

Incorporation of lyophilized leaves and pods into artificial diets to assess the antibiosis component of resistance to pod borer *Helicoverpa armigera* (Lepidoptera: Noctuidae) in chickpea

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Abstract. Host-plant resistance is one of the major components of integrated pest management programmes against the noctuid pod borer *Helicoverpa armigera* (Hübner) in chickpea. Survival and development of *H. armigera* on nine chickpea genotypes were compared using two food substrates, namely fresh leaves and pods, and artificial diets containing lyophilized leaf or pod powder of the same genotypes. Among the genotypes used, six showed different levels of resistance to *H. armigera*, while three were used as susceptible checks. Using leaves and pods, five of the resistant genotypes yielded lower larval weights compared to one of the susceptible checks used. Significant differences between four of the resistant and two of the susceptible genotypes were also observed when using artificial diets containing leaf or pod powder, but the rankings were different from that on the fresh leaves and pods. On both substrates, four resistant genotypes resulted in lower larval survival, pupation, adult emergence and fecundity when compared to one of the susceptible checks. A similar trend was also observed for larval survival and development when using F₁ hybrids based on four of the resistant genotypes. Survival and development of *H. armigera* on the two food substrates, fresh leaves and pods and artificial diets with lyophilized leaf or pod powder, were highly correlated, suggesting that incorporation of lyophilized leaves or pods into the artificial diet can be used to assess antibiosis to *H. armigera* in chickpea.

Key words: *Helicoverpa armigera*, chickpea, biology, resistance, artificial diet

Introduction

The legume pod borer *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) is the most important pest on a wide variety of crops such as cotton *Gossypium* spp. (Malvaceae), pigeonpea *Cajanus cajan* (L.) Mill

sp. (Fabaceae), chickpea *Cicer arietinum* L. (Leguminosae), tomato *Lycopersicon esculentum* Mill sp. (Solanaceae) and a range of fruit and vegetable crops (Sharma, 2005). It is widely distributed in Asia, Africa, Oceania and Europe (IIE, 1993). Its significance as a pest is based on the peculiarities of its

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biology such as high mobility, polyphagy, high reproductive rate and diapause (Fitt, 1989). Its preference for flowering/fruiting parts of high-value crops confers a high socio-economic cost to subsistence farmers in the tropics and subtropics. Monetary losses result from the direct reduction in crop yield and the cost of monitoring and control, particularly the cost of insecticides. The extent of losses in chickpea has been estimated at over US\$ 328 million in the semi-arid tropics (ICRISAT, 1992). Total losses due to *H. armigera* in cotton, legumes, vegetables and fruits may exceed US\$ 2 billion in the semi-arid tropics, and the cost of insecticides used to control *H. armigera* may be over US\$ 500 million annually (Sharma, 2005).

Chickpea germplasm accessions with resistance to *H. armigera* have been identified by several workers (Lateef, 1985; Chhabra *et al.*, 1990; Lateef and Sachan, 1990; Das and Kataria, 1999; Singh and Yadav, 1999a,b). However, the genotypic responses have been found to be quite variable across seasons and locations (Sharma *et al.*, 2003). There are large differences in the flowering times of different chickpea genotypes (35 to >90 days), whereas *H. armigera* infestation varies over space and time. *H. armigera* infestations in chickpea are either too high and cause complete damage to the crop or too low to result in significant differences among test genotypes. The onset of infestation also varies over seasons and locations, resulting in differential crop response to damage by *H. armigera*.

To increase the levels of and to diversify the bases of resistance, it is important to identify chickpea genotypes with different mechanisms of resistance to *H. armigera*, and combine the resistance genes from diverse sources (gene pyramiding) in the same genetic background. However, it is difficult to assess antibiosis to *H. armigera* under natural infestation because of staggered flowering of different genotypes, and the difficulty in locating eggs and small larvae on the plants (Sharma *et al.*, 2005a). Also, a proportion of the larvae is lost because of parasitism, predation and cannibalism. Therefore, the present studies were undertaken to assess the usefulness of incorporating lyophilized leaf or pod powder into the artificial diet to assess the antibiosis component of resistance to *H. armigera* in chickpea.

