Indian Journal of Agricultural Sciences **86** (4): 481–4, April 2016/Article

Mechanisms of physical and biochemical basis of resistance against leaf-hopper (*Amrasca biguttula biguttula)* in different okra (*Abelmoschus esculentus*) genotypes

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Received: 17 March 2015; Accepted: 23 November 2015

ABSTRACT

Different biophysical parameters, viz. trichomes, leaf length, angle between the mid-ribs, angle between the mid-ribs and vein, mid-rib thickness, leaf angle, number of leaves/plant, plant height and biochemical parameter like total phenol content of leaves were studied in relation to the expression of reaction towards leaf hopper (*Amrasca biguttula biguttula*) in 10 okra [*Abelmoschus esculentus* (L.) Moench] genotypes. It was observed that genotype SB-6 had relatively lower number of trichomes on leaf lamina (10.11), mid-rib (7.17) and vein (8.05) as showed highly susceptible to jassids (17.57 jassids/leaf) as compared to tolerant genotype VROB-181 (5.43 jassids/leaf) which had 11.85, 9.17 and 9.95 trichomes per cm^2, respectively. Susceptible cultivar SB-10 possessed higher mid-rib thickness (1.75 mm) and leaf length (25.65 cm) as compared to tolerant genotype VROB-181 (1.58 mm and 22.43 cm, respectively). Similarly, higher number of total leaves (31.9) per plant was also recorded from SB-6 as compared to other tolerant lines (VROB-181, VROB-178, VROR-160). Leaf length, plant height, angle between mid-ribs and total leaves showed a strong positive correlation (r value = 0.493, 0.499, 0.723 and 0.474, respectively) with jassid incidence. Amongst the biochemical parameter, total phenol content was highest in VROB-181 (75.04 mg/100 g) and the incidence of jassids was lowest than those of susceptible genotypes, viz. SB-6 (42.61 mg/100 g) and SB-10 (48.35 mg/100 g) and thereby establishing the significant negative correlation (r = - 0.577) with the jassid incidence and total phenol content.

Key words: *Amrasca biguttula biguttula*, Morphological and biochemical parameters, Okra germplasm, Varietal screening

Okra [*Abelmoschus esculentus* (L.) Moench], also known as lady's finger, is one of the most important traditional vegetable in India. It is an important source of vitamin A, B, C and is also rich in protein, carbohydrates, fats, iron and iodine and plays a pivotal role in human diet (Halder *et al*. 2015). Beside these, it is also a rich source of dietary fiber, antioxidants, ascorbic acid and folate. Increasing crop loss owing to insect pests is a major constrain in agricultural production and productivity. Okra jassid, *Amrasca biguttula biguttula* Ishida (Hemiptera: Jassidae) is among the most important sucking insect attack on okra crop (Kakar and Dobra 1988, Singh *et al*. 1993, Dhandapani *et al*. 2003). According to Sharma and Singh (2002) okra was the most suitable host in terms of oviposition, nymphal survival and feeding of leafhopper. Both the nymphs and adults of the leafhopper can cause damage right from early seedling stage to till fruit maturity of the crop by sucking the cell sap thereby causing typical hopper burn (phytotoxemia); resulting in 40-50% yield reduction (Bindra and Mahal 1981). At present, management of this nefarious sucking pest has largely been relied on chemical control. However, the demands for clean and ecologically sound environmental envisages, careful planning for rationalizing the insecticides interventions are need of the hour. With the environmental friendly pest management approach, Host Plant Resistance (HPR) is one of the most cost-effective and safe methods. Development of suitable resistant/tolerant varieties is an ideal component at no additional cost, compatible with other methods of pest control and free from environmental pollution against buildup of pest population. Various biophysical and biochemical characters of the plants play an important role by providing resistance against number of insect pests (Halder *et al*. 2006 b, Halder and Srinivasan 2011). Therefore, an attempt was made to identify the response by different available genotypes of okra in order to determine resistance/susceptibility.

