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Effects of Cucumber Mosaic Virus on Yield Components and Yield of Bambara Groundnut

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Abstract: Bambara groundnut (*Vigna subterranean* L. Verdc) is a leguminous plant and its seeds are highly nutritious and can be considered as complete food. In addition to being a neglected crop; its production is limited by several constraints among which virus diseases are the most implicated. In order to contribute to the improvement of bambara groundnut in Nigeria, landraces of bambara groundnut were screened for resistance to *Cucumber mosaic virus* (CMV), one of the most damaging viruses in grain legumes. Treatments consisted of five landraces namely Black, Brown, Cream, Red nav, and Zebra coloured got from local farmers. These were arranged in Randomised Complete Block Design, with three replicates. Seedlings were infected with CMV isolate three weeks after sowing (WAS), during the 2019 and 2020 trials. The infected plants exhibited various symptoms of chlorosis, leaf deformation, growth retardation, and plant stunting. The sCMV caused significant reduction in plant height, the number of branches, leaves, pods, and seeds weight. In the 2019 and 2020 trials, 100 % incidence with the highest severity was observed in the Bambara groundnut landraces. Disease incidence varied between 41.6% in “Black” and 95.8 % in “Cream,” while disease severity was higher in “Cream” and “Red nav” with 5.0 respectively. “Black” and “Zebra” were the tallest with 16.8 cm among the infected as well as disease-free with 22.6 cm tall plants. “Black” landrace produced the highest number of pods in both CMV infections with 21 and disease-free with 33 pods respectively. Conversely, the number of days to flowering of 43 days was highest in infected plants of “Black”. The “Black” and “Zebra” landraces recorded uniform seed weights and the highest seed weight under CMV infection with 1.7 g per plant. The impact of the virus on yield loss was lowest in “Black” and “Zebra” landraces which indicated their possible use in the management of *Cucumber mosaic virus* disease in Bambara groundnut.

Keywords: Bambara Groundnut, Cucumber Mosaic Virus, Disease Incidence and Severity Impact, Growth and Yield Loss

1. Introduction

Bambara groundnut (*Vigna subterranean* L. Verdc) is a leguminous plant and its seeds are highly nutritious and can be considered as complete food [1]. It is a significant grain legume in Nigeria, especially in rural areas [2, 3]. The crop is suited to a variety of climatic and ecological circumstances, and it thrives in the country's semi-arid climate. It contributes to soil fertilization as a leguminous plant by symbiotically fixing atmospheric nitrogen [2]. West Africa is the main Bambara groundnut production region in SSA, where Burkina Faso, Niger, and Cameroon are the leading producers, contributing to 74% of global production [3].

Except for riverine and swampy places, the crop is now grown throughout the country [4]. Bambara groundnut is commonly intercropped with maize (*Zea mays* L.) and Guinea corn (*Sorghum bicolor* L.) in Nigeria by subsistence farmers on small plots of land to supply nutrients to the intercrops [2-4].

After cowpea (*Vigna unguiculate* **L.** Walp) and groundnut (*Arachis hypogaea* L.), the crop is now widely dispersed in sub-Saharan Africa's semi-arid zone, and most authors appear to agree that it is the third most significant food legume [5]. It is known as "Okpa" by the Igbo, **"**Kwaruru**"** or "Gurjiya" by the Hausa, "Epa-roro" by the Yoruba, "Edzu" by the Nupe, and "Abwui" by the Gbagi. It is an annual herbaceous crop that grows to a height of 0.30 **-** 0.35 m and comes in both upright and prostrate varieties [4].

It's used to prepare a paste out of dried seeds, which is then used to make the fried or steamed delicacies "Moin-moin" and "Akara" [6]. This crop has been used in Nigeria to make a tasty delicacy called "Okpa," which is a doughy mixture wrapped in banana leaves and boiled. The demand for Bambara groundnut is growing as a result of its multiple uses, strong nutritional and, use as a primary source of sustenance for diabetic patients. Consumption of Bambara groundnut seed is safe as long as it is properly prepared [7]. It shares high nutritive values like other popularly consumed African legumes and, capable of growing in arid land where groundnut, maize and guinea corn have failed [8]. It is an excellent supplement to cereals and tubers that are the main food sources in sub-Saharan Africa. Despite its multiple economic benefits, Bambara groundnut is underutilized; landraces are unimproved as a result of scientists' disregard of the crop [9].

Bacteria, fungi, insect pests, nematodes, and viruses are among the biotic agents that limit output [10]. *Cucumber mosaic virus* (CMV, genus *Cucumovirus*) is a common legume virus that has been reported in Bambara groundnut farms [11]. Although insecticidal control of the virus's insect vectors can reduce the CMV disease incidence and severity, the strategy's numerous unfavorable consequences exceed its benefits. These include insect resistance, contamination of the environment and soil, health risks for applicators, and pesticide residues in the food supply. High costs and a lack of application knowledge are also stumbling blocks. The practice of close spacing and early planting has also been recorded as part of the virus management process. Furthermore, intercropping Bambara ground nut with tall cereals like maize or millet, as well as keeping the land weed-free, has been suggested [12]. Also [13] proposed that resistant cultivars be planted. Only virus-free seeds should be used, and quarantine precautions should be followed [14]. The most effective technique for managing legume virus infections is certainly host-plant resistance.

