



# **Evaluation of Cowpea F2 Population for Resistance to Flower Bud Thrips (*Megalurothrips sjostedti*)**

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## **Authors' contributions**

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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## **ABSTRACT**

A study to screen F2 populations obtained from IT10K-837-1 x Sanzi for resistance to *M. sjostedti*. A total of 351 F2 plants were screened under field conditions and each line rated visually for thrips damage score, flower abortion rate, number of pods per plant and number of thrips per flower. The study was conducted during the 2020 cropping seasons where F2 seeds were planted under field conditions. Thrips damage score, flower abortion rate, number of pods per plant, and number of thrips per bloom was all visually scored for each accession. The data from the field and laboratory were analyzed using GENSTAT edition 12 to determine if there were any major variations among the cowpea genotypes. The approximate correlations among all measured parameters, including damage scores at 45 DAP, 55 DAP, 65 DAP, using were established using Breeding View software

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and heritability values were also calculated. There was a strong and significant correlation between thrips population and damage rating during the planting season. Thrips resistance levels identified in most genotypes in this study are interesting compared to the resistant check Sanzisabinli and can be further exploited for future studies to develop resistant varieties to improve cowpea yields.

**Keywords:** Cowpea; accessions; effective peduncles; correlation analysis; flower thrips.

## 1. INTRODUCTION

“Cowpea (*Vigna unguiculata* (L.) Walp.) is a significant grain-legume particularly found within the dry savannah agro-ecologies of sub-Saharan Africa (SSA)” [1]. “In Africa, cowpea is a component of the normal cropping systems, and it is considered an important crop because of its different uses, especially for human food. Cowpea is used as a human food and cattle feed, as well as hay, silage, pasture, soil cover, and green manure” [2]. “It is an important source of nitrogen for soil reclamation, and income for resource-poor farmers, even as small-scale processors” [3]. “Cowpea is a valued component of farming systems in many areas because of its ability to restore soil fertility for successive cereal crops grown in rotation with it” [4]. “Cowpea grains represent a major source of protein (20% – 32%), minerals, and vitamins within the diet of the bulk of rural and semi-urban communities” [5]. “The crops supplements staple low-protein cereal is a valued and responsible product that produces income for farmers and traders” [6]. “The delicate leaves, soft stems, and green pods are likewise eaten as vegetables in Asian and East African communities” [7]. “About 95 % of the world cowpea production comes from the West African sub-region” [8]. First maturing cowpea varieties can provide the first food from the current harvest sooner than any other crop, thereby shortening the hunger period that often occurs just before harvest of the current season’s crop in farming communities in the developing world [9].

“Cowpea is both receptive to satisfactory growing conditions and adapts to growing under drought, heat and other abiotic strains. As much as 1000 kg/ha of dry grain has been produced in a Sahelian environment with only 181 mm of rainfall and high evaporative conditions” Hall and Patel [10] as cited in Alfonso and Brent [11]. Currently, existing cultivars of other crop species cannot produce an important amount of grain under these conditions. The crop is more tolerable to low soil fertility, due to its high rates of nitrogen fixation [12] active symbiosis with *Mycorrhizae* [13], and capable to restore

tolerable soils over a wide range of pH when related to other popular grain legumes [14]. Aphids, flower bud thrips, pod borer, pod bugs, and *bruchids* are the principal insect pests of cowpea in Africa [15]. Thrips (*Megalurothrips sjostedti*) are responsible for significant financial loss concerning cowpea production in the world. They infect buds, racemes, and flowers, causing the reproductive organs to abort prematurely [16]. A variety of insect pests and diseases, inadequate agronomic techniques, and the use of low yielding cultivars are all implicated for the low output [17]. Diverse insect pests damage cowpea in the field, with *M. sjostedti* being the most devastating in Africa [18]. Their destruction could also lead to changes in the nutritive value of cowpeas [19].