Materials and methods

Test material

Nine chickpea genotypes (eight desi-grain with brown seed coat and one kabuli-type-grain with white seed coat) were selected to assess antibiosis to *H. armigera* based on their reaction to this pest under field conditions (Lateef, 1985; Sharma *et al.*, 2005a). Among these, international chickpea cultivar (ICC

506EB (ICC 12475), ICC 12476, ICC 12477, ICC 12478, ICC 12479 and ICCV 2 (ICC 12968) had shown different levels of resistance to *H. armigera* (Lateef, 1985). The genotypes ICC 37 (ICC 12426), ICC 3137 and ICC 4918 were used as susceptible checks. These lines were also mated in all possible combinations, and the parents and their F₁ hybrids were studied for antibiosis to *H. armigera* by incorporating the lyophilized leaf powder into an artificial diet. The test genotypes were grown in the greenhouse/field conditions during the 2003/2004 and 2004/2005 post-rainy seasons (November–March) to obtain leaf/pod material for the bioassays.

In the greenhouse, the chickpea genotypes were raised on a sterilized mixture of black soil (Vertisols), sand and farmyard manure (2:1:1). The soil was filled into pots of 30 cm in diameter and 30 cm in depth. The seeds were sown 5 cm below the soil surface and watered when required. Ten seeds were sown in each pot, and five plants with uniform growth were retained at 10 days after seedling emergence. The plants were fertilized with diammonium phosphate of 20 g per pot at day 15 after seedling emergence. The plants were raised in the greenhouse, which was cooled by desert coolers (27 ± 5 °C and 65–90% RH). The parents and their F₁ hybrids were also grown under field conditions. There were three replications in a randomized complete block design, with a plot size of four rows of 2 m long (4 × 2 m). The rows were 60 cm apart, and plant-to-plant distance within a row was 10 cm. The crop was raised under irrigated conditions during the post-rainy season (November–March). There was no insecticide application in the experimental plot.

Insect culture

The insects were obtained from the laboratory culture maintained on a chickpea flour-based artificial diet (Armes *et al.*, 1992). The neonates were reared for 5 days in groups of 200–250 in 200 ml plastic cups containing a 2–3 mm layer of artificial diet on the bottom and sides of the cup. Thereafter, the larvae were transferred individually to six-cell well plates (each cell well 3.5 cm in diameter and 2.0 cm in depth) to avoid cannibalism. Each cell well had sufficient amount of diet (7 ml) to support larval development until pupation. The pupae were removed from cell wells, sterilized with 2% sodium hypochlorite solution, and kept in groups of 50 in plastic jars containing moist vermiculite. Upon emergence, 10 pairs of adults were released inside an oviposition cage (30 × 30 × 30 cm). Adults were provided with 10% honey solution on a cotton swab for feeding. Diaper liners, which have a rough surface, were provided as

a substrate for egg laying. The liners were removed daily, and the eggs were sterilized in 2% sodium hypochlorite solution. The liners were dried under a table fan and then placed inside the plastic cups with diet. The liners were removed after 4 days. Freshly emerged neonates were used for assessing antibiosis component of resistance on fresh leaves/pods or on artificial diets containing lyophilized leaf or pod powder of the test genotypes.

Survival and development of H. armigera on leaves and pods of different chickpea genotypes

Survival and development of *H. armigera* were studied on chickpea leaves and pods. The neonates were fed on the leaves for the first 7 days, and then on the pods to simulate natural feeding behaviour. The larvae were individually confined to chickpea plants with pods grown in the greenhouse at $27 \pm 5^\circ\text{C}$ and 65–90% RH using the no-choice cage technique (Sharma *et al.*, 2005b). The experiment was conducted in a completely randomized design, and there were five replications, each replication having 10 larvae.

