Research Article

Relative Resistance of Seed of Advance Genotypes of Cicer arietinum Against Callosobruchus Chinensis During Storage

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Abstract | Pulse beetle *(Callosbruchuschinesis)* causes serious damage to the seed of chickpea, and other pulses during storage. Seed viability and other quality parameters are affected by its infestation. This pest is controlled by phosphine fumigation and synthetic insecticides. Unfortunately, this pest has developed resistance against these pesticides. Moreover, with application of these pesticides, chemical residues may cause severe damage to non-target organisms. Host plant resistance against insect pests is considered best ecological approach to pest management. Keeping this in consideration, the current research activity is planned to screen out available germplasm of chickpea against dhora beetle *(Callosobruchuschinisensis)*. For this purpose, 8 genotypes of chickpea genotypes were taken from Ayub Agricultural Research Institute, Faisalabad. 1 kg of each genotype was taken in plastic jars. There were three replications of each genotype for each insect. Twenty adults of dhora beetle were discharged in each container which were enclosed with muslin cloth with rubber bands. The Jars were placed at room temperature. Data of egg laying were observed after five days of release of insects. Data of emergence F1 adults were observed after 30 days of storage. At this time percent weight loss and Seed viability was also analyzed. The Same data were observed after two- and three-month storage period. The results revealed that all the varieties/lines differed significantly from one another for percent weight loss and viability. On the basis of percent weight loss, the variety Bittal-2016 was found to be the most susceptible. Whereas the variety Niab-CH-2016 was also found to be the least susceptible. The current research found that none of the chickpea cultivars were totally resistant to *C.chinensis*, however their vulnerability varied significantly. Finally, the acquired data will be statistically examined for the study of variance via statistical software. Means of significant treatments will be examined at = 5% using the HSD-Tuckey test.

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Introduction

Pulses are valuable crops that are cultivated in most regions of the world in many forms (Alemayehu and Getu, 2015). Among the world's pulses, chickpea ranks second in terms of growing area and third in terms of productivity (CGIAR, 2017). Pakistan produces 0.5 million tonnes of dry seed per year from an area of around one million hectares (FAO, 2016). Pulses are a good source of protein (20-40%), carbs (50-60%), and thiamin, niacin, calcium, and iron (Bhalla *et al*., 2008), and is well-regarded as a source to reduce cholesterol levels by a wide variety of preparations in our kitchen (Pittaway *et al*., 2006). Chickpeas are divided into two types based on their features and seeds, such as Desi and Kabuli (Meuhlbauer and Singh, 1987). Kabali chickpeas are significantly larger, have a creamy colour with a smooth surface, and have white blossoms, however they lack anthocyanin. The chickpeas are one of the most popular in most continents and have been consumed across the world for more than 7,500 years. Because of its nitrogen fixation capabilities and as a source of significant nutritional protein for human consumption, this plant is critical to maintaining soil fertility (Kantar *et al*., 2007).

During storage, losses include both quantity and quality, which can be attributed to insects, rodents, mites, birds, and microbiological agents, as well as moisture, etc. The insect-pest in the store inflict substantial harm in the tropical and temperate climates approximately 20-35% and 5-10% accordingly (Nakakita, 1998). The pulse beetle, *Callosobruchus chinensis L.* (*Bruchidae: Coleoptera*), also is a major insect pest of preserved chickpeas (Rajasri and Rao, 2012). Due to the assault of the pulse beetle, *Callosobruchus chinensis*, on understorages, considerable post-harvest losses have been observed (Zia *et al*., 2011), causing around 10% damage and rendering grains unsuitable for human consumption (Aslam, 2004). The beetle in storage where beetle pass from seed to seed is responsible for the severe loss of quality and value in seeds (Bhattacharya and Banerjee, 2001). It is amongst the most destructive pest due to storage of up to 40%-50% (Gosh and Durbey, 2003). Both in the field and in storage, *Callosobruchus chinensis* is vulnerable to pulses that damage chickpea seeds.

The agricultural industries are presently gaining impetus in their efforts to investigate grain resistance to insect pest storage and the usage of plant products. In the past years an alternative, eco-friendly environment has gained pace to decrease the use of chemical insecticides to manageinsect pests of stored grain. Efforts are being made to develop solutions for the management of hazardous pesticides which do not impact people, animals or profitable insects, except in terms of the safety, ease of biodegradation, cost and availability of these to farms. Therefore, new resistence/tolerance sources for the cultigens and the ancient chickpea races against bruchids can also be identified to help integrate these factors in the creation of new cultivars of resistant/tolerant.

