Biological and Environmental Sciences Journal for the Tropics
Volume 21, Number 3, December 2024. Pp 67 - 81
https://dx.doi.org/10.4314/bestj.v21i3.6

Research article

Identification of Cowpea Genotypes with Enhanced Resistance to *Callosobruchus maculatus* (F.) for Sustainable Storage in Pest Management

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Submission: 16/07/2024
Accepted: 17/11/2024

Abstract

Callosobruchus maculatus (F.) is a devastating storage pest of cowpea, causing significant losses if left uncontrolled. This study evaluated the response of 411 cowpea genotypes to *C. maculatus* infestation to identify novel sources of resistance and investigate associations between bruchid growth, damage parameters, and resistance metrics. The experiment was conducted at Entomology Laboratory, Department of Zoology, Ahmadu Bello University, Zaria using completely randomized design (CRD). A no-choice assay was employed to screen cowpea genotypes, and data were collected on bruchid growth, damage parameters, and seed morphometric traits. Statistical analysis revealed significant differences ($p < 0.05$) among genotypes for all measured traits, indicating substantial variability for bruchid resistance. Five genotypes (ABU_Vu16, ABU_Vu39, ABU_Vu225, ABU_Vu239, and ABU_Vu309) showed high bruchid tolerance (>70%) and low Dobie susceptibility index (DSI) values (<3). Negative associations were observed between percentage pest tolerance and adult emergence, growth index, and DSI, suggesting that selecting for lower values of these traits can enhance bruchid resistance. Our findings identify reliable indicators of bruchid resistance in cowpea and highlight the potential of these genotypes for developing sustainable storage pest management strategies using a multi-trait approach. This approach is essential, as various factors influence the bruchid resistance trait.

Keywords: Cowpea**,** *Callosobruchus maculatus*, bruchid resistance, storage pest management, sustainable agriculture.

Introduction

Cowpea (*Vigna unguiculata* (L. Walp.) is a vital grain legume in sub-Saharan Africa, serving as a primary source of dietary protein for low-income households (Boukar *et al*., 2018). Its ability to fix nitrogen enables it to thrive in nutrient-deficient soils, making it a valuable crop for smallholder farmers (Kpoviessi *et al*., 2021). However, cowpea production faces numerous challenges, including biotic (Aliyu *et al*., 2023) and abiotic factors (Amusa and Ogunkanmi, 2021, Ahmed *et al*., 2023; Sakariyahu *et al*., 2023; Indabo *et al*., 2023; Ossai *et al*., 2024). One major post-harvest pest is *Callosobruchus maculatus* (Fabricius, 1775), which can cause up to 100% grain loss in stored cowpea seeds (Horn *et al*., 2020). The use of synthetic pesticides to control this pest is expensive, harmful to human health, and environmentally unfriendly (Tengey *et al*., 2023). Cultivating bruchid-resistant varieties offers a safer and more cost-effective alternative (Affram *et al*., 2022). This can only be achieved by screening available genotypes for breeding, aiming to produce bruchid-resistant varieties for farmers to cultivate.

The importance of cowpea as a food and income source for smallholder farmers in sub-Saharan Africa cannot be overstated. However, its production is constrained by various factors, including insect pests and diseases (Amusa and Ogunkanmi, 2021). Bruchid resistance in cowpea is crucial for sustainable production and storage (Carrillo-Perdomo *et al*., 2019). Despite earlier successes in identifying bruchid-resistant cowpea genotypes (Singh *et al*., 1985; Singh and Singh, 1990; Shade *et al*., 1996), some have lost their resistance over time (Amusa *et al*., 2013).

Locally adapted farmers' varieties remain valuable resources for modern breeding (Tripathi *et al.*, 2020). This study aimed to evaluate cowpea genotypes for bruchid resistance, assess associations between bruchid growth, damage parameters, and seed morphometric characters, and identify new sources of resistance.

MATERIALS AND METHODS

Experimental materials

A total of four hundred and eleven (411) cowpea genotypes obtained from the Plant Resilience Research for Economic Sustainability group of the Department of Botany, Ahmadu Bello University (ABU) Zaria were used in this study (Table 1**)**.

Table 1: List of cowpea accessions, entry number and state of collection for bruchid resistance study

Accession Name	Entry name	State
ABU_Vu001 to ABU_Vu014	Bosop_Gombe, Bosop 2_Gombe, Dan Misra, Iron Beans, Iron Beans 2, Iron Brown_Gombe, Jan Wake, Sea, Wake Mai Bakin Kono, Wake Mai Borgo, Wake Mai Borgo Ja, Yebbereru_Gombe, Yebbereru 2, Yebbereru Mai Farin Hanci	Gombe
ABU_Vu015 to ABU_Vu027	Yar 40, Anannadi, Bosop_Taraba, Brown Beans_Taraba, Dan Ogoja, Iron Beans_Taraba, Kanannado_Taraba, Ogoja, Silver_Taraba, Wake Mai Yaduwa, Warwarbashi, Yar Malaysia, Yebbereru_Taraba	Taraba
ABU_Vu028 to ABU_Vu001, 033, 035	Kwankwaso_Benue, Yar Misra, Tiligali, Komcall 1, Oloyin	Benue
ABU_Vu032, ABU_Vu034	Gbako 3, Bida 2	Niger
ABU_Vu036 to ABU_Vu037, 039-047	Hannun Marini_Zamfara, Bahaushe Zamfara, Dan Emir, Dan Dam, Kanana, Dan Zafi, Kanannado_Zamfara, Dan Misra Zamfara, Mai Bakar Kowa, Ife Brown Zamfara, Dan Agaji	Zamfara
ABU_Vu048 to ABU_Vu062	Bahaushen Wake, Bahaushen_Sokoto, Kanannado Sokoto, Kalabas, Sababba Sata, Dan Galankawa, Iron Beans_Small Sokoto, Dan Misira_Sokoto, Farin Wake_Sokoto, Iron Beans_Big Sokoto, Olowin(Milk), Drum, Olowin(Red), Zafa_Sokoto	Sokoto
ABU_Vu063 to ABU_Vu074, ABU_Vu298	Wake Dan Yagau, Farin Wake_Kebbi, Sa Babba Sata, Kanannado_Kebbi, Milk Sobo, Farin Sobo, Ka Ki Ganin Shono, Jan Sobo, Dan Hausa, BaAre, Kalan Madara, Olanyo, Kananando Kebbi 2	Kebbi
ABU_Vu075 to ABU_Vu084	Bakin Hantsi, Kanannado_Adamawa, Waken Gombe, Jan Wake_Adamawa, Oloto, Geila, Iron Beans Adamawa, Kili Banjaram, Banjaram_Adamawa, Kere-Kere	Adamawa
ABU_Vu085 to ABU_Vu096, ABU_Vu264	Ci Kai Shiru Red, Ci Kai Shiru White, Farin Wake_Manya, Farin Wake Qanana, Farin Wake Matsakaita, Honey Beans Bauchi, Kanannado_Bauchi, Red Beans-Big, Red Beans-Medium, Red Beans-Small, Silver_Bauchi, Yabbareru Bauchi, Medium Beans Bauchi	Bauchi
ABU_Vu097 to ABU_Vu115	Aloka Borno, Bangara Borno, Borno Yasu, Bornoji, Dinka, Genchein, Gwalam, Iron Beans Borno, Jan Baki, Kanannado Borno, Kolobe, Mai Madara Borno, Oho, Rangem, Warwara Bashi, Banjiram Kanannado, Dimairi, Kiri-Kiri, Sulpha	Borno
ABU_Vu116 to ABU_Vu124	Honey Beans Jos, Honey Brown Beans, Iron Beans Jos, Medium Beans Plateau, Yabbarere Plateau, Dan Zaria, Mai- Toka, Achi Shiru-Fari, Achi Shiru	Plateau
ABU_Vu125 to ABU_Vu140	Jan Wake Yobe, Yabbereru Yobe, Wake Mai Bargo, Aloka Yobe, Jangau, Banjara Yobe, Iron Beans Yobe, Fari Manyan Kwaya, Dan Umaru, Yambari, Wake Dan Chadi, Karaduwa Fari, Bosho Yobe, Silver Yobe, Karaduwa Yobe, Olotu	Yobe
ABU_Vu141 to ABU_Vu142	Tvu 2027, Tvu 17470	
ABU_Vu162 to ABU_Vu166	Dan Sokoto, Sa-Babba-Satah, Sobo_Kebbi, Dan Rima, Dan Ilela	Kebbi

