Inheritance of trichomes and resistance to pod borer (*Helicoverpa armigera*) and their association in interspecific crosses between cultivated pigeonpea (*Cajanus cajan*) and its wild relative *C. scarabaeoides*

Rupakula Aruna[1,2,*], D. Manohar Rao[2], L.J. Reddy[1], Hari D. Upadhyaya[1] & Hari C. Sharma[1]
[1]*International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru 502 324, Andhra Pradesh, India;* [2]*Department of Genetics, Osmania University, Hyderabad 500 007, Andhra Pradesh, India*
(*author for correspondence: e-mail: a.rupakula@cgiar.org)

Summary

The legume pod borer, *Helicoverpa armigera*, is one of the most devastating pests of pigeonpea. High levels of resistance to pod borer have been reported in the wild relative of pigeonpea, *Cajanus scarabaeoides*. Trichomes (their type, orientation, density and length) and their exudates on pod wall surface play an important role in the ovipositional behavior and host selection process of insect herbivores. They have been widely exploited as an insect defense mechanism in number of crops. In the present investigation, inheritance of resistance to pod borer and different types of trichomes (A, B, C and D) on the pod wall surface in the parents (*C. cajan* and *C. scarabaeoides*) and their F_1, F_2, BC_1 (*C. cajan* × F_1), and F_3 generations has been studied. Trichomes of the wild parents (high density of the non-glandular trichomes C and D, and glandular trichome B and low density of glandular trichome A) were dominant over the trichome features of *C. cajan*. A single dominant gene as indicated by the segregation patterns individually will govern each trait in the F_2 and backcross generation. Segregation ratio of 3 (resistant): 1 (susceptible) for resistance to pod borer in the F_2 generation under field conditions was corroborated with a ratio of 1:1 in the backcross generation, and the ratio of 1 non-segregating (resistant): 2 segregating (3 resistant: 1 susceptible): 1 non-segregating (susceptible) in F_3 generation. Similar results were obtained for pod borer resistance under no-choice conditions. Resistance to pod borer and trichomes associated with it (low density of type A trichome and high density of type C) are governed individually by a dominant allele of a single gene in *C. scarabaeoides*. Following backcrossing, these traits can be transferred from *C. scarabaeoides* into the cultivated background.

Introduction

Pigeonpea (*Cajanus cajan* (L.) Millspaugh), an important legume crop, is cultivated on about 4.8 million ha of dry lands in south Asia, Africa and Latin America. The traditional long-duration cultivars, with insect management are capable of producing over $3\,\mathrm{Mg\,ha^{-1}}$ grain yields but the realized yields are as low as $0.7\,\mathrm{Mg\,ha^{-1}}$ which vary across locations and seasons (Saxena, 2000). One of the main reasons for low and unstable yields is the damage caused by a number of insect pests, of which the pod borer, *Helicoverpa*

armigera, is the most devastating, causing losses to the tune of more than US $ 300 million annually (ICRISAT, 1992). More than 14,000 pigeonpea germplam accessions have been screened to identify the sources of resistance to *H. armigera*, but only a few accessions with low levels of resistance have been detected (Lateef, 1992; Sachan, 1992). Wild relatives of *Cajanus cajan*, especially *C. scarabaeoides*, have been identified as a potential source of resistance to pod borer (Pundir & Singh, 1987; Saxena et al., 1990; Shanower et al., 1997). The wild species have antixenosis, antibiosis and non-preferential oviposition among the

mechanisms of resistance against pod borer (Sharma et al., 2001). Attempts have also been made to exploit wild relatives as sources of resistance to pod borer, pod wasp and *Phytophthora* blight in pigeonpea (Reddy et al., 1981; Saxena et al., 1996; Sharma et al., 2001).

Trichomes and trichome exudates on the pod wall surface play an important role in the ovipositional behavior and host selection process of insect herbivores (Bernays & Champman, 1994). Trichomes have been widely exploited as an insect defense mechanism in a number of crop plants including *Leonotis* (Ascensao et al., 1995), *Brassica rapa* (Kole, 1996), potato (Bacchetta et al., 1993; Lai et al., 2000), soybean (Lam & Pedigo, 2001), alfalfa (Ranger & Hower, 2001), *Arabidopsis* (Karkkainen & Agren, 2002) and tomato (Simmons et al., 2004; Simmons & Geoff, 2004). The types of trichomes, their orientation, density and length have been correlated with reduced insect damage in several crops (Levin, 1973; Webster et al., 1975; Jeffree, 1986; Peter et al., 1995; David & Easwaramoorthy, 1998). Trichomes minimize the insect loads and/or damage by repellent activity of the exudates, limiting contact with the surface of the plant, physical and/or chemical entrapment, increasing exposure time to biotic and abiotic agents, inhibiting larval growth and deterring oviposition (Webster et al., 1975; Stipanovic, 1983; Peter et al., 1995). Mortality of arthopod pests resulting from glandular trichomes has been hypothesized to be a result of physical entrapment (Muigai et al., 2002) as opposed to the toxic effects of compounds produced by the trichomes (Kennedy, 2003).