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MATERIALS AND METHODS

A total of 10 genotypes, viz. SB-6, SB-8, SB-10, VROB-

178, VROB-179, VROB-181, VROT-108, VROR-157, VROR-159 and VROR-160 were undertaken for the studies on infestation of jassid on okra at experimental farm of ICAR-Indian Institute of Vegetable Research (82°52' E longitude and 25°12' N latitude), Varanasi, Uttar Pradesh, India during 2011-14. Two rows of 50 hills of each genotype were sown. A spacing of 60 × 30 cm was adopted and crops were raised following all recommended agronomic practices. Two rows of okra (cv. Pusa Sawani) were maintained to serve as infester lines after 5 test entries to favour the buildup of the population of jassids. Ten randomly selected plants of each row were taken at regular intervals. Ten plant parameters, viz. trichomes on leaf lamina (cm^2), mid-rib (cm) and veins (cm), leaf length (cm), leaf angle (°), angle between the mid-ribs (as the okra leaf is a compound leaf), angle between mid-rib and veins, mid-rib thickness (mm), number of leaves per plant and plant height were studied for their role in expression of varietal reaction to jassid. In addition to these, the content of total phenol in green leaves as biochemical parameter was also evaluated.

The uniformly developed leaves (5-days after opening) of all test genotypes were collected from randomly selected plants at 60 days after sowing (DAS) and trichome density from different parts of the leaves, viz. leaf lamina, mid-rib and veins were measured. The leaves of each genotypes were cut into bits of 1 × 1 cm and number of trichomes present on the epidermis of these bits were counted and then length was measured under a binocular microscope (10 - 100X) (Nikon SMZ-10A).

Angle between the okra leaves (five days after opening) and the main stem/branch, between the mid-ribs and leaf vein and mid-rib was measured by using a protractor. Total numbers of leaves were counted twice, i.e. 60 DAS and at physiologically matured stage; whereas, plant heights were taken at physiological matured stage of the crop. Similarly, twenty fully developed mature leaves of uniform age were collected and their lengths from base to apical tip were recorded.

Biochemical parameter, i.e. total phenols in fresh green leaves were estimated for their role in expression of varietal reaction against jassid. Leaf samples of each genotype (five days after emergence) were collected and analyzed for total phenols and anthocyanin contents in fruits to indicate existence of any variations among the entries. Estimation of total phenols was done by using Folin-Ciocalteau reagent (FC reagent) as per the procedure outlined by Malick and Singh (1980).

The data were subjected to analysis of variance using SAS package and the means were separated by LSD test at 5% level of probability. The data regarding physico-morphic plant characters were correlated with jassid population to find out their relation with leaf hopper incidence.

RESULTS AND DISCUSSION

Among the 10 test genotypes, highest jassid incidence (17.57/leaf) was recorded on SB-6 and SB-10 (15.57/leaf). Further, dense trichomes were observed on the ventral surface of leaf lamina of VROT-108 (16.35/cm^2) while trichome density was lower in SB-10 (8.51) which had relatively higher jassid incidence (Table 1). A significant negative correlation (r = - 0.343) was observed between trichome density on leaf lamina and jassid incidence. High trichome density might be imparting the physical barrier for the jassids rendering their non-preference over the low-trichomes genotypes. Earlier, Iqbal *et al.* (2011) reported a significant negative correlation between trichome density of leaf lamina

Table 1 Morphological and biochemical parameters of different okra genotypes with jassid incidence

