Gene Action Controlling Seed Size in Cowpea (Vigