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Dissecting antibiosis and antixenosis mechanisms conferring resistance to *C. chinensis* in chickpea

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Abstract

Pulse beetle, *Callosobruchus chinensis* (L.), is a serious storage pest of chickpea causing extensive grain damage and percent weight loss. The present study evaluated the resistance of different chickpea genotypes to *C. chinensis* by assessing antibiosis and antixenosis mechanisms under laboratory conditions. Antibiosis was studied using parameters such as adult emergence, number of holes per grain, percentage of adult emergence, seed weight loss, growth index, and adult weight. Significant differences were observed among the genotypes. PI 599066 exhibited complete resistance, as no adult emergence, seed damage, or weight loss was recorded. Genotypes IG 72953 and IG 72933 showed high levels of resistance with reduced adult emergence, minimal grain damage, lower adult emergence percentage, and lighter adults, indicating strong antibiosis effects. In contrast, ICCV 2, KAK 2, VIHAR, and JGK 2 were highly susceptible and supported greater insect development and damage. Antixenosis studies revealed significant variation in adult preference, with minimum attraction observed in IG 72953 and maximum preference in ICCV 2. The results indicate that resistance to *C. chinensis* in chickpea is mediated through both antibiosis and antixenosis mechanisms. The resistant genotypes identified can be effectively utilized in breeding programmes aimed at improving post-harvest pest resistance in chickpea.

Keywords: Chickpea, *Callosobruchus chinensis*, antibiosis, antixenosis and host plant resistance

Introduction

The chickpea, *Cicer arietinum* L. (Fabales: Fabaceae), native to southeast Turkey is the major food legume worldwide. It is a good source of energy i.e. 416 calories/100g of chickpea (Shrestha, U. K., 2001) ^[16], along with proteins (18-22%), carbohydrates (52-70%), fats (4-10%), minerals (calcium, phosphorus, iron) and vitamins (Ali SI, *et al.*, 2002) ^[1]. Chickpea is used in wide range of different preparations in our cuisine, and also helps in lowering the cholesterol levels (Pittaway JK, *et al.*, 2006) ^[12]. Cultivated chickpeas are mainly divided into two main groups based on characteristics and seed size, shape and coloration as Desi and Kabuli (Meuhlbaauer and Singh 1987) ^[17]. The Kabuli chickpeas have relatively large creamy colored seeds, white flowers and do not contain anthocyanin. While, the desi chickpeas have small seeds of various colors, purplish flowers and presence of anthocyanin pigment. Beyond its nutritional importance, chickpea improves soil health through symbiotic nitrogen fixation and fits well into diverse cropping systems, particularly in semi-arid regions (Singh *et al.*, 2014) ^[17]. However, post-harvest constraints continue to limit the effective utilization of chickpea, with insect infestation during storage representing a major cause of quantitative and qualitative losses.

The pulse beetle, *Callosobruchus chinensis* (L.) (Coleoptera: Bruchidae) is recognized as one of the most damaging pests of stored chickpea. The larvae develop inside the grain, resulting in exit holes, loss of seed weight,

decline in nutritional quality, and poor seed viability (Shaheen *et al.*, 2006; Jha *et al.*, 2007) ^[13, 6]. In severe infestations, grain losses may exceed 50 per cent within a short storage period, rendering the produce unsuitable for consumption or planting (Gujar, 1976; Lambrides and Imrie, 2000) ^[4, 8].

Although chemical control measures are commonly adopted for bruchid management, their continued use has raised serious concerns related to resistance development, food contamination, and environmental safety (Arthur, 1996; Zettler and Arthur, 2000) ^[2, 18]. As a result, emphasis has increasingly shifted towards eco-friendly alternatives. Host plant resistance offers a durable and cost-effective strategy, particularly through mechanisms such as antibiosis, which impairs insect growth and reproduction, and antixenosis, which deters insect preference and infestation (Sharma and Thakur, 2014) ^[15].

Considerable variability in chickpea genotypes for resistance to *C. chinensis* has been reported, often associated with seed coat characteristics, grain hardness, and biochemical factors (Shaheen *et al.*, 2006; Kuldeep Tripathi *et al.*, 2015) ^[13, 7]. However, comprehensive evaluation of resistance mechanisms in diverse germplasm remains limited. Therefore, the present study was undertaken to characterize chickpea genotypes for antibiosis and antixenosis resistance against *C. chinensis*, with the aim of identifying promising donor lines for use in breeding bruchid-resistant chickpea cultivars.