Resistant cultivars have been an important component in the success of many ongoing insect pest management efforts. One of the promising approaches to reducing pesticide reliance in agriculture, particularly in Pakistan, is to plant insect resistant cultivars, which is among the best, practical, temperate, and ecologically safe insect pest administration (Pedigo,1996) and completely legitimises the upcoming WTO guidelines.

The good seed storage in future is dependent on the host grain resistance to overcome losses from insect pest infestation and it can be included in upcoming breeding programmes. Ahmed *et al*. (1993) reported that cultivars with hard seed surface indicated protection from beetle. Coefficients of phenotypic and genotypic assortments were exceptionally emphatically related with hurt seeds and development holes. The present examination is planned to discover chickpea cultivars having resistance against pulse beetle, which further could be utilized for hybridization to limit pesticide use against pulse beetle. The current study was conducted to attain the following objectives. (i) To check the relative resistance and susceptibility of eight genotypes of chickpea against *Callosbruchuschinesis*. (ii) Of compare the quantitative and qualitative losses to genotypes of pulses caused to *C. chinensis* infestation.

Materials and Methods

Testing Varieties

Ayub Agriculture Research Institute in Faisalabad provided uninfested seeds of approved varieties of chickpea. The Department of Entomology at the University of Agriculture Faisalabad conducted research on the relative resistance of eight genotypes of chickpea to the attack of *Callosobruchus chinensis* during storage. Genetic materials were comprised chickpea genotypes Balkassar-200, Noor-2013, Bhakkar-2011, Niab-CH-2016, Bittal-2016, Tamman-2013, Punjab-2008 and Wanhar-2000. The seeds of each genotype were examined under a binocular microscope to check that no insects had previously infested them and that no eggs had been placed on the seeds of the selected genotypes. All of the test cultivars' sound grains were conditioned in the laboratory for two weeks at the same temperature and relative humidity as the experiment. Before experimentation the moisture content and grain size of all the test varieties were determined.

Testing Insect

The adults of *Callosobruchus chinensis* were collected from infested pulsed stored in grain markets and farmer storages. For the experiment, seeds from each assortment (each comprising 1kg of seeds) were placed separately in glass jars with a capacity of 250 ml. Each container was considered one replication, and three replicates of each genotype were used in this test. These beetles were released on un infested sterilized seeds of chickpea to get homogeneous insect culture. For this purpose, the beetles were separated from seeds after three days. The seeds having eggs of these beetles were again put into the jars and will be placed in cooled incubators at optimum growth conditions (25°C and 75 % R.H) for one month to get population of the same age. The newly grown-up adult was checked day by day and the recently grown-up insects were utilized for the trial. This fundamental population was referred to as the stock culture.

Experimental protocol

During the free choice testing, the processes described in all chickpea varieties undergoing a free *C.chinensis* attack were exposed to small alterations in accordance with the protocols indicated by (Raina,1971). Each variety's seeds are placed in a plastic container for this test. Three repetitions with different genotypes for each plastic basin were carried out as a single replication for that test. Using an aspirator apparatus, ten pairs of adulthood of *C. chinensis* aged 0–24 hours were collected and discharged in every plastic basin. To prevent adults from escape *C. chinensis* and to allow for air circulation, the basins are covered in plastic and the edge of the lid is placed in the bottom. These beetles may stay in it for up to a week before being removed. The genotype was evaluated on a biweekly base in order to record by visual inspection the number of harmed seeds in each variety and all experimentation was conducted at a relative humidity of 28.0 ±5°C 70.0 ±5 percent and photoperiods atL:12 & D:12 hours. *C. chinensis*-injured seeds may be seen in openings inside the seed coat "flap" generated by immature adults (Ahmed *et al*., 1989; Riaz *et al*., 2000).

Statistical analysis

The results for weight loss and viability % were statistically examined using analysis of variance techniques. The Tukey-HSD test was used to determine the significance of treatment means at a 5% probability level. The coefficient of correlations was worked out to study the relationship between *Callosobruchus chinensis* susceptibility and various grain characteristics.

Table 1A: *Analysis of variance of different varieties of chickpea for percent weight loss.*

SOV.	D.F.	S.S.	M.S.	F.Value	p. Value
Variety	7	5694.48	813.49	8966.88	0.0000
Error	16	13.4	60.841		
Total	235707.95				

Significant at 5% level of probability.