Trichomes could, therefore, provide a potential resistance mechanism against *H. armigera* and other insect pests of pigeonpea. Bisen and Sheldrahe (1981), and Navasero and Ramaswamy (1991) studied the trichomes in *C. cajan* while Romies et al. (1999) studied different types of trichomes in *C. cajan*, *C. scarabaeoides* and *C. platycarpus.* Four morphologically distinct types of trichomes (A, B, C and D) were identified on the pods of *C. scarabaeoides* accessions and pigeonpea genotypes by light and scanning electron microscopy (Romies et al., 1999). The high density of erect, non-glandular trichomes (types C and D), predominantly on the pods of wild *C. scarabaeoides* accessions, confers high level of resistance against pod borer (Shanower et al., 1997; Romies et al., 1999; Sharma et al., 2001). There is no information on the inheritance of different types of trichomes in *Cajanus*, except for a report on the inheritance of pubescence on the pods

of *Cajanus* (Reddy et al., 1981; Pundir & Singh, 1985; Singh et al., 2000). There are no studies on the inheritance of resistance against pod borer except for a report on the antixenosis mechanism of resistance by Verulkar et al. (1997). Knowledge of the inheritance of resistance to pod borer and different types of trichomes on the pod surface will enhance their utilization in breeding for resistance against pests in pigeonpea. Keeping this in view, an attempt was made to study the inheritance of different types of trichomes and resistance against *H. armigera* in the interspecific crosses of *C. cajan* and *C. scarabaeoides*.

Materials and methods

Three accessions of *C. scarabaeoides*, ICPW 94, ICPW 125 and ICPW 130, resistant to *H.armigera* (Sharma et al., 2001), and two susceptible cultivated types; ICP 26 (T-21) and ICP 28 (Pusa Ageti), were used in the study. Three crosses between the wild, as male, and the cultivated, as female, parents were made during the 2000 rainy season. The F_1 hybrids of these crosses were used to raise F_2 and F_3 generations besides producing BC_1 plants by crossing the F_1s to *C. cajan* parents. Two rows each of parents, F_1s (ICP 28 × ICPW 94, ICP 28 × ICPW 130 and ICP 26 × ICPW 125), 25 rows of 96-F_2 single plants and eight rows of BC_1 plants were raised in the field (alfisols) at ICRISAT, Patancheru, Andhra Pradesh, India (17°N and 78°E, 545 m above the mean sea level) during the 2002 rainy season. Plants selected for recording observations on trichome types and their densities were protected through appropriate measures from insect damage. However, none of these measures was taken for the plants in the field meant for screening for resistance against pod borer. Di-ammonium phosphate ($100 \, Kg \, ha^{-1}$) was applied to the experimental field and the recommended package of practices was followed to raise the crop. Plants were raised in 4-m long rows, with intra row spacing of 30 cm and inter-row spacing of 75 cm. Similarly, the F_3 population was grown in the next season.

Trichome types and their density

Of the three crosses made, only two (ICP 28 × ICPW 94 and ICP 26 × ICPW 125) were used to study the inheritance of trichomes. Randomly selected 20 plants from each parent and their F_1s, 250 plants of F_2 and 75 plants of BC_1 populations were evaluated for the type and density of trichomes. Fully developed green pods

were observed under a light microscope with $100\times$ magnification. The pods were also scanned under the scanning electron microscope (Phillips XL 30) to visualize the trichomes, using the methodology described by Reddy et al. (1995). Four different types of trichomes were identified as Trichomes A, B, C and D (Figures 1 and 2). Glandular trichomes; type A has a long tubular segmented neck from which viscous liquid is secreted, while the trichome type B is unsegmented globular sac. Non-glandular trichomes were separated into short, type C and long, type D. Type D was 10 times longer than type C. Trichome counts were taken under a light microscope with an ocular measuring grid. The differences among the trichomes were counted in an area of $4.84 \, mm^2$ (Types A, B and D) or $1.21 \, mm^2$ (Type C) (Romies et al., 1999). In the absence of previous information on the density classification, a scale was worked out in consultation with entomologists and cell biologists. Density of A, B and D trichome types was classified as high when the counts (in $1 \, mm^2$) exceeded 20 and as low when they were 20 and below. In the case of C type, the counts exceeding 50 were classified as high density while 50 and below as low density. For data analysis, means of five pods per plant, at three microscopic fields, were used. χ^2 test was applied to test the goodness of fit of the observed ratio for trichome density (high: low) in F_2 and BC_1 generations (Panse & Sukhatme, 1967).

Pod borer resistance

Plants from all the three crosses were screened for pod borer resistance under multi-choice and no-choice conditions in the laboratory (Sharma et al., 2001). During the 2001 rainy season, one row each of the parents and their F_1s, 20 rows of F_2s of each cross and five rows of BC_1 were grown. Crop was maintained without any insecticidal spray during the reproductive phase and the plants were exposed to natural infestation. Similarly, during the 2002 rainy season 125, 116 and 109 F_3 families were screened in crosses, ICP 28 × ICPW 94, ICP 28 × ICPW 130 and ICP 26 × ICPW 125, respectively. Two inflorescences of 25–30 cm length per plant were tagged on the 15th day after flowering with plastic ribbons. Observations on total number of buds, flowers, and pods present and the number of buds, flowers, and pods infected by pod borer, per inflorescence, were recorded on 5th, 7th, 11th, 21st and the 31st day of tagging. The average of percentage damage was used in correlation studies. The material was also evaluated visually for *H. armigera* pod damage on a 1–5 scale (Sharma et al., 2001) (1 = plants with no
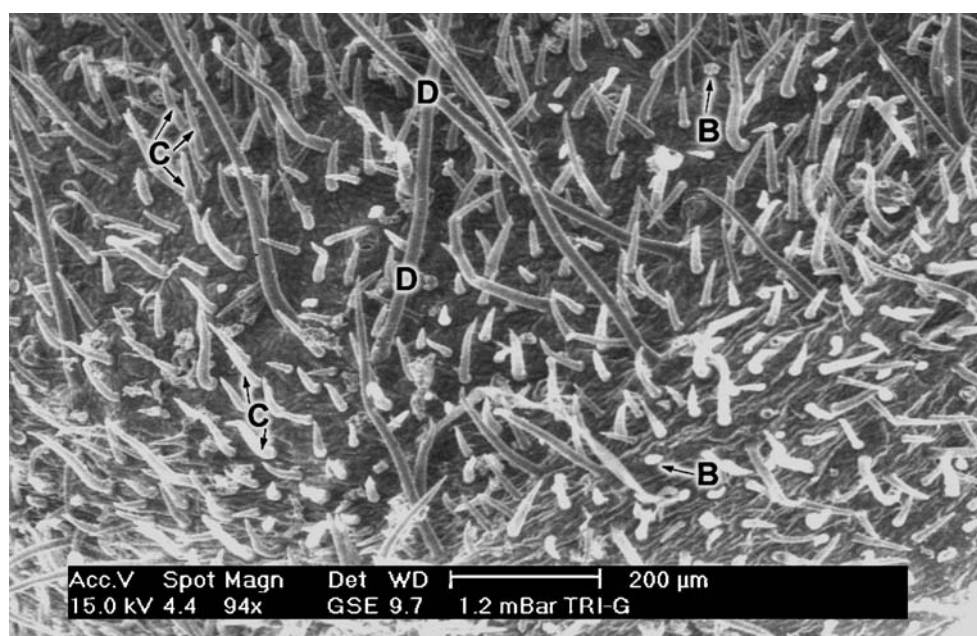

