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Biochemical Changes Of Chickpea Genotypesbefore and After Infestation of Pulse Beetle, *Callosobruchus Chinensis* L. (Coleoptera: Bruchidae) During Storage

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Abstract

The pulse beetle is a field-to-store pest as its infestation on pulses often begins in the field itself as adults lay eggs on mature pods and when such seed is harvested and stored, the pest population increases rapidly and results in total destruction within a short period of 3-4 months. Keeping in view,varietal screening of fifty chickpea genotypes was carried outin the storage laboratory, Department of Entomology, OUAT, BBSR and the performance of the genotypes was assessed based on various biological parameters of test insect, damage and infestation by C. chinensis. The results indicated that none of the genotypes was completely resistant to pest attack whereas 4 genotypes (Himachal Chana 1, Dheera (NBeG-47), JG-14 and Dilaji) were found moderately resistant, 8 genotypes (Phule Vikram, JG 11, ICCV-181108, ICCV-181107, ICCV-181605, C-18203, C-18205 and C-18252) were moderately susceptible,11 genotypes (RVG-204, RVG-203, JAKI-9218, Pratap Chana, Bharati, ICCC 4, ICCV-181106, ICCV-181612, C-18206, ICCV-181101 and Radhey) were susceptible and 27 genotypes (NBeG-49, Himachal Chana 2, JG-16, JG-130, CO 4, Vishal, Kranthi, NBeG-3, ICCV-14102, ICCV-171117, C-18175, ICCV-181611,ICCV-14106, Kalahandi Local, ICC 3137, ICCL 86111, C-19162, C-19168,GNG 2207, BG 3043, GG 3, Birsa Chana 3, C 19199, RSG 963, C 19200, KPG 59and NBeG 119) were noticed to be highly susceptible. The bio-chemicalconstituents analyzed in the present studies viz., protein, phenol, ash and fibre contents of the genotypes contributed to the resistance / susceptibility of C. chinensis. Among the biochemical parameters, protein exerted significant positive effect whereas phenol, ash and fibre contents exhibited negative influence on pest infestation and development.

Keywords: *Callosobruchus Chinensis L., Chickpea Accessions, Protein, Phenol, Ash Content*

Introduction

Pulse crops occupy a unique position in Indian agricultural economy. These are annual leguminous crops from the family (Leguminoasae) yielding between one and twelve grains or seeds of variable size, shape and colour within a pod, used for both food and feed. All pulses are nutrient-dense, providing a rich source of fiber, protein, minerals (e.g., iron), and vitamins (e.g., folate). Pulses account for around 20 per cent of the area under foodgrains and contribute around 7-10 per cent of the total foodgrains production in the country. Pulse beetle, *Callosobruchus chinensis* L. (Coleoptera: Bruchidae) is widely distributed and known as a major destructive insect of stored chickpea (Park *et al*., 2003; Aslam, 2004). The economic loss of this bruchid in various pulses ranged from 30-40 per cent within a period of six months and when left un-attended, losses could be up to 100per cent (Dongre *et al*., 1996; Akinkurolere*et al*., 2006; Sharma *et al*., 2011). It is a field-to-store pest as its infestation on pulses often begins in the field itself as adults lay eggs on mature pods (Huignard*et al*., 1985) and when such seed is harvested and stored, the pest population increases rapidly and results in total destruction within a short period of 3-4 months (Rahman and Talukder, 2006). This pest exhibits definite varietal preferences, and hence it is considered as a vital component of the present studies on pulse beetle, *Callosobruchus chinensis* L.

Materials And Methods

The present study on the screening of fifty chickpea genotypes, were procured from the AICRP on Chickpea, Keonjhar Centre, OUAT, ICRISAT, Hyderabad and IIPR, Kanpur. After cleaning of genotypes, hundred numbers of healthy and disinfested seeds of fifty chickpea test genotypes were kept in glass jars of 500 g capacity separately. Five pairs of freshly emerged adult chickpea bruchids were released into each of the jars and the mouth of the jars were covered with muslin clothes and secured through rubber bands for better aeration. The adult bruchids were removed from the jars after 10 days and these jars containing the seeds were kept undisturbed for the emergence of F_1 adults.Performance of the test chickpea genotypes was assessed on the basis of bio-chemical parameters.