Table 1B: *Comparison of Means regarding of different varieties of chickpea for percent weight loss.*

Varieties	**Mean**
Balkassar-200	25.46c
Noor-2013	53.13a
Bhakkar-2011	19.55d
Niab-CH-2016	18.81d
Bittal-2016	55.24a
Tamman-2013	51.45b
Punjab-2008	23.52c
Wanhar-2000	49.10ab
H.S.D. value for varieties = 2.5965	

Means of sharing similar letters are not substatially different from the DMR test at P = 5%

Results and Discussion

Percent weight loss

The results in Table 1A show a significantly substantial difference across varieties/lines. The means were compared using the Tukey HSD test at the 0.05 level of probability, and the results are shown in Table 1B. The maximum weight loss after 60 days was found in 55.24% in Bittal-2016 and was statistically at par with Noor-2013 (53.13%), Tamman-2013 (51.45%), Wanhar-2000 (49.10%). The minimum weight loss (18.81%) was observed in Niab-CH-2016 that was statistically at par with Bhakkar-2011 (19.55%), Punjab-2008 (23.52%) and Balkassar-200 (25.46%).

Percent Viability

The data given in Table 2 show different trends percentage viability in different varieties/lines at 60 days after release of insect. Maximum viability percentage (62%) was noted in Niab-CH-2016 and was followed by those of Punjab-2008 (60.33%), Bhakkar-2011 (60%), Balkassar-200 (55.33%), Tamman-2013 (53.66%), Wanhar-2000 (52.66%) and Noor-2013 (50.66%). The minimum viability percentage was noted in Bittal-2016(40.33%).

Table 2: *Analysis of variance of different varieties/lines of chickpea for percent of viability.*

SOV.	D.F.	S.S.	M.S.	F. Value	p Value
Variety	7	410.000	58.5714	10.19	0.0001
Error	16	92.000	5.7500		
Total	23	502.000			

Significant at 5% level of probability

Varieties	Mean
Balkassar-200	55.33b
Noor-2013	50.66b
Bhakkar-2011	60.0a
Niab-CH-2016	62.0a
Bittal-2016	40.33c
Tamman-2013	53.66ab
Punjab-2008	60.33a
Wanhar-2000	52.66b
H.S.D. value for varieties = 6.7878	

Means of sharing similar letters are not substantially different from the DMR test at P = 5%

Comparison of the means of percent weight loss and viability in various chickpea varieties

The table compares the means of % weight loss and viability in several chickpea varieties and indicates that none of the types were fully resistant to pest attack, but their reaction varies enormously. The percentage weight reduction in Niab-CH-2016 was much lower, but it was significantly greater in Bittal-2016. The results regarding viability percentage showed that higher in variety Niab-CH-2016 while significantly lower in variety Bittal-2016.

Table 3: *Comparison of the means of percent weight loss and viability in different chickpea varieties.*

Variety	Weight loss (%)	Viability (%)
Balkassar-200	25.46 d	55.33 abc
Noor-2013	53.13 ab	50.66 ab
Bhakkar-2011	19.55 e	60 ab
Niab-CH-2016	18.81 e	62 a
Bittal-2016	55.24 a	40.33 a
Tamman-2013	51.45 bc	53.66 bc
Punjab-2008	23.52 c	60.33c
Wanhar-2000	49.10 c	52.66 c

Means sharing similar letters are not substantially different from the DMR test T P= 5%

Coefficient of correlation in percent weight loss and percent viability in various chickpea varieties

The coefficient of correlation between percent weight loss and percent viability in several chickpea cultivars. The table depicts the relation between the variables. The correlation coefficient value for percent weight loss and viability was 0.9206.

Coefficient of correlation in percent weight loss and viability in different chickpea varieties

Parameters	1	2
Wt. loss	1.0000	
Percent viability	0.9206**	1.0000

** = significant at the 1% level of probability.

Study was carried out to check the response of eight approved varieties of chickpea viz., Balkassar-200, Noor-2013, Bhakkar-2011, Niab-CH-2016, Bittal-2016, Tamman-2013, Punjab-2008 and Wanhar-2000 against Dhora, *Callosobruchus chinensis L.* The findings demonstrated that all of the kinds differed considerably in terms of % weight loss and viability.