Data on larval weights were recorded on day 10 using a microbalance. For this purpose, the larvae were removed from the rearing cups, cleaned, weighed and then placed back on the respective plants. The pupal weights were recorded 1 day after pupation. Pupae from each replication were placed in a 1 l plastic jar containing moist vermiculite. Percentage larval survival on day 10, pupation and adult emergence were computed in relation to number of neonate larvae released in each replication. Data were also recorded on larval and pupal periods. The adults were collected with an aspirator from the jars, and three pairs of adults emerging on the same day on a particular genotype were placed inside an oviposition cage (30 × 30 × 30 cm), and provided with inflorescences of the respective chickpea genotypes for oviposition to record data on fecundity of insects reared on different chickpea genotypes. The chickpea inflorescences were kept in 250 ml conical flasks containing water. The adults were provided with 10% honey solution on a cotton swab as a food. There were five replications for each genotype and the experiment was laid out in a completely randomized design. The numbers of eggs laid on each genotype were counted and the chickpea branches were changed daily.

Survival and development of H. armigera on artificial diets containing lyophilized leaf or pod powder of different chickpea genotypes

Chickpea terminals at 30 days after seedling emergence were collected from the pots and placed in an icebox. The leaves were freeze-dried,

powdered in a Willey Mill and used for incorporating into the artificial diet to assess antibiosis to *H. armigera*. The pods were collected from the field-grown plants at 12–15 days after flowering and freeze-dried for use in bioassays. To study antibiosis to *H. armigera* in chickpea, 20 g of freeze-dried powder of leaves or pods (as a replacement for part of the flour of a susceptible kabuli chickpea variety, used in the artificial diet (KAK 2)) were mixed with artificial diet (having ingredients sufficient for 250 ml artificial diet) for rearing *H. armigera* (Armes *et al.*, 1992). Diet of 7 ml was poured into each cell well in a six-cell well plate. The neonate larvae were released individually into the cell wells. There were three replications for each genotype, and each replication had 10 larvae. Antibiosis was also assessed in 72 hybrids based on nine parents (in all possible combinations) using the leaf powder in the diet incorporation assays. The plants were grown under field conditions as described above. Data were recorded on larval and pupal weights, survival, and larval and pupal development periods, adult emergence and fecundity as described above.

Statistical analysis

Genstat release 10.1 was used for data analysis. The data were subjected to ANOVA to test for significance of differences among the genotypes. The significance level was set at $P \leq 0.05$, and the treatment means were compared using the least significant difference (LSD) test. Correlation coefficients between larval survival and development on fresh leaves/pods and on artificial diets containing lyophilized leaf and pod powder were computed to assess the relevance of diet impregnation assay to assess antibiosis to *H. armigera* in chickpea.

Results

Weights of H. armigera larvae and pupae on different chickpea genotypes

Weights of the 10-day-old larvae reared on leaves/pods of different chickpea genotypes differed significantly among the genotypes tested and ranged from 298.1 to 396.3 mg on ICC 506EB and ICC 4918 (Table 1), respectively. Larval weights were significantly lower in larvae reared on leaves/pods of ICC 12475, ICC 12476, ICC 12477 and ICCV 2 as compared to those reared on the susceptible check, ICC 37. Pupal weights were lower in insects reared on ICC 12476, ICC 12478, ICCV 2 and ICC 506EB as compared to the insects reared on ICC 37.