Materials and Methods

Experimental location

The research experiments were conducted in the Department of Entomology, ICRISAT, Patancheru, India. Geographically, Patancheru is located on the longitude of 78.27° east, the latitude of 17.53° north and at an average elevation of 522 meters (1712 feet) from mean sea level (MSL) in Sangareddy district of Telangana.

Collection of test genotypes

The seeds of fifteen cultivars each of chickpea were procured from the Department of Plant Breeding and Entomology at ICRISAT. Seeds were cleaned, washed under tap water, oven-dried at 45°C and stored in cold chamber to prevent further insect pests and microbial attack.

Culturing of test insects

The test insect, *Callosobruchus chinensis* (L.), culture was maintained in a Bio-Oxygen Demand (BOD) incubator at the Department of Entomology, ICRISAT, using healthy chickpea grains as food. Prior to infestation, grains were cleaned, sieved, and sterilized at 65 ± 5 °C for 5 h to eliminate hidden insect and mite infestations. The sterilized grains were conditioned for one week at 28 ± 2 °C and $70 \pm 5\%$ relative humidity to stabilize seed moisture content. Sex differentiation of *C. chinensis* adults was carried out before release based on morphological characters described by Halstead (1963) [5]. Males were smaller with pectinate antennae and a reduced pygidium, whereas females were larger, serrate, and possessed a broader, darkly pigmented pygidium. The beetles were reared in plastic containers, and newly emerged adults were periodically transferred to fresh grains to maintain a continuous culture for 3-4 generations throughout the experimental period.

Releasing of test insects

C. chinensis were allowed access to seeds of a single genotype. The seeds were placed in a plastic cup and each cup was considered as one replication for each genotype. This experiment was conducted in triplicates for the chickpea genotypes separately. Five pairs of 0-24 hr. old adults of *C. chinensis* were released into each cup in each replication.

Observations recorded

Observations on damaged grains were recorded after all the F1 adults emerged from the release of *C. chinensis*. The following of different parameters were recorded on number of adults emerged, number of holes/grain, adults emergence percentage, weight loss percentage, growth index and adult weight (mg).

Data analysis

The experimental data were statistically analyzed by analysis of variance (ANOVA). The percentage values of the data were converted into angular transformed values. The Critical difference (CD) values at 5 per cent was determined.

Results and Discussion

The resistance of chickpea genotypes to pulse beetle, *Callosobruchus chinensis* (L.), was assessed through

antibiosis and antixenosis mechanisms were evaluated using different parameters viz. mean number of adults emerged (adults/ 50 seed), mean number of holes/grain, adults emergence percentage, weight loss percentage, growth index and adult weight (mg).

Mean number of adults emerged

There were significant differences in mean number of adults emerged from different accessions of chickpea (Table 1). The lowest mean number of adults/50 grains was observed in IG 72953 (3.60) followed by IG 72933 (3.95), JAKI 9268 (5.10), ICC 506 EB (5.70), JG 14 (5.92), ICC 37 (7.50), NBeG 47 (8.21), NBeG 119 (8.76), NBeG 3 (10.23), and RVG 204 (12.01). The highest mean number of adults were observed in ICCV 2 (18.20), followed by KAK 2 (16.30), VIHAR (14.50), and JGK 2 (12.01). There was no adult emergence in PI 599066 (0.00) and it was statistically superior to the rest of the genotypes. These results are in conformity with the findings of Kuldeep Tripathi *et al.* (2015) [7], Shafique, M *et al.* (2005) [14] and Shaheen *et al.* (2006) [13] who stated that those varieties, which had smooth, soft and thin seed coat, exhibited maximum adult emergence. The results of present study also showed considerable findings of adult emergence from the bold seeded varieties.

