Role of physico-morphic and biochemical characters of different okra genotypes in relation to population of okra shoot and fruit borer, *Earias vittella* (Noctuidae: Lepidoptera)

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ABSTRACT

Ten promising genotypes of okra were field screened under natural infestation of shoot and fruit borer, *Earias vittella* Fabricius to assess its reaction. Ten biophysical parameters, *viz*. trichomes, fruit length, fruit width, fruit weight, fruit angle, total number of fruits and branches, leaf length, plant height, stem diameter and biochemical parameters like total phenol and anthocyanin content of fruits were studied in relation to the expression of varietal reaction towards shoot and fruit borer. It was observed that highly susceptible genotype SB 8 had relatively lower number of trichomes (24) as compared to tolerant genotype SB 6 which had 51.8 trichomes/cm². Fruit length, width and fruit weight showed a positive correlation (r value = 0.376, 0.034, 0.026, respectively) with borer incidence. Susceptible genotype SB 8 possessed lower fruit angle (23.4°) and stem thickness (1.17 cm) as compared to tolerant genotype SB 6 (26.4° and 1.28 cm, respectively). Similarly, higher number of total leaves (38.8), fruits (15.75) and branches (3.6) per plant were also recorded from SB 8 as compared to other tolerant lines (SB 6, VROR 160, SB 10). Amongst the biochemical parameters, total phenol and anthocyanin showed negative correlation with the borer incidence. Free-choice arena and ovipostional tests also confirmed that highest larval orientation and egg laying were in susceptible genotype SB 8 (53.3 and 19.7%, respectively) than the other tolerant genotypes.

Key words: Biophysical and biochemical parameters, *Earias* spp., Okra germplasm, Olfactometer test, Ovipostional test, Varietal screening.

Okra (Abelmoschus esculentus L. Moench), also known as lady's finger, is an important vegetable crop of tropics and subtropics. 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. Beside these, it is also a rich source of dietary fiber, antioxidants, ascorbic acid and folate. The crop is attacked by a variety of pests throughout its growth stages (Rai et al. 1993, Rao et al. 2002). Amongst them, okra shoot and fruit borer (Earias vittella Fabricius; Noctuidae, Lepidoptera) is of much significance (Prasad and Prasad 2004, Priya and Mishra 2007, Gautam et al. 2014) and causes extensive damage to fruits resulting in 69% yield loss (Atwal and Singh 1990). Its infestation started from the beginning of the crop growth and damage due to the borer varied from 21.33 to 43.99% in shoots and 21 to 51.3% in case of fruits (Singh et al. 2007). At present day, management of Earias spp. has

¹Scientist (e mail: jaydeep.halder@gmail.com); ²Senior Scientist (e mail: satishsanwal@rediffmail.com); ³Senior Research Fellow (e mail: ajeetiivr@gmail.com); ⁴Head and Principal Scientist (e mail: abraiiivr@gmail.com); ⁵Director (e mail: bsinghiivr@gmail.com); ⁶Scientist (e mail: bksinghkushinagar@ yahoo.co.in) largely been relied on chemical control. However, the demands for clean and ecologically sound control envisages, careful planning for rationalizing the insecticides interventions. Development of resistant varieties is an ideal component against buildup of pest population at no additional cost, compatible with other methods of pest control and free from control pollution. Various biophysical and biochemical characters of the plants play an important role by providing resistance against this pest. However, literature on role of these biophysical and biochemical parameters imparting resistance towards different okra genotypes against *E. vittella* is scanty. Therefore, the present study was undertaken to find out the role of different morphochemical properties of the plants on the incidence of the borer, if any, and the results reported herein.

MATERIALS AND METHODS

Studies on infestation of this borer pest was undertaken on ten genotypes, viz. SB 6, SB 8, SB 10, VROB 178, VROB 179, VROB 181, VROT 108, VROR 157, VROR 159 and VROR 160 at experimental farm of Indian Institute of Vegetable Research (82°52' E longitude and 25°12' N latitude), Varanasi, Uttar Pradesh, India. 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 shoot and fruit borer. Ten randomly selected plants of each row were taken at regular intervals. Ten plant parameters, viz. trichomes, fruit length, fruit diameter, fruit weight, fruit angle, number of leaves, fruits and branches per plant, leaf length, plant height and stem thickness were studied for their role in expression of varietal reaction to shoot and fruit borer. In addition to these, the content of total phenol and anthocyanin in fruits as biochemical parameters of the fruits were also evaluated.