Multiple factors are responsible for storing grains susceptibility against insects. Chemical nature (crude protein) and moisture content proved positive factors in decreasing and increasing percent weight loss (Aslam *et al*., 2006). Some varieties/lines are damaged at higher rates and are relatively more susceptible than others while other comparatively resistant and are damaged at lower rates. According to Kamble *et al*. (2016), medium size seeds with thin seed coat characteristics were found to be the least preferred for oviposition when compared to bold seeded varieties, while lentil and mungbean seeds were smaller in size than blackgram seeds, but egg deposition was higher on lentil and mungbean seeds. All these factors, singly or in combination provide positive or negative susceptibility index, which is physically expressed in terms of damage and losses. In this way, the status of a variety could be judged in a better way to achieve the desirable objectives of screening programs. Percent weight loss of the variety has been considered to be the main index of their susceptibility to Dhora (Aslam, 2006; Shaheen *et al*., 2006).

These parameters were used to determine the susceptibility of chickpea genotypes in the current investigations (percent weight loss and percent viability). As a consequence, these data totally reflect the prior personnel' research findings for their assessment parameters. The physic-chemical characteristics of the grains, such as size, shape, texture, colour, and chemical contents, are essential in addition to the fundamental variables responsible for the observed differences in susceptibility of gramme varieties. According to Chakraborty and Mondal (2016), the pulse beetle placed the most eggs on the bigger surface area of the seed, and piercing and initial damage resulted in grain weight loss. Demnati and Allache (2014) discovered chickpea seed weight reduction as a result of chickpea beetle infestation. These findings also show that percent weight loss has a positive and significant role in the attack of that pest on gram. According to Bharathi *et al*. (2016), *C. chinensis* was tested on eight different host-grains, including green gram *Vigna radiate L*., blackgram *Vigna mungo L*., Bengal gram *Cicer arietinum L*., redgram *Cajanus cajan L*., and cowpea. *Vigna sinensis L*., soybean *Glycine max L*., pea Pisum *sativum L*., pillipesara *Phaseolus trilobus L*., and bengal gram had the highest percentage of grain weight loss (58.55 percent).

Based on the overall findings, it is determined that the eventual sensitivity of gramme to *C.chinensis* cannot be attributable to a single cause, but that numerous variables are involved in some way. A little change in any of these variables can have an additive, complementary, or antagonistic impact. The current studies demonstrated that none of the gramme varieties/lines were entirely resistant to C.chinensis, however their susceptibility varied significantly. These findings are comparable to those of earlier researchers (Radha and susheela, 2014; Osman *et al*., 2015; Hossain *et al*., 2014; Shivanna *et al*., 2011; Badii *et al*., 2013).

To sum up, the comparative susceptibility of these varieties was found to be the following order: Balkassar-200 > Noor-2013 > Bhakkar-2011 > Niab-CH-2016 > Bittal-2016 > Tamman-2013 > Punjab-2008 > Wanhar-2000.

In the light of the above results, it is suggested that the breeders focus more on the evolution of insect-pest resistant cultivars. To withstand assault during storage, certain genetic alterations must be integrated by induced mutation. This will significantly assist to reduce insect losses. Furthermore, the provided information on insect sensitive varieties/lines will serve as a reference for planning and implementing effective control actions against storage insects such as *C. chinensis*.

Conclusions and Recommendations

The findings demonstrated that none of the varieties were totally resistant to *C. chinensis* L.'s attack. The variety Bittal-2016 was found to be the most sensitive, followed by Noor-2013, Tamman-2013, and Wanhar-2000, whereas the variety Niab-CH-2016 was found to be the least susceptible, followed by Bhakkar-2011, Punjab-2008, and Balkassar-200. There were positive and substantial correlations between percent weight loss and percent viability.

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Novelty Statement

This is the first study to look at the resistance of advanced genotypes of *cicer arietinum* seed to *callosobruchus chinensis* during storage. The study is one of the few to investigate the possible resistance of chickpea varieties to the dhora beetle in Pakistan, which should contribute to the enhancement of chickpea productivity in the Pakistan.

Author's Contribution

Muhammad Waseem Akhtar: Carried out the experiments, collected data and wrote the manuscript.
Khalid Mahmood Khawar: Contribute in planning the experiment and supervised.
Muhammad Naeem Akhtar and Muhammad Rafique: Assisted in Laboratory.
Khalid Hussain, Sharmin Ashraf and Muhammad Fida Hassan: Helped in statistical analysis figures drawing and revision of the manuscript.

Conflict of interest

The authors have declared no conflict of interest.

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