Mean number of holes/grain

The lowest mean number of holes/grain were observed in IG 72953 (0.767), IG 72933 (0.90), JAKI 9268 (1.23), ICC 506 EB (1.30), JG 14 (1.36), ICC 37 (1.43), NBeG 47 (1.56), NBeG 119 (1.74), NBeG 3 (1.77), and RVG 204 (1.79). The highest mean number of holes/grain were observed in ICCV 2 (2.33), followed by KAK 2 (2.11), VIHAR (2.06) and JGK 2 (1.92) while PI 599066 (0.00) had no holes and was statistically superior among the rest of genotypes. These findings are similar to Muhammad *et al.* (2013) [10].

Adult emergence%

The percent adult emergence was highest in ICCV 2 (31.14) followed by KAK 2 (30.63), VIHAR (29.28), JGK 2 (26.78), RVG 204 (26.50), NBeG 3 (23.40), NBeG 119 (20.98), NBeG 47 (20.50), ICC 37 (18.93), JG 14 (17.55), ICC 506 EB (17.06), and JAKI 9218 (16.72). The lowest percent adult emergence was observed in IG 72953 and IG 72933 (15.60 and 15.37 respectively). There was no adult emergence in PI 599066 (0.00), which was found to be statistically superior among the test genotypes. These results are similar to Sharma and Thakur (2014) [15] and also Panzario *et al.* (2011) [11] who evaluated the susceptibility of six genotypes of *Cicer arietinum* L. (Fabaceae) to *Callosobruchus maculatus* (Fabr.) (Coleoptera: Bruchidae) through comparative laboratory bioassays.

Weight loss percentage

The test chickpea genotypes differed significantly in terms percent weight loss due to *C. chinensis* in various degrees. The weight loss percent was minimum in IG 72953 (1.51%) while it was maximum in ICCV 2 (23.40%) followed by KAK 2 (22.18). The weight loss percent in IG 72933 was 3.65 followed by ICC 506 EB (4.02), JAKI (5.83), JG 14 (7.60), NBeG 47 (7.70), ICC 37 (8.59), RVG 204 (9.54),

NBeG 119 (9.73) and NBeG 3 (14.33). In the remaining genotypes, the mean percent weight loss were 16.62 (JGK 2) and 18.04 (VIHAR). In other words, PI 599066 was the only genotype that exhibited complete resistance in terms of the mean percent weight loss by *C. chinensis*. The present results are in corroboration with the findings of Jha *et al.* (2007) [6], Shaheen *et al.* (2006) [13] and Lambrides and Imrie (2000) [8] who reported that the tolerant varieties exhibited least weight loss due to could be attributed to the small size and the presence of well-formed texture layer on the seed. Gujar (1976) [4] while studying the weight loss of chickpea concluded that *C. chinensis* was more injurious to seeds than *C. maculatus*.

Adult weight (mg)

Significant differences were noticed in *C. chinensis* adult weights, emerged (F1) from different accessions of chickpea during the experiment. The lowest average adult weights were observed in IG 72953 (5.02 mg) and IG 95733 (5.06mg), which were at par with JAKI 9218 (5.10mg), whereas highest adult weight was recorded on ICCV 2 (7.69 mg). Adult weights of the remaining genotypes ranged from 5.11 mg in ICC 506 EB to 7.24 mg in KAK 2. Whereas, PI 599066 was the only genotype that did not recorded any weight due to complete the inhibition of adult emergence, it was the best accession among all the genotypes and was

statistically superior over others. These results are similar to the findings of Shaheen *et al.* (2006) [13] who studied the adult weight (mg) of *C. chinensis* in chickpea genotypes.

Antixenosis mechanism for bruchids *C. chinensis* on different genotypes of chickpea: In Antixenosis test, the preference and non-preference response of pulse beetle was observed and data showed significant differences between the test genotypes (Table 2). The minimum average of 1.62 pulse beetle adults were attracted towards chickpea genotype IG 72953 which was significantly different from IG 72933 (2.33 adults), JAKI 9218 (3.06 adults), ICC 506 EB (3.31 adults), JG 14 (3.80 adults), ICCV 37 (4.10 adults), NBeG 47 (4.60 adults), NBeG 119 (4.90 adults), NBeG 3 (5.20 adults), and RVG 204 (5.80 adults). The preference response ranged from 1.62 to 5.80. The maximum preference was observed in grains of ICCV 2 (7.50), which was statistically inferior, followed by KAK 2 (6.50 adults), VIHAR (6.31 adults), and JGK 2 (6.00 adults). In these cultivars, the adult's attraction ranged from 6.00 to 7.50. The results are similar with findings of F. A. Shaheen *et al.* (2006) [13] who reported antixenosis in chickpea, wherein the minimum adults (2.96) of pulse beetle were attracted towards Parbat grains and the maximum adults (5.07) were attracted towards Flip 97-192 C.