The uniformly developed leaves of all test genotypes were collected from randomly selected plants at 60 days after sowing (DAS) and trichome density were measured and expressed in number per cm². The leaves of each line 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 ($10 \times 100X$) (Nikon SMZ-10A).

Twenty hand cut cross-sections of stems of each genotype (50 days after sowing) were taken and the stem thicknesses were measured by using a slide caliperse and expressed in centimeter. Twenty randomly selected fruits (five days after anthesis) were harvested from each test genotypes and fruit length and diameters were measured with the help of a graph paper, whereas fruits weights were recorded by using an electronic balance (Shimadzu AUX220). Similarly, twenty fully developed mature leaves of uniform age were collected and their lengths from base to apical tip were recorded.

Angle between the okra fruits (five days after anthesis) and the main stem/branch was measured by using a protractor. Total number of leaves were counted twice, i.e. 60 DAS and at physiologically matured stage; whereas total number of fruits, branches and plant heights were taken at physiological matured stage of the crop. Freechoice test for ovipositional preference of E. vittella was conducted under laboratory conditions at $28\pm2^{\circ}$ C, 70-80% relative humidity and a photoperiod of 13:11 (L:D) hour in caged conditions. Five-day old okra fruits of different genotypes were harvested, brought to the laboratory and kept in reagent bottles (Schott Duran, 50 ml capacity) filled with water in such a way that only peduncle portion of the fruits were dipped into the water inside the cage $(36 \times 30 \times$ 40 cm). Ten pairs of 4-day old pupae were also released in the cage along with sugar solution (10%) as adult diet and ovipositional preference was determined after three days in terms of number of eggs laid on each fruit.

The free-choice arena test (olfactometer test) was conducted to assess the relative preference of third instar larvae of *E. vittella* among the different okra test germplasm. The basic component of the olfactometer assay is a round plastic container "*arena*" (18.7 cm diameter \times 5.1 cm depth) and ten test tubes (2.5 cm diameter and 9.5 cm length)

whose both sides were opened and one open side were plugged with cotton. These test tubes were inserted into the plastic container maintaining equidistance with each other. Later in each test tube, one five-day old fruit of respective okra genotypes were kept. Thirty third instar larvae of *E. vittella* starved for six hours were introduced at the centre of the arena and its cover was placed in its position. At the junctions between arena and its cover and test tubes and arena were sealed with Vaseline. After 3 hours, the numbers of larvae reaching the host genotypes in each test tube were counted. Three replicates for each study were maintained and larval preference in different okra genotypes were expressed in terms of per cent.

Two biochemical parameters, viz. total phenols and anthocyanin in fresh fruits were estimated for their role in expression of varietal reaction to shoot and fruit borer. Fruit samples of each genotype (five days after anthesis) 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 were done by using Folin-Ciocalteau reagent (FC reagent) as per the procedure outlined by Malick and Singh (1980), whereas anthocyanin contents in fruits of each genotypes were estimated as per the methodology described by Mahadevan and Sridhar (1986) and expressed in terms of optical density (OD) values.

RESULTS AND DISCUSSION

Among the ten test genotypes, highest borer incidence (7.2/plant) was recorded on SB 8 followed by VROR 159 and VROB 178 (6.6 and 5.5 larvae/plant, respectively). Further, dense trichomes were observed on the leaves of SB 6 (51.8/cm²) while trichome density was lowest in VROR 159 (6.8) had relatively higher borer incidence (Table 1). A significant negative correlation was observed between trichome density and borer incidence (Table 2). High trichome density might be imparting the physical barrier for the borers rendering their non-preference over the lowtrichomes genotypes. Earlier, Sharma and Singh (2010) reported a significant negative correlation between trichome density and borer incidence in okra from Rajasthan. Similar observations were also documented by Halder et al. (2006) who observed significant (P<0.05) negative correlation between trichome density in pods and pod borer infestation and damage severity in mungbean.