Figure 1. Different types of trichomes on the podwall of *C. scarabaeoides* accession ICPW 94; B: trichome type B (unsegmented, globular sac), C: trichome type C (short, non-glandular) and D: trichome type D (long, non-glandular).
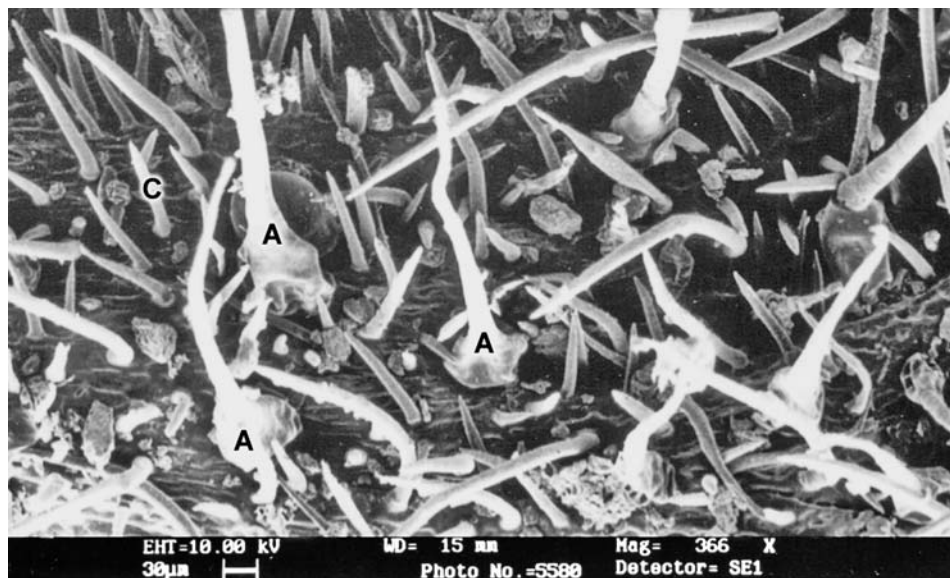
Figure 2. Different types of trichomes on the podwall of *C. cajan* genotype ICP 28. A: trichome type A (glandular, long) and C: trichome type C (short, non-glandular).

damage to the pods and no eggs or larvae on inflorescences, $2 = \leq10\%$ damage with no eggs and larvae on inflorescences, $3 = 11$ to $\leq20\%$ damage to pods with a minimum of two egg masses and one larvae, $4 = 21$ to $\leq 40\%$ damage to pods with more than three egg masses and two larvae and $5 = \geq41\%$ damage to pods with more than five egg masses and five larvae). Plants with a score of ≤2 were rated as resistant and >2 were rated as susceptible. χ^2 test was used to test the goodness of fit of the observed ratio of segregation in the F_2 and BC_1 generations in all the three crosses. The results were further confirmed by analyzing the data on F_3 families following a similar approach.

Fully developed green pods, of only one cross (ICP 28 × ICPW 94), were screened in the laboratory to study the antibiosis mechanism of resistance against pod borer. Pods from 20 plants each of the two parents (ICP 28 and ICPW 94), 10 plants of F_1, 250 plants of F_2 and 70 plants of BC_1 population were collected in polythene bags in the morning and brought to laboratory for screening. *H. armigera* larvae were reared on chickpea diet at 27 °C (Armes et al., 1996). Pods were screened under no-choice conditions, where in a third instar larva was released on each pod (Sharma et al., 2001). Experiment was repeated three times for each plant, with five pods per plant.

Pods without any damage and the dead insects were given a rating of 1, whereas those with >10% increase in body weight of insects without mortality were given the damage rating of 9 (Sharma et al., 2001). The others were rated from 2 to 8 depending on the proportionate damage caused to the pods. The plants with pods rated between 1–3 were clustered as the resistant types, while those from 4–9 were categorized as susceptible. The χ^2 test was applied to test the goodness of fit of the observed ratio of resistant: susceptible to the expected ratio of segregation for the damage rating in the F_2 and backcross generations. Correlations between the density of each type of trichome and resistance to pod borer were computed for the F_2 population.