Larval weights were highest on the standard artificial diet, followed by the larvae reared on

Table 1. Survival and development of *Helicoverpa armigera* on leaves and pods of nine chickpea genotypes (ICRISAT, Patancheru, 2003/2004 post-rainy season)

Genotype	Larval weight on 10th day (mg)	Larval period (days)	Pupal period (days)	Pupal weight (mg)	Larval survival on 10th day (%)	Pupation (%)	Adult emergence (%)	Fecundity (eggs laid/female)
ICC 12476	320.5 ^{ab}	16.2 ^b	11.8 ^{cd}	274.2 ^a	76.0 ^{bc}	66.0 ^{ab}	60.0 ^a	839.5 ^b
ICC 12477	340.8 ^{bc}	16.4 ^{bc}	11.8 ^{cd}	302.6 ^{cd}	74.0 ^b	70.0 ^{ab}	60.0 ^a	882.9 ^c
ICC 12478	367.5 ^{cde}	16.5 ^c	11.0 ^b	292.3 ^b	78.0 ^{bcd}	74.0 ^{abc}	62.0 ^a	907.1 ^d
ICC 12479	359.8 ^{cd}	16.5 ^c	11.1 ^b	317.8 ^d	78.0 ^{bcd}	72.0 ^{ab}	60.0 ^a	901.3 ^{cd}
ICCV 2	329.7 ^b	16.5 ^c	12.0 ^a	300.0 ^{bc}	84.0 ^{cde}	76.0 ^{bcd}	70.0 ^a	1170.1 ^f
ICC 4918	396.3 ^e	15.5 ^a	10.9 ^b	323.9 ^e	86.0 ^{de}	84.0 ^{cd}	84.0 ^b	1270.7 ^g
Controls								
ICC 506EB (R)	298.1 ^{de}	17.8 ^d	11.7 ^c	286.2 ^{ab}	66.0 ^a	64.0 ^a	62.0 ^a	785.0 ^a
ICCC 37 (S)	382.9 ^{de}	15.5 ^a	8.8 ^a	316.6 ^{de}	88.0 ^e	86.0 ^d	86.0 ^b	1291.2 ^f
ICC 3137 (S)	361.8 ^{cd}	16.4 ^{bc}	10.6 ^b	324.5 ^e	88.0 ^e	84.0 ^d	84.0 ^b	1066.5 ^e
Fp	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.001	<0.001
LSD ($P = 0.05$)	29.00	0.27	0.27	16.00	9.40	11.20	13.10	20.80

R, Resistant check; S, susceptible check. Figures followed by the same letter in a column are not significantly different at $P \leq 0.05$.

diets having the leaf powder of ICC 4918 and ICC 37 (Table 2). Larval weight was lowest in larvae reared on artificial diets with leaf powder of the resistant check, ICC 506EB, followed by those reared on diets with leaf powder of ICC 12478 and ICC 12476. Larvae fed on artificial diet with lyophilized pod powder of ICC 506EB, ICC 12476 and ICC 12479 weighed significantly lower than those fed on the standard artificial diet and the diet containing pod powder of ICC 37 (Table 3). The pupal weights were lower on artificial diets with leaf powder of ICC 12476, ICC 12477, ICC 12478 and ICC 506EB as compared to the diet with leaf powder of ICC 37 (Table 2). The highest pupal weight was recorded in insects reared on artificial diet having pod powder of ICC 37, followed by those reared on standard artificial diet; they were low in diets containing pod powder of ICC 506EB, ICC 12478, ICC 12477, ICC 12479 and ICC 12476 (Table 3).

Survival and development of H. armigera on different chickpea genotypes

The larval period was prolonged when the larvae were reared on fresh leaves/pods of ICC 506EB as compared to those reared on ICC 37 and ICC 4918. The pupal period was longer on ICCV 2, ICC 12477, ICC 12476 and ICC 506EB as compared to the insects reared on the susceptible check, ICC 37 (Table 1). When the larvae were reared on artificial diet with lyophilized leaf powder, larval period ranged from 14.9 to 17.0 days on artificial diet and ICCV 2, respectively. The larval period was also prolonged in insects reared on artificial diets with leaf powder of ICC 12478, ICC

12479, ICCV 2 and ICC 506EB (Table 2), while in diets with pod powder, the larval period was longer on ICC 506EB, ICCV 2 and ICC 12479 as compared to that on ICC 37 (Table 3). The pupal period ranged from 9.2 to 12.0 days on ICC 37 and ICC 506EB, respectively. The pupal period was prolonged in insects reared on diets with lyophilized pod powder of ICC 506EB, ICC 12479, ICC 12476 and ICC 12478 as compared to that on ICC 37 (Table 3).