Table 1: Evaluation of antibiosis mechanism for *C. chinensis* on different genotypes of chickpea

Genotypes	Number of adults emerged	Number of holes /grains	Adults' emergence%	Weight loss%	Growth Index	Adult weight (mg)
JAKI 9218	5.10 (13.05)	1.233 (6.38)	16.72 (24.14)	5.831 (13.97)	0.040 (1.15)	5.10 (13.05)
NBeG 119	8.76 (17.22)	1.74 (7.58)	20.98 (27.26)	9.733 (18.18)	0.046 (1.23)	5.88 (14.03)
JGK 2	12.64 (20.83)	1.92 (7.96)	26.78 (31.16)	16.62 (24.06)	0.049 (1.27)	6.90 (15.23)
IG 72933	3.95 (11.46)	0.93 (5.53)	15.60 (23.26)	3.65 (11.01)	0.041 (1.16)	5.06 (13.00)
RVG 204	12.01 (20.28)	1.79 (7.69)	26.50 (30.98)	9.535 (17.99)	0.046 (1.23)	6.71 (15.01)
JG 14	5.92 (14.08)	1.367 (6.71)	17.55 (24.77)	7.604 (16.01)	0.044 (1.20)	5.13 (13.09)
PI 599066	0.00 (0.00)	0 (0.00)	0.00 (0.00)	0.792 (5.11)	0.000 (0.00)	0 (0.00)
NBeG 47	8.21 (16.65)	1.56 (7.17)	20.50 (26.92)	7.703 (16.11)	0.046 (1.23)	5.54 (13.61)
NBeG 3	10.23 (18.65)	1.77 (7.65)	23.40 (28.93)	14.33 (22.24)	0.046 (1.23)	5.91 (14.07)
VIHAR	14.50 (22.38)	2.06 (8.25)	29.28 (32.76)	18.04 (25.13)	0.048 (1.26)	6.99 (15.33)
KAK 2	16.30 (23.81)	2.11 (8.35)	30.63 (33.60)	22.18 (28.10)	0.048 (1.26)	7.24 (15.61)
ICC 506 E B	5.70 (13.81)	1.33 (6.62)	17.06 (24.40)	4.021 (11.57)	0.042 (1.17)	5.11 (13.06)
ICCC 37	7.50 (15.89)	1.43 (6.87)	18.93 (25.79)	8.587 (17.04)	0.053 (1.32)	5.27 (13.27)
IG 72953	3.60 (10.94)	0.767 (5.02)	15.37 (23.08)	1.511 (7.06)	0.038 (1.12)	5.02 (12.95)
ICCV 2	18.20 (25.25)	2.33 (8.78)	31.14 (33.92)	23.400 (28.93)	0.049 (1.27)	7.69 (16.10)
SE d±	1.10	0.31	0.81	0.60	0.09	0.66
C.D @0.05%	2.26	0.64	1.67	1.51	0.22	1.36

Figures in the parentheses are angular transformed values

Table 2: Evaluation of antixenosis mechanism for bruchids *C. chinensis* on different genotypes of chickpea

Sl. No.	Genotypes	Antixenosis
1	JAKI 9218	3.06 (10.09)
2	NBeG 119	4.90 (12.79)
3	JGK 2	6.00 (14.18)
4	IG 72933	2.33 (8.79)
5	RVG 204	5.80 (13.94)
6	JG 14	3.80 (11.24)
7	PI 599066	0.16 (2.33)
8	NBeG 47	4.60 (12.38)
9	NBeG 3	5.20 (13.18)
10	VIHAR	6.31 (14.55)
11	KAK 2	6.50 (14.77)
12	ICC 506 E B	3.31 (10.49)
13	ICCC 37	4.10 (11.68)
14	IG 72953	1.622 (7.32)
15	ICCV 2	7.50 (15.89)
16	SE d±	0.272
17	C.D @ 0.05%	0.55

Figures in the parentheses are angular transformed values

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