Bio-Chemical Parameters

Estimation of Crude Fibre

2 g of ground material with ether or petroleum ether was extracted to remove fat (initial boiling temperature 35-38^0 C and final temperature 52^0 C). After extraction with ether, 2g of dried material was boiled with 200ml of sulphuric acid for 30 minutes with bumping chips. Filtered through muslin cloth and washed with boiling water until washings were no longer acidic. Then boiled with 200ml of sodium hydroxide solution for 30 minutes. Filtered through muslin cloth again and washed with 25ml of boiling 1.25% H_2SO_4, three 50ml portions of water and 25ml alcohol. The residue was removed and transferred to washing dish (W_1). The residue was dried for 2 hours at 130 ± 2^0 C. The dish was cooled in a desiccator and weighed (W_2). Finally, the residue was ignited for 30 minutes at 600 ± 15^0 C,cooled in a desiccator and reweighed (W_3).

% crude fibre in ground sample= Loss in weight in ignition (W_2-W_1) – (W_3-W_1) / weight of the sample × 100

Estimation of total ash content

Ten gram of dry and homogenized sample was taken and spread evenly as a thin layer in a previously tarred and weighed silica dish (W_1). The material in the dish was ignited on a low flame of a burner or under low heat on an electric hot plate till the smoke/fumes ceased to appear. Then the dish was transferred into a muffle furnace; ignited at 550 ± 25^{0}C for 5-6 hours. The dish, suitably covered with the corresponding lid transferred into a desiccator only after the muffle temperature reached less than 100^0 C followed by cooling and then weighed. The process of ignition in the furnace followed by cooling and weighing was repeated until concordant weights were obtained. The final weight was noted (W_2). Total ash content (Percent by mass, on dry weight basis) – W_2-W_1/10× 100

Estimation of Protein content

Protein content was estimated by Lowry's Method developed by Lowry *et al.* (1951). In this method 0.2g of chickpea seeds were taken. Then it was macerated by pestle and mortar in 10 ml TCA (10%) solution. Each sample was transferred to separate centrifuge tube and rest was discarded. 10 ml of 1N NaOH was added to each tube and mixed well with the help of glass rod. Again, tubes were centrifuged at 10,000 rpm for 10 minutes. Supernatants were collected for true protein estimation. In this estimation process 0, 0.2, 0.4, 0.6, 0.8 and 1.0 ml of the working standards were pipette out into a series of test tubes. Then 0.2 ml of the sample extract were pipette out into the test tubes. The volume was made upto 1ml in all test tubes with water including the working standard. 10 ml of reagent 'C' was added to each tube including the blank, mixed well and allowed to stand for 10 minutes. (Reagent 'C'=50 ml of reagent 'A' mixed with 1 ml of reagent 'B'. Reagent A=2% Na_2CO_3 in 0.1 N NaOH solution, Reagent B = 0.5% $CuSO_4$ .5 H_2O in 1% K-Na-tartarate solution). Then 0.5 ml reagent D (1 N Folin - Cicalteau reagent) was added, mixed well and incubated at room temperature in the dark for 30 minutes till the blue colour was developed. Optical Density (O.D.) of samples and standards was noted at 660 nm. A standard graph was drawn and protein content was calculated. Protein content was calculated by following formula. Protein content = W × O.D value × dilution factor.

Estimation of total phenol

The method as described by Bray and Thorpe (1954) was used for the assay of total phenol in the seed sample. Exactly 0.5g to 1g of the sample was weighed and grinded with a pestle and mortar in ten times volume of 80% ethanol. The homogenate was centrifuged at 10,000 rpm for twenty minutes and supernatant was collected. The residue was re-extracted with five times the volume of 80% ethanol. The supernatant was centrifuged, pooled and evaporated to dryness. Then the residue obtained was dissolved in 5ml of distilled water.Different aliquots (0.2-2ml) pipetted out into test tubes and made up the volume in each tube to 3 ml with distilled water. Then 0.5 ml of 0.1 N Folin - Ciocalteu reagent was added and incubated for 3 minutes. After 3 minutes, 2 ml of 20% Na_2CO_3 solution was added to each tube and mixed thoroughly. The tubes were incubated in boiling water bath for one minute, cooled and the absorbance of the sample was measured at 650 nm against blank. A standard curve using different concentrations of catechol was prepared. From the standard calibration curve, the concentration of phenols in the test sample was found out and expressed as mg phenols per gram of seeds sample.

Results And Discussion

Table 1: Biochemical parameters analyzed in chickpea genotypes before infestation

Chickpea genotype	Category (As per IS)	Biochemical parameters analyzed before infestation			
		Protein (%)	Phenol (mg/100g fw)	Ash (%)	Fibre (%)
RVG-204	S	22.77	223.12	2.99	3.23
Phule Vikram	MS	20.92	248.58	3.22	4.22
NBeG-49	HS	25.14	182.31	2.27	2.58
Dheera (NBeG-47)	MR	18.67	271.65	3.61	5.58
RVG-203	S	21.92	220.43	3.04	3.67
JG-14	MR	19.45	256.78	3.44	5.27
JAKI-9218	S	22.31	218.58	3.07	4.02
Pratap Chana	S	22.96	212.24	2.88	3.58
Himachal Chana 2	HS	25.07	191.38	2.43	2.94
JG-16	HS	25.27	178.65	2.21	2.47
Dilaji	MR	19.13	264.32	3.53	5.40
JG-130	HS	23.42	201.31	2.58	2.79
CO 4	HS	24.11	185.42	2.44	2.91
JG 11	MS	20.13	245.25	3.15	4.64
Himachal Chana1	MR	18.46	294.53	3.76	5.91
Vishal	HS	23.86	192.43	2.62	3.15
Bharati	S	23.01	211.39	3.00	4.25
Kranthi	HS	24.12	189.32	2.70	3.07
ICCC 4	S	23.25	220.33	3.02	3.98
NBeG-3	HS	24.21	196.22	2.73	4.21
ICCV-181108	MS	21.33	250.09	3.30	4.35
ICCV-181106	S	22.45	215.07	2.78	3.73
ICCV-14102	HS	23.99	193.66	2.49	4.16
ICCV-171117	HS	24.17	189.79	2.38	2.74
ICCV-181107	MS	20.84	241.75	3.25	4.11
ICCV-181605	MS	21.66	230.65	3.19	4.32
C-18203	MS	20.56	235.34	3.42	3.99

ICCV-181612	S	23.13	199.97	3.10	3.89
C-18175	HS	24.22	188.28	2.75	3.97
C-18205	MS	20.85	228.71	3.27	4.13
C-18206	S	22.71	201.56	3.07	4.04
C-18252	MS	20.11	230.27	3.16	4.66
ICCV-181611	HS	23.91	202.11	2.42	3.04
ICCV-181101	S	22.79	214.44	2.97	3.86
ICCV-14106	HS	24.02	191.35	2.48	3.34
Kalahandi Local	HS	23.79	185.69	2.46	2.73
ICC 3137	HS	25.01	184.22	2.29	2.68
ICCL 86111	HS	24.78	191.26	2.54	3.09
C-19162	HS	24.43	186.25	2.34	2.82
C-19168	HS	25.00	187.18	2.32	2.62
GNG 2207	HS	24.44	200.04	2.74	3.53
BG 3043	HS	24.31	194.88	2.37	2.86
GG 3	HS	23.83	188.33	3.07	3.17
Birsa Chana 3	HS	24.28	190.81	2.73	3.36
C 19199	HS	23.95	185.89	2.31	2.71
Radhey	S	23.15	217.15	2.92	3.81
RSG 963	HS	24.37	197.73	2.51	2..96
C 19200	HS	23.98	188.56	2.58	3.23
KPG 59	HS	24.58	192.37	2.66	3.34
NBeG 119	HS	24.81	185.11	2.36	2.85
SE(m) ±	-	0.673	6.168	0.083	0.109
CD (p=0.05)	-	1.89	17.31	0.23	0.31

IS- Index of Susceptibility, MR- Moderately Resistant, MS-Moderately Susceptible, S- Susceptible, HS-Highly susceptible.

Table 2: Biochemical parameters analyzed in chickpea genotypes after infestation

Chickpea genotype	**Category (As per IS)**	**Biochemical parameters analyzed after infestation**			
		Protein (%)	**Phenol (mg/100g fw)**	**Ash (%)**	**Fibre (%)**
RVG-204	S	24.58	221.69	3.84	2.41
Phule Vikram	MS	22.61	247.56	4.11	3.37
NBeG-49	HS	28.36	180.25	3.32	1.60
Dheera (NBeG-47)	MR	19.56	271.10	4.23	4.99
RVG-203	S	23.91	218.69	3.87	2.73
JG-14	MR	19.99	256.04	4.18	4.71
JAKI-9218	S	24.25	217.24	3.87	3.01
Pratap Chana	S	25.49	210.54	3.72	2.68
Himachal Chana 2	HS	28.19	189.49	4.13	2.01
JG-16	HS	28.56	176.35	3.26	1.48
Dilaji	MR	19.78	263.57	4.22	4.79
JG-130	HS	26.28	199.34	3.85	1.78
CO 4	HS	27.04	183.38	3.63	1.98
JG 11	MS	21.89	244.34	4.34	3.63
Himachal Chana 1	MR	19.29	293.95	4.43	5.32
Vishal	HS	26.47	190.51	3.83	2.17
Bharati	S	25.83	209.61	4.06	3.23
Kranthi	HS	26.86	187.48	3.85	2.09
ICCC 4	S	25.06	218.61	3.72	2.95
NBeG-3	HS	27.19	194.36	4.04	2.98

ICCV-181108	MS	23.55	248.89	4.03	3.40
ICCV-181106	S	24.75	213.33	3.65	2.62
ICCV-14102	HS	26.84	191.76	3.62	3.12
ICCV-171117	HS	26.99	187.95	3.60	1.80
ICCV-181107	MS	22.49	240.47	4.05	3.08
ICCV-181605	MS	23.42	229.80	3.94	3.33
C-18203	MS	22.63	234.30	4.09	3.03
ICCV-181612	S	25.33	198.97	3.93	3.05
C-18175	HS	27.13	186.38	3.86	2.87
C-18205	MS	22.70	227.52	4.07	3.12
C-18206	S	25.07	199.67	3.88	3.03
C-18252	MS	21.76	228.96	3.82	3.88
ICCV-181611	HS	26.82	200.05	3.62	2.04
ICCV-181101	S	25.01	212.53	3.84	2.93
ICCV-14106	HS	26.74	189.30	3.63	2.32
Kalahandi Local	HS	26.83	183.67	3.41	1.74
ICC 3137	HS	28.22	182.18	3.40	1.79
ICCL 86111	HS	27.70	189.27	3.60	2.12
C-19162	HS	27.36	184.29	3.55	1.81
C-19168	HS	28.06	185.25	3.58	1.72
GNG 2207	HS	27.05	197.94	3.92	2.52
BG 3043	HS	26.99	192.95	3.81	1.87
GG 3	HS	26.28	186.58	3.68	2.09
Birsa Chana 3	HS	26.85	188.90	3.78	2.30
C 19199	HS	26.84	183.90	3.56	1.75
Radhey	S	25.56	215.61	3.75	2.96
RSG 963	HS	27.14	195.89	1.00	1.95
C 19200	HS	26.55	186.84	3.70	2.24
KPG 59	HS	27.34	190.43	3.69	2.32
NBeG 119	HS	27.87	183.09	3.58	1.86
SE(m) ±	-	0.744	6.122	0.111	0.083
CD (p=0.05)	-	2.09	17.18	0.31	0.23

IS- Index of Susceptibility, MR- Moderately Resistant, MS-Moderately Susceptible, S- Susceptible, HS- Highly susceptible.