Results and discussion

Type A trichomes are present on *C. cajan* pods but are absent on the pods of *C. scarabaeoides*. The density of other three types of trichomes (B, C and D) was higher on *C. scarabaeoides* pods than on the *C. cajan* pods (Table 1). Significant positive correlation was observed between the density of trichome A, which is longer than type B (both glandular types), and flower and pod damage in both the crosses (Table 2). The type B trichome appears to give no protection against *H. armigera* damage. However, they are the source of characteristic pigeonpea fragrance (Bisen & Sheldrahe, 1981). The high density of type A trichomes increases

Table 1. Trichome density (number mm^{-2}) in parents, F$_1$s and segregating generations of the interspecific crosses between *C. scarabaeoides* (ICPW 94 and ICPW 125) and *C. cajan* (ICP 28 and ICP 26) under field conditions, ICRISAT, Patancheru, rainy season 2002

Test material	Trichome A	Trichome B	Trichome C	Trichome D
ICP 28	5.69 ± 1.000[*]	2.42 ± 0.542	31.76 ± 1.471	2.74 ± 0.542
ICPW 94	0.00 ± 0.000	6.49 ± 0.501	174.20 ± 10.291	10.21 ± 1.132
F$_1$ (ICP 28 × ICPW 94)	0.90 ± 0.639	4.07 ± 0.623	163.35 ± 13.472	9.61 ± 0.962
F$_2$ (ICP 28 × ICPW 94)	0.87 ± 1.031	3.56 ± 1.812	29.71 ± 62.503	8.73 ± 3.341
BC$_1$ (ICP 28 × F$_1$)	2.09 ± 1.700	3.00 ± 2.673	94.54 ± 64.781	5.01 ± 2.941
ICP 26	4.39 ± 0.641	0.81 ± 0.112	25.54 ± 1.124	2.26 ± 0.593
ICPW 125	0.00 ± 0.000	7.57 ± 0.824	157.23 ± 6.192	5.85 ± 0. 593
F$_1$ (ICP 26 × ICPW 125)	0.66 ± 0.105	7.69 ± 0.761	155.57 ± 2.742	5.83 ± 0.432
F$_2$ (ICP 26 × ICPW 125)	1.32 ±1.128	5.26 ± 2.812	111.17 ± 54.791	3.31 ± 1.708
BC$_1$ (ICP 26 × F$_1$)	2.79 ± 1.433	3.09 ± 2.730	104.76 ± 61.209	5.17 ± 3.001

[*]Density measurements based on mean observations on five pods.

Table 2. Correlations between the densities (number mm^{-2}) of different trichome types with percent damage in F$_2$ populations of the interspecific crosses, ICRISAT, Patancheru, rainy season, 2002

		Bud damage	Flower damage	Pod damage	No. of eggs	No. of larvae
Trichome A	Cross 1	0.12	0.36**	0.25**	0.09	0.16*
	Cross 2	0.19**	0.34**	0.45**	0.06	0.14*
Trichome B	Cross 1	0.05	0.02	0.05	0.06	0.09
	Cross 2	0.06	0.02	0.03	0.02	0.07
Trichome C	Cross 1	−0.51**	−0.53**	−0.22**	−0.42**	−0.15**
	Cross 2	−0.49**	−0.35**	−0.74**	−0.21**	−0.09
Trichome D	Cross 1	−0.19**	−0.06	−0.10	−0.06	−0.04
	Cross 2	−0.07	−0.28**	−0.17*	−0.05	−0.19*

**,*Significant at $P = 0.05$ and 0.01, respectively; Cross 1 = ICP 28 × ICPW 94; Cross 2 = ICP 26 × ICPW 125.

the vulnerability of the host plant to *H. armigera* to feed on the pod as indicated by high damage observed in the present study in the case of *C. cajan*. There is an indication that the exudates from type A trichome contain a feeding stimulant for *H. armigera* larvae (Shanower et al., 1997; Green et al., 2003). However, in *L. esculentum*, *L. hirsutum* and *L. pennellii* a highly significant positive relationship between mortality of tomato pests and higher density of glandular trichome type IV is reported (Simmons et al., 2003; Simmons & Geoff, 2004).

In contrast to the above, significant negative correlations were observed between the density of trichome C, a non-glandular type, with damage to buds, flowers and pods, number of eggs and larvae on the inflorescences (Table 2). The density of type C trichomes on *C. scararbaeoides* was 5 times more than that of *C. cajan* (Table 1). Shanower et al., 1997 reported significant mortalility of small larvae on the pods of

C. scarabaeoides compared to the *C. cajan* pods. Although, there were a few significant negative correlations observed between type D trichomes and different sites of pod borer damage, their magnitude was low to have any significant contribution on pod borer damage resistance (Table 2). It appears that the higher densities of the non-glandular type C trichomes impart resistance to pod borer damage. Selection of plants bearing pods with higher density of type C trichome and lower density of type A trichomes will be useful in breeding for resistance to pod borer.

Inheritance of trichomes A, B, C and D

Density of type A trichomes of F$_1$s of both the crosses; ICP 28 × ICPW 94 (Table 3) and ICP 26 × ICWP 125 (Table 4), was very low, very similar to *C. scarabaeoides*. The F$_2$ plants in both the crosses segregated into 3 (low density): 1 (high density) ratio

Table 3. χ^2 test for segregation for trichome types in F_2 and backcross populations of ICP 28 × ICPW 94 under field conditions, ICRISAT, Patancheru, rainy season 2002

	Population size	No. of plants		Excepted ratio	χ^2 value	P-value
		Low**	High**			
Trichome A						
ICP 28	10	–	10	–	–	–
ICPW 94*	10	–	–	–	–	–
F_1 (ICP 28 × ICPW 94)	10	10		–	–	–
F_2 (ICP 28 × ICPW 94)	250	186	64	3:1	0.033	0.75–0.90
BC_1 (ICP 28 × F_1)	75	41	34	1:1	0.654	0.25–0.50
Trichome B						
ICP 28	10	10		–	–	–
ICPW 94	10	–	10	–	–	–
F_1 (ICP 28 × ICPW 94)	10		10	–	–	–
F_2 (ICP 28 × ICPW 94)	250	70	180	1:3	1.181	0.10–0.25
BC_1 (ICP 28 × F_1)	75	41	34	1:1	0.654	0.25–0.50
Trichome C						
ICP 28	10	10	–	–	–	–
ICPW 94	10	–	10	–	–	–
F_1 (ICP 28 × ICPW 94)	10	–	10	–	–	–
F_2 (ICP 28 × ICPW 94)	250	71	179	1:3	1.204	0.25–0.50
BC_1 (ICP 28 × F_1)	75	40	35	1:1	0.123	0.50–0.75
Trichome D						
ICP 28	10	10	–	–	–	–
ICPW 94	10	–	10	–	–	–
F_1 (ICP 28 × ICPW 94)	10	–	10	–	–	–
F_2 (ICP 28 × ICPW 94)	250	69	181	1:3	0.901	0.25–0.50
BC_1 (ICP 28 × F_1)	75	39	36	1:1	0.121	0.50–0.70