The presence of virus infections in legume populations is of enormous economic importance since it results in a loss of seed output and infected seeds, which serve as a channel for germplasm exchange [11]. The prevalence, epiphytotic potential, general severity, and high vulnerability of locally cultivated landraces all make CMV disease problematic to legume crops in Nigeria. The virus can be found in any field where legumes are planted. It is especially dangerous when it is combined with other infections. This pathogen can cause full crop loss in several Nigerian Bambara groundnut landraces.

Virus infections, unlike other plant pathogens, cannot be managed directly with chemical application. As a rapid response to vector resurgence and long-range virus transmission, farmers apply synthetic insecticides. Apart from being costly for smallholder farmers, this frequently results in pest resistance and detrimental repercussions on humans, livestock, and the environment. Insecticidal residues on Bambara groundnut leaves, pods, and grains are increasing as a result of indiscriminate and excessive application of synthetic insecticides, which has become a major concern. If insecticidal residue in seeds exceeds the Maximum Residue Limit (MRL)., seed businesses may face significant financial losses and their germplasm may be rejected outright on the foreign market. For example, the European Union (EU) has prohibited Nigeria from exporting cowpea seeds due to unacceptably high pesticide residual levels [14].

One of the biggest impediments to Bambara groundnut production, according to (15) is virus infections. Virus infection might result in a 60 % reduction in output. Extensive study has been done on screening legumes for resistance to CMV, such as cowpea, groundnut, and soyabeans, but little has been done on Bambara groundnut. This study was set up to bridge this gap in knowledge where the identified resistant Bambara groundnut landraces will play a key role in the development of high-yielding, virus-tolerant germplasm. Also identified seed transmission of CMV in infected seeds will give baseline data on the pathogen's epiphytology and possible mechanisms of long-range transmission. Identified resistant Bambara groundnut land races from this study will form genetic traits in hybridization studies for the development of CMV - resistant varieties for the crop's growers.

2. Materials and Methods

2.1 Description of the Study Location

During the 2019 cropping season, the experiment was carried out in the Teaching and Research Farm of the Department of Crop Production, Federal University of Technology, Minna, Nigeria. The site's coordinates are 9º.51' N, 6º.44' E, and 212 m above sea level, as measured with Geographical Position System (GPS- 4300). For the past four years, the site has been planted with maize and millet.

2.2 Collection and Sterilization of Soil Samples

Soil samples were taken from the Teaching and Research Farm in the following proportions: 60 river sand, 30 loamy soil, and 10 thoroughly decomposed cow dung. In order to eliminate soil microorganisms and insect pests, the collected soil samples were mixed and sterilized. The steam method was used to sterilize the soil, as described by [16]. Upper and lower metal components made up the trough. The soil was held in the upper metal piece, which was perforated at the bottom, while the water was held in the lower metal piece. To assemble the trough, the bottom component was placed on a metal platform supported by three legs and half-filled with water. The upper component was positioned on top of the lower portion, which was filled with soil and covered with thick sacking, and then a relatively tight fit was constructed through which a hole was made to allow the thermometer to be placed into the top of the soil. The covering was done to assure sterilization down to the soil's surface. Dry woods were stacked between the metal stand and set ablaze. The steam generated by the boiling water in the lower piece passed through the perforations at the bottom of the upper piece, sterilizing the soil until it reached 100 °C.

2.3 Land Preparation and Ridging

The previous harvest was removed from the land during the first week of August 2019. At a 0.75 m inter-row spacing, ridges measuring 2 m long were constructed. For each landrace in each replicate, four ridges were constructed.

2.4 Treatments and Experimental Design

The treatments consisted of five landraces of Bambara groundnut viz: Black, Brown, Cream, Red nav, and Zebra commonly grown in Minna. *Cucumber mosaic virus*-inoculated and uninoculated (control) plots were used to assess each landrace Set up in Randomised Complete Block Design (RCBD) with three replicates was used.

2.5 Source of Seeds

Bambara groundnut seeds were collected from the local farmers in Gidan Kwano Village, Minna, Niger State. These were as indicated under treatment and experimental design section above.

2.6 Preparation of Virus Extraction Buffer

In distilled water, 0.1M sodium phosphate dibasic (NaHPO4), 0.1M potassium phosphate monobasic (K2PO4), 0.01M EDTA (Ethylene diamine tetra acetic acid), and 0.001M L-cysteine were used to make a virus extraction buffer with a pH of 7.2. Each reagent was placed in a plastic cylinder, which was then filled with 600 mL of distilled water. The contents of the cylinder were swirled on a magnetic stirrer until the salts were entirely dissolved. The buffer's pH was adjusted to 7.2 by adding HCl or NaOH as necessary as described by [17].

