

## Original Research Article

# Effects of Secondary Exposure of *Cry1Ac* Protein on Predatory Insect *Coccinella arcuata* Fabri of BPH Fed on Different IR-64 Bt Rice Events

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## ABSTRACT

The experiment was undertaken at the transgenic containment facility, Department of Plant molecular biology and Biotechnology, College of Agriculture, Raipur during 2014 and 2015. Secondary exposure of Bt protein was assessed for predatory insect, *Coccinella arcuata* number of BPH consumed by each adult beetle was recorded daily. Feeding potential of *Coccinella* beetle was obtained on the basis of two years mean, the maximum average consumption rate (15.30 nymph / beetle) was noticed in TN-1 followed by IR64-1 (13.90 nymph / beetle) and minimum in IR64-2 (12.70 nymph / beetle). Whereas, test weight of beetle on the basis of two years pooled mean of beetle was noticed maximum (3.15mg) in TN-1 followed by IR64-C (3.04mg) and minimum in IR64-2(2.47mg). On the basis of present investigation, the effect of *Cry1Ac* protein expressed in different IR-64 Bt rice events on non-target and its secondary exposure on predatory insects. The *Cry1Ac* protein was expressed significantly on non- target (BPH) and its predator *Coccinellid* beetles differ non -significantly. There is an urgent need to generate biosafety data for Bt. rice under controlled conditions for taking policy decision about its cultivation in the country.

### Keywords

IR-64 Bt rice,  
*Cry1Ac* protein on  
*Coccinella*,  
secondary  
exposure of Bt  
protein

## Introduction

Rice is commonly carried out worldwide as it is used as a basic diet by almost half of the world population especially eastern communities and it is by far the greatest cash crop. Major Rice growing countries includes India, China, Bangladesh, Thailand, Myanmar, Philippines, Japan, Pakistan, USA, Indonesia, Korea and Vietnam. Rice (*Oryza sativa* L.) is one of the world's most important crops, providing a staple food for nearly half of the global population (FAO, 2004). Rajendran *et al.*, (1989) studied that

predatory potential of the adult staphylinid beetle, *Paederus fuscipes* Curtis. on the adults of Brown plant hopper (BPH), *Nilaparvata lugens* Stal., White backed plant hopper (WBPH), *Sogatella furcifera* Horvath. and Green leaf hopper (GLH), *Nephotettix virescens* Distant. They observed that each staphylinid beetle consumed on an average 8.7 BPH or 8.3 WBPH or 8.4 GLH per day. Chandel (2008) assessed the potential effect of Bt protein expressed in transgenic Bt rice on two non-

target insect species, *N. lugens* and *C. lividipennis*. A transgenic *Bt* rice line expressing high amount (0.812 to 1.279 µg/g TSP) of *Bt* protein was used for this study. The ELISA results clearly showed no detectable biologically active *Bt* protein in the body tissues of *N. lugens* after feeding on the transgenic rice lines expressing a high amount of *Bt* protein. It may be possible that either toxic protein was not present in the xylem or phloem saps or degradation of toxin protein after ingestion by *N. lugens* adults. Manley, G.V. (1977) conducted laboratory and field trial for staphylinid beetle, *Paederus fuscipes* Curtis to determine natural field density level, feeding patterns, behaviour and biology.

He observed that *P. fuscipes* was found to be an aggressive leaf hopper predator in rice fields and feeding on leaf hopper nymphs appears to be density dependent within certain population levels.

### Materials and Methods

*Coccinella arcuata* beetle was collected from the BPH rice field. The colony of beetle was maintained on culture TN-1 rice plants infested with *N. lugens* eggs. Newly hatched *N. lugens* nymphs, reared on control rice plants for more than 48 h were provided to *Coccinella arcuata* daily at the rate of ten nymphs of BPH to first and second-instar grubs and 15 nymphs to third, fourth and fifth-instar of *Coccinella* grubs into petri dish (90 x 100 mm). Survival and molting of *Coccinella* grubs were recorded daily under a binocular microscope until the grub died or reached at the adult stage. The number of prey eaten was recorded daily and uneaten nymphs were removed from the petri dish and fresh nymphs were provided. Twenty petri dish (representing replicates) containing one *Coccinella arcuata* Fabri in each petri dish were arranged in CRBD.

### Results and Discussion

The effects of *Bt* toxins on life-history parameters of brown planthopper and *Coccinella* beetle feeding on *Bt* rice and control non-*Bt* rice did not differ significantly with respect to their feeding behaviour, development and weight.