Table 3: Changes in biochemical parameters after infestation

Chickpea genotype	Category (As per IS)	Protein (%)	Phenol (mg/100g fw)	Ash (%)	Fibre (%)
RVG-204	S	+7.99	-0.64	+28.42	-25.38
Phule Vikram	MS	+8.23	-0.41	+31.06	-20.14
NBeG-49	HS	+12.83	-1.13	+53.81	-40.08
Dheera (NBeG-47)	MR	+3.32	-0.21	+17.17	-10.57
RVG-203	S	+9.09	-0.79	+27.30	-25.61
JG-14	MR	+6.73	-0.29	+19.48	-10.63
JAKI-9218	S	+8.68	-0.61	+26.06	-25.12
Pratap Chana	S	+11.03	-0.80	+29.17	-27.37
Himachal Chana 2	HS	+12.45	-0.99	+44.85	-31.63
JG-16	HS	+13.01	-1.35	+63.80	-44.65
Dilaji	MR	+4.65	-0.28	+18.41	-11.29
JG-130	HS	+12.21	-0.98	+49.22	-26.44
CO 4	HS	+12.17	-1.10	+48.77	-31.21
JG 11	MS	+8.79	-0.32	+28.89	-21.76
Himachal Chana 1	MR	+2.21	-0.19	+15.43	-9.98

Vishal	HS	+10.93	-1.00	+46.48	-31.11
Bharati	S	+12.24	-0.84	+37.83	-24.00
Kranthi	HS	+11.36	-0.97	+42.59	-31.92
ICCC 4	S	+7.78	-0.78	+23.19	-25.88
NBeG-3	HS	+12.30	-0.95	+47.99	-31.12
ICCV-181108	MS	+10.41	-0.48	+30.69	-21.84
ICCV-181106	S	+10.26	-0.81	+31.29	-29.76
ICCV-14102	HS	+11.88	-0.98	+44.57	-25.00
ICCV-171117	HS	+11.66	-0.97	+41.88	-34.31
ICCV-181107	MS	+7.92	-0.53	+24.62	-25.06
ICCV-181605	MS	+8.12	-0.37	+23.51	-22.92
C-18203	MS	+10.07	-0.44	+33.33	-24.06
ICCV-181612	S	+9.52	-0.66	+26.77	-21.59
C-18175	HS	+12.02	-1.01	+40.73	-27.71
C-18205	MS	+8.85	-0.52	+25.08	-24.46
C-18206	S	+10.37	-0.94	+26.38	-25.01
C-18252	MS	+7.21	-0.57	+22.78	-16.74
ICCV-181611	HS	+12.18	-1.02	+49.59	-32.89
ICCV-181101	S	+9.76	-0.89	+29.29	-24.09
ICCV-14106	HS	+11.32	-1.07	+43.55	-30.54
Kalahandi Local	HS	+12.33	-1.09	+48.91	-36.23
ICC 3137	HS	+12.82	-1.11	+53.01	-33.21
ICCL 86111	HS	+11.78	-1.06	+41.73	-31.39
C-19162	HS	+12.19	-1.05	+50.41	-35.82
C-19168	HS	+12.25	-0.93	+51.00	-34.35
GNG 2207	HS	+10.69	-1.05	+43.07	-28.61
BG 3043	HS	+11.04	-0.99	+44.07	-34.12
GG 3	HS	+10.27	-1.03	+37.43	-34.07
Birsa Chana 3	HS	+10.57	-1.00	+38.46	-31.55
C 19199	HS	+12.06	-1.07	+49.79	-37.98
Radhey	**S**	+10.41	-0.71	+28.42	-22.31
RSG 963	HS	+11.36	-0.97	+39.84	-34.62
C 19200	HS	+10.71	-0.91	+26.53	-30.65
KPG 59	HS	+11.22	-1.01	+38.72	-30.54
NBeG 119	HS	+12.77	-1.09	+50.42	-34.74

IS- Index of Susceptibility, MR- Moderately Resistant, MS-Moderately Susceptible, S- Susceptible, HS- Highly susceptible.