Genotype	Jassids/ Leaf	Trichomes			Leaf length (cm)	Angle between the mid ribs (°)	Angle between the leaf vein and and mid rib (°)	Mid rib thickness (mm)	Leaf angle (°)	Total leaf/ plant	Plant height (cm)	Phenol (mg/ 100 g)
		Mid rib	Lamina	Vein								
SB-6	17.57	7.17	10.11	8.05	22.57	59.7	45.3	1.53	57.86	31.9	84	42.61
SB-8	9.71	8.33	9.95	6.67	20.42	43.2	47.1	1.59	59.71	29.8	101.7	54.43
SB-10	15.57	5.17	8.51	5.13	25.65	43.6	48.8	1.75	57.14	34.6	92.6	48.35
VROB-178	5.71	13.5	15.98	9.83	21.05	41.3	43.1	1.51	60.14	28.1	55.3	59.57
VROB-179	6.57	10.5	13.83	10.83	22.04	46.4	46	1.54	54.00	30.5	62.3	49.74
VROB-181	5.43	9.17	11.85	9.95	22.43	35.7	45.2	1.58	55.57	25.2	67.8	75.04
VROR-157	7.71	12.17	15.39	11.67	23.61	39.6	42.5	1.47	55.29	32.5	55	53.57
VROR-159	5.55	9.6	8.45	9.75	20.1	32.4	38.1	1.55	60.57	32.1	84.7	51.13
VROR-160	8.43	6.33	9.33	7.27	22.15	40.2	46.5	1.57	59.57	25.4	59.2	58.61
VROT-108	10.57	7.85	16.35	12.65	26.28	36.7	38.7	1.68	62.43	36.6	62	63.13
SEM	1.39	0.86	0.91	0.78	0.46	1.16	0.53	0.07	0.35	0.67	1.98	1.22
CD (P=0.05)	3.47	2.15	2.28	1.95	1.15	2.89	1.33	0.19	1.01	1.83	4.95	3.08
F-test		S	S	S	S	S	NS	S	NS	S	S	S
Correlation coefficient (r) with jassid		-0.678	-0.343	-0.507	0.493	0.723	0.378	0.458	0.056	0.491	0.499	-0.577

S, Significant; NS, Non-significant.

and jassid incidence in okra from Pakistan. Similar observations were also documented by Murugesan and Kavitha (2010) who observed significant ($P<0.05$) negative correlation between trichome density in on the ventral surface of the leaves and damage and oviposition by leaf-hopper on cotton.

Similarly, the differences in trichome density in mid ribs among different genotypes were also found to influence leaf hopper population. It was seen that the cultivars SB-6 and SB-10 harboured highest number of leaf hopper per leaf than the rest of the genotypes and number of trichomes on mid-ribs were significantly lower than the rest of the genotypes. The correlation ($r = - 0.678$) was significant and negative. Higher number of trichomes on mid-ribs might be imparting oviposition inhibitor to the jassid. Present study was in accordance with Mohankumar (1996) who reported similar findings on cotton leafhopper, *Amrasca devastans* (Distant) and also reported that susceptibility to leaf-hopper decreased as the hair bases were closer to each other.

The results pertaining to trichome distribution on leaf vein among all 10 genotypes of okra revealed that leaf veins of VROT-108 were significantly hairy (12.65 number/cm of length) as compared to other test genotypes. Negative and significant correlation ($r = - 0.507$) existed between hairs on veins and incidence of okra jassid. From the present finding it has been observed that the selection of genotypes with higher number of trichomes on leaf lamina, leaf vein and mid-rib will help to minimize the jassid infestation in okra. Sivasubramanian *et al*. (1991) and Mohankumar (1996) also reported this host physical property on the degree of jassid infestation.

Varied angle existed among mid-ribs of okra compound leaf of the 10 test genotypes and it was observed that varieties with wide angled genotypes suffered high jassid incidence. Among the test genotypes, SB-6 (17.57 jassid/ leaf) had the highest angles (59.7°) between the mid-ribs (Table 1) whereas, lowest (32.4°) was on VROR-159, harbored relatively lower jassid population. This was probably because jassid could get ample feeding space to congregate as the first and second instar nymphs feed near the bases of leaf vein. The correlation was observed between mid-rib angle and jassid incidence was significant and positive ($r = 0.723$) (Table 1). Similar observation was also noted in case of angle between the leaf vein and mid-ribs. However, leaf angle showed positive and non-significant correlation ($r = 0.056$) with jassid infestation.

The data also revealed that lengthy leaf and more number of leaves per plant were observed in varieties which suffered high jassid incidence and vice versa. The okra genotype SB-6 which showed average 17.57 jassids/leaf had the higher leaf length of 22.57 cm (Table 1). Similarly, lowest number of leaves (25.2/plant) was observed on relatively tolerant genotype VROB-181, had the lowest jassid population (5.43/ leaf). Thus the correlations between the leaf length and total leaf/plant and jassid population were positive and significant (0.493 and 0.491, respectively). The present findings corroborate with Shinde *et al.* (2014) who revealed that jassid infestation recorded positive association with total number of leaves. Similarly, leaf length also showed a significant and positive correlation ($r = 0.493$) with jassid preference. From the present findings, it is evident that the genotypes had broader leaves and dense foliage favored the jassid incidence.