Table 1 continue

ABU_Vu145, ABU_Vu148 ABU_Vu151 ABU_Vu167 to ABU_Vu171 ABU_Vu240, ABU_Vu289, ABU_Vu294	Chanchaga 2, Zungeru, Bida1, Kanannado Niger, Maifitila, Kwankwasiya Niger, Jan Wake Niger, Chanchaga 1, Kontagora , Gbako 1 , Gbako 2	Niger
ABU_Vu172, ABU_Vu174 to ABU_Vu175	Horicot Bean, Shell Bean, Pea Beans	Nassarawa
ABU_Vu173	Aye Talba	Kwara
ABU_Vu268	Ewa pupa	
ABU_Vu176 to ABU_Vu181	Bakin Acishiru, Mada (Small), Bikara_Plateau, Bakin Kanannade, Ruwan Kasan Kanannade, Mada (Large)	Plateau
ABU_Vu181 to ABU_Vu187	Mada (Large), Jan Bosok, Farin Bosok, Bakin Hanci_Adamawa, Kanannado_Adamawa, Mai Madara_Adamawa, Silva_Adamawa	Adamawa
ABU_Vu188 to ABU_Vu194	Jan Wake_Jigawa, Kanannado_Jigawa, Aloka_Jigawa, Dankaka_Jigawa, Yozka 1, Bakolo_Jigawa, Dan Wuri_Jigawa	Jigawa
ABU_Vu195 to ABU_Vu198 ABU_Vu301	Iron Beans_Zamfara, Oloru, Dan Wari_Zamfara, Medial, Dan Misra 2_Zamfara	Zamfara
ABU_Vu199, ABU_Vu200, ABU_Vu284, ABU_Vu201	Belata_Benue, Ewa funfun, Ndakosode , Shiswa	Benue, Kwara, Benue
ABU_Vu038, ABU_Vu202 to ABU_Vu223	TVu7778, Dan Malan, Dan Kwari, Dan Masar, Yan Barere_Kano, Iron Beans_Kano, Siver_Kano, Dan Eka, Gama Gari Mai Yado, Dan Wuri_Kano, Kwankwaso, Hannun Marini_Kano, Dan Tsaye, Kanannado Dan Kaka, Dan Kaka_Kano, Dan Ilan, Karaduwa, Dan Arewa, Mai Kasa, Oluka, Dan Ringin, Dan Misra_Kano, Dan Feshi	Kano
ABU_Vu224 to ABU_Vu239	Iron Beans_Kaduna, Medium Beans_Kanuna, Kanannado_Kaduna, Milk_Kaduna, Honey Beans_Kaduna, Dan Giwa, Dan Kaya_Kaduna, Dan Shika_Kaduna, Dan Misra_Kaduna, Dan Muzakkar_Kaduna, Bakin Wake Dan Ghana, Jan Wake_Kaduna, Wake Mada Brown, Wawa Mata, Waken Rumfa Brown	Kaduna
ABU_Vu241 to ABU_Vu253	Patiskum Medium Beans, Jan Wake_Nassarawa, Namu Beans, Yan Barere_Nasarawa, Iron Beans_Nassarawa, Kwana Arbain, Si Beans, Lafia Beans 1, Lafia Beans 2, Kwankwasiya_Nassarawa, Oshiki Ja, Silver Beans_Nasarawa, Oshiki Fari	Nassarawa
ABU_Vu254 to ABU_Vu259	Kanannado_Abuja, Brown Beans Abuja, Hot Beans Abuja, Honey Beans Abuja, Butter Beans Abuja, Iron Beans_Abuja,	Abuja
ABU_Vu261 to ABU_Vu263	Kanannado_Nassarawa Hot Beans_Nasarawa, Iron Beans	Nassarawa
ABU_Vu265 to ABU_Vu267 , ABU_Vu269	Iron Beans_Big_Plateau, Iron Beans_Medium_Plateau, Iron Beans_Small_Plateau, Aloka Dan Gombe	Plateau
ABU_Vu271 to ABU_Vu274 , ABU_Vu295	Dan Gwambi, Aloka Yabbarere, Kanannado_Jigawa 2, Ja Kanannado, Yozka 2	Jigawa
ABU_Vu275 to ABU_Vu278	Bakolo_Kano, Dan Harisu, Komfita, Kyanbas	Kano
ABU_Vu279 to ABU_Vu283 ABU_Vu285 to ABU_Vu288 ABU_Vu290 to ABU_Vu293 ABU_Vu296- ABU_Vu297	Dan Misrah 2_Katsina, Dan Muzakkar_Katsina, Dan Barari, Farin Wake_Katsina, Bakin Wake Honey Brown_Katsina, Olonyi, Butter Beans_Kastsina, Iron Beans_Katsina Dan Misra Katsina, Dan Shika_Katsina, Zafa_Katsina, Medium Bakin Baki Sobo_Katsina, Kanan Nado	Katsina
ABU_Vu299 to ABU_Vu300, ABU_Vu302 to ABU_Vu308	Dan Eka Kano 2, Big Brown _Kano, Butter Beans_Kano, Pure Pure, Drone, Butter Beans Kanana_Kano, Honey Beans_Kano, Black Beans_Kano, Uloyi_Kano	Kano

Table 1 continue

ABU_Vu309 to ABU_Vu405 ABU_Vu407 to ABU_Vu419	Tvu-16921, Tvu-16924, Tvu-16928, Tvu-16929, Tvu-16934, Tvu-16935, Tvu-16936, Tvu-16937, Tvu-16941, Tvu-16942, Tvu-16943, Tvu-16946, Tvu-16947, Tvu-16948, Tvu-16949, Tvu-16950, Tvu-16952, Tvu-16954, Tvu-16955, Tvu-16956, Tvu-16958, Tvu-16961, Tvu-16962, Tvu-16963, Tvu-16964, Tvu-16966, Tvu-16967, Tvu-16968, Tvu-16969, Tvu-16970, Tvu-16971, Tvu-16972, Tvu-16973, Tvu-16974, Tvu-16976, Tvu-16977, Tvu-16978, Tvu-16979, Tvu-16980, Tvu-16982, Tvu-16984, Tvu-16985, Tvu-16987, Tvu-16988, Tvu-16989, Tvu-16990, Tvu-16991, Tvu-16992, Tvu-16994, Tvu-16995, Tvu-16996, Tvu-16997, Tvu-16998, Tvu-16999, Tvu-17000, Tvu-17001, Tvu-17002, Tvu-17003, Tvu-17004, Tvu-17005, Tvu-17006, Tvu-17007, Tvu-17008, Tvu-17009, Tvu-17010, Tvu-17011, Tvu-17012, Tvu-17013, Tvu-17014, Tvu-17015, Tvu-17016, Tvu-17017, Tvu-17018, Tvu-17019, Tvu-17020, Tvu-17022, Tvu-17023, Tvu-17024, Tvu-17025, Tvu-17026, Tvu-17027, Tvu-17028, Tvu-17029, Tvu-17030, Tvu-17031, Tvu-17032, Tvu-17034, Tvu-17035, Tvu-17037, Tvu-17038, Tvu-17039, Tvu-17040, Tvu-17041, Tvu-17042, Tvu-17043, Tvu-17045, Tvu-17046, Tvu-17047, Tvu-17048, Tvu-17049, Tvu-17051, Tvu-17360, Tvu-17461, Tvu-17462, Tvu-17464, Ife Brown, Danila, IT84S-2246-4	IITA, Ibadan

Experimental site
Bruchid Screening assay was performed at the Entomology Laboratory, Department of Zoology, A.B.U. Zaria.