Assessment of biochemical parameters of chickpea genotypes

The data pertaining to chemical constituents viz., protein, phenol, ash and fibre content were assessed in fiftygenotypes of chickpea before and after infestation and the per cent changes in these chemical constituents were presented in Table 1 to 3.

Biochemical Parameters Before Infestation

Protein content

The data presented in Table 1 revealed that among the different genotypes of chickpea, the highest protein content was observed in highly susceptible genotype JG 16 (25.27 per cent).The initial percentage of protein (before *C. chinensis* infestation) in test chickpea genotypes ranged from 18.46 to 25.27 per cent. High percentage of protein was noticed in the highly susceptible genotypes *viz.,* JG 16 (25.27 per cent) followed by NBeG-49 (25.14 per cent), Himachal Chana 2 (25.07 per cent) and ICC 3137 (25.01per cent). which were more preferred by the pulse beetle as higher amount of protein supplemented its growth and development, whereas less protein content was recorded in moderately resistant genotypes *viz.* Himachal Chana 1 (18.46 per cent) followed by NBeG-47 (18.67 per cent), Dilaji (19.13 per cent) and JG-14 (19.45 per cent). which were comparatively less preferred by the test insect. Lower protein content might have acted as ovipositional deterrents leading to less egg laying. The results are in concurrence with Kancherela e*t al.,* (2018) who reported that the genotype JG 11 with high per cent seed damage recorded high protein content (22.70%) and the genotype JG 315 with low per cent seed damage recorded with low protein content (15.33%), The results are also in agreement with Singh *et al.,* (2016) and Nandini *et al.,* (2018).

Phenol content

The phenol content of the fifty test genotypes before infestation of *C. chinensis*ranged from 178.65 to 294.53 mg/100g fresh weight (Table 1). Moderately resistant genotypes viz., Himachal Chana 1 (294.53 mg/100g fw) and NBeG-47 (271.65 mg/100g fw) recorded the highest phenol content and were at par, whereas the highly susceptible genotypes viz., JG 16 (178.65 mg/100g fw), NBeG-49 (182.31 mg/100g fw) exhibited the lowest phenol content. The phenol content in other moderately resistant genotypes viz., Dilaji, JG 14, ICCV-181108 and JG 11 were 264.32, 256.78, 250.09 and 245.25 mg/100g fw respectively. The phenol content in the rest of the chickpea genotypes varied between 184.22 to 241.75 mg/100g fw.The observations on higher amount of phenol content in resistant genotypes is in agreement with Augustine *et al.,* (2018) who screened 15 cowpea genotypes *viz.,* GC-3, IC 202702, IC 259065, DC-15, IC 6202, IC 998326, IC 259106, IC 91556, IC 219871, IC 198383, IC 253272, IC219594, DCS 47-1, IC 253276 and IC 257407 to *Callosobruchus chinensis* L. in storage. He reported that there is a negative and significant correlation between per cent damage and phenol content of the seeds whereas the moisture and protein of seeds were positively correlated with per cent damage at 90 days after release. Gopala Swamy *et al.,*(2020) recorded that less susceptible chickpea genotypes viz., NBeG 511, JAKI 9218 and JG 11 possessed higher amounts of total phenols when compared to the highly susceptible genotypes viz., NBeG 458, NBeG 471 and KAK 2. The results are also in agreement with Bhattacharya and Banerjee (2001) and Mainali *et al.,* (2015).

Ash content

The data presented in Table 1 revealed that the ash content of the test genotypes varied from 2.21 to 3.76 per cent. In the present investigation, the highest ash content was noticed in moderately resistant genotypes *viz.,* Himachal Chana 1, NBeG-47, Dilaji, JG-14 and C-18203. The lowest ash content was recorded in the highly susceptible genotype JG-16, NBeG-49,ICC 3137, C 19199, C-19168, C-19162 and NBeG 119. High ach content in the seeds might leave some toxic effect on the test insect and also injure digestive as well as respiratory organs. This might also play an important role in delaying the developmental period of the pulse beetle by interfering with digestion and making it a non-preferred host for development and provide unfavourable conditions for feeding, growth and development (Kancherla *et al.,* 2020). The findings are in accordance with *Mogbo et al.,* (2014). He reported that high levels of ash content in cowpea varieties conferred high resistance to *C. maculatus* infestation. Holay*et al.,* (2018) reported that out of 25 pigeonpea genotypes, PKV-TARA was found most superior, which was recorded less ovipositional preference (10.25 eggs/10 seeds), adult emergence (71.01%), adult longevity (7.50 days) of *C. maculatus* and growth index (2.56) due to having higher ash content with lower content of protein.