### 1.1 Problem Statement and Justification

“Cowpea is an attractive host to numerous insect pests that diminish its grain yield and quality. Among them, the flower bud thrips causes the most serious damage during the crop’s flowering stage” [19]. The insect lays eggs on cowpea flower buds and therefore the nymphs/adult’s prey on the reproductive structures of the plant [20], bringing about drying out and browning of the stipules, flower bud abscission, flower discoloration, distortion, or abortion [21]. Due to premature flower abortion, the peduncles of susceptible plants are stunted as no pods develop in them. Grain yield reduction occurs as a result of flower bud thrips infestation which ranges from 20% to 80% [22] and could reach 100% under high pervasion [20,23].

Insecticide application is the prevalent method for controlling this pest on cowpea. A few selective control measures including cultural practices [24], biological control.

Mfuti et al. [25]; (Tamo’ et al. 2003) and the utilization of bio-pesticides, for instance, neem extract [26], have been examined to regulate this insect. “Host plant resistance appears to be the most economical and environmentally well-disposed approach to scale back thrips damage to cowpea. Unfortunately, most of the cowpea varieties grown in West Africa are exceptionally

susceptible to *M. sjostedti*. Only a few cowpea genotypes are accounted for to point out low degrees of resistance to the pest" [27]. Hence, a field evaluation of the currently available germplasm accessions could lead to the identification of more lines with more significant levels of protection from this pest. Genotypes recognized as thrips resistant are going to be used for the advancement of improved cowpea varieties, which might limit the necessity for insecticide application by farmers while likewise guaranteeing increased grain yield.

The objective of the present research was aimed to evaluate cowpea F2 populations for resistance to flower bud thrips through classifying F2 genotypes according to the phenotypic classes observed, testing the goodness of fit of selected traits of the F2 population and finally determining the impact of thrips on flowers and raceme damage.

## 2. MATERIALS AND METHODS

### 2.1 Description of the Experimental Site

The fieldwork was carried out at the CSIR-Savanna Agricultural Research Institute (SARI). The area has an average minimum temperature of 25°C and a maximum temperature of 35 °C [28] with an annual average rainfall of 900 and 1000 mm. The site is characterized by natural vegetation dominated by grasses with few shrubs. The soils of the area are brown, moderately drained sandy loamy and free from concretions. They are very shallow with a hard pan under the top few centimeters and were derived from Voltarian sandstone classified as Nyankpala series (Plinthic Acrisol) according to [29]. The location has been identified as a hotspot for flower bud thrips from previous field experiments by the Cowpea Improvement Program of the CSIR-SARI.

### 2.2 Development of F2 Population

The trials were carried out during the 2020 main cropping seasons (July–September) when cowpea is mostly grown by farmers in the area. Two parents namely IT10K-837-1 and Sanzi were used in developing the F2 populations. Sanzi is a landrace material in Ghana which has been reported to have resistance to aphids but does not have the marketable characteristics of cowpea. IT10K-837-1 has the farmer preferred characteristics, but it is susceptible to flower bud thrips. A cross was therefore made between IT10K-837-1 x Sanzi in 2019 to obtain F1's. F1's was selfed the same year to obtain F2 seeds.

Single seeds of bulked F2 seeds coming from 10 F1 plants were planted.

### 2.3 Planting and Design of the Experiment

The 351 F2 seeds were space planted at 40 cm between plants in a single row plot each measuring 12 m each. Since this is a segregating population, one seed per hill was planted. Insecticides were not applied during the pre-flowering stages as this is the stage that the cowpeas are most susceptible to flower bud thrips. However, at the podding stage, one insecticide spraying was done to control *Maruca*. One manual weeding was carried out using hoes in the growing season. A total of 702 flower samples and racemes were collected and preserved in 70 % ethanol and screened under field conditions. The cowpea samples were collected to determine their resistance to thrips.

### 2.4 Data Collection

Data were collected on the number of thrips in flower buds and raceme at 49 and 54 days after planting (DAP) respectively. Two flowers and a raceme were randomly collected during these stages. The flowers were collected between 8.0 and 10.0 a.m. and placed separately in label labelled vials containing 70% ethanol and subsequently dissected in the laboratory to count the number of thrips.