*There were no type A trichomes on the pod wall of *C. scarabaeoides* accession ICPW 94.
**Density of A, B and D trichome types was classified as high when the counts exceeded $20 \, \text{mm}^{-2}$ and as low when they were 20 and below. The corresponding values for C type were above $50 \, \text{mm}^{-2}$ and $50 \, \text{mm}^{-2}$ and below, respectively.

indicating that the low density of trichome A was governed by a single dominant gene. Further, the BC_1 plants, in both the crosses, segregated into 1 (low density): 1 (high density) ratio confirming the single gene control of density of type A trichomes (Tables 3 and 4). The density of type C trichome of F_1s of both the interspecific crosses was similar to that of type C trichome in *C. scarabaeoides* suggesting the dominance of high density of this trichome type (Tables 3 and 4). Further, the density of the C type trichomes on the pods of F_2 and BC_1 plants in both the crosses gave a good fit of 1 (low density):3 (high density) and 1 (low density):1 (high density), respectively, confirming the monogenic inheritance of C trichome also. Trichome types B and D, though not involved in protection against pod borer damage, had the similar inheritance pattern as that of type C. The test of homogeneity indicated similar seg-

regation patterns in all the F_2 and backcross populations for all the four trichome types in both the crosses (Table 5).

The two types of trichomes, A (lower density) and C (higher density), are associated in giving protection against pod borer damage. They are also negatively associated with each other (−0.49** to −0.55**). Both are governed individually by a single dominant gene. It would be interesting to study the relationship between these two genes and bring them together for higher level of protection against pod borer damage.

Inheritance of resistance to pod borer

Identification and transfer of gene(s) for pod borer resistance from wild accessions to the cultivated background is one of the major steps to create an inbuilt

Table 4. χ^2 test for segregation for trichome types in F$_2$ and backcross populations of ICP 26 × ICPW 125, under field conditions, ICRISAT, Patancheru, rainy season 2002

| | | No. of plants | | | | |
	Population size	Low**	High**	Excepted ratio	χ^2 value	*P*-value
Trichome A						
ICP 26	10	–	10	–	–	–
ICPW 125*	10	–	–	–	–	–
F$_1$ (ICP 26 × ICPW 125)	10	10		–	–	–
F$_2$(ICP 26 × ICPW 125)	250	183	67	3:1	0.432	0.50–0.75
BC$_1$(ICP 26 × F$_1$)	75	35	40	1:1	0.334	0.50–0.75
Trichome B						
ICP 26	10	10	–	–	–	–
ICPW 125	10	–	10	–	–	–
F$_1$(ICP 26 × ICPW 125)	10		10	–	–	–
F$_2$(ICP 26 × ICPW 125)	250	60	190	1:3	0.133	0.50–0.75
BC$_1$(ICP 26 × F$_1$)	75	34	41	1:1	0.654	0.25–0.50
Trichome C						
ICP 28	10	10	–	–	–	–
ICPW 125	10	–	10	–	–	–
F$_1$(ICP 26 × ICPW 125)	10	–	10	–	–	–
F$_2$(ICP 26 × ICPW 125)	250	73	177	1:3	2.352	0.10–0.25
BC$_1$(ICP 26 × F$_1$)	75	38	37	1:1	0.012	0.90–0.95
Trichome D						
ICP 26	10	10	–	–	–	–
ICPW 125	10	–	10	–	–	–
F$_1$(ICP 26 × ICPW 125)	10	–	10	–	–	–
F$_2$(ICP 26 × ICPW 125)	250	91	159	1:3	17.328	–
BC$_1$ (ICP 26 × F$_1$)	75	41	34	1:1	0.654	0.25–0.50

*There were no type A trichomes on the pod wall of *C. scarabaeoides* accession ICPW 125.

**Trichome density of A, B and D trichome types was classified as high when the counts exceeded $20\,\mathrm{mm}^{-2}$ and as low when they were 20 and below. The corresponding values for C type were above $50\,\mathrm{mm}^{-2}$ and 50 and below, respectively.

defense mechanism in the plants to control the damage of this devastating pest. The three accessions of *C. scarabaeoides* (ICPW 94, ICPW 125 and ICPW 130) used as parents did not exhibit any pod damage in the field and were rated as 1, whereas, the cultivated, ICP 28 and ICP 26, parents showed 67.65% and 65.45% pod damage, respectively, and were rated as 5. Plants with ≤2 rating were classified as resistant and those ≥2 rating as susceptible. The F$_1$ plants from these crosses exhibited a mean pod damage rating of 1.5 ± 0.01 in ICP 28 × ICPW 94, 1.6 ± 0.03 in ICP 28 × ICPW 130 and 1.8 ± 0.01 in ICP 26 × ICPW 125 and were classified as resistant. These results indicated that the resistance to pod borer was a dominant character.

