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Relative susceptibility of chickpea varieties to pulse bruchid, *Callosobruchus maculatus* (F.)

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Abstract

An experiment was conducted to know the relative susceptibility of chickpea varieties to pulse bruchid under laboratory conditions. A total of 16 varieties were screened for their susceptibility and based on the parameters *viz.*, oviposition, adult emergence and grain damage, they were categorized into less susceptible, moderately susceptible and highly susceptible groups. The varieties; NBeG 511, JAKI 9218 and JG 11 had less number of eggs and adult emergence, with low per cent grain damage were categorized as "Less susceptible" varieties under free-choice conditions. Similarly the varieties; NBeG 458, NBeG 471, NBeG 732 and KAK 2 which recorded higher numbers of eggs, adult emergence and high per cent grain damage were placed in "Highly susceptible" group.

Keywords: Callosobruchus maculatus, chickpeas, relative susceptibility

Introduction

Pulses constitute a major source of dietary protein for majority of the vegetarian population of India. Chickpea (Cicer arietinum L.) is one of the most important pulse crops extensively grown in dry and rain-fed areas of India. India is the largest producer of chickpea with a share of about 70 % in area and 67 % in the production of chickpea in the world ^[1]. During 2016-17, a total of 3.81 Lakh tonnes of chickpea was produced in Andhra Pradesh from an area of 3.97 Lakh ha ^[2]. The total loss of chickpea produce at national level during harvest and post-harvest handling was 8.41% with an estimated monitory loss of Rs. 2453 Crore which includes 1.18% loss in storage [3]; while the storage losses were mainly attributed to bruchids. *Callosobruchus* sp (Bruchidae: Coleoptera) commonly designated as seed weevils/bruchids are the major insect-pests associated with stored pulses [4]. The Callosobruchus maculatus (F.) is a cosmopolitan pest that can cause substantial losses in stored chickpeas ^[5], even up to 100 per cent as reported in tropical countries like India^[6] and render the grain unsuitable for food or seed within 4–6 months ^[7]. These insects multiply at a rapid rate in suitable environmental conditions such as high humidity and optimum temperature conditions^[8]. The storage insect pest management is mostly relied on the use of synthetic insecticides and fumigants for the past several years; however, their continued usage has led to a number of problems including insect resistance, toxic residues in food grains, and environmental pollution. Breeding legume crops to improve their resistance against storage insect pests, is an environment-friendly technology ^[9], the development and use of resistant chickpea cultivars offers a simple, cheap and attractive approach for the reduction of bruchid damage. Pulse bruchids exhibit preference for oviposition among various legume seeds ^[10], while the seed characters such as; seed type, seed coat, texture and colour have been reported to affect oviposition [11]. However, the association between preferences for oviposition and further survival of the offspring would help in identifying the leguminous seed that probably could be used against the beetle as a component of integrated pest management ^[12]. With this in view, the present investigation was made to identify the resistant and susceptible chickpea varieties based on the varied response of the pulse bruchid.

Materials and Methods

During 2016-17, a total of eight varieties of chickpea; NBeG 3, NBeG 47, NBeG 49, NBeG 119, Vihar, JAKI 9218, JG 11 and KAK 2 were screened against pulse bruchid under nochoice and free-choice conditions. During 2017-18, as many as 16 chickpea varieties including the earlier set, *viz;* NBeG 3, NBeG 47, NBeG 49, NBeG 119, NBeG 399, NBeG 452, NBeG 458, NBeG 471, NBeG 507, NBeG 511, NBeG 732, Vihar, ICCV 10, JAKI 9218, JG 11 and KAK 2 were screened for resistance to pulse bruchid under free-choice conditions. All these varieties were obtained from Regional Agricultural Research Station, Nandyal. Initially, they were disinfested by fumigating with aluminium phosphide for seven days and were well aerated before the use.

Rearing of the test insect: The insect culture was established on fresh produce of chickpea introducing few pairs of pulse beetles collected from the local godowns. About 250 g of chickpea grains were taken in plastic jars (500 ml capacity) and 50 freshly emerged adults of *C. maculatus* were released into each jar. After allowing oviposition by females for a week the adults were removed. The containers were covered with muslin cloth and kept at room temperature $(27\pm3^{\circ}C)$ and 75% relative humidity throughout the period of study. The adults emerged from this culture were utilized for the experiments.

No-choice test: Healthy chickpea grains (100 g) of each variety were taken in a plastic jar and five pairs of newly emerged *C. maculatus* beetles were introduced into each jar. The adult beetles were removed after 3 days and the number of eggs laid on grains of each variety was counted by observing under illuminated magnifying lens. Later the jars were kept under laboratory conditions till the emergence of adults. The adults of *C. maculatus* that emerged from different varieties were counted at 40 days after release of insects.