Data on days to first pod maturity, days to 95 % pod maturity, pod and grain yield per plant was also taken. The thrips damage scores were recorded on each test line using a visual scoring was based on a 1–9 scale [21], where 1 = no browning or drying of stipules and flower buds with no flower bud abscission; 3 = initiation of browning of stipules and/or flower buds but no flower bud abscission; 5 = distinct browning/drying of stipules and/or flower buds with some flower bud abscission; 7 = serious flower bud abscission accompanied by browning/drying of stipules and buds; and 9 = very severe flower bud abscission, heavy browning with drying of stipules and stunted peduncles.

Number of pods and peduncles per plant were counted at plant maturity. The number of pods per plant, number of peduncles with or without pods, the flowering time, stem /pod colour were measured. The number of pods and number of peduncles were counted on all the three hundred and fifty-two plants. Based on the

number of peduncles and pods, the percentage of effective peduncles (%Eff pdcl) and the flower bud abortion rates (AR) were also calculated as:

$$\%Eff\ pdcl = \frac{No.\ of\ peduncles\ with\ pods}{Total\ peduncle\ no.} \times 100$$

$$AR = \frac{No.\ of\ peduncles\ without\ pods}{Total\ peduncle\ no.} \times 100$$

## 2.5 Definition of the Resistance Status

Based on the damage score ratings [21], accessions with scores less than five (i.e., 1–4) were considered resistant while those with scores of five and seven to nine indicated a moderate level of resistance and susceptible, respectively. The observations in the F2 genotypes were genetically grouped into homozygous resistant, segregating and homozygous susceptible. Those displaying patterns equal to or higher than the susceptible parent IT10K-873-1 were considered susceptible.

## 2.6 Data Analysis

The data from the field and laboratory were analyzed using GENSTAT edition 12 to determine if there were any major variations among the cowpea genotypes. To separate the means, the LSD test was used at a 5 % significance stage. A simple correlation analysis was used to examine relationships between the damage score at 65 DAP (DS3) and a variety of variables of interest, including the number of pods per plant (Podplt), the number of peduncles with pods (Pedclwpd), the flower bud abortion rate (AR), and the percent effective peduncles (% Effpdcl). To estimate the number of genes conditioning thrips resistance among these populations, a Chi-square test was done. The Chi-square analysis was used to test the goodness of fit of observed segregations to the expected genetic ratios of 1 homozygous dominant, 2 heterozygous and 1 homozygous recessive.

$$\chi^2 = \sum \frac{(O_i - E_i)^2}{E_i}$$

The approximate correlations among all measured parameters, including damage scores (at 45 DAP, 55 DAP, 65 DAP), number of pods per plant, number of peduncles per plant, number of peduncles with pods, flower bud abortion rate, percent effective peduncles, and thrips population/flower, were established using Breeding View software. These parameters' heritability values were also calculated.

## 3. RESULTS

### 3.1 Thrips Damage Score and Population

The summary statistics of Thrips in flowers and raceme, damage due to thrips, and Maruca have been presented in Table 1. Thrip population ranged from 0-7 for flowers with a mean of 2.27 and for raceme, it ranges from 0-9 with a mean of 2.9. The results also showed a maximum and minimum damage rate of 9 and 1 respectively. The number of maruca also ranged from 0-4 with a mean of 0.31.

### 3.2 Pods per Plant and Percent Effective Peduncles

F2 population evaluated had a minimum and the maximum number of pods per plant of 0 and 42 respectively, with a mean of 12.41. Damage per plant showed a minimum of 0 and a maximum of 80 with an average of 66.65. The results also showed a maximum and minimum pod yield of 0 and 82.5g/plant respectively, with an average of 26.52g/plant. The minimum and maximum grain yields were 0 and 56 respectively, with an average of 15.13 g/plant (Table 2).

Table 3 shows peduncles with or without pods, number of peduncles, flower abortion rate and percentage of effective pods of cowpea accessions. The average peduncle with and without pods were 7.146 and 2.702 respectively and a maximum 24 and 14 respectively. The number of peduncles registered was 30 with a mean number of 9.84. A maximum rate of flower abortion of 83.33 was recorded and an average of 27.39. Comparatively, at 25 % and 75% of effective pods was 81.82 and 63.64 respectively with a mean of 75.