The F$_2$ population of all the three crosses gave a good fit for 3 (resistant):1 (susceptible) ratio, indicating a single dominant gene responsible for resistance to pod borer (Table 6). These results were further supported by the segregation ratios in backcross generation with *C. cajan* with a good fit for 1 (resistant):1 (susceptible) ratio in all the three crosses. In F$_3$ generation also, the segregation pattern was consistent with a monogenic segregation in terms of number of progenies which bred true and those which segregated for 1 non-segregating resistant: 2 segregating:1 non-segregating susceptible in all the three crosses (Table 6). Further, the segregating F$_3$ progenies, showed similar segregation patterns of 3 resistant:1 susceptible ratio in all the three crosses. On the whole, the data confirmed the hypothesis that a single dominant gene was involved in imparting resistance to pod borer under field conditions. However, the allelic relationships for pod borer's non-preference to

Table 5. χ^2 test of heterogeneity in F_2 and backcross populations for trichome density in interspecific crosses, ICRISAT, Patancheru, rainy season 2002

Cross	Generation	Total number of pods	No. of plants		Segregation ratio	χ^2 value	P-value
			Low*	High*			
Trichome A							
ICP 28 × ICPW 94	F_2	250	183	67	3:1	0.432	0.50–0.75
ICP 26 × ICPW 125	F_2	250	186	64	3:1	0.033	0.50–0.75
Total		500	369	131	3:1	0.384	0.50–0.75
χ^2 Heterogeneity = 0.015 P value = 0.90–0.95							
ICP 28 × ICPW 94	BC_1	75	35	40	1:1	0.334	0.50–0.75
ICP 26 × ICPW 125	BC_1	75	41	34	1:1	0.654	0.25–0.50
Total		150	76	74	1:1	0.027	0.75–0.90
χ^2 Heterogeneity = 0.961 P value = 0.25–0.50							
Trichome B							
ICP 28 × ICPW 94	F_2	250	70	180	1:3	1.181	0.10–0.50
ICP 26 × ICPW 125	F_2	250	60	190	1:3	0.133	0.50–0.75
Total		500	130	370	1:3	0.266	0.50–0.75
χ^2 Heterogeneity = 1.048 P value = 0.25–0.50							
ICP 28 × ICPW 94	BC_1	75	41	34	1:1	0.654	0.25–0.50
ICP 26 × ICPW 125	BC_1	75	34	41	1:1	0.654	0.25–0.50
Total		150	75	75	1:1	0.000	0.995–1.00
χ^2 Heterogeneity = 1.308 P value = 0.25–0.50							
Trichome C							
ICP 28 × ICPW 94	F_2	250	69	181	1:3	1.204	0.25–0.50
ICP 26 × ICPW 125	F_2	250	73	177	1:3	2.352	0.10–0.25
Total		500	142	358	1:3	3.083	0.05–0.10
χ^2 Heterogeneity = 0.473 P value = 0.25–0.50							
ICP 28 × ICPW 94	BC_1	75	39	36	1:1	0.123	0.50–0.75
ICP 26 × ICPW 125	BC_1	75	38	37	1:1	0.012	0.90–0.95
Total		150	77	73	1:1	0.104	0.50–0.75
χ^2 Heterogeneity = 0.031 P value = 0.75–0.90							
Trichome D							
ICP 28 × ICPW 94	F_2	250	69	181	1:3	0.901	0.25–0.50
ICP 26 × ICPW 125	F_2	250	91	159	1:3	17.328	<0.005
Total		500	160	340	1:3	13.067	<0.005
χ^2 Heterogeneity = 5.162 P value = 0.010–0.025							
ICP 28 × ICPW 94	BC_1	75	39	36	1:1	0.121	0.50–0.70
ICP 26 × ICPW 125	BC_1	75	41	34	1:1	0.654	0.25–0.50
Total		150	80	70	1:1	0.625	0.50–0.75
χ^2 Heterogeneity = 0.15 P value = 0.50–0.75							

*Trichome density of A, B and D trichome types was classified as high when the counts exceeded 20 mm^{-2} and as low when they were 20 and below. The corresponding values for C type were above 50 mm^{-2} and 50 mm^{-2} and below, respectively.

Table 6. Reaction of parents and different generations to pod borer in three interspecific crosses between *C. scarabaeoides* (ICPW 94, ICPW 130 and ICPW 125) and cultivated pigeonpea (ICP 28 and ICP 26) under field conditions, ICRISAT, Patancheru, 2002

Cross	Population	No. of plants		Expected ratio	χ^2 value	*P*-value
		Resistant*	Susceptible*			
ICP 28	10	–	10	–	–	–
ICPW 94	10	10	–	–	–	–
F_1 (ICP 28 × ICPW 94)	10	10	–	–	–	–
F_2 (ICP 28 × ICPW 94)	250	185	65	3:1	0.133	0.50–0.75
BC_1 (ICP 28 × F_1)	112	53	59	1:1	0.322	0.50–0.75
ICP 28	10	–	10	–	–	–
ICPW 130	10	10	–	–	–	–
F_1 (ICP 28 × ICPW 130)	10	10	–	–	–	–
F_2 (ICP 28 × ICPW 130)	250	183	67	3:1	0.432	0.50–0.75
BC_1 (ICP 28 × F_1)	106	51	55	1:1	0.151	0.50–0.75
ICP 26	10	–	10	–	–	–
ICPW 125	10	10	–	–	–	–
F_1 (ICP 26 × ICPW 125)	10	10	–	–	–	–
F_2 (ICP 26 × ICPW 125)	216	158	58	3:1	0.395	0.50–0.75
BC_1 (ICP 26 × F_1)	75	36	39	1:1	0.121	0.50–0.75

F_3 population

Cross	F_3 families	F_3 families			Expected ratio	χ^2 value	*P*-value
		R	Seg	S			
ICP 28 × ICPW 94	116	32	56	28	1:2:1	0.414	0.75–0.90
ICP 28 × ICPW 130	120	28	62	30	1:2:1	0.200	0.90–0.95
ICP 26 × ICPW 125	96	26	48	22	1:2:1	0.250	0.75–0.90

*Damage rating was given on a scale of 1–5. Plants with a score of ≤2 were rated as resistant and >2 as susceptible.

oviposit in these *C. scarabaeoides* parents need to be determined.