From the Table 2 it is also evident that mid-rib thickness had positive and significant correlation (r value = 0.458) with the jassid incidence. Varieties (SB-10, VROT-108 and SB-6) having higher mid-rib thickness suffered more jassid incidence than the varieties had narrowly thickened mid-ribs as in case of VROR-157. Higher mid-rib thickness provided ample space for jassid egg laying which might be serving as cue for the jassid infestation. Earlier, Shinde *et al.* (2014) also documented that cotton genotypes which had lower mid-rib thickness were found less susceptible to jassid infestation while the genotypes with higher midrib thickness showed high infestation of jassid. The results obtained during present investigation are in agreement with Sharma and Kalra (1996), who reported that thickness of midrib was positively correlated with leaf hopper population.

Varied differences were observed in the plant height of different okra genotypes and it ranged from 55 (VROR-157) – 101.7 cm (SB-8). A significant and positive correlation ($r = 0.499$) was found between plant height and jassid

Table 2 Forward stepwise regression models showing effect of different leaf characters of okra genotypes on jassids density/leaf

Number of jassid/leaf	Variable entered	Model R^2	Partial R^2
$Y = -7.80442 + 0.40799X_1$	Angle between the mid ribs (X_1)	0.52	0.519
$Y = -54.65 + 0.45X_1 + 28.66X_2$	Mid rib thickness (X_2)	0.82	0.304
$Y = -53.54 + 0.43X_1 + 23.42X_2 + 0.26X_3$	Total number of leaves (X_3)	0.86	0.040
$Y = -17.77 + 0.34X_1 + 4.71X_2 + 0.36X_3 + 0.68X_4$	Trichomes on Mid rib (X_4)	0.92	0.055
$Y = -12.49 + 0.31X_1 + 1.73X_2 + 0.44X_3 + 0.59X_4 + 0.27X_5$	Trichomes on veins (X_5)	0.93	0.010
$Y = -2.63 + 0.38X_1 + 22.69X_2 + 1.06X_3 + 0.72X_4 + 1.00X_5 + 0.27X_6$	Phenol (X_6)	0.97	0.045
$Y = 6.68 + 0.37X_1 + 28.44X_2 + 1.26X_3 + 0.78X_4 + 1.43X_5 + 0.31X_6 + 0.05X_7$	Plant height (X_7)	0.989	0.015
$Y = -12.55 + 0.48X_1 - 24.44X_2 + 1.52X_3 - 0.32X_4 - 1.42X_5 + 0.43X_6 - 0.10X_7 - 0.59X_8$	Trichomes on lamina (X_8)	0.998	0.008

No other variable met the 0.500 significance level for entry into the model, so the final model is: $Y = -12.55 + 0.48X_1 - 24.44X_2 + 1.52X_3 - 0.32X_4 - 1.42X_5 + 0.43X_6 - 0.10X_7 - 0.59X_8$.

incidence among the 10 test genotypes. Shinde *et al.* (2014) reported cotton accessions taller in height (166.5 - 168 cm), harbored more jassid infestation.

Biochemical parameters also imparted resistance to jassid incidence in okra. Highest total phenol content in VROB-181 was 75.04 mg/100 g followed by VROT-108 (63.13 mg/100 g) which harboured lower jassid population whereas the highly susceptible the genotype SB-6 had lowest phenol content (42.61). Thus, correlation established between total phenol content and jassid incidence was negative and significant (r = - 0.577). Phenolics in a fairly large concentration could have direct toxicity to the insect. These findings are in close conformity with those of Hooda *et al.* (1997) who reported that resistance to okra leaf-hopper correlated with higher concentration of total phenol content in leaves. Similar findings are also noted by Halder *et al*. (2006 a, 2007), Iqbal (2011), Shinde *et al.* (2014) and Venkatesha (2014).

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