Larval survival at 10 days after release of the larvae was 66% on the resistant check, ICC 506EB and 88% on the susceptible check, ICC 37. More than 80% larval survival was recorded on ICC 3137, ICCV 2, ICC 4918 and ICC 37 as compared to 66% survival on the resistant check, ICC 506EB. Pupation was lowest in insects reared on ICC 506EB, followed by those reared on ICC 12476 and ICC 12477. Adult emergence was 60–62% on ICC 12476, ICC 12477, ICC 12478 and ICC 506EB compared to 86% survival on ICC 37 (Table 1).

Larval survival was 70–75% on artificial diets with leaf powder of ICC 12476, ICC 12477, ICC 12479 and ICC 506EB compared to 91.7% survival on ICC 37 and 98.0% on the standard artificial diet. Pupation and adult emergence were lower on ICC 506EB, ICC 12476, ICC 12477 and ICC 12478 as compared to that on ICC 37 and the standard artificial diet (Table 2). In diets with pod powder, larval survival was lower on ICC 12476, ICC 12477, ICC 12478 and ICC 506EB as compared to that on ICC 37 and artificial diet (Table 3). Pupation and adult emergence were lower in insects reared on diets with pod powder of ICC 12476, ICC 12477, ICC 12478, ICC 12479 and ICC 506EB as compared to that on ICC 37 (Table 3).

Table 2. Survival and development of *Helicoverpa armigera* on artificial diet containing lyophilized leaf powder of nine chickpea genotypes (ICRISAT, Patancheru, 2003–2005 post-rainy seasons)

Genotype	Larval weight on 10th day (mg)	Larval period (days)	Pupal period (days)	Pupal weight (mg)	Larval survival on 10th day (%)	Pupation (%)	Adult emergence (%)	No. of eggs laid/female
ICC 12476	321.8 ^a	16.2 ^{bc}	10.4 ^d	298.9 ^a	75.0 ^a	71.7 ^{abc}	66.6 ^{ab}	730.7 ^b
ICC 12477	366.9 ^{bc}	16.2 ^{bc}	10.0 ^c	300.2 ^a	75.0 ^a	66.6 ^{ab}	66.6 ^{ab}	839.8 ^c
ICC 12478	355.6 ^b	16.5 ^{cd}	10.8 ^f	297.3 ^a	76.6 ^a	71.0 ^{abc}	70.0 ^{abc}	899.7 ^d
ICC 12479	375.8 ^{bc}	16.7 ^{cd}	11.6 ^d	342.5 ^{de}	75.0 ^a	73.3 ^{bc}	71.7 ^{bc}	854.5 ^c
ICCV 2	378.7 ^{bc}	17.0 ^d	10.7 ^{ef}	317.0 ^{bc}	80.0 ^{ab}	76.6 ^{cd}	76.6 ^c	975.7 ^e
ICC 4918	417.5 ^d	16.3 ^b	10.5 ^{de}	355.7 ^{ef}	89.0 ^{bc}	86.6 ^e	86.6 ^{de}	1015.0 ^f
Controls								
ICC 506EB (R)	307.2 ^a	17.4 ^e	11.4 ^g	311.2 ^{ab}	70.0 ^a	63.3 ^a	63.3 ^a	675.0 ^a
ICCV 37 (S)	415.6 ^d	16.0 ^b	9.0 ^a	342.7 ^{de}	91.7 ^{bc}	89.0 ^{ef}	87.0 ^e	1150.0 ^g
ICC 3137 (S)	383.8 ^c	16.2 ^{bc}	9.3 ^b	329.0 ^{cd}	87.0 ^{bc}	83.3 ^{de}	80.0 ^d	1025.0 ^g
Artificial diet	445.8 ^e	14.9 ^a	8.9 ^a	365.8 ^f	98.0 ^c	97.0 ^f	95.0 ^f	1220.0 ^h
Fp	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
LSD ($P = 0.05$)	24.55	0.31	0.26	14.00	12.00	9.10	6.80	18.91

R, Resistant check; S, susceptible check. Figures followed by the same letter in a column are not significantly different at $P \leq 0.05$.