Under no-choice conditions in the laboratory, ICP 28, the cultivated susceptible parent, recorded a mean damage rating of 6.25 ± 1.064 and the resistant parent, ICPW 94 a damage rating of 0.42 ± 0.116 on a 1–9 scale where scores ≤3 were grouped as resistant and those above 4 as susceptible. The F_1 plants of these parents showed a damage rating of 0.72 ± 0.511 and were classified as resistant to pod borer damage. Segregation in F_2 and backcross population confirmed the control of a single dominant gene in resistance to pod borer (Table 7). Results from both the field and laboratory confirm that the antixenosis mechanism of resistance to pod borer is governed by the dominant allele of a single gene. Verulkar et al. (1997)

Table 7. Reaction of parents and different generations to pod borer in interspecific cross (ICP 28 × ICPW 94) under laboratory conditions, ICRISAT, Patancheru, 2002

Cross	Population	Resistant*	Susceptible*	Expected ratio	χ^2 value	*P*-value
ICP 28	10	–	10	–	–	–
ICPW 94	10	10	–	–	–	–
F_1 (ICP 28 × ICPW 94)	10	10	–	–	–	–
F_2 (ICP 28 × ICPW 94)	250	184	66	3:1	0.261	0.50–0.75
BC_1 (ICP 28 × F_1)	75	36	39	1:1	0.121	0.50–0.75

*Damage rating was given on a scale of 1–9. Plants with a score ≤3 were rated as resistant and >4 as susceptible.

also reported similar results for pod borer resistance in the interspecific crosses, involving *C. cajan* and *C. scarabaeoides*, using dual choice arena test. Other mechanism of resistance and their inheritance has to be studied in future to completely understand this complex problem.

The resistance factors, low density of type A trichome, high density of type C trichome; low damage by *H. armigera* under field conditions and low damage under no-choice laboratory conditions are all governed by dominant alleles of single genes. The backcrossing should be used to incorporate pod borer resistance gene(s) into the adapted high-yielding pigeonpea varieties.

Acknowledgement

References

Armes, N.J., D.R. Jadhav & K.R. DeSouza, 1996. A Survey of insecticide resistance in *Helicoverpa armigera in* the Indian sub-continent. Bull Ent Res 86: 499–514.

Ascensao, L., N. Marques & M.S. Pais, 1995. Glandular trichomes on vegetative and reproductive organs of *Leonotis leonurus* (Lamiaceae). Ann Bot 75: 619–626.

Bacchetta, L., P.C. Remotti, A. Lai, S. Arnone, V. Beinat & A. Sonnino, 1993. Glandular trichomes as a possible defense mechanism against Colorado potato beetle (*Leptinotarsa decemlineata*, Say) and late blight (*Phytophthora infestans* Mont. De Bary). In Abstracts of the 12th Triennial Conference of the European Association for Potato Research, 18–23 July 1993, pp. 397–398, Paris, France.

Bernays, E.A. & R.F. Champman, 1994. Host-plant selection by phytophagopus insects. Chapman & Hall, New York.

Bisen, S.S. & A.R. Sheldrahe, 1981. The anatomy of the pigeonpea. Research Bulletin No. 5. International Crop Research Institute for the Semi –Arid Tropics (ICRISAT), Patancheru 502 324, A.P India.

David, H. & S. Easwaramoorthy, 1998. Physical resistance mechanisms in insect plant interaction. In: Ananthakrishna, T.N. & Raman, A. (Eds.), Dynamics of Insect-plant Interactions recent Advances and Future Trends, pp. 45–70. Oxford & IBH publishing, New Delhi, India.

Green, P.W.C., P.C. Stevenson, M.S.J. Simmonds & H.C. Sharma, 2003. Phenolic compounds on the pod-surface of pigeonpea, *Cajanus cajan*, mediate feeding behavior of *Helicoverpa armigera* larvae. J Chem Ecol 29: 811–821.

ICRISAT, 1992. The Medium Term Plan, Vol. 1. International Crop Research Institute for the Semi-Arid Tropics. Patancheru 502324, Andhra Pradesh, India.

Jeffree, C.E., 1986. The cuticle, epicuticular wax and trichomes of plants, with reference to their structure, functions, and evolution. In: Juniper, B.E. & Southwood, T.R.E. (eds.), Insects and plant surfaces, pp. 23–64. Edward Arnold publishers Ltd, London, UK.

Karkkainen, K. & J. Agren, 2002. Genetic basis of trichome production in *Arabidopsis lyrata*. Hereditas 136: 219–226.

Kennedy, G.G., 2003. Tomato, pests, parasotoids and predators: tritrophic interactions involving the genus *Lycopersicon*. Ann Rev Ent 48: 51–72.

Kole, C., 1996. Molecular mapping of a locus controlling resistance to *Albuga candida* in *Brassica rapa*. Phytopathol 86: 367–369

Lai, A., V. Cianciolo, S. Chiavarini & A. Sonnino, 2000. Effects of glandular trichomes on the development of *Phytophthora infestans* infection in Potato (*Solanum tuberosum*). Euphytica 114(3): 165–174.

Lam, W.-K.F. & L.P. Pedigo, 2001. Effect of trichome density on soybean pod feeding by adult Bean leaf Beetles (Coleoptera: Chrysomelidae) J Econ Ent 94: 1459–1463.