Bruchid culture preparation.
Bruchid culture were prepared according to Beck and Blumer (2014) with modifications, where cowpea genotypes was infested with freshly emerged adult bruchids (males and females) at 28-30ºC and 55-60% relative humidity.

Seeds disinfection
Seeds of the 411 cowpea genotypes collected were kept at 4ºC for 24 hours to kill off any bruchid eggs or larvae that may have infested the seeds from the field, according to Beck and Blumer (2014).

Bruchid bioassay
The bruchid screening experiment was conducted using a completely randomized design (CRD) following the method described by Singh *et al*. (2014) with some modifications. Ten healthy, well-dried seeds of each genotype were placed in separate petri dishes with covers (90 x15mm). Each petri dish was infested with two freshly emerged males and three females, all of which were fully covered with the petri dish cover. The male and female adults were identified based on their morphology described by Raina (1970). One week after infestation, the parental insects were removed, and the seeds in the petri dishes were kept on shelves at a temperature of 28 to 30°C and a relative humidity of 55-60%. Ten days after infestation, when the eggs became clearly visible, the total number of eggs laid on 10 seeds in each petri dish were counted using magnifying lens. The petri dishes were placed in a tray and kept on the shelves until the adult emergence began, which occurred 25 to 28 days after the seeds were infested. The emerged adults in each petri dish were counted and removed on a daily basis. The counting of emerged adults was stopped 50 days after infestation to ensure that second-generation adults were not included. The bioassay was replicated three times.

Data were recorded according to Beck and Blumer, (2014). Initial seed weight (g) was taken as the weight of the seeds measured before infestation; residual seed weight (g) was measured as the weight of the seeds measured after the larvae perforated the seeds at the end of the experiment. The number of eggs laid was the total number of eggs deposited on all the ten seeds in each petri dish. The number of adult emerged was taken as the total number of adults that emerged. Days to first emergence was the number of days before the first adult emerged.

The median developmental period was taken as the time in days from the middle of the oviposition period to the emergence of 50% of the adults. Total developmental time (days) was taken as the time taken in days for the complete emergence of adults in susceptible genotypes. The number of damaged seeds was the number of seeds perforated by the larvae. Number of exit holes was the total number of holes perforated by the larvae. Percentage weight loss (PWL), percentage adult emergence (PAE), percentage seed damage (PSD) and percentage pest tolerance (PPT) were calculated according to Singh *et al.* (2014) as follows:

$$PWL\ (\%) = \frac{Initial\ seed\ weight - Residual\ seed\ weight}{Initial\ seed\ weight} \times 100$$

$$PAE\ (\%) = \frac{Number\ of\ adults\ emerged}{Total\ number\ of\ eggs\ laid} \times 100$$

$$PSD\ (\%) = \frac{Number\ of\ damaged\ seeds}{Total\ number\ of\ seeds} \times 100$$

$$PPT\ (\%) = \frac{Number\ of\ undamaged\ seeds}{Total\ number\ of\ seeds} \times 100$$

Growth index (GI) was calculated as:

$$\mathrm{GI} = \frac{Percentage\ adult\ emergence}{Median\ development\ period}$$

The susceptibility index for each line was calculated according to Dobie (1974) using the formula below

$$SI = \frac{LogeF}{D} \times 100$$

Where Log_e = the natural logarithm of numbers, F = the total number of F_1 progeny emerged (emerged adults), D = Median development period.

The susceptibility index which is a scale of 0 to 11 was used, to classify the cowpea genotypes according to Dobie, (1974), where 0-3 was resistant, 4-7 was moderately resistant, 8-10 was susceptible and ≥10 was highly susceptible.

Validation test of the screened resistant genotypes

A subset of five (5) genotypes classified as resistant were further subjected to validation test according to Beck and Blumer, (2014).

Morphometric Characterization of Cowpea Seeds

The morphometric features of cowpea seeds were determined using a combination of visual observation and instrumental measurements (Arotolu *et al.*, 2018; Aliyu *et al.*, 2022). Seed color was observed visually and classified into four categories: white, mottled, intermediate, or brown. Eye color was also observed visually and classified as either black or brown. Additionally, the following qualitative features were determined for each genotype: Seed shape: oval, rhomboid, globose, or kidney-shaped (Singh *et al.*, 1985), Seed luster: dull or shiny, and seed texture: wrinkled or smooth (Mei *et al.*, 2009). These morphometric characteristics were used to describe the physical attributes of each cowpea genotype.

Statistical Analysis

Data collected on insect growth, damage and seed morphometric traits were subjected to one-way analysis of variance (ANOVA). Significant means were separated using the Fisher's least significant difference (LSD) test at 5% probability level. Box plots and stacked bar charts were used to visualize the means for seed morphometric traits.

Principal Component Analysis (PCA) was used to visualize the association between evaluated parameters. All statistical analyses were carried out using R version 4.0.5 for Windows (RStudio Team, 2021)

RESULTS

Response of diverse cowpea genotypes to artificial infestation of bruchid

The response of diverse cowpea genotypes to artificial infestation of bruchid revealed significant differences ($p < 0.05$) among the genotypes for all measured traits (Table 2). The frequency distribution of the 411 genotypes based on their resistance status showed a range of responses, with 24.8% exhibiting high susceptibility, 42.7% showing susceptibility, 29.3% displaying moderate resistance and 3.2% demonstrating resistance (Figure 1).

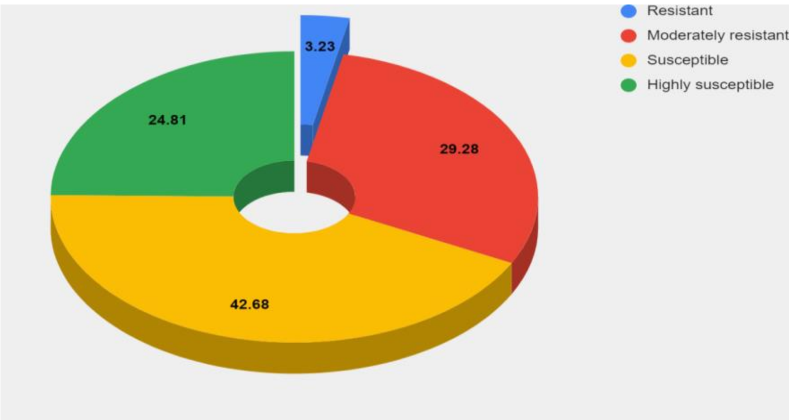


Figure 1: Percentage of Genotypes by Resistance Status of the 411 Cowpea Landraces Studied

Table 2: Mean squares of insect-growth and damage parameters of 411 cowpea genotypes screened for *C. maculatus* infestation in northern Nigeria.