Free-choice test: The relative preference of C. maculatus to the chickpeas of different varieties was observed under free choice condition, where the insects were allowed to choose their preferred variety of grains. Hundred grains of each chickpea variety were taken in individual plastic cups and all the eight cups with grains were arranged equidistantly in a circle in a plastic tray. Thirty newly emerged pulse beetles were released in the middle to choose the grains of their choice variety; this was closed immediately with another tray and secured both the trays tightly with binder clips. After allowing for three days, the adults were carefully removed and the grains were transferred into individual jars. Oviposition on chickpea grains was observed under the magnifying lens and counted. Later the jars were kept undisturbed till the emergence of adults under laboratory conditions. Adult emergence was recorded at 40 DAR. Similarly in the following year, fifty newly emerged pulse beetles were released in the centre of sixteen varieties of chickpeas which were placed equidistantly. Data on adult emergence at 40 and 80 days after release of insects were recorded and was pooled to get the total number of adults emerged. At 80 days after release of insects, the damaged grains were separated from the sample, counted and expressed as per cent grain damage. The number of exit holes per 10 randomly selected grains was also recorded. All the experiments were conducted in completely randomized block design (CRBD) and were replicated thrice. The data obtained were suitably transformed and analysed for comparison.

The categorization of chickpea varieties into less susceptible, moderately susceptible and highly susceptible groups ^[13] was done based on oviposition, adult emergence, grain damage and number of exit holes per 10 grains. Grain characters such as grain type (*desi/kabuli*), size, test weight, colour, testa nature (wrinkled /smooth) and thickness were also recorded.

For measuring the thickness of testa, the grains were soaked for 2 hours to peel off the seed coats, then hot air (50 °C) dried in a tray dryer for 24 hrs for ensuring complete removal of moisture and measured in using a digital vernier callipers.

Results and Discussion

The data on oviposition and adult emergence of pulse bruchid in different chickpea grains under no-choice and free choice conditions are presented in Table 1. The results indicated that the insects showed varied response to chickpea grains of different varieties for oviposition and consequent population buildup. None of the varieties was free from oviposition by pulse bruchid. The number of eggs laid varied from 3.70 to 40.30 per 100 grains in different varieties. Lowest number of eggs was laid in NBeG 3 (3.70) and was at par with JAKI 9218 (5.30). The highest number of eggs was recorded in NBeG 119 (40.30) and was followed by Vihar (37.70 eggs/100 grains).

There were significant differences in the number of adults emerged from the grains of different chickpea varieties under no-choice. The number of adults emerged at 40 days after release of insects was lowest in NBeG 3 (0.67) and the highest number of adults emerged recorded in NBeG 119 (23.33) which was followed by Vihar (21.33 adults). Under free-choice condition, the number of eggs laid by C. maculatus varied from 3.80 to 42.50 eggs/100 grains. The lowest number of eggs per 100 grains was recorded in JAKI 9218 (3.80) which were at par with NBeG 49 (4.30) and NBeG 3 (6.0). The highest oviposition was observed in NBeG 119 (42.50) which was followed by Vihar (36.80 /100 grains). Similar trend was observed with adult emergence also at 40 days after release of insects. The minimum number of adult emergence was noticed in varieties; JAKI 9218 (0.75) and NBeG 3 (0.75) which were at par with NBeG 49 (1.75). The maximum number of adult emergence was recorded in Vihar (45.5) followed by NBeG 119 (24.25) which was at par with KAK 2 (18.25). Thus based on population build up of pulse bruchid in both no-choice and free-choice conditions, the chickpea varieties; JAKI 9218 and NBeG 3 were found less susceptible while the varieties; NBeG 119 and Vihar were found to be highly susceptible.

Under free-choice conditions during 2017-18, oviposition by C. maculatus was maximum on KAK 2 (82.33 eggs/100 grains) followed by NBeG 732 (36.33) and ICCV 10 (34.33 eggs/100 grains) (Table 2). Whereas, the varieties; NBeG 511 (3.33) and JAKI 9218 (3.67) recorded the lowest oviposition followed by JG 11 (6.0). Similarly, the total adult emergence after 80 days of insect release and the percent grain damage were recorded significantly less in the three varieties; JAKI 9218 (0.33), JG 11 (1.0) and NBeG 511 (1.33). Pulse bruchid adults were emerged in maximum numbers from KAK 2 (152.67) and NBeG 732 (132.67) followed by NBeG 458 (112.0) and NBeG 471 (110.33). Similar differential reaction of C. chinensis to chickpea cultivars was observed in terms of oviposition, adult emergence and developmental period ^[14]. Likewise, chickpea variety PKG 1 was the most susceptible to C. chinensis as it recorded the highest preference for oviposition and the highest number of adult emergence ^[15]. The percent grain damage recorded was ranged from 92.0 to 97.0 while the numbers of insect exit holes per 10 grains ranged from 39.0 to 46.33 in those four varieties. The varieties; JAKI 9218 (0.33), JG 11 (0.67) and NBeG 511 (0.67) recorded the minimum numbers of exit holes. In the same way, a chickpea variety Bittle-98 was found to be