**Table 1. Damage due to Thrips and Maruca**

	Flower thrips	Raceme thrips	Damage rating	Maruca
Minimum	0	0	1	0
1st Quartile	1	2	1	0
Median	2	3.5	7	0
Mean	2.27	2.85	5.31	0.31

3rd Quartile	3	4	7	0
Maximum	7	9	9	4

**Table 2. Thrips damage to pods**

	Number of pods/ plant	Damage per plant	Pod Yield/plant (g)	Grain Yield/plant (g)
Minimum	0	0	0	0
1st Quartile	6	65	16	7.63
Median	11	68	24.5	13
Mean	12.41	66.65	26.52	15.13
3rd Quartile	17	75	33.38	19.88
Maximum	42	80	82.5	56

**Table 3. Number of peduncles with pods and flower abortion rate**

	Peduncle with pods	Peduncles without pods	Number of peduncles	Flower abortion rate	Percent of effective pods
Minimum	0	0	0	0	16.67
1st Quartile	4	1	6	18.18	63.64
Median	6	2	9	25	75
Mean	7.15	2.70	9.84	27.39	72.61
3rd Quartile	10	4	13	36.36	81.82
Maximum	24	14	30	83.33	100

The number of peduncles with pods ranged from 0-24 with a mean of 7.15 whilst the number of peduncles without pods ranged from 0-14 with a mean of 2.7. The mean number of peduncles was 9.8 with the highest being 30. The mean flower abortion rate was 27.4% and mean percentage effective pods for the population was 72.61% (Table 4).

The results from Table 4 shows, the seed coat colour of the F2 cowpea accessions. The heterozygote black, mottled brown and white homogenous dominant recorded 5.754, 5.033 and 1.320 respectively (Table 5).

The least and greatest number of days for the cowpea to reach maturity after planting were 76-80 DAP (0.108) and 52-65 DAP (1.458) (Table 6). The population perfectly fits the expected 1:2:1 for the maturity classes.

Genotypes IT10K-837-1 and Sanzi showed a positive correlation relationship between damage levels. The damage rating of thrips in flowers ( $r = 0.34$ ) and thrips in racemes ( $r = 0.31$ ). There was a correlation between thrips in flowers and thrips in racemes but no correlation between thrips in raceme and damage rating (Table 7). On the other hand, the correlation between thrips in flowers and thrips in raceme was ( $r = 0.75$ ).

**Table 4. Percent effective pods and flower abortion of cowpea lines**

<b>Effective pods</b>				
Percent effective pods	16-59 % effective pods	60-84 % effective pods	85-100 % effective pods	Total
Chi-square	5.313	8.247	3.084	16.645
<b>Flower abortion</b>				
Percent flower abortion	41-83 % flower abortion	16-40 % flower abortion	0-15 % flower abortion	Total
Chi-square	5.313	7.807	2.711	15.831

**Table 5. Seed coat colour of F2 individuals**

	Homozygous dominant	Heterozygous	Homozygous recessive	Total
Seed coat colour	White	Black	Mottle brown	
Chi-square	1.320	5.754	5.033	12.11

**Table 6. Maturity period of cowpea lines**

Maturity period	76-80 DAP	66-75 DAP	52-65 DAP	Total
Chi-square	0.108	0.386	1.458	1.952

**Table 7. Correlation among study traits**

	Thrips in flowers	Thrips in Raceme
Thrips in Raceme	0.75***	
Damage rating	0.34***	0.31***

#### 4. DISCUSSION

As stated by Visschers et al. [30], the observed strong correlation in thrips damage scores from 49 to 54 DAP in this study can be attributed to a steady increase in the insect population density in the field from flower bud initiation to when the final scores were collected. This indicates the presence of a genetic resistance component that is helpful against a variety of thrips species.

Flower bud thrips (*M. sjostedti*) infests cowpea plants from the pre-flowering stage onwards [31], and due to the insect's short developmental period of about 18 days (IRAC), it can produce up to four generations between flower bud initiation and flowering. The higher insect population at 49 to 54 DAPS led to increased insect pressure, resulting in more plant damage. "The signs and symptoms of a thrips infestation on cowpea plants are well-known. Browning of stipules and flower buds, numerous stunted peduncles with no pods and flower bud abscission are among the symptoms" [21,32]. Plants that are severely infested appear diseased and contain a small number of flowers that enter anthesis.