Source of variation	d.f	Mean square									
		NEL	MDP (%)	DFE	PAE (%)	PSD (%)	NEH	PPT (%)	PWL (%)	GI	DSI
Genotypes	411	2519.7	37.4	29.6	1351	1144	0.27	1144	362	1.5	24
Reps	3	691.5	696.6	725.0	154935	158841	60.95	158841	63604	384.0	12442
Residuals	766	1862.9	30.5	18.7	383	944	0.21	944	234	0.5	21
P-values		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

Significant differences ($p < 0.001$) were observed in various parameters, including; Number of eggs laid (NE) by *C. maculatus*, Median time to adult bruchid emergence (MDP), Total adult emergence (TAE), Percentage adult bruchid emergence (PAE), Percentage seed damage (PSD), Number of exit holes per ten seeds (NEH), Percentage pest tolerance (PPT), Percentage weight loss (PWL), Insect growth index (GI) and Dobie susceptibility index (DSI) among the 411 cowpea genotypes studied, indicating substantial genetic variation in bruchid resistance.

Validation of Resistant Cowpea Genotypes

The screening experiment identified 13 resistant genotypes, which accounted for 3.2% of the total genotypes screened. Further validation confirmed the resistance of five genotypes. For instance, the validated resistant genotype ABU_Vu039 showed a significantly lower number of eggs laid on it compared to the resistant check TVu-2027 (Table 3). The validated resistant genotypes exhibited a longer median development period (25 days), whereas other resistant genotypes had a shorter median development period (21 days). The first adult emergence occurred in the susceptible check Ife-Brown 24 days after infestation, followed by highly susceptible genotypes (25 days after infestation). In contrast, the validated resistant genotypes showed significantly delayed adult emergence (33 days after infestation). The validated resistant genotypes displayed the lowest growth index, while highly susceptible genotypes recorded the highest growth index. The percentage of adult emergence ranged from 15% in the validated resistant genotypes to 55% in the highly susceptible genotypes (Table 4).

The percentage seed damage varied from 49% in the validated resistant genotypes to 90% in Ife Brown. The validated resistant genotypes exhibited fewer exit holes than the susceptible check and highly susceptible genotypes. The percentage of pest tolerance increased from 10% in highly susceptible genotypes to 60% in validated resistant genotypes, surpassing the resistant check (25%). A consistent decrease in percentage weight loss was observed from highly susceptible genotypes (45%) to validated resistant genotypes (20%). The validated resistant genotypes had the lowest Dobie Susceptibility Index (DSI) (<3), while highly susceptible genotypes recorded the highest DSI (>10). These findings confirm the resistance of the validated genotypes and their potential for improving cowpea breeding programs.

Table 3: Means of insect growth parameters on the validated resistant cowpea genotypes evaluated at Samaru.
Note:

Genotype	NEL	MDP	DFE	GI
ABU_Vu039	7.33±2.67^{b}	25.67±4.06^{ab}	30.67±2.33^{a}	0.34±0.18^{ab}
ABU_Vu225	38.33±28.39^{ab}	23.00±2.00^{ab}	17.67±8.84^{b}	0.42±0.40^{ab}
ABU_Vu239	46.67±4.70^{ab}	19.00±0.00^{b}	32.67±16.33^{a}	0.00±0.00^{b}
ABU_Vu309	42.00±30.57^{ab}	20.33±1.33^{ab}	24.33±0.33^{ab}	0.18±0.18^{ab}
ABU_Vu016	64.00±26.96^{ab}	26.67±1.20^{ab}	28.67±1.20^{ab}	0.20±12.45^{ab}
Tvu_2027 (+ve Control)	52.33±31.76^{ab}	27.33±4.41^{a}	27.33±3.38^{ab}	1.50±0.87^{a}
Ife Brown (-ve Control)	100.33±3.96^{a}	23.00±1.07^{ab}	23.33±0.44^{ab}	1.03±0.64^{ab}

Values with the same superscripts along the columns are not significantly different ($p<0.05$)
Key: NEL- Number of exit holes, MDP- Median time to adult bruchid emergence, GI- Insect growth index

Table 4: Means of insect damage parameters of the validated resistant cowpea genotypes studied

Genotype	PAE (%)	PSD (%)	NEH	PPT (%)	PWL (%)	DSI
ABU_Vu039	10.00±5.77^{b}	33.33±28.48^{c}	5.67±5.17^{c}	66.67±28.48^{a}	5.75±1.83^{d}	0.67±0.67^{d}
ABU_Vu225	10.61±10.09^{ab}	40.00±30.55^{c}	11.67±10.68^{b}	60.00±30.55^{a}	19.89±18.93^{c}	2.00±2.00^{c}
ABU_Vu239	0.00±0.00^{ab}	33.33±33.33^{c}	10.00±10.00^{b}	66.67±33.33^{a}	10.73±9.01^{c}	0.00±0.00^{d}
ABU_Vu309	4.17±4.17^{b}	33.33±18.56^{c}	6.33±4.84^{c}	66.67±18.56^{a}	14.16±4.65^{c}	0.00±0.00^{d}
ABU_Vu01	5.54±3.90	60.00±30.55	25.67±7.51	40.00±30.55	28.80±12.45^{b}	0.20±0.88^{d}
Tvu_2027 (+ve Control)	45.94±25.43^{a}	76.67±18.56^{a}	25.00±18.08^{a}	23.33±18.56^{c}	25.29±21.20^{b}	7.00±4.73^{b}
Ife Brown (-ve Control)	25.67±3.18^{ab}	90.00±2.40^{a}	36.67±3.17^{a}	10.00±2.40^{d}	37.33±2.91^{a}	10.00±1.75^{a}

Note: Values with the same superscripts along the columns are not significantly different ($p<0.05$).
Key: PAE- Percentage adult bruchid emergence, PSD- Percentage seed damage, NEH- Number of exit holes per ten seeds, PPT- Percentage pest tolerance, PWL- Percentage weight loss, DSI- Dobie susceptibility index.

The results shows significant variation ($P<0.05$) in the mean number of eggs laid among the screened genotypes, with the validated resistant genotypes having the lowest number of eggs laid even lower than the positive check (TVu-2027) (Figure 2). The median development period (Figure 3) also varied significantly among the screened genotypes, with the validated resistant genotypes having a longer median development period (25 days) compared to the resistant check (21 days). The mean days to first adult emergence (Figure 4) showed a progressive delay from the highly susceptible genotypes (25 days) to the validated resistant genotypes (33 days). The percentage adult emergence (Figure 5) was highest in the highly susceptible genotypes (55%) and lowest in the validated resistant genotypes (15%). The percentage seed damage (Figure 6) was highest in the highly susceptible genotypes (90%) and lowest in the validated resistant genotypes (49%). The number of exit holes (Figure 7) was fewer in the validated resistant genotypes and highest in the highly susceptible genotypes.

The percentage pest tolerance (Figure 8) showed an ascending trend from highly susceptible genotypes (10%) to validated resistant genotypes (60%), which is higher than the positive check (25%). The percentage weight (Figure 9) loss showed a decreasing trend from highly susceptible genotypes (45%) to validated resistant genotypes (20%). The growth index (Figure 10) was lowest in the validated resistant genotypes and highest in the highly susceptible genotypes.

The Dobie susceptibility index (Figure 11) showed a similar trend, with the validated resistant genotypes having the lowest DSI (<3) and the highly susceptible genotypes having the highest DSI (>10). These results demonstrate the effectiveness of the validated resistant genotypes in resisting bruchid infestation and damage.