Lateef, S.S., 1992. Scope and limitations of host plant resistance in pulses for the control of *Helicoverpa armigera*. In: J.N. Sachan (ed.), Helicoverpa management Current status and future statergies, pp. 31–37 Indian Institute of Pulses Research, Kanpur.

Levin, D.A., 1973. The role of trichomes in plant defense. Q. Rev Biol 48: 3–15.

Muigai, S.G., D.J. Schuster, J.W. Scott, M.J. Basset & H.J. McAuslane, 2002. Mechanisms of resistance in lycopersicon germplasm to the white fly *Bemisia argentofoli*. Phytoparasitica 30: 347–360.

Navasero, R.C. & S.B. Ramaswamy, 1991. Morphology of leaf surface trichomes and its influence on egg laying by *Heliothis virescens*. Crop Sci 31: 324–353.

Panse, V.G. & P.V. Sukhatme, 1967. Statistical methods for agricultural workers. Indian Council of Agricultural research, New Delhi.

Peter, A.J., T.G. Shanower & J. Romies, 1995. The role of plant trichomes in insect resistance: A selective review. Phytophaga 7: 41–64.

Pundir, R.P.S. & R.B. Singh, 1985. Biosystematic relationship among *Cajanus*, *Atylosia* and *Rhynchosia* species and evolution of Pigeonpea (*Cajanus cajan* (L.) Millspaugh). Theor Appl Genet 69: 531–534.

Pundir, R.P.S. & R.B. Singh, 1987. Possibility of genetic improvement in pigeonpea utilising the wild genetic resourses. Euphytica 36: 33–37.

Reddy, L.J., J.M. Green & D. Sharma, 1981. Genetics of *Cajanus cajan* (L.) Millsp. × *Atylosia* spp. In: Proceedings of the International Workshop on Pigeonpea, Volume 2, 15–19 December 1980, pp 39–50. ICRISAT Centre, India. Patancheru, A.P., India.

Reddy, M.V., V.K. Sheila, A.K. Murthy, & N. Padma, 1995. Mechanism of resistance to *Aceria cajani* in pigeonpea. Int Trop Plant Dis 13: 51–57.

Romies, J., T.G. Shanower & A.J. Peter, 1999. Trichomes on pigeonpea [*Cajanus cajan* (L.) Millspaugh] and two wild *Cajanus* spp. Crop Sci 39: 564–569.

Sachan, J.N., 1992. Present status of *Helicoverpa armigera* in pulses and strategies for its management. In: J.N. Sachan (ed.), *Helicoverpa* management: Current status and future strategies, pp. 7–23. Indian Institute of Pulses Research, Kanpur.

Saxena, K.B., L. Singh, M.V. Reddy, U. Singh, S.S. Lateef, S.B. Sharma & P. Remanandan, 1990. Inter species variation in *Atylosia scarabaeoides* (L.) Benth. a wild relative of pigeonpea [*Cajanus cajan* (L.) Millsp.]. Euphytica 49: 185–191.

Saxena, K.B., M.V. Reddy, V.R. Bhagwat & S.B. Sharma, 1996. Prelimnary studies on the incidence of major diseases and insects in *Cajanus platycarpus* germplasm at ICRISAT Asia center. Intl Chickpea Pigeonpea Newsl 3: 51–52.

Saxena, K.B., 2000. Pigeonpea.. In S.K. Gupta (ed.) Plant Breeding: Theory and Techniques, pp. 82–112. Agrobios, Jodhpur, India.

Shanower, T.G., M. Yoshida, & A.J. Peter, 1997. Survival, growth, fecundity, and behaviour of *Helicoverpa armigera* (Lepidoptera: Noctuidae) on pigeonpea and two wild Cajanus species. J Econ Entomol 90: 837–841.

Sharma, H.C., P.W.C. Green, P.C. Stevenson & M.J. Simmonds, 2001. "What makes it so tasty for the pest?" In: Identification of *Helicoverpa armigera* (Hubner) feeding stimulants and location of their production on the pod surface of Pigeonpea [*Cajanus cajan* (L.) Millspaugh], pp. 85. Final Technical Report, Competitive Research Facility (CRF) Project R7029C, ICRISAT, Patancheru, India.

Simmons, A.T. & M.G. Geoff, 2004. Trichome-based host plant resistance of *Lycopersicon* species and the biocontrol agent *Mallada signata*: Are they compatible? Entomologia Experimentalis et Applicata 113: 95–101.

Simmons, A.T., M.G. Geoff, D. Mc Granth, H.I. Nicol & M.M. Peter, 2003. Trichomes of *Lycopersicon* spp. and their effect on *Myzus persicae* (sulzer) (Hemiptera: Aphididae). Austr J Entomol 42(4): 373–378.

Simmons, A.T., M.G. Geoff, D. Mc Granth, M.M. Nicol & H.I. Peter, 2004. Entrapment of *Helicoverpa armigera* (Hubner) (Lepidoptera:Noctuidae) on glandular trichomes of *Lycopersicon* species. Austr J Entomol 43: 196–200.

Singh, I.P., D.P. Srivastava & N.P. Singh, 2000. Inheritance of certain morphological characters in interspecific crosses of *Cajanus* species. Indian J Agric Sci 70: 667–670.

Stipanovic, R.D., 1983. Function and chemistry of plant trichomes and glands in insect resistance. In: P.A. Hedin (ed.), Plant Resistance to Insects, American Chemical Society Symposium Series 208, Washington, DC, U.S.A.

Verulkar, S.B., D.P. Singh & A.K. Bhattacharya, 1997. Inheritance of resistance to podfly and podborer in the interspecific cross of pigeonpea. Theor Appl Genet 95: 506–508.

Webster, J.A., D.H. Smith, H. Rathke & C.E. Cress, 1975. Persistence to cereal leaf beetle in wheat: Density and strength of leaf surface pubescence in four wheat lines. Crop Sci 15: 199–202.