The differences in fruit length among different genotypes were also found to influence fruit damage by *Earias*. It was seen that the fruit length of VROT 108 and VROR 159 were greater than the rest of the genotypes. The correlation (r = 0.348) was positive. Lengthy fruits were found more suitable for damage by *Earias* as they harbored more larvae/fruit. Present study was in accordance with Halder and Srinivasan (2011) who reported similar findings on *Maruca vitrata* infesting cowpea and also found that pod infestation was higher in genotypes that had highest pod length than those genotypes with shorter pods. The results pertaining to fruit width among all ten genotypes of

			Tab	Table 1 Biophysical an	sical and biod	nd biochemical parameters of different okra genotypes with borer incidence	neters of diff	erent okra ge	motypes wi	ith borer incide	ence			
Genotypes	Borers/	_			Biopl	Biophysical parameters	sters						Biochemic	Biochemical parameters
	plant	Trichomes (cm ²)	Fruit length (cm)	Trichomes Fruit length Fruit width Fruit (cm ²) (cm) (cm) (cm)	Fruit angle (°)	Number of fruits/plant	Ten fruit weight (g)	Number of leaves	Leaf length (cm)	Number of branches/ plant	Plant height (cm)	Stem diameter (cm)	Phenol (mg/100g)	Phenol Anthocyanin (mg/100g) (OD value)
SB 8	7.2	24	14.30	1.55	21.6	15.75	185	38.8	14	3.6	84	1.17	37.91	0.04
SB 6	3.2	51.8	14.47	1.73	26.4	13.67	140	20	10.2	1.4	101.67	1.28	34.78	0.047
SB 10	4	44.6	13	1.75	29.6	14.2	165	25.2	11.4	2	92.6	1.4	33.5	0.040
VROB 178	5.5	39	13.57	1.47	23.6	17.17	205	30.8	12	3.8	55.33	1.17	48.17	0.036
VROB 179	4.8	45.4	12.63	1.57	29.4	15.33	180	36.8	13	2	62.33	1.13	44.33	0.035
VROB 181	4.5	20.6	13.33	1.73	25.2	14.67	190	21.6	8	1.4	67.83	1.67	46.2	0.038
VROT 108	5.4	23.4	16.47	1.37	28	17.55	156	36.4	9.8	1.8	55	1.13	35.75	0.075
VROR 157	4.6	26.2	13.1	1.47	21.2	13.33	160	40.2	10	1.2	84.67	1.03	62.26	0.030
VROR 159	6.6	6.8	14.67	1.63	22	19.17	126	43.4	11.2	2	59.17	1.17	34.78	0.037
VROR 160	3.7	38.5	13.33	1.6	26.6	14.33	175	25.4	9.8	1	62	1.2	48.35	0.054
SEm (±)	0.56	2.47	0.31	0.10	0.59	0.49	1.84	1.50	0.463	0.57	2.001	0.163	1.37	0.064
CD (P=0.05)	1.47	6.50	0.82	0.26	1.55	1.29	4.84	3.95	1.22	1.50	5.26	0.43	3.60	0.17
F-test		S	NS	NS	S	S	NS	S	S	S	NS	NS	NS	NS
S = Significant; NS = Non-significant	int; NS	= Non-signi	ficant											

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okra revealed that fruits of SB 10 were significantly broader as compared to other test genotypes. Negative and significant correlation (r = -0.429) existed between fruit width and incidence of okra shoot and fruit borer. Kamashi and Srinivasan (2008) also reported this host physical property on the degree of infestation.

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Varied fruit angle existed among the test genotypes and it was observed that varieties with narrow angled genotypes suffered high fruit damage. Among the test genotypes of okra, SB 8 (7.2 larvae/plant) had the angles between fruits was only 21.6° (Table 1). This was probably because larvae unable to adequately conceal themselves easily fall prey to parasites, predators and desiccation in wide angle. In addition, considerable amount of energy was also expended in migrating from one fruit to another (Halder and Srinivasan 2005). The correlation was observed between fruit angle and borer incidence was significant and negative (-0.568) (Table 2). According to Oghiakhe *et al.* (1992) damage to the susceptible genotype, IT 82D-176 was significantly reduced by widening the pod angle.