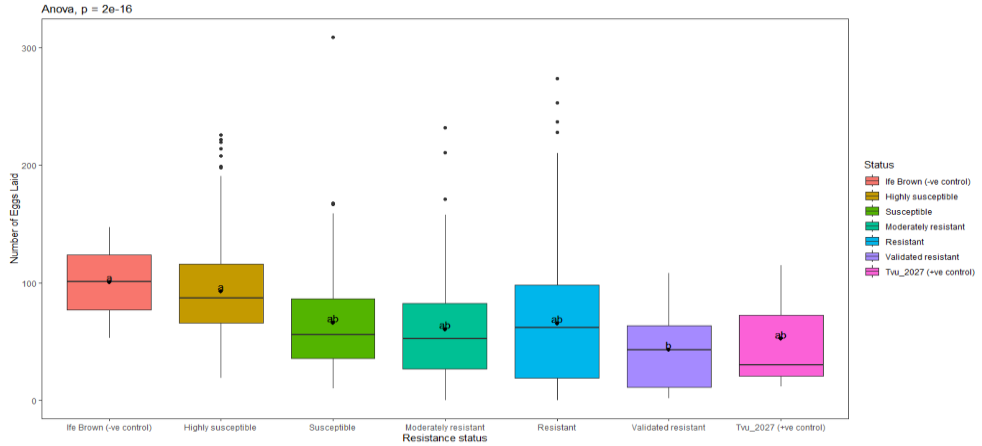


Figure 2: Number of Eggs Laid by *Callosobruchus maculatus* on Cowpea Varieties with Varying Resistance Levels

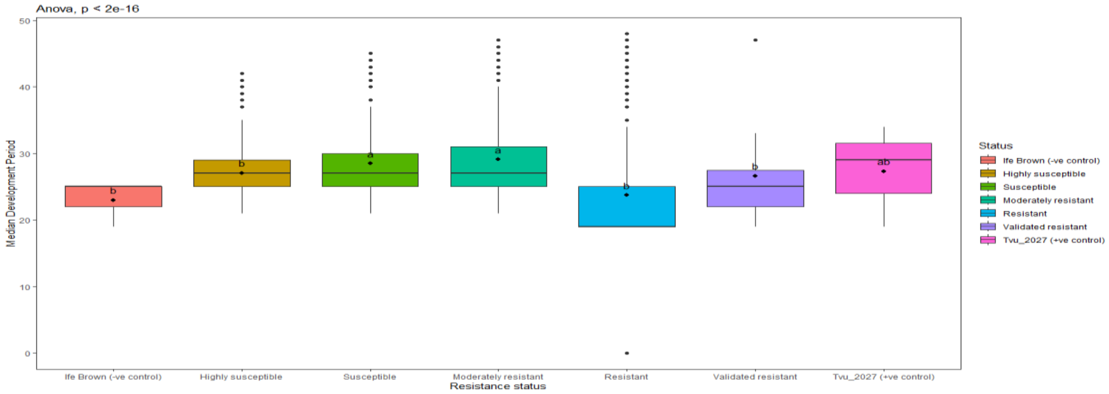


Figure 3: Median Development period of *Callosobruchus maculatus* on Cowpea Genotypes with varying Resistance Levels

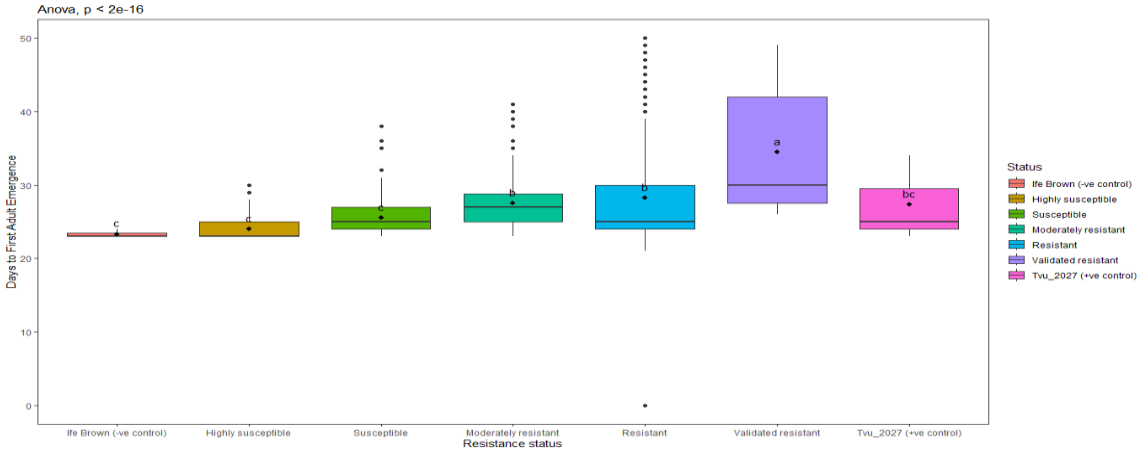


Figure 4: Mean Days to First Adult Emergence of *Callosobruchus maculatus* on Cowpea Genotypes with varying Resistance Levels

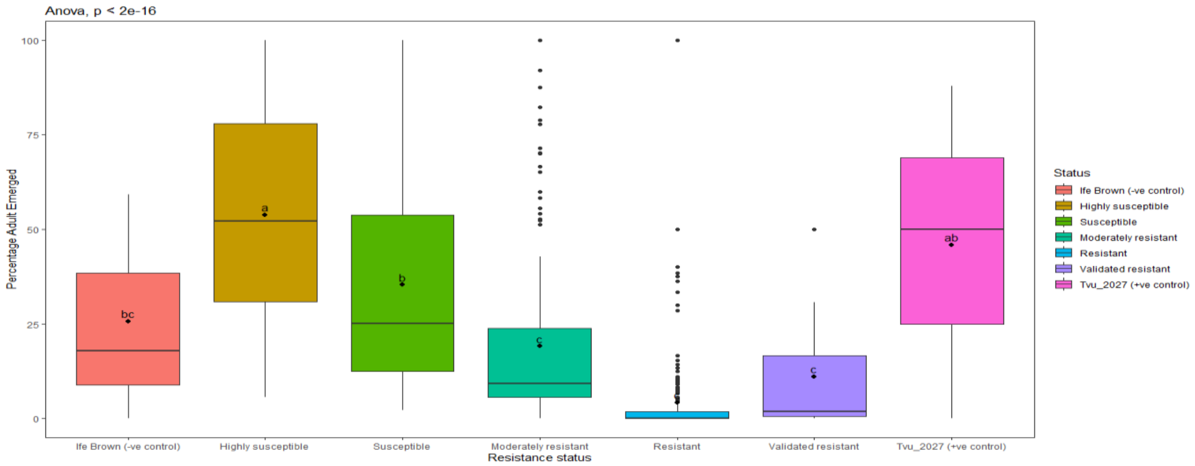


Figure 5: Percentage Adult Emergence of *Callosobruchus maculatus* on Cowpea Genotypes with varying Resistance Levels

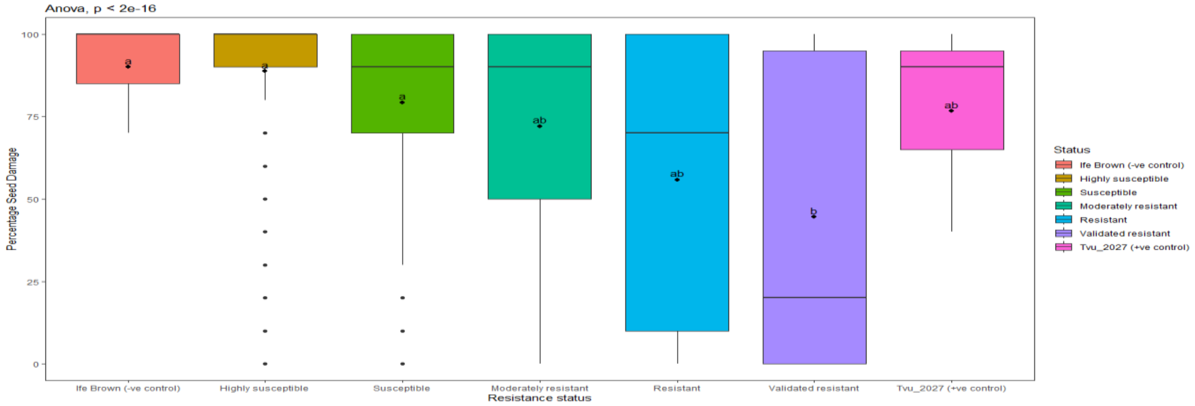


Figure 6: Percentage of Seed Damage by *Callosobruchus maculatus* on Cowpea Genotypes with varying Resistance Levels

