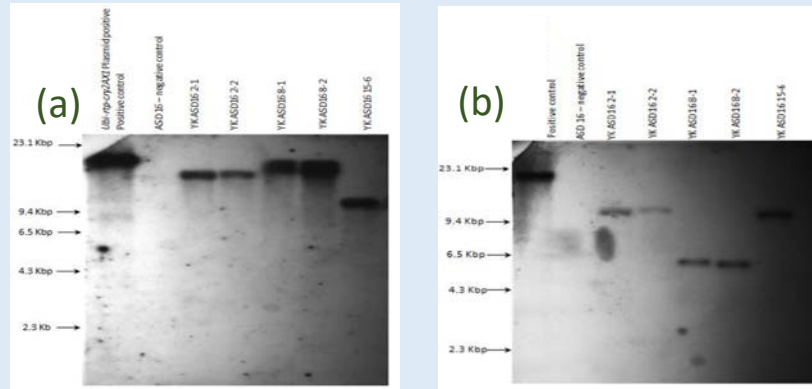
h. Established putative transgenic plants in transgenic greenhouse

# WHAT?

PCR confirmation of T<sub>0</sub> progeny for presence of (a) *hpt* gene, (b) *cry2AX1* gene



Southern blot hybridization analysis of T<sub>0</sub> transgenic rice plants with (a) *cry2AX1* probe; (b) *hpt* probe



Quantitative ELISA for marker-free T<sub>1</sub> progeny.

T <sub>1</sub> transgenic rice lines	Cry2AX1 Concentration (ng/g of fresh leaf tissue)
YK ASD16 8-1-1	8.8
YK ASD16 8-1-27	12.9
YK ASD16 8-1-57	25.7

20 events positive to *gus* and *hpt*

10 events positive to *cry2AX1*

3 best *cry2AX1* expressing events with single locus integration

2 events for segregation study

1 event with 8 marker-free plants

# ***BENEFITS?***

<b>Yield</b>	<b>29 % increase</b>
Pesticide Use	Decrease by 2/3rd
Cost	Reduced by 7 times per ha.
Pesticide poisoning	No incidence reported

(Chen *et al.*, 2011)