The test lines had a strong pod load (24) and a high percentage of successful peduncles (72.61 %) that were comparable to the resistant check, "Sanzi." As a result, these lines should be considered *M. sjostedti* immune. "Pod load and percent active peduncles are important components of cowpea yield, and they can be used to determine cowpea genotypes' resistance or susceptibility to flower bud thrips" [33,34]. These are characteristics that breeders can depend on when choosing resistant lines in the field. There were major differences in the number of peduncles with pods and flower abortion rates among the cowpea genotypes. The resistant line had more peduncles with pods and a lower rate of flower abortion, while the vulnerable line had the opposite tendency.

Many yield-related traits, such as the number of pods per plant and the percentage of effective peduncles, were significantly and positively correlated with the damage scores reported at 49 to 54 DAP, as predicted. This means that thrips damage scores obtained at the late flowering stage (65 DAP) are more accurate than scores obtained at 49 and 54 DAP in determining the resistance status of cowpea genotypes to *M. sjostedti*. At 49 to 54 DAP, there was a positive association between flower abortion rate and harm scores. In addition to damage scores at the late flowering stage, pod weight, and percentage of successful peduncles, the number of peduncles with pods and flower abortion rates may be used to determine cowpea resistance to *M. sjostedti*. In contrast to the non-effective form, peduncles with pods are usually elongated, as observed in our research. We found no major differences in thrips populations in flowers among the test entries, but we did note that the damage reported on the susceptible lines was greater than the damage recorded on the resistant lines. There is a connection between damage rating on thrips in flowers and racemes, as shown in Table 7. This finding supports that of Salifu et al. [36], who discovered heavy thrips infestation on susceptible lines at the flower bud level, resulting in flower abortion. Based on their moderate damage scores (which ranged from 0.59 to 1.78) and average pod number per plant (12.41). Since these genotypes were able to generate some pods despite having reasonably high levels of thrips harm in the field, the mechanism of resistance operating in them may be tolerance. The resistant and moderately resistant genotypes in this study had high pod load and effective peduncles. These are yield-related traits in thrips resistant/tolerant cultivars because of low flower abortion. Cowpea cultivars that can resolve or compensate for insect pest damage can yield more pods, according to Togola et al. [36]. "Early and late flowering plants

have phenological traits that can help cultivars avoid pest damage”, according to Asante et al. [37]. To confirm the exact mechanism present in each of the established resistant cowpea lines, further research is required.

Some of the assessed traits such as damage scores at 52 to 65 DAP, number of pods per plant, number of peduncles with pods, and percentage of active peduncles, had high heritability values (>50%), meaning that these traits are heritable and environmental influences on them are minimal. By using these characteristics as selection criteria, breeders will make progress in developing improved cowpea varieties that are resistant to flower thrips. This is especially true when low damage ratings, low flower abortion rates, and a high number of successful peduncles are used in the selection process. These results confirm the resistance of Sanzi to thrips as the resultant population generally had low thrips damage.

## 5. CONCLUSION

The need to identify sources of *M. sjostedti* resistance in cowpea has remained a top priority in SSA's cowpea breeding programs.

With the discovery of many promising lines with high levels of resistance to flower bud thrips, improved cowpea with resistance to this pest appears to be within reach shortly. Results of the assessment studies will enable breeders develop genotypes that are resistant to flower bud thrips

## 6. RECOMMENDATION

With the increasing demand for cowpea and the growing population of consumers, chemical tolerance is a distinct possibility. Cowpea, *Vigna unguiculata*, is a high-protein legume that is widely used for food and feed around the world. It is critical to assist in ensuring that customers have access to poison-free products. As a result, producers should be urged to grow resistant crop varieties. The cost of producing resistant cultivars is low, and chemical remnants on cowpea are reduced. Experiments involving insect pests, particularly flower bud thrips, considering the time and location of the investigation. To evaluate the influence of the different climatic zone on the research, future works and research should be done in multiple areas at a given time. Also, rearing of thrips and artificially inoculating them under screenhouse

conditions should also be done to evaluate cowpea genotypes resistant to flower Thrips

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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