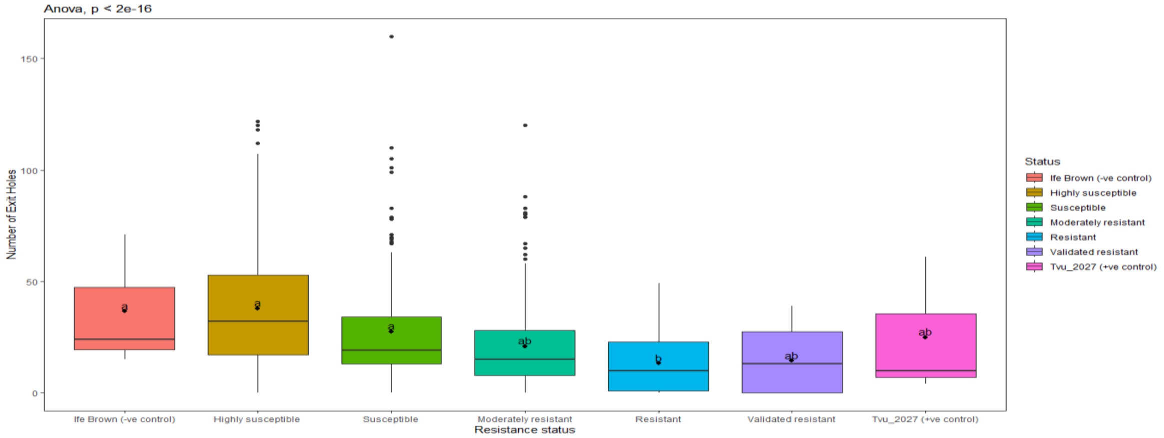


Figure 7: Number of Exit Holes on Cowpea genotypes from *Callosobruchus maculatus* infestation