Fibre content

The highest fibre content was recorded in moderately resistant chickpea genotypes viz., Himachal Chana 1(5.91%) which was at par with other moderately resistant genotypes viz., Dheera (NBeG-47) (5.58%), Dilaji (5.40%) and JG-14 (5.27%) whereas the lowest fibre content was recorded in susceptible variety viz., JG-16 (2.47%) which was at par with C-19162 (2.28%), NBeG 49 (2.58%), C-19168 (2.62%), ICC 3137 (2.68%) and C 19199 (2.71%). The fibre content recorded in Kalahandi Local, NBeG-119, RSG 963, Phule Vikram, ICCV-181108, ICCV-181605, JG 11 and C-18252 varied from 2.73 to 4.66 per cent.The results are in accordance with the findings of Jyothsna (2014). She reported high percentage of crude fibre in moderately resistant varieties viz., K9 (2.72per cent) and ICGV86015 (2.70per cent) and these varieties were comparatively less preferred by the bruchid. Similar results were also found with Ahmad *et al.* (2015) and Nandini *et al.* (2018).

Changes In Biochemical Parameters After Infestation

Protein content

The data presented in Table 2 revealed that after infestation of *C. chinensis*in the different genotypes of chickpea, the highest protein content was observed in highly susceptible genotype JG 16 (28.56 per cent) which was at par with NBeG-49 (28.36per cent), whereas the lowest protein content was found in moderately resistant genotype Himachal Chana 1 (19.29 per cent) which was at par with NBeG-47 (19.56 per cent), Dilaji (19.78

per cent) and JG-14 (19.99 per cent). The per cent change in protein content of different chickpea test genotypes after infestation was presented in Table 3. It was observed that the protein content of these genotypes increased post infestation. The highest increase in protein content was noticed in highly susceptible genotype JG 16 (13.01 per cent) followed by NBeG-49 (12.83 per cent), ICC 3137 (12.82 per cent) and NBeG-119 (12.77 per cent) whereas the lowest increase in protein content was recorded in moderately resistant genotype Himachal Chana 1 (2.21 per cent) followed by NBeG-47 (3.32 per cent), Dilaji (4.65 per cent), JG-14 (6.73 per cent) and C-18252 (7.21 per cent). In rest of the genotypes the per cent increase in protein content varied from 7.78 to 12.45 per cent.The findings are in line with Ashish *et al.* (2019) who reported that the percentage of crude protein significantly increased with the duration of storage period of 30 days (25.42 per cent), 90 days (25.71 per cent) and 180 days (26.01 per cent). Similarly, Nandini *et al.,* (2018) reported that in different split legumes there was a significant increase in protein content at 90 days after storage except for black gram dhal and kesar dhal where the per cent increase in protein content was not significant due to rice weevil infestation. Similar results had been reported by Shanti *et al.,* (2015) and Gadeware*t al.* (2016).