2.7 Source of virus inoculum and inoculation procedure

From the stock in the Department of Crop Production, Federal University of Technology, Minna, a severe isolate of CMV inoculum that is, CMV-infected cowpea leaf as reported by [18] was obtained. In a screenhouse, the inoculum was increased by mechanical propagation onto seedlings of an indicator cowpea cv. Ife Brown, through sap transmission according to [17]. Ife Brown seeds were sown in sterilized soil in 10 plastic pots measuring 30 cm diameter and 40 cm deep. The isolate was ground at a rate of 1 g/mL in virus extraction buffer. The upper leaf surface of the indicator cowpea plants was dusted with carborundum powder got from 600-mesh 10 days after sowing (DAS) to produce an opening on the leaf surface.

In order to aid virus penetration into the plant, one microlitre of β-mercaptoethanol was added to the virus extract. The upper leaf surface was gently rubbed with a piece of cheesecloth dipped in the extract. The inoculated plants were rinsed with sterile distilled water and kept in the screenhouse to monitor their symptoms. The inoculated plants were then sprayed with lambda-cyhalothrin (Karate 5 EC) insecticide at a rate of 60 mL/20L water. A 20 L backpack sprayer (Cooper Pegler, Burgess Hill, Sussex, and UK) was used to apply the insecticide at high pressure. At two weeks after inoculation (WAI), symptomatic leaves were taken from the inoculated plants and stored in vial bottles over self-indicating silica gels for use in the main experiment.

2.8 Sowing and Crop Management

Two seeds of Bambara groundnut plants were sown at a 0.25 m intra-row spacing in the second week of August 2019. The emerged seedlings were thinned to one plant per stand. Weeds were manually controlled at three and six weeks after sowing.

2.9 Seedling Inoculation with CMV

Inoculation was done with the CMV-infected leaves recovered from the stored CMV-infected leaves. The grinding, inoculation period and inoculation technique were all done as described above.

2.10 Seed Transmission Trial

2.10.1 Source of Bambara seeds

Bambara seeds harvested during the 2019 trial were used in the field establishment and seed transmission studies.

2.10.2 Field Establishment and Seedling Inoculation

The 2020 field trial was cited in Minna, Niger State, at Gidan-Mangoro (9º 56'N; 6º 48'E; 214 m above sea level). The techniques for land preparation, treatments and experimental design, planting, and crop management were as described above. The plants were not inoculated with CMV, but lambda-cyhalothrin insecticide was sprayed two weeks after sowing (WAS), as specified above.

2.10.3 Data collection and analysis

Data were collected on disease incidence, disease severity, growth and yield components. Using the Statistical Analysis System, the data were subjected to analysis of variance (ANOVA). Duncan's Multiple Range Test (DMRT) was used to rank the treatment means at a 5 % probability level [19].

3. Results

3.1 Incidence and Severity of *Cucumber Mosaic Virus* Disease

The first symptoms of CMV infection were observed at 1 WAI during the 2019 crop season. At varying levels, the infected plants displayed typical leaf symptoms. Mild leaf chlorosis, vein chlorosis, and yellow patches on the secondary leaves of infected plants were the symptoms. The disease incidence did not differ significantly (p>0.05) among the landraces at 1 WAI, however, the "Cream" landrace recorded the highest incidence of 33.9 %. The "Zebra" landrace was next with 33.3 %, and the "Black" landrace was third with 25.3 % infection (Table 1). The disease incidence of the "Brown" and "Red nav" landraces, however, was found to be similar with 29.4 %.

Disease incidence varied significantly (p<0.05) among the five landraces at 2 WAI, with the "Cream" landrace recording the highest percentage of 70.8 disease incidence compared to the "Zebra" landrace recording the lowest percentage of 40.2 disease incidence. The "Red nav" landrace recorded a disease incidence of 62.4 %, whereas the "Brown" landrace recorded a level of infection of 54.1 %, and the "Black" landrace had a disease incidence of 46.1 %. There were significant (p<0.05) differences among the landraces at 3 WAI, with the "Cream" landrace recording the highest incidence of 95.6 %. The "Red nav" landrace was next, with an 87.4 % disease incidence. The "Black" and "Brown" landraces both recorded 58.1 % disease incidence, whereas the "Zebra" landrace recorded the least incidence (45.9 %).

Healthy plants were symptomless in 2020, but inoculated plants had foliar symptoms comparable to those observed in the 2019 study. The "Cream" landrace recorded the highest disease incidence of 45.8%, followed by the "Red nav" landrace with 37.5 %, which did not differ significantly (p>0.05) from the "Cream" landrace at 1 WAS. The "Brown" and "Zebra" landraces recorded 33.3 % and 29.1 %, disease incidence respectively which were not statistically different from that of the "Black" landrace with 20.8 %. The landraces differed significantly (p<0.05) at 2 WAS among themselves in disease incidence. The “Cream” landrace recorded the highest incidence of CMV infection of 75.6 %, which did not differ significantly (p>0.05) from the “Brown” landrace's incidence of 62.5 %. The disease incidence of the "Red nav" landrace was greater (58.3%) than that of the "Zebra" landrace (41.6 %). The "Black" landrace recorded the least of infection with 33.3 %.

There were significant (p<0.05) differences in disease incidence among the landraces at 3 WAS, with the “Cream” landrace eliciting 100 % disease incidence. Between the "Brown" and "Red nav” landraces with disease incidence of 79. 1 and 75.0 % respectively, there was no significant difference.

Table 1: Disease incidence on Bambara groundnut plants inoculated with Cucumber mosaic virus disease in Minna during 2019 and 2020 cropping seasons

Landrace colour ID	Disease incidence (%) 2019			Disease incidence (%) 2020		
	1 WAI	2 WAI	3 WAI	1 WAS	2 WAS	3 WAS
Black	25.3^{a}	46.1^{cd}	58.1^{b}	20.8^{c}	33.3^{c}	41.6^{d}
Brown	29.4^{a}	54.1^{bc}	58.1^{b}	33.3^{bc}	62.5^{a}	79.1^{b}
Cream	33.9^{a}	70.8^{a}	95.6^{a}	45.8^{a}	75.6^{a}	100.0^{a}
Red nav	29.4^{a}	62.4^{ab}	87.4^{a}	37.5^{ab}	58.3^{b}	75.0^{b}
Zebra	33.3^{a}	40.2^{d}	45.9^{b}	29.1^{bc}	41.6^{bc}	54.1^{c}
±SEM	3.6	3.4	7.7	4.3	4.3	3.7

Means with dissimilar letter (s) within the column differ significantly (p≤0.05) using Duncan Multiple Range Test (DMRT)

WAI = Week (s) after inoculation; WAS = Week (s) after sowing

Table 2. Disease severity on Bambara groundnut plants inoculated with Cucumber mosaic virus disease in Minna during 2019 and 2020 cropping seasons

Landrace colour ID	Disease severity 2019			Disease severity 2020		
	3 WAI	6 WAI	9 WAI	3 WAS	6 WAS	9 WAS
Black	2.6^{a}	4.0^{b}	4.6^{a}	2.3^{ab}	3.0^{c}	3.8^{b}
Brown	2.3^{a}	3.6^{c}	4.6^{a}	3.3^{a}	3.6^{bc}	4.6^{a}
Cream	3.3^{a}	4.6^{a}	5.0^{a}	3.0^{a}	4.3^{a}	5.0^{a}
Red nav	3.0^{a}	5.0^{a}	5.0^{a}	2.6^{a}	4.0^{ab}	4.4^{ab}
Zebra	2.3^{a}	3.2^{c}	4.3^{b}	2.4^{a}	3.5^{bc}	4.4^{ab}
±SEM	0.2	0.2	0.2	0.2	0.2	0.2

Means with dissimilar letter (s) within the column differ significantly ($p \leq 0.05$) using Duncan Multiple Range Test (DMRT)

WAI = Week (s) after inoculation; WAS = Week (s) after sowing

There were significant ($p<0.05$) differences in disease incidence among the landraces at 3 WAS, with the “Cream” landrace eliciting 100 % disease incidence. Between the "Brown" and "Red nav” landraces with disease incidence of 79. 1 and 75.0 % respectively, there was no significant difference. The "Zebra" landrace recorded 54.1 % incidence rate, whereas the "Black" landrace recorded 41.6 % incidence. The severity of CMV infection on inoculated plants did not differ significantly at 3 WAI during the cropping season of 2019. (Table 2). The "Cream," landrace on the other hand, exhibited the highest severity of 3.3. The severity scores for the “Red nav" and "Black" landraces were 3.0 and 2.6 respectively. The severity score for the “Brown” and “Zebra” landraces was 2.3. The five landraces showed significant variations ($p<0.05$) at 6 WAI. Except for the “Cream” landrace with severity score of 4.6, the “Red nav” landrace recorded the highest value of 5.0, which was significantly different ($p<0.05$) from all the other landraces, followed by the “Black” landrace with 4.0. The severity scores of the "Brown" and "Zebra" landraces, of 3.6 and 3.3, respectively, were not significantly different ($p>0.05$) from each other. The "Cream" landrace and "Red nav" landrace recorded the highest disease severity of 5.0 at 9 WAI, but their severity did not differ significantly from that of the "Black" and "Brown" landraces, which was 4.6. The "Zebra" landrace, on the other hand, recorded least severity of 4.3.