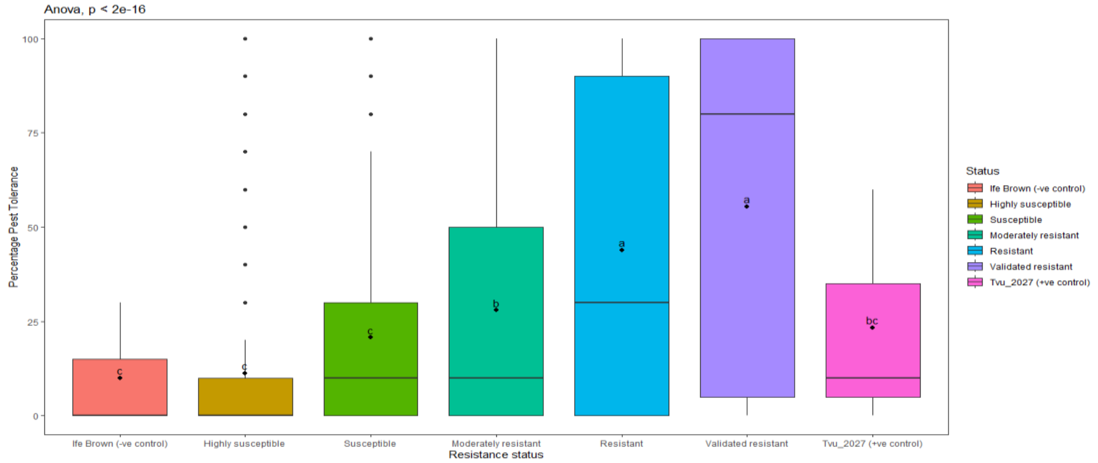


Figure 8: Percentage Pest (Bruchid) Tolerance by Resistance Levels

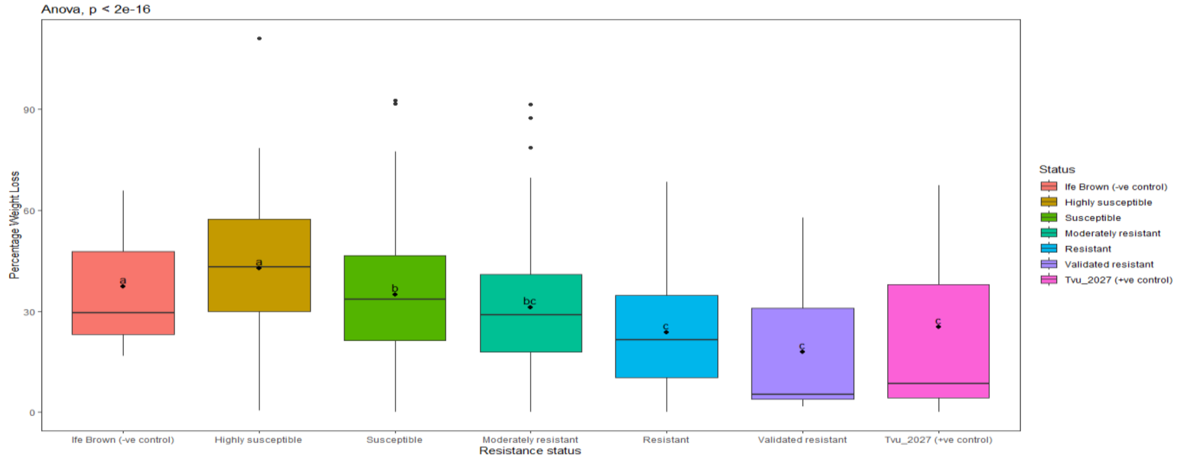


Figure 9: Percentage Weight Loss of cowpea Genotypes by Resistance Levels

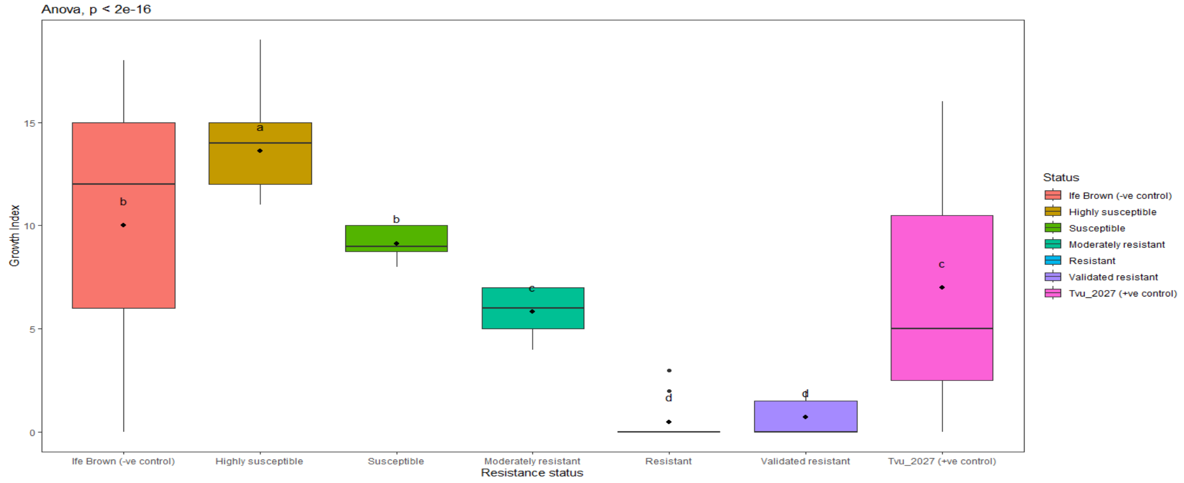


Figure 10: Growth Index by Resistance Levels Status of the Cowpea Genotypes

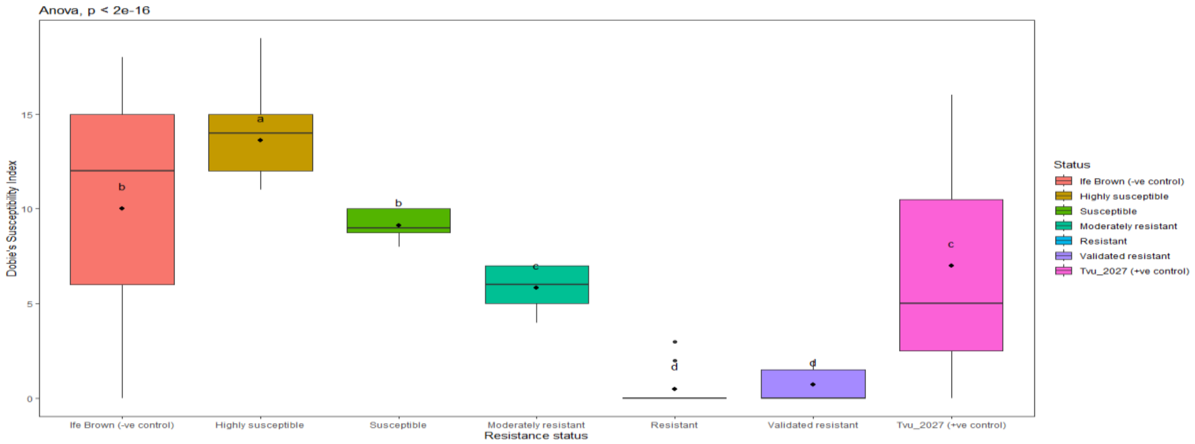
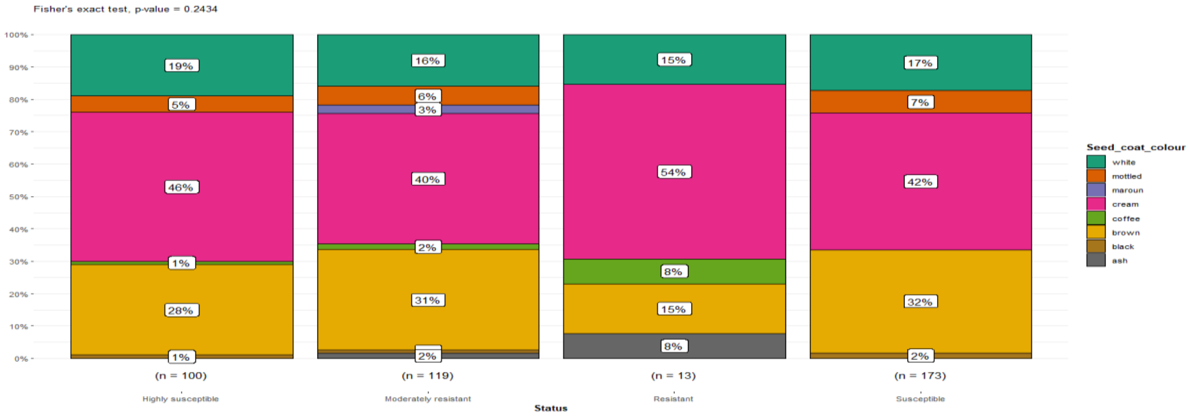


Figure11: Dobie Susceptibility Index of the Cowpea Landraces based on Bruchid Infestation

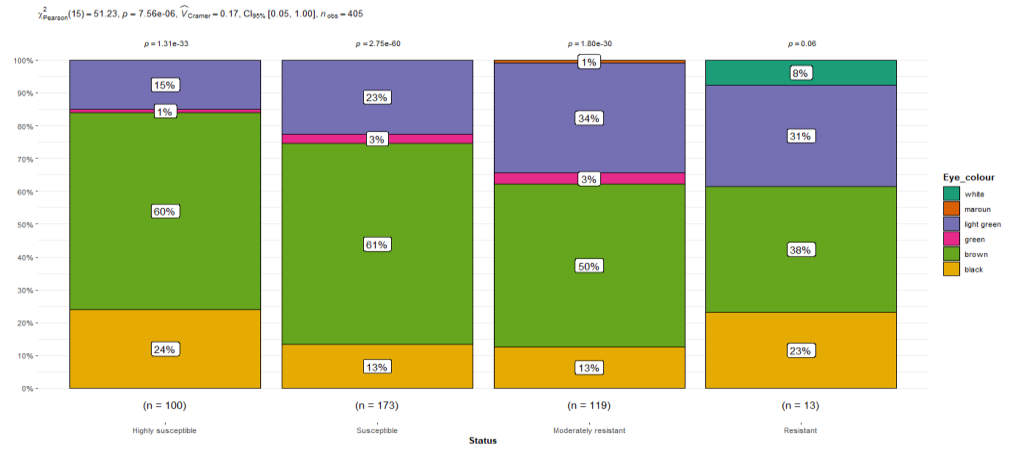
Effect of Seed Morphometric Traits on Bruchid Resistance

The relationship between seed morphometric traits and bruchid resistance was investigated in a collection of cowpea genotypes. The results showed that there was no significant association between bruchid resistance and several seed morphometric traits, including: seed coat color, eye color), seed shape, seed luster and seed texture (Figures 12 and 13). These findings suggest that these seed morphometric traits do not play a significant role in determining bruchid resistance in cowpea. The result shows no significant association between bruchid resistance and several seed morphometric traits in the cowpea genotypes studied. This suggests that these traits may not be reliable predictors of bruchid resistance in cowpea.

A



B



C

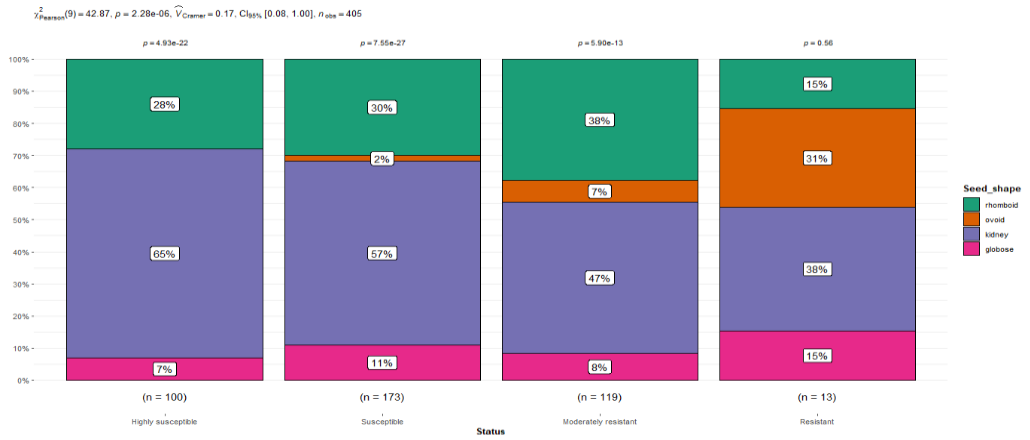


Figure 12 (A-C): Relationship between seed coat colour(A) eye color(B), seed shape (C) and bruchid resistance