Table 3. Survival and development of *Helicoverpa armigera* on artificial diet containing lyophilized pod powder of nine chickpea genotypes (ICRISAT, Patancheru, 2003–2005, post-rainy seasons)

Genotype	Larval weight on 10th day (mg)	Larval period (days)	Pupal period (days)	Pupal weight (mg)	Larval survival on 10th day (%)	Pupation (%)	Adult emergence (%)	No. of eggs laid/female
ICC 12476	285.4 ^b	15.6 ^b	10.5 ^f	249.5 ^a	76.6 ^a	70.0 ^{ab}	60.0 ^a	672.5 ^b
ICC 12477	359.1 ^c	16.2 ^c	8.9 ^b	262.4 ^a	80.0 ^{ab}	73.3 ^{ab}	63.3 ^{ab}	860.5 ^d
ICC 12478	334.9 ^c	16.5 ^c	10.7 ^f	245.7 ^a	76.6 ^a	70.0 ^{ab}	60.0 ^a	901.6 ^e
ICC 12479	288.3 ^b	17.6 ^e	11.6 ^g	233.8 ^a	80.0 ^{ab}	76.6 ^{ab}	66.6 ^{ab}	842.0 ^c
ICCV 2	420.2 ^a	17.6 ^e	9.5 ^e	274.7 ^{ab}	83.3 ^{abc}	80.0 ^b	66.6 ^{ab}	1051.5 ^f
ICC 4918	413.9 ^a	16.9 ^d	9.3 ^{cd}	327.9 ^c	90.0 ^{bcd}	86.6 ^c	80.0 ^c	1198.1 ^g
Controls								
ICC 506EB (R)	253.3 ^a	18.3 ^f	12.0 ^h	244.1 ^a	76.0 ^a	63.3 ^a	60.0 ^a	632.8 ^a
ICCV 37 (S)	443.8 ^{df}	15.4 ^a	9.2 ^c	351.4 ^c	93.3 ^{cd}	86.6 ^c	83.3 ^c	1241.2 ^h
ICC 3137 (S)	424.1 ^d	16.6 ^a	8.5 ^a	315.8 ^{bc}	86.6 ^c	80.0 ^b	70.0 ^b	1092.9 ^g
Artificial diet	468.8 ^f	14.8 ^a	8.8 ^b	342.1 ^c	100.0 ^d	100.0 ^c	100.0 ^d	1290.2 ⁱ
Fp	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
LSD ($P = 0.05$)	31.60	0.35	0.26	41.80	12.20	14.30	9.10	12.4

R, Resistant check; S, susceptible check. Figures followed by the same letter in a column are not significantly different at $P \leq 0.05$.

Table 4. Survival and development of *Helicoverpa armigera* on artificial diet containing lyophilized leaf powder of chickpea hybrids (72 hybrids) based on nine parents (ICRISAT, Patancheru, 2004/2005 post-rainy season)