In 2020, the severity of disease among infected plants differed significantly ($p<0.05$) (Table 2). The "Brown" landrace with score of 3.3 was the highest disease severity, followed by "Cream" with score 3.0, "Red nav" with score of 2.6, "Zebra" landrace with score of 2.4, and "Black" landrace with 2.3s. The “Cream” landrace with score of 4.3was the highest disease severity at 6 WAS but did not differed significantly from the “Red nav” landrace ($p>0.05$) with 4.0 severity. There was also no significant ($p>0.05$) difference in disease severity between the “Brown” and “Zebra” landraces with scores of 3.6 and 3.5, respectively. The "Black" landrace, with a severity score of 3.0, recorded the least severity of infection. The "Cream" landrace's disease severity peaked at 5.0 at 9 WAS, but the value did not differ significantly ($p>0.05$) from that of the "Brown" landrace with 4.6. The "Red nav" and "Zebra" landraces, on the other hand, recorded similar levels of disease severity of 4.4, while the "Black" landrace recorded the least disease severity of 3.8. Plate I depict the plants' reactions at 3 WAI.

3.2 Effect of Cucumber Mosaic Virus Disease on Plant Height

The CMV infection induced a considerable reduction in the height of inoculated plants compared to uninoculated plants of Bambara groundnut landraces during the 2019 cropping season (Table 3). Throughout the trial period, the inoculated plants showed poor growth, low vigor, thin stems, and short internodes. The uninoculated plants, on the other hand, were taller than the infected plants (Table 3). When only uninoculated plants were considered at 3 WAI, the "Zebra" landrace produced the tallest plants of 20.1 cm, whereas the "Red nav" landrace produced the shortest of 17.3 cm tall. In addition, the "Brown," "Black," and "Cream" landraces were 19.6 cm, 18.7 cm, and 17.6 cm tall, respectively. The "Black" landrace, with a height of 15.8 cm, was the tallest among the CMV-inoculated plants, followed by the "Zebra" landrace, with a height of 15.7 cm. Furthermore, "Red nav"with 14.7 cm plants were taller than "Brown" landrace with 13.4 cm, while "Cream" landrace infected plants were the shortest with 11.5 cm. The "Red nav" landrace recorded the least height reduction of 15.0 %, followed by the "Black" landrace, which had a height reduction of 15.5 %. The remaining landraces, on the other hand, showed less than 40 % height reductions (Table 3).

Uninoculated plants recorded a minimum height of 18.7 cm in the "Cream" landrace and a maximum of 21.6 cm in the "Zebra" landrace at 6 WAI. The CMV-inoculated plants on the other hand, ranged in height from 12.4 cm in Cream landrace to 16.9 cm in Zebra landrace (Table 3). Infected plants of the "Red nav" landrace recorded the least height reduction of 15.3 %, followed by infected plants of the "Black" landrace, with a mean height reduction of 18.0 %. The “Cream” landrace, on the other hand, recorded the highest height reduction of 33.7 %. The height reduction in infected plants of the "Brown" and "Zebra" landraces was 30.6 and 21.8 %, respectively (Table 3).Uninoculated plants of the "Zebra" landrace were the tallest at 9 WAI, with 22.6 cm in height, followed by "Brown” landrace with 21.8 cm, and "Black" landrace with 20.9 cm, while "Cream" and "Red nav" landraces were the shortest with 19.3 cm. The inoculated plants, on the other hand, were between 13.0 cm in Cream landrace and 17.3 cm in Zebra landrace tall. Furthermore, the CMV-inoculated plants of the “Black” and “Red nav” landraces grew to the same height of 16.8 cm, while the “Brown” landrace grew to 15.1 cm (Table 3). The height decreases were moderate, ranging from 13.0 % in the "Red nav" landrace to 32.6 % in the "Cream" landrace. The infected "Black" landrace's height reduction of 19.6 % was less than the "Zebra" landrace's of 23.5 cm, but the inoculated "Brown" landrace's plants recorded a 30.7 % height reduction (Table 3).

In 2020, CMV infection caused significant ($p<0.05$) height reductions in infected plants, whereas healthy plants grew rapidly. The diseased plants grew slowly and had low vigour (Table 4). Healthy plants ranged in height from 17.7 cm in the "Cream" and "Red nav" landraces to 19.8 cm in the "Black" landrace at 3 WAS. Healthy "Brown" and "Zebra" plants, on the other hand, grew to a consistent height of 19.5 cm. The heights of the CMV-inoculated plants ranged from 12.0 cm in “Cream” landrace to 15.8 cm in “Zebra” landrace. The average height of the “Black” landrace was 15.1 cm, while the “Brown” and “Red nav” landraces were uniformly 14.9 cm tall. The "Red nav" landrace recorded the least height reduction of15.8 %, while the "Zebra" landrace recorded the highest reduction of 19.0 %. Infected plants of the “Black” and “Brown” landraces lost 23.7 and 23.6 % of their heights respectively, whereas infected plants of the “Cream” landrace suffered the highest height reduction of 32.2 %.