resistant against C. chinensis^[16].

Based on the response of bruchids viz., oviposition, population build up, grain damage and number of insect exit holes to different varieties chickpeas under free-choice conditions, the varieties were categorized into three groups *i.e.*, less susceptible, Moderately susceptible and highly susceptible varieties. Thus, three varieties namely; NBeG 511, JAKI 9218 and JG 11 were categorised as less susceptible varieties as the insect emergence, grain damage and number of insect exit holes were significantly minimum compared to other varieties (Table 3). Whereas the chickpeas of NBeG 458, NBeG 471, NBeG 732 and KAK 2 were placed in highly susceptible category due to observance of higher insect emergence and their damage. The remaining varieties were categorized in "Moderately susceptible" group. The varieties; NBeG 511 and JAKI 9218 were least preferred for oviposition by pulse bruchids while NBeG 732 and KAK 2 were highly preferred compared other varieties. Though JG 11 was a moderately susceptible variety in terms of oviposition preference, the insect emergence and grain damage were bare minimum placing it in the resistant group. The test (100 grain) weight ranged from 18.52 g (ICCV 10) to 43.52 g (NBeG 458), while the testa thickness varied between 0.02 mm to 0.12 mm (Table 4). The seed coat thickness was measured as the least (0.02 mm) for the extra large sized grain variety, NBeG 458. The three less susceptible varieties; NBeG 511, JAKI 9218 and JG 11 were brown coloured "desi" varieties with medium sized grain covered by wrinkled testa. Their test weight varied between 21.08 and 28.04 g and the thickness of testa ranged from 0.09 to 0.11 mm. Whereas. the varieties in the highly susceptible category; NBeG 458,

NBeG 471, NBeG 732 and KAK 2, were white coloured "kabuli" type with test weights ranging from 37.49g (NBeG 471) to 43.52 g (NBeG 458). The present studies are in conformity with earlier researchers ^{[17], [18]} who also found that the genotypes of chickpea had smooth, soft, thin seed coat, light colour, bigger grain size and supported higher emergence of adult beetles. However, the ICCV 10 which is a desi brown variety with small sized grain (test weight of 18.52 g) and thick seed coat (0.09 mm) showed moderate susceptibility to bruchid. This finding was in accordance with the report that ICCV 10 was the most susceptible whereas ICCV 03311 was the least susceptible variety among the twelve varieties of chickpea screened ^[19]. The observance of more number of exit holes (41.33 to 46.33 per 10 grains) in susceptible varieties signifies the preference by females of C. maculatus for oviposition on the large sized grains probably because sufficient food was available for the individual larvae developing within the seed without competition. This is in agreement with the observation that increased surface area account for overall increase in egg deposition ^[20]. The number of eggs laid per seed was positively correlated with the surface area of cowpea seeds ^[21]. Similar results were with Muhammad and Maqbool (2005) who reported The preference of the bruchid for oviposition on a host was based on sensory nature as lower of eggs were laid on the wrinkled and black grain genotypes of chickpea [22]. Nevertheless, it was concluded that in addition to egg counts, adult emergence, growth index and per cent weight loss are the most reliable indicators for resistance of cowpea to damage by bruchid^[20].

Table 1: Oviposition and adult emergence o	f pulse bruchid in different chickpea varieties (2016-17)