Genotype	Larval weight on 10th day (mg)	Larval period (days)	Pupal period (days)	Pupal weight (mg)	Larval survival on 10th day (%)	Pupation (%)	Adult emergence (%)
F ₁ s based on ICC 506EB	318.9 ^a	15.8 ^b	9.8 ^{ab}	279.5 ^a	75.0 ^a	61.3 ^a	56.3 ^a
F ₁ s based on ICC 12476	394.3 ^{cd}	15.5 ^{ab}	9.7 ^{ab}	317.9 ^{abc}	76.3 ^a	71.3 ^a	63.8 ^{abc}
F ₁ s based on ICC 12477	369.4 ^{bc}	15.8 ^b	10.1 ^b	317.7 ^{abc}	73.8 ^a	66.3 ^a	61.3 ^{ab}
F ₁ s based on ICC 12478	353.8 ^{abc}	15.9 ^b	10.1 ^b	294.1 ^a	76.3 ^a	71.3 ^a	65.0 ^{abc}
F ₁ s based on ICC 12479	319.8 ^a	16.0 ^b	9.4 ^{ab}	300.4 ^{ab}	73.8 ^a	66.3 ^a	61.3 ^{ab}
F ₁ s based on ICC 3137	319.9 ^a	16.0 ^b	10.1 ^b	287.1 ^a	76.3 ^a	71.3 ^a	61.3 ^{ab}
F ₁ s based on ICC 4918	329.0 ^a	15.9 ^b	9.8 ^a	285.4 ^a	77.5 ^a	75.0 ^a	67.5 ^{bc}
F ₁ s based on ICC 37	333.5 ^{ab}	15.9 ^b	9.9 ^b	305.6 ^{ab}	80.0 ^a	76.3 ^a	72.5 ^c
F ₁ s based on ICCV 2	326.2 ^a	15.8 ^b	9.9 ^b	318.0 ^{abc}	73.8 ^a	68.8 ^a	65.0 ^a
Controls							
ICC 506EB (R)	356.6 ^{abc}	16.8 ^c	11.9 ^c	338.6 ^b	70.0 ^a	63.3 ^a	63.3 ^{abc}
ICCC 37 (S)	434.6 ^{de}	15.5 ^{ab}	9.0 ^a	345.6 ^c	93.3 ^b	90.0 ^b	88.0 ^d
Artificial diet	468.9 ^e	15.1 ^a	8.9 ^a	351.5 ^c	98.0 ^b	98.0 ^b	96.0 ^d
F _p	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
LSD (<i>P</i> = 0.05)	42.00	0.65	0.82	39.00	13.00	16.00	11.10

R, Resistant check; S, susceptible check. Figures followed by the same letter in a column are not significantly different at *P* ≤ 0.05.

Fecundity of H. armigera females reared on different chickpea genotypes

Reduced fecundity was observed in insects reared on the leaves/pods of ICC 12476, ICC 12477 and ICC 506EB as compared to that on the susceptible check, ICC 37 (Table 1). Fecundity was also lower in insects reared on artificial diet containing leaf or pod powder of ICC 12476, ICC 12477, ICC 12479 and ICC 506EB than in insects reared on standard artificial diet and on diets containing leaf or pod powder of ICC 37.

Survival and development of H. armigera on artificial diets with leaf powder of F₁ hybrids and their parents

Larval and pupal weights were lower in insects reared on diets with leaf powder of the hybrids

based on ICC 506EB, ICC 12479, ICC 3137, ICC 4918 and ICCV 2 as compared to the insects reared on the susceptible check, ICC 37 (Table 4). The larval period in diets containing lyophilized leaf powder of F₁ hybrids did not vary much, and ranged from 15.5 to 16 days in hybrids based on ICC 12476, and ICC 12479 and ICC 3137, respectively. The pupal period ranged from 9.7 days in hybrids based on IC 12476 to 11.9 days in those reared on ICC 506EB (Table 4). Larval survival ranged from 70 to 93.3% in diets with leaf powder of ICC 506EB and ICC 37, respectively, and 98% on the artificial diet. Pupation ranged from 61 to 76% on F₁ hybrids compared to 90% on the susceptible check, ICC 37; while adult emergence was 56.3–72.5% on F₁ hybrids compared to 88% on ICC 37 (Table 4).