All healthy plants were significantly taller ($p>0.05$) than CMV-infected plants at 6 WAS. Among the healthy plants, heights ranged from 18.2 cm in “Red nav” landrace to 21.5 cm in “Zebra” landrace. The "Cream" landrace's healthy plants were 18.4 cm tall on average, whereas the "Brown" and "Black" landraces were 21.0 and 21.3 cm tall, respectively. The "Cream" landrace recorded a plant height of 12.5 cm, while the "Black" landrace recorded a height of 16.2 cm. Plant heights in the "Brown," "Red nav," and "Zebra" landraces, on the other hand, averaged 15.1 cm, 15.7 cm, and 16.0 cm, respectively (Table 4). The "Red nav" landrace recorded the least height reduction of 13.7 %, while the "Cream" landrace recorded the highest reduction by 32.1 %. Furthermore, the "Black," "Brown," and "Zebra" landraces exhibited height reductions of 23.9 %, 28.1 %, and 25.6 %, respectively (Table 4). At 9 WAS, healthy plants ranged in height from 19.8 cm in “Cream” and “Red nav” landraces to 22.1 cm in “Zebra” landrace, with the "Black" and "Brown" landraces recording a uniform height of 21.9 cm. Infected plants' heights, on the other hand, ranged from 12.6 cm in the "Cream" landrace to 16.2 cm in the "Zebra" landrace. Furthermore, the "Black" landrace's infected plants were taller with16.3 cm than "Red nav," which was taller with 15.7 cm plant than the "Black" landrace with15.1 cm. The height reductions in virus-infected plants ranged from 20.7 % in "Red nav" to 36.4 % in the "Cream" landrace. The "Black," "Brown," and "Zebra" each lost an average of 25.6 %, 31.1 %, and 26.7 % of their heights.

3.3 Effect of Cucumber Mosaic Virus Disease on A Number of Days To 50 % Flowering

In 2019, the number of days to 50 % flowering did not differ between uninoculated and inoculated plants of any landrace (Table 5). Uninoculated and CMV-inoculated plants of the "Cream" landrace attained 50 % flowering first at 35 DAS, followed by healthy and infected plants of the "Red nav" landrace at 38 DAS. Plants of the "Brown" and "Zebra" landraces reached 50 % flowering at 40 DAS, whereas those of the "Black" landrace reached 50 % flowering at 43 DAS. The number of days to 50 % flowering of uninoculated plants in 2020 was the same as in 2019. Infected plants, on the other hand, reached 50 % flowering at 44 and 41 DAS for the “Black” and “Brown” landraces, respectively. CMV-infected plants of the “Cream,” “Red nav,” and “Zebra” landraces, on the other hand, attained 50 % flowering at 38, 40, and 42 DAS, respectively. Days to 50 % flowering increased by one day in the “Black” and “Brown” landraces, but by two days in the “Red nav” and “Zebra” landraces, and three days in the “Cream” landrace (Table 5).

Table 3. Plant heights from Bambara groundnut landraces inoculated and uninoculated with Cucumber mosaic virus disease in Minna during 2019 cropping season

Landrace colour ID	Plant height (cm)											
	3 WAI				6 WAI				9 WAI			
	Uninoculated	Inoculated	SEM	Reduction (%)	Uninoculated	Inoculated	SEM	Reduction (%)	Uninoculated	Inoculated	SEM	Reduction (%)
Black	18.7[a]	15.8[b]	0.1	15.5	20.0[a]	16.4[b]	0.1	18.0	20.9[a]	16.8[b]	0.2	19.6
Brown	19.6[a]	13.4[b]	0.5	31.6	20.9[a]	14.5[b]	0.5	30.6	21.8[a]	15.1[b]	0.6	30.7
Cream	17.6[a]	11.5[b]	0.2	34.7	18.7a	12.4[b]	0.1	33.7	19.3[a]	13.0[b]	0.1	32.6
Red nav	17.3[a]	14.7[b]	0.4	34.2	18.9[a]	16.0[b]	0.4	15.3	19.3[a]	16.8[b]	0.3	13.0
Zebra	20.1[a]	15.7[b]	0.1	21.9	21.6[a]	16.9[b]	1.1	21.8	22.6[a]	17.3[b]	0.1	23.5

Means with dissimilar letter (s) within the row differ significantly (p≤0.05) according to the Least Significant Difference WAI = Week (s) after inoculation

Table 4. Plant heights from Bambara groundnut landraces inoculated and uninoculated with Cucumber mosaic virus disease in Minna during 2020 cropping season