The laboratory studied of predation potential of common predators, *Coccinella* showed that common predator consumed BPH nymphs when offered in a test tube feeding situation. The single prey i.e. grubs which thirty numbers of BPH nymphs consumed or killed per day/test tube and their weight was recorded. The results are presented in table-1 fig.-1.

The BPH nymph consumption by *Coccinellid* beetle revealed that the average feeding potential rate ranged from 11.60 to 15.40 nymph /beetle. The maximum average consumption rate (15.40 nymph / beetle) was noticed in TN-1 followed by IR64-2, IR64-C and Ptb-33-C (12.80 nymph / beetle), respectively and minimum in IR64-3 (11.60 nymph / beetle) during 2014. The maximum average consumption rate (15.20 nymph / beetle) was noticed in TN-1 followed by IR64-1 (14.60 nymph / beetle) and minimum in IR64-2 (12.60 nymph / beetle) during 2015.

On the basis of two years mean, the maximum average consumption rate (15.30 nymph / beetle) was observed in TN-1 followed by IR64-1 (13.90 nymph / beetle) and minimum in IR64-2 (12.70 nymph / beetle). The weight of beetles measured which consumed BPH nymphs into the tube feeding situation on different transgenic rice lines including control under controlled condition. Mean test weight of beetle ranged from 2.24 to 3.17mg when consumed 11.60 to 15.40 nymph /beetle.

**Table.1** Feeding potential of *Coccinella* beetle on different transgenic rice lines (including control) during 2014 and 2015

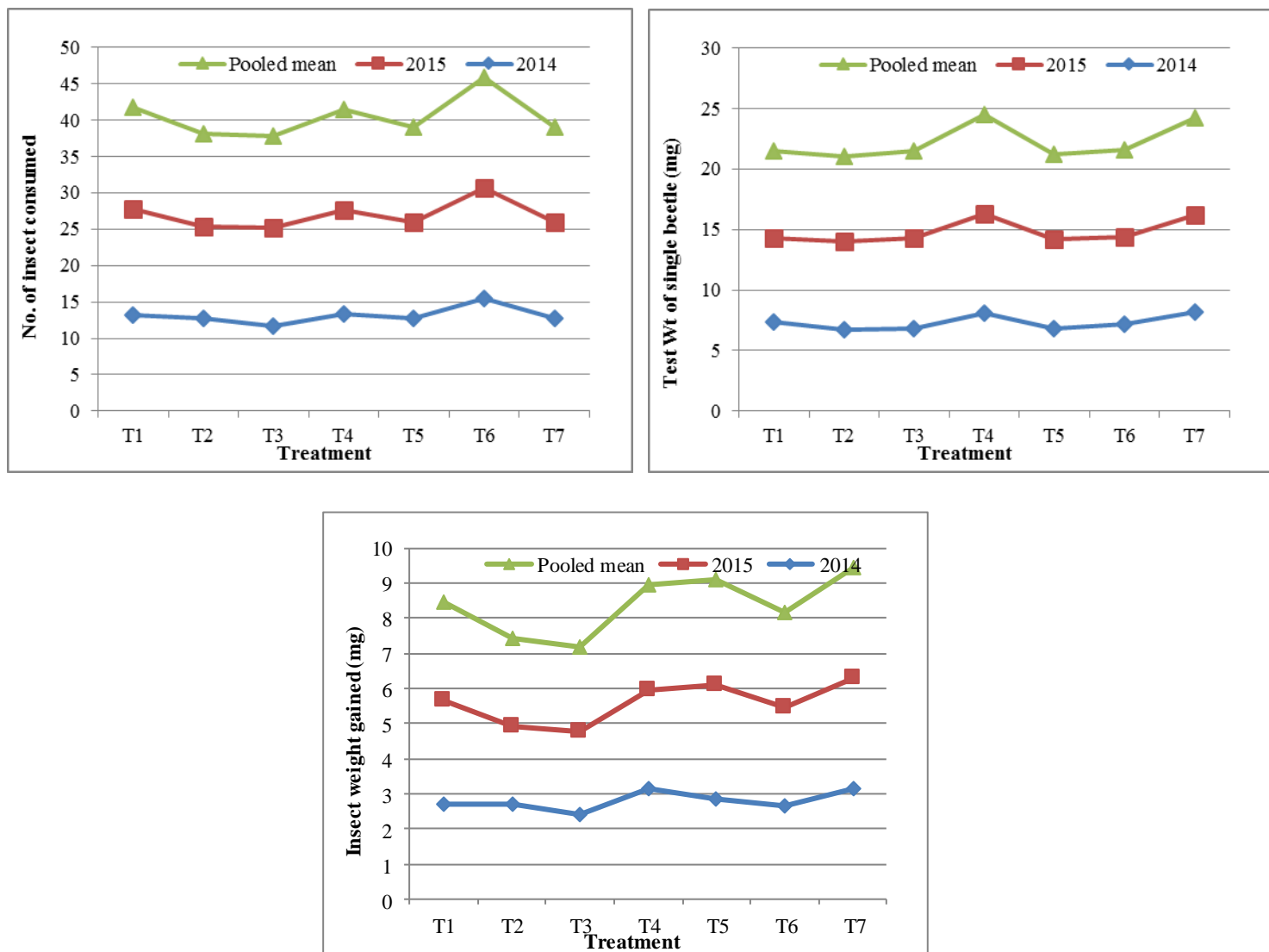
Transgenic events	Number of insect consumed			Test weight of single beetle (mg)			Insect weight gained (mg)		
	2014	2015	Pooled Mean	2014	2015	Pooled Mean	2014	2015	Pooled Mean
IR64-1	13.20	14.60	<b>13.90</b>	7.38	6.93	<b>7.155</b>	2.71	2.93	<b>2.82</b>
IR64-2	12.80	12.60	<b>12.70</b>	6.72	7.32	<b>7.02</b>	2.70	2.24	<b>2.47</b>
IR64-3	11.60	13.60	<b>12.60</b>	6.85	7.45	<b>7.15</b>	2.42	2.36	<b>2.39</b>
IR64-4	13.40	14.20	<b>13.80</b>	8.13	8.21	<b>8.17</b>	3.12	2.84	<b>2.98</b>
IR64-C	12.80	13.20	<b>13.00</b>	6.86	7.30	<b>7.08</b>	2.87	3.21	<b>3.04</b>
TN-1(C)	15.40	15.20	<b>15.30</b>	7.18	7.21	<b>7.195</b>	2.65	2.80	<b>2.725</b>
PTB-33(C)	12.80	13.20	<b>13.00</b>	8.17	8.00	<b>8.085</b>	3.13	3.17	<b>3.15</b>
<b>SEM</b>	<b>0.160</b>	<b>0.146</b>		<b>0.420</b>	<b>0.537</b>		<b>0.193</b>	<b>0.268</b>	
<b>CD (P=0.05)</b>	<b>(0.462)NS</b>	<b>(0.422)NS</b>		<b>(1.217)NS</b>	<b>(1.557)NS</b>		<b>(0.559)NS</b>	<b>(0.777)NS</b>	