Table 5. Association between larval survival and development on fresh leaves/pods and on artificial diet containing lyophilized leaf or pod powder of nine chickpea genotypes (ICRISAT, Patancheru, 2003–2005 post-rainy seasons)

Survival/development parameter	Artificial diet with leaf powder	Artificial diet with pod powder
Larval weight 10th day	0.88**	0.64*
Larval period (days)	0.82**	0.67*
Pupal period (days)	0.63*	0.31
Pupal weight (mg)	0.83**	0.63*
Larval survival 10th day (%)	0.92**	0.85**
Pupation (%)	0.95**	0.91**
Adult emergence (%)	0.93**	0.88**
Number of eggs laid/female	0.91**	0.96**

*, **Correlation coefficients significant at *P* = 0.05 and 0.01, respectively.

Correlation between survival and development of H. armigera on leaves/pods and in diets with leaf or pod powder of different chickpea genotypes

Larval and pupal periods as well larval survival, pupation and adult emergence in insects reared on fresh leaves/pods, and on artificial diet containing lyophilized leaf or pod powder of different genotypes were positively correlated. Larval and pupal weights ($r = 0.63-0.88$) and fecundity ($r = 0.91-0.96$) on different substrates were also positively correlated (Table 5). Except for pupal period in diets with pod powder, the rest of the correlation coefficients were significant and positive. The results suggest that lyophilized leaf or pod powder incorporated into artificial diet can be used to assess antibiosis against *H. armigera* in chickpea.

Discussion

There are considerable differences in numbers of *H. armigera* larvae on different genotypes under field conditions (Lateef, 1985; Lateef and Sachan, 1990). Antibiosis to *H. armigera* in chickpea is expressed in terms of larval mortality, decreased larval and pupal weights, prolonged larval and pupal development, failure to pupate and reduced fecundity and egg viability (Yoshida *et al.*, 1995; Cowgill and Lateef, 1996). Srivastava and Srivastava (1990) assessed antibiosis in terms of larval survival, larval and pupal weights, adult longevity and fecundity, while Sharma and Yadav (2000) used life-table analysis to assess antibiosis to *H. armigera*.

In the present studies, larval and pupal weights and larval survival were greater in larvae reared on artificial diet containing lyophilized leaf or pod powder of different chickpea genotypes as compared to that on fresh leaves and pods. This may be because of more nutrients available in the artificial diet compared to that in the plant material *per se* (Schoonhoven, 1990). Antibiosis expressed in terms of reduced larval and pupal weights and prolonged larval period was observed when the insects were reared on the fresh leaves/pods of ICC 12476, ICC 12477, ICC 12478 and ICC 12479. Similar results were also obtained when the insects were reared on artificial diets containing leaf or pod powder of these genotypes. Larval survival, pupation and adult emergence were lower when the insects were reared on the fresh leaves/pods of ICC 12476, ICC 12477, ICC 12478 and ICC 506EB as compared to those reared on the susceptible checks, ICC 37 and ICC 4918. Similar results were also observed in insects reared on artificial diets with leaf or pod powder of these genotypes. Larval survival and development were also adversely affected on F₁ hybrids based on ICC 12476, ICC 12477, ICC 12478

and ICC 506EB, suggesting that antibiosis mechanism of resistance is transferred to the progeny from the resistant parents, and there is a distinct possibility of developing varieties with resistance to *H. armigera*.

Survival and development of *H. armigera* on fresh leaves/pods, and on diets with lyophilized leaf and pod powder of different chickpea genotypes were highly correlated, except pupal period in diets with pod powder, suggesting that incorporation of lyophilized leaves and pods into artificial diet can be used to assess antibiosis to *H. armigera* in chickpea. Growth inhibitor and/or antifeedant substances in chickpea leaves/pods might contribute to antibiosis to *H. armigera* in chickpea (Yoshida and Shanower, 2000). Slower larval growth, which results in prolonged development, may also increase the probability of predation, parasitism and infection by pathogens, resulting in reduced survival of *H. armigera*. The genotypes showing antibiosis *in vivo* and in diets with lyophilized leaf or pod powder can be used to develop chickpea cultivars with resistance to *H. armigera*.

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