Landrace colour ID	Plant height (cm)											
	3 WAS				6 WAS				9 WAS			
	Healthy	Inoculated	SEM	Reduction (%)	Healthy	Inoculated	SEM	Reduction (%)	Healthy	Inoculated	SEM	Reduction (%)
Black	19.8[a]	15.1[b]	0.2	23.7	21.3[a]	16.2[b]	0.1	23.9	21.9[a]	16.3[b]	0.1	25.6
Brown	19.5[a]	14.9[b]	0.2	23.6	21.0[a]	15.1[b]	0.2	28.1	21.9[a]	15.1[b]	0.3	31.1
Cream	17.7[a]	12.0[b]	0.1	32.2	18.4[a]	12.5[b]	0.1	32.1	19.8[a]	12.6[b]	0.1	36.4
Red nav	17.7[a]	14.9[b]	0.1	15.8	18.2[a]	15.7[b]	0.2	13.7	19.8[a]	15.7[b]	0.1	20.7
Zebra	19.5[a]	15.8[b]	0.1	19.0	21.5[a]	16.0[b]	0.1	25.6	22.1[a]	16.2[b]	0.1	26.7

Means with dissimilar letter (s) within the row differ significantly (p≤0.05) according to the Least Significant Difference WAS = Week (s) after sowing

Table 5. Number of days to 50 % flowering from Bambara groundnut plants inoculated and uninoculated with Cucumber mosaic virus disease in Minna during 2019 and 2020 cropping seasons

Landrace colour ID	Days to 50 % flowering (no.), 2019				Days to 50 % flowering (no.), 2020			
	Uninoculated	Inoculated	SEM	Increased days	Healthy	Inoculated	SEM	Increased days
Black	43^{a}	43^{a}	0	0	43^{b}	44^{a}	0	1
Brown	40^{a}	40^{a}	0	0	40^{b}	41^{a}	0	1
Cream	35^{a}	35^{a}	0	0	35^{b}	38^{a}	0	3
Red nav	38^{a}	38^{a}	0	0	38^{b}	40^{a}	0	2
Zebra	40^{a}	40^{a}	0	0	40^{b}	42^{a}	0	2

Means with dissimilar letter (s) within the row differ significantly ($p \leq 0.05$) according to the Least Significant Difference

Table 6. Seed weight per plant from Bambara groundnut landraces inoculated and uninoculated with Cucumber mosaic virus disease in Minna during 2019 and 2020 cropping seasons

Landrace colour ID	Seed weight per plant (g), 2019				Seed weight per plant (g), 2020			
	Uninoculated	Inoculated	SEM	Reduction (%)	Healthy	Inoculated	SEM	Reduction (%)
Black	3.6^{bc}	2.2^{b}	0.0	38.9	3.8^{a}	1.7^{b}	0.0	55.3
Brown	3.4^{c}	1.9^{b}	0.0	44.1	3.2^{a}	1.6^{b}	0.0	50.0
Cream	3.4^{a}	2.0^{b}	0.1	41.2	3.4^{a}	1.3^{b}	0.0	61.8
Red nav	4.0^{a}	3.1^{b}	0.1	22.5	3.9^{a}	1.4^{b}	0.0	64.1
Zebra	3.1^{c}	2.4^{b}	0.0	22.6	3.0^{a}	1.7^{b}	0.0	43.3

Means with dissimilar letter (s) within the row differ significantly ($p \leq 0.05$) according to the Least Significant Difference

3.4 Effect of Cucumber Music Virus Disease on Seed Weight

The *Cucumber mosaic virus* disease lowered seed weight in infected cucumber plants to varying degrees (Table 6). In contrast to large and normal seeds produced by uninoculated plants, the majority of seeds produced by inoculated plants were small, shrivelled, and distorted. The healthy plants of each landrace had significantly ($p<0.05$) heavier seed weight than the CMV-infected ones during the 2019 cropping season. The "Red nav" landrace yielded the heviest seed weight of 4.0 g in the infected plants, followed by the "Black" landrace with 3.6 g. The “Brown” and “Cream” landraces, on the other hand, recorded uniform seed weight of 3.4 g per plant, while the “Zebra” landrace recorded the least of 3.1 g per plant (Table 6). The “Red nav” landrace recorded the highest seed weight of 3.1 g per plant in CMV-infected plants, followed by the “Zebra” landrace with a mean seed weight of 2.4 g per plant. Furthermore, the "Black," "Brown," and "Cream" landraces had seed weights of 2.2 g, 1.9 g, and 2.0 g, respectively. The "Red nav" landrace recorded the least seed weight reduction of 22.5 %, followed by the "Zebra" landrace with 22.6 %. Seed weight reductions in the “Black,” “Brown,” and “Cream” landraces, on the other hand, averaged 38.9 %, 44.1 %, and 41.2 %, respectively.