Phenol content

The phenol content of the different test genotypes post infestation by *C. chinensis*ranged from 176.35 mg/100g fresh weight to 293.95 mg/100 g fresh weight (Table 2). The moderately resistant genotypes *viz.,* Himachal Chana 1, NBeG-47, Dilaji and JG 14 recorded the highest phenol content of 294.53, 271.65, 263.57 and 256.04mg/100g fw, respectively which were at par with each other. Likewise, the highly susceptible genotypes viz., JG 16, NBeG-49 and ICC 3137 were found with less phenol content (176.35, 180.25 and 182.18 mg/100g fw, respectively). The per cent change in phenol content of different test genotypes after chickpea bruchid infestation was presented in Table 3. It was noticed that there was a decrease in phenol content of the genotypes post infestation. The highly susceptible genotypes viz., JG 16 (1.35per cent), NBeG-49 (1.13per cent), ICC 3137 (1.11per cent) exhibited higher reduction in phenol content, whereas the moderately resistant genotypes viz., Himachal Chana 1 (0.19per cent) and NBeG-47 (0.21per cent) recorded less reduction in phenol content. However, moderately susceptible genotypes viz., JG 11and ICCV-181605 also exhibited lesser reduction in phenol content.The findings are in line with Ashish *et al.* (2019) who reported that the percentage of total phenol significantly decreased with the duration of storage period of 30 days (2.90 per cent), 90 days (2.60 per cent) and 180 days (2.14 per cent). Similar results had been reported by Shanti *et al.,* (2015) and Gadeware*t al.* (2016).

Ash content

The data indicating to the ash content after chickpea pulse beetle infestation in the test genotypes varied from 3.26 to 4.43per cent (Table 2). Moderately resistant genotype Himachal Chana 1 recorded the highest (4.43per cent) ash content post infestation which was at par with Dheera (NBeG-47) (4.43per cent) while, the susceptible genotypes viz., JG 16, NBeG-49,C-19199, Kalahandi Local, C-19162, NBeG-119, C-19168 andICC 3137 were found with less ash content and were at par with each other. It was noticed that there was an increase in ash content in the chickpea genotypes after pulse beetle infestation (Table 3). The maximum increase in ash content was observed in the susceptible variety JG 16 (63.80 per cent), NBeG-49 (53.81 per cent) and ICC 3137 (53.01 per cent) whereas minimum increase in ash content was found in moderately resistant genotypes viz., Himachal Chana 1 (15.43per cent) and Dheera (NBeG-47) (17.17per cent). Ashish *et al.* (2019) also revealed that there was significant increase in ash contents of the chickpea seeds after infestation by *C. chinensis* with the duration of storage period of 30 days (4.92 per cent), 90 days (5.46 per cent) and 180 days (6.02 per cent). Similar results had been reported by Shanti *et al.,* (2015) and Gadeware*t al.* (2016).

Fibre content

The fibre content after infestation of *C. chinensis*in the test genotypes varied from 1.48 to 5.32 per cent (Table 2). The highest fibre content post infestation was observed in moderately resistant genotype viz., Himachal Chana 1(5.32 per cent) which was at par with moderately resistant variety Dheera(NBeG-47)(4.99 per cent), whereas the lowest fibre content was recorded in susceptible genotypes viz., JG-16 (1.48per cent) which was at par with NBeG 49 (1.60 per cent). The per cent change in fibre content of different chickpea test genotypes after infestation is presented in Table 3. It was observed that the fibre content of these genotypes decreased post infestation. Maximum decrease in fibre content was recorded in susceptible genotype JG 16 (44.65per cent) whereas minimum decrease was found in moderately resistant genotype Himachal Chana 1(9.98per cent). Other susceptible genotypes viz., NBeG-49, C 19199, Kalahandi Local, C-19162, NBeG-119, C-19168 andICC 3137 recorded 40.08per cent, 37.98per cent, 36.23per cent, 35.82per cent, 34.74per cent, 34.35per cent and 33.21per cent reduction in fibre content.The findings are in agreement with Nandini *et al.* (2018). They reported that there was a significant decrease in fibre content (40.46 per cent) at 90 days after storage in the legumes post infestation. Similar results had been reported by Shanti *et al.,* (2015) and Ashish *et al.* (2019).

Conclusion

Among the biochemical parameters, high phenol, ash and fibre content offered resistance to the pest attack while protein content favoured high adult emergence in susceptible varieties. Study of these factors would help in better understanding of the resistance mechanisms and subsequent development of resistant varieties to the bruchid infestation. This information can be used to select better parent and crossed to raise more resistant recombinants for the development of resistant chickpea varieties.

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