S. No.	Variety	No of eggs/100 grain	Adult emergence (No.) at 40 DAR	No of eggs/100 grain	Adult emergence (No.) at 40 DAR	
		No-c	choice test	Free-choice test		
1	NBeG 3	3.70 (1.92) ^a	0.67 (1.05) ^a	6.0 (2.44) ^a	0.75 (1.09) ^a	
2	JG 11	16.70 (4.08) ^c	2.0 (1.56) ^{ab}	26.0 (5.09) ^b	7.75 (2.85) ^b	
3	NBeG 47	11.0 (4.59) ^{cd}	8.67 (3.01) ^c	33.80 (5.76) ^c	16.0 (4.05) ^c	
4	NBeG 49	8.0 (2.83) ^b	3.0 (1.86) ^b	4.30 (2.04) ^a	1.75 (1.41) ^a	
5	KAK 2	28.0 (5.26) ^d	18.67 (4.37) ^d	22.50 (4.75) ^b	18.25 (4.33) ^{cd}	
6	Vihar	37.70 (6.11) ^e	21.33 (4.66) ^d	36.80 (6.05) ^{cd}	45.5 (6.78) ^e	
7	JAKI 9218	5.30 (2.30) ^{ab}	3.67 (2.03) ^b	3.80 (1.92) ^a	0.75 (1.05) ^a	
8	NBeG 119	40.30 (6.33) ^e	23.33 4.86) ^d	42.50 (6.52) ^d	24.25 (4.92) ^d	
	CD (p=0.05)	0.82	0.61	0.56	0.69	

The values in parentheses are transformed values; DAR: Days after release of insects. In each column values with similar alphabet do not vary significantly at P=0.05

C No Voriety		No. of acce/100 cmoine	Adult emergence (No.)			Grain	No. of exit	
S. No.	Variety	No. of eggs/100 grains	At 40 DAR At 80 DAR		Total	damage (%)	holes/10 seeds	
1	NBeG 3	7.0(2.61) ^{bc}	1.67(1.46) ^b	5.0(2.34) ^b	6.67(2.66) ^b	11.33(19.64) ^{bc}	6.33(2.60) ^{de}	
2	NBeG 47	17.33(4.17) ^e	4.0(2.10) ^c	13.33(3.69) ^c	17.33(4.19) ^c	15.67(23.22)bc	3.67(2.03) ^{cd}	
3	NBeG 49	9.0(3.0) ^{cd}	4.0(2.04) ^c	20.67(4.59) ^d	24.67(4.98) ^c	22.67(28.39) ^{cd}	4.67(2.25) ^{de}	
4	NBeG 119	22.33(4.73) ^f	17.67(4.26) ^f	55.67(7.48) ^f	73.33(8.59) ^e	92.67(76.77) ^f	29.0(5.42) ^h	
5	NBeG 399	8.67(2.92) ^{bcd}	8.33(2.97) ^d	40.67(6.40) ^e	49.0(7.02) ^d	33.67(37.23) ^d	14.67(3.89) ^g	
6	NBeG 452	8.33(2.88) ^{bcd}	4.0(2.10) ^c	18.0(4.28) ^{cd}	22.0(4.72) ^c	7.33(15.65) ^b	2.33(1.56)bc	
7	NBeG 458	27.67(5.25) ^g	26.0(5.14) ^g	86.0(9.30) ^{gh}	112.0(10.61) ^f	94.33(76.99) ^f	41.33(6.46) ⁱ	
8	NBeG 471	24.33(4.92) ^{fg}	19.67(4.47) ^f	90.67(9.55) ^{hi}	110.33(10.52) ^f	92.0(76.28) ^f	43.33(6.62) ⁱ	
9	NBeG 507	11.33(3.36) ^d	6.67(2.66) ^d	39.0(6.27) ^e	45.67(6.78) ^d	23.0(28.58) ^{cd}	8.0(2.91) ^{ef}	
10	NBeG 511	3.33(1.82) ^a	1.33(1.35) ^{ab}	$0.0(0.70)^{a}$	1.33(1.35) ^a	1.33(3.97) ^a	0.67(1.05) ^{ab}	
11	NBeG 732	36.33(6.02) ^h	26.67(5.22) ^g	106.0(10.31) ^j	132.67(11.53) ^g	97.0(80.28) ^f	46.33(6.83) ⁱ	
12	Vihar	8.33(2.88) ^{bcd}	7.0(2.74) ^d	43.0(6.59) ^e	50.0(7.10) ^d	54.67(47.71) ^e	23.0(4.83) ^h	
13	ICCV 10	34.33(5.85) ^h	12.33(3.57) ^e	74.33(8.63) ^g	86.67(9.32) ^e	66.33(54.78) ^e	11.0(3.36) ^{fg}	
14	JAKI 9218	3.67(1.92) ^a	0.33(0.88) ^a	$0.0(0.70)^{a}$	0.33(0.88) ^a	0.33(2.19) ^a	0.33(0.88) ^a	