In 2020, there was a significant ($p<0.05$) difference in seed weight between healthy and infected plants in all the landraces (Table 6). The "Black" landrace recorded the heaviest seed weight of 1.9 g per plant in CMV infected plants, followed by the "Zebra" landrace with 1.7 g per plant, the "Brown" and "Red nav" landraces recorded identical seed weights of 1.6 g, and the "Cream" landrace recordeds the least seed weight of 1.4 g. The "Red nav" landrace recorded the heaviest seed weight of 3.9 g while the "Zebra" landrace recorded the least seed weight of 3.0 g in healthy plants. The "Black" landrace, on the other hand, produced an average seed weight of 3.8 g per plant, while the "Brown" and "Cream" landraces recorded 3.2 and 3.4 g, respectively (Table 6). The “Black” and “Zebra” landraces recorded the heaviest seed weight of 3.8 g per plant, followed by the “Brown” landrace with 1.6 g per plant, and the “Red nav” landrace with 1.4 g per plant, with the “Cream” landrace recording the least seed weight of 1.3 g. The “Red nav” landrace recorded the most severe seed weight loss by 64.1 %. The "Cream" landrace was next with 61.8 %, followed by the "Black" the "Brown" the "Zebra" landraces with 58.3 %, 50 %, and 43.3 % respectively.

4. Discussion

One of the key viruses impacting legume productivity has been identified as the cucumber mosaic virus [20]. Cultivation of virus-resistant or tolerant types has long been an efficient and long-term control technique for mitigating the challenges posed by virus infections [13]. The present study discovered a lot of genetic diversity among the test Bambara groundnut landraces. However, as various gene activities are required for each trait, none of the landraces showed consistent performance for morphological and yield attributes. Quantitative features are governed by two or more genes that may work synergistically or otherwise [21]. Some of the landraces had 100 % infection, indicating their susceptibility to *Cucumber mosaic virus* disease. The fact that the “Black” and “Zebra” landraces maintained a steady and less than 50 % disease incidence at 1 and 2 WAI in both the 2019 and 2020 cropping seasons demonstrate their tolerance to CMV.

The high disease incidence in all the landraces at 3 WAI agrees with the report by [22] where some soyabean lines inoculated with CMV at the primary leaf stage showed up to 100% infections. This shows the sensitivity of plants when attacked by virulent viral strains during their early stages of development. Seedlings are more vulnerable at this stage of development due to lack of immunity, especially in susceptible landraces and cultivars. Furthermore, the symptoms observed on CMV-inoculated plants in the present study were comparable to those reported by [17] who registered chlorosis and necrotic patches along the margins of older leaves of CMV-infected cucumber plants. Due to the genetic makeup of the infected landraces, disease severity differed significantly among them. When several cowpea accessions were infected with CMV, [23] found that the severity of infection increased with time in the various landraces. The infected landraces' low to moderate severity symptom at 3 WAI may be attributed to an initial battle between viral particles and innate plant defence mechanisms, which corroborate the report of [24]. At 9 WAI, disease severity rose in certain infected plants, indicating that some viruses can get beyond the plant's defenses [24]. Plants use mechanisms such as antiviral RNA silencing, but viruses fight back with silencing repressors in the plant-virus pathotype which is what might have happened at this growth stage in the study.

Variation in the genetic make-up and responsiveness of the landraces to CMV infection may explain the differences in growth and yield characteristics across the affected plants. That the healthy control plants were substantially taller than the inoculated ones in this study agrees with the report of [23] on a CMV infection experiment with cowpea plants. In cases where healthy plants developed more leaves, it appears that leaf production may be hampered under virus disease conditions, which can reduce photosynthesis and ultimately grain yield [23]. The pattern of reduction in infected plants suggests that none of the landraces was completely superior to the other.

The susceptible and low resistant genes of the "Brown" and "Cream" landraces may explain their high incidence of infection and resultant reductions in the number of pods, pod weight, and seed weight. Conversely, landraces expressing low-level infection, such as "Red nav" and "Zebra," recorded the least t reduction in the number of pods, pod and seed weight per plant, indicating that they may contain CMV-resistant genes. Plants undoubtedly have genes known as susceptibility genes whose products are vital in their regular physiology, but these genes may also aid pathogen invasion and colonization in some ways [25]. Increased disease resistance can be achieved by changing such genes through conventional and molecular breeding which can be exploited by plant breeders using some of the identified resistant landraces in the present study [26].

Plants produced from CMV-infected seeds displayed symptoms of CMV disease, suggesting that the virus is seed-borne and seed-transmitted. This agrees with the report of [27] who showed CMV survival in soyabean. Seed transmission is a typical occurrence in legume crops, and it has been linked to a long-distance and cross-border viral transmission. This emphasizes the need of planting virus-free Bambara groundnut seed, particularly in the management of CMV infections. The fact that both healthy and CMV-infected plants reached 50 % flowering on the same day shows that the virus had no effect on this parameter. Also, the fact that days to 50 % flowering in infected plants were identical to healthy plants even during the 2020 trial implies that the virus has a slight impact on this characteristic. This result contradicts the findings of [28] who showed that CMV delayed flowering in *Arabidopsis thaliana* [28]. Seed weight is a critical agronomic feature that is usually prioritized during breeding. As a result of infection, seed weight reduction was severely induced in the "Cream" landrace, resulting in small, malformed seeds. This is in line with the report by [29] who recorded a significant reduction in seed weight in cowpea plants infected with the CMV.

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