\*Data based on the five replications and each replicate consists 30 adult/nymph in one glass tube with single predator.

\*C-control

\*NS-non-significant.

**Fig.1** Feeding potential of *Coccinella* beetle on different transgenic rice lines (including control) during 2014 and 2015



The maximum mean test weight of beetle (3.17mg) was noticed in TN-1 followed by IR64-C (2.87mg) and minimum in IR64-3 (2.42mg) during 2014. Whereas, during 2015, the maximum mean the weight of beetle (3.17mg) was noticed in TN-1 followed by IR64-C (3.21mg) and minimum in IR64-2 (2.24mg). On the basis of two years, pooled mean test weight of beetle was noticed maximum (3.15mg) in TN-1 followed by IR64-C (3.04mg) and minimum in IR64-2(2.47mg).

Controversy about the benefits and ecological risks of transgenic crops has existed since their advent, and became fiercer after the first three Bt crops were commercialized in 1996 (Shelton and Sears, 2001). Potential impact, especially on non-target organisms, was a major concern with the deployment of transgenic crops (Romeis *et al.*, 2006). Not accurate but similar finding gave by Laba (1999) found that *P. fuscipes* and *Ophionea sp.* consumed 4.9 and 2.7 brown plant hopper per day respectively,

in field condition. This was showed that they had lower predation ability rather than *M. vittaticollis*, *A. longipennis*. The consumption rate of *Paederus fuscipes* were observed on an average 8.7 BPH per day in the laboratory condition (Rajendran and Gopalan, 1989). No significantly negative or unintended effects of transgenic rice on non-target arthropods were found compared with non-transgenic rice (Chen *et. al.*, 2006). Many field studies shown negligible impact on non-target organisms (Romeis *et al.*, 2006; Marvier *et al.*, 2007).

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