The data also revealed that high fruit weight and higher number of fruits/plant were observed in varieties which suffered high fruit damage and vice versa. The okra genotype SB 6 which showed only 3.2 borers/plant had the lower fruit weight of 140 g /ten fruits (Table 1). Similarly, lowest number of leaves (20/plant) and fewer branches (1.4 /plant) and lower leaf length (10.2 cm) were observed on relatively tolerant genotype SB 6, had the lowest borer damage (3.2 larvae/plant). The genotype SB 6 also measured as tallest genotype (101.67 cm) amongst the all ten test genotypes and established a weak and negative correlation for plant height and borer incidence. The present findings corroborate with Balakrishnan and Sreenivasan (2010) who revealed that borer infestation recorded negative association with plant height and suggested that selection of genotypes with reduced growth habit and minimum fruit length will help to minimize the shoot and fruit borer infestation. Similarly, stem diameter also showed a weak and negative correlation (r = -0.230) with borer preference. From the present findings, it is evident that the genotypes had dense foliage and bushy architecture favored the borer incidence.

From the Table 1 it is also evident that number leaves per plant, leaf length and number of branches had positive and significant correlation (r value = 0.346, 0.448 and 0.572, respectively) with the borer incidence. Varieties (VROR 159, VROT 108) having higher number of leaves and more leaf length suffered more borer incidence than the varieties had fewer leaves, narrow leaf length and less number of branches as in case of SB 6. More number of broad leaves and higher branches imparting more greenery and bushy appearance of the plants which might be serving as cue for the borer.

Varied differences were observed in the stem diameter of different okra genotypes and it ranged from 1.03 (VROR 157) – 1.67 cm (VROB 181). A negative and weak correlation (r = -0.311) was found between stem diameter and shoot and fruit borer incidence among the ten test

(unucles	Correlatio coefficien	n negression
Trichome (x) vs Borer (y)	-0.692	Y = 6.939 - 0.062X
Fruit angle (x) vs Borer (y)	-0.568	Y = 10.675 - 0.226X
Number of fruits / plant (x) vs. Borer (y)	0.725	Y = -2.492 + 0.479X
Number of leaves (x) vs Borer (y) 0.765	Y = 1.292 + 0.115X
Leaf length (x) vs Borer (y)	0.556	Y = 0.599 + 0.397X
Number of branches (x) vs Borer (y)	0.682	Y = 3.139 + 0.897X

Table 2Correlation studies of different variables on fruit damage
due to *E. vittella* on okra

genotypes. Thick stems might be reducing the tenderness and there by restricting the borer preference.

Biochemical parameters also imparted resistance to borer incidence in okra. Highest total phenol content in VROR 157 was 62.26 mg/100 g followed by VROR 160 (48.35) which haboured lower borer population whereas the highly susceptible the genotype SB 8 had relatively lower phenol content (40.43). Thus, correlation established between total phenol content and borer incidence was negative and non-significant (Table 2). Phenolics in a fairly large concentration could have direct toxicity to the insect. These findings are in close conformity with those of Sharma and Singh (2010) who reported that resistance to fruit borer may be correlated with higher amount phenol content in fruits. Similar findings are also noted by Halder *et al.* (2006), Halder and Srinivasan (2007) and Jat and Pareek (2003).

The role of water soluble pigment 'Anthocyanin' on borer incidence was also worked out and a negative correlation (r value = -0.109) was established. The genotype SB 8 suffered highest borer infestation (8.2 larvae / plant) had relatively lower anthocyanin pigmentation (0.04 ODvalue). Our present findings are in conformity with the observations of Musmeci *et al.* (2005) and Karageorgou and Manetas (2006).

Olfactometer test (free-choice arena test) with fiveday old fruit of respective okra genotypes also revealed that

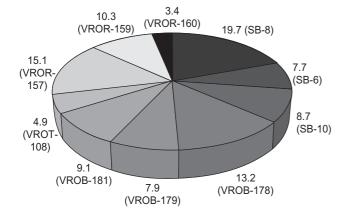


Fig 1 Olfactometer test

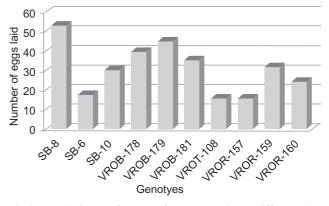


Fig 2 Egg-laying preference of *Earias* against different okra germplasms

highest population of larvae reached to SB 8 (19.7%) whereas least number was on VROR 160 (3.4%) (Fig 1). Ovipositional preference of gravid *Earias* females indicated their preference of egg laying towards the genotype SB-8 (53.3) followed by VROB 178 (39.3) and lowest of only 16.1 eggs on VROR 157 (Fig 2). Earlier Gautam *et al.* (2014) field screened one hundred germplasm of okra and classified the SB 8 as susceptible against shoot and fruit borer which is close proximity with our present findings.

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