15	JG 11	6.0(2.45) ^b	1.0(1.23) ^{ab}	$0.0(0.70)^{a}$	$1.0(1.23)^{a}$	$1.0(3.97)^{a}$	0.67(1.05) ^{ab}
16	KAK 2	82.33(9.07) ⁱ	49.33(7.04) ^h	103.33(10.17) ^{ij}	152.67(12.35) ^g	95.0(78.63) ^f	39.0(6.27) ⁱ
	CD (p=0.05)	0.49	0.48	0.75	0.84	9.72	0.66

The values in parentheses are square root and angular transformed values; DAR: Days after release of insects. In each column values with similar alphabet do not vary significantly at P=0.05

Parameter	Less Susceptible (<mean-sd)< th=""><th>Moderately Susceptible (Mean-SD to Mean+SD)</th><th>Highly Susceptible (>Mean+SD)</th></mean-sd)<>	Moderately Susceptible (Mean-SD to Mean+SD)	Highly Susceptible (>Mean+SD)
Oviposition /100 grains (No.) *Mean = 3.89 SD = 1.89	NBeG 511 and JAKI 9218	NBeG 3, NBeG 47, NBeG 49, NBeG 119, NBeG 399, NBeG 452, NBeG 458, NBeG 471, NBeG 507, Vihar, ICCV 10 and JG 11	NBeG 732 and KAK 2
Adult emergence (No.) Mean = 55.33 SD = 49.77	NBeG 511, JAKI 9218 and JG 11	NBeG 3, NBeG 47, NBeG 49, NBeG 119, NBeG 399, NBeG 452, NBeG 507, Vihar and ICCV 10	NBeG 458, NBeG 471, NBeG 732 and KAK 2
Grain damage (%) Mean = 44.40 SD = 38.75	NBeG 511, JAKI 9218 and JG 11	NBeG 3, NBeG 47, NBeG 49, NBeG 399, NBeG 452, NBeG 507, NBeG 511, NBeG 732, Vihar and ICCV 10	NBeG 119, NBeG 458, NBeG 471, NBeG 732 and KAK 2
Exit holes/10 grains (No.) *Mean = 3.62 SD = 2.14	NBeG 511, JAKI 9218 and JG 11	NBeG 3, NBeG 47, NBeG 49, NBeG 119, NBeG 399, NBeG 452, NBeG 507, Vihar and ICCV 10	NBeG 458, NBeG 471, NBeG 732 and KAK 2

*Mean and standard deviation were worked out with square root transformed values.

Table 4: Grain characters of chickpea varieties

S. No.	Variety	Туре	Colour	Skin type	Size	Test Weight (g)	Testa thickness (mm)
1	NBeG 3	Desi	Brown	Wrinkled	Medium	28.36 ^g	0.12ª
2	NBeG 47	Desi	Brown	Wrinkled	Medium	26.63 ^h	0.11 ^{ab}
3	NBeG 49	Desi	Light Brown	Wrinkled	Medium	27.19 ^{gh}	0.12 ^a
4	NBeG 119	Kabuli	White	Wrinkled	Bold	36.94 ^d	0.06 ^e
5	NBeG 399	Kabuli	White	Wrinkled	Bold	39.27 ^{bc}	0.05 ^{ef}
6	NBeG 452	Desi	Brown	Wrinkled	Medium	22.52 ^{ij}	0.10 ^{bc}
7	NBeG 458	Kabuli	White	wrinkled	Extra Large	43.52 ^a	0.02 ^h
8	NBeG 471	Kabuli	White	Semi-wrinkled	Bold	37.49 ^d	0.05^{fg}
9	NBeG 507	Desi	Brown	Wrinkled	Medium	31.35 ^f	0.11 ^a
10	NBeG 511	Desi	Brown	Wrinkled	Medium	21.08 ^j	0.09 ^d
11	NBeG 732	Kabuli	White	Wrinkled	Bold	39.95 ^b	0.04 ^g
12	Vihar	Kabuli	White	Wrinkled	Medium	33.55 ^e	0.05^{fg}
13	ICCV 10	Desi	Brown	Wrinkled	Small	18.52 ^k	0.09 ^d
14	JAKI 9218	Desi	Brown	Wrinkled	Medium	28.04 ^{gh}	0.11 ^a
15	JG 11	Desi	Brown	Wrinkled	Medium	22.82 ⁱ	0.10 ^{cd}
16	KAK 2	Kabuli	White	Wrinkled	Medium	38.33 ^{cd}	0.04^{g}
	CD (p=0.05)					1.51	0.01

The values in parentheses are transformed values; DAR: Days after release of insects

Conclusion

The information generated may be useful in programming grain protection schedules during storage and also the varieties; NBeG 511, JAKI 9218 and JG 11, which were categorised as less susceptible group can be used in genetic improvement of chickpea crop in terms of stored grain insect pest resistance.

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