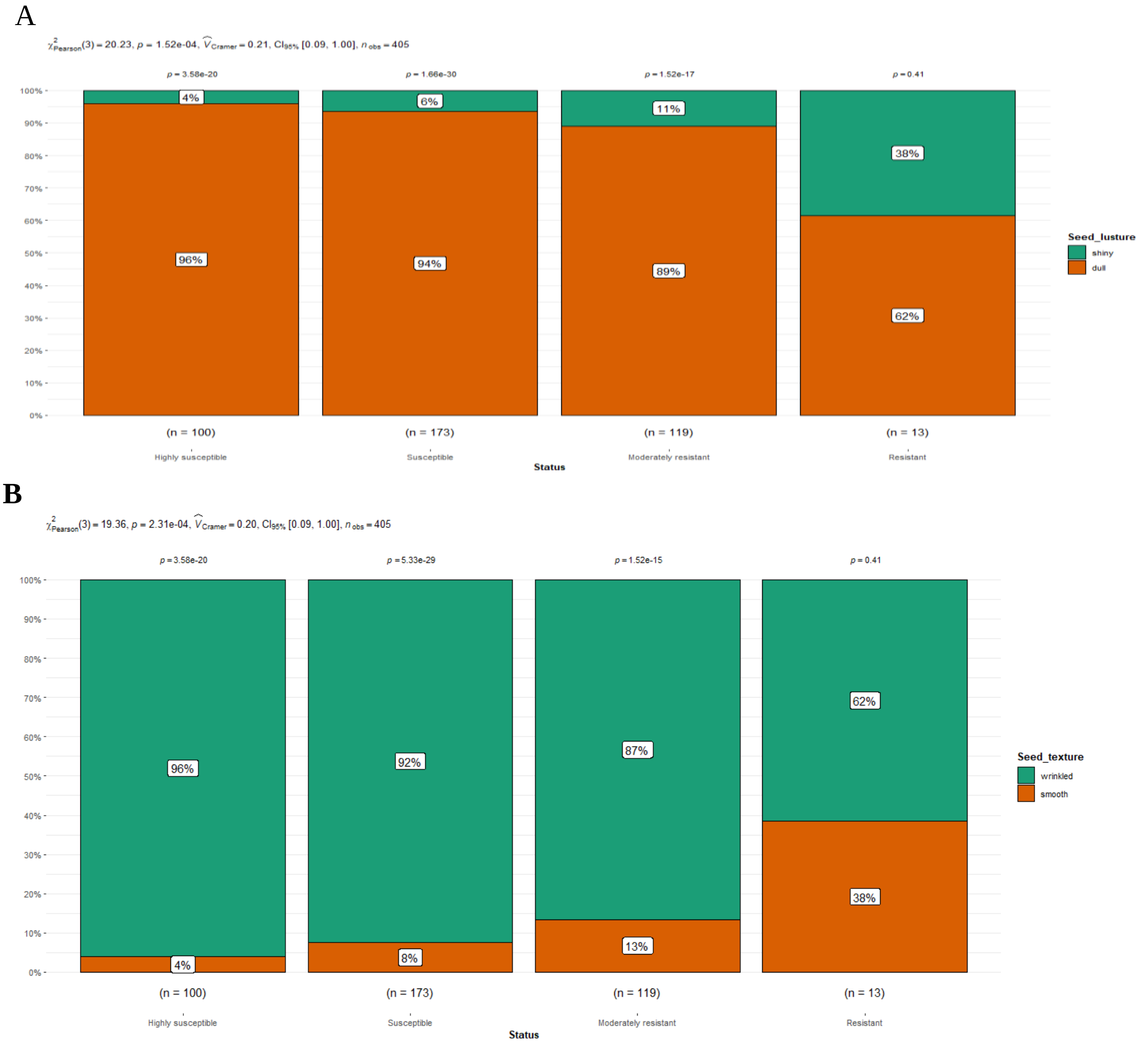


Fig. 13(A-B): Relationship between seed lustre (A), tecture (B) and bruchid resistance

DISCUSSION

The significant differences observed among the cowpea genotypes for insect growth and damage parameters suggest the presence of genetic variability for these traits, which is essential for crop improvement (Miesho *et al*., 2018). This variability provides a valuable resource for breeding programs aimed at improving cowpea resistance to bruchid infestation. However, the non-significant association between bruchid resistance and seed morphometric traits indicates that seed characters are not reliable identifiers for bruchid resistance (Pankaj & Singh, 2011; Maina *et al*., 2012; Cruz *et al*., 2016). This suggests that bruchid resistance in cowpea is a complex process involving biochemical and morphological mechanisms that cannot be solely predicted by seed morphometric traits (Singh, 2002).

Studies have attributed bruchid resistance in cowpea seeds to morphological traits such as seed coat texture, seed size, seed color, and seed thickness (Mei *et al*., 2009; Amusa *et al*., 2013; Chakraborty & Mondal, 2016). Our findings suggest that these traits do not significantly influence bruchid resistance. Instead, the variability in bruchid growth and damage parameters observed in this study agrees with previous reports by Amusa *et al*. (2021).

The percentage adult emergence (PAE) is a critical measure of larval mortality, which is a reliable indicator of seed susceptibility (Amusa *et al*., 2018). The significant positive correlation among the percentage weight loss (PWL), number of exit holes (NEL), and PAE is consistent with previous findings (Tripathi *et al*., 2012). These results suggest that the susceptibility of cowpea seeds to bruchid attack is related to the rate of larval development and median development period (Kosini *et al*., The lack of association between seed morphometric traits and bruchid resistance highlights the importance of exploring multiple factors contributing to resistance. As noted by Kell *et al*. (2017), a single-trait approach may not be sufficient to improve bruchid resistance in cowpea, and a more comprehensive understanding of the factors influencing resistance is necessary.

CONCLUSION

This study revealed significant genetic variability for bruchid resistance in the cowpea genotypes evaluated and indicate potential for genetic improvement. A subset of 13 genotypes (approximately 3.2%) demonstrated resistance to bruchid infestation, with five genotypes (ABU_Vu16, ABU_Vu39, ABU_Vu225, ABU_Vu239, and ABU_Vu309) consistently exhibiting high levels of bruchid tolerance (>70%) and low Dobie Susceptibility Index (DSI) values (<3) upon validation. The results identified percentage adult emergence (PAE), growth index (GI), and DSI as reliable indicators of bruchid resistance. Notably, cowpea seed morphometric traits showed no significant 2017). Overall, our findings highlight the importance of considering multiple parameters when evaluating bruchid resistance in cowpea genotypes. The significant genetic variability observed in this study provides a valuable resource for breeding programs aimed at improving cowpea resistance to bruchid infestation.

Seed morphometric traits may not be reliable predictors of bruchid resistance in cowpea. Studies have shown that bruchid resistance in cowpea is a complex trait influenced by multiple factors, including biochemical composition (Mei *et al*., 2009), protein content (Singh *et al*., 1985), and genetic factors (Tengey *et al*., 2023). Therefore, relying solely on seed morphometric traits may not be effective in improving bruchid resistance. Breeding programs have successfully used a multi-trait approach to improve bruchid resistance in cowpea. Amusa *et al*. (2018) identified cowpea genotypes with high levels of bruchid resistance and desirable seed traits through a comprehensive screening program. Similarly, Tripathi *et al*. (2020) developed cowpea varieties with improved bruchid resistance by selecting multiple traits (seed protein content and genetic markers).

association with bruchid resistance, suggesting that relying solely on seed morphometric traits may not be effective in improving bruchid resistance in cowpea. Instead, a multi-trait approach that considers multiple factors, including biochemical composition, protein content, and genetic factors, may be necessary to develop cowpea varieties with improved bruchid resistance. Overall, this study contributes to the understanding of the factors (percentage adult emergence, growth index, and Dobie susceptibility index) influencing bruchid resistance in cowpea and provides insights for the development of effective breeding strategies to improve cowpea resistance.

The findings of this study highlight the importance of exploring multiple factors and support the use of a comprehensive breeding program that selects for multiple traits to improve bruchid resistance in cowpea.

Acknowledgement

This research was funded by TETFUND under the National Research Fund, 2020, with grant number TETFund/DR&D/CE/NRF/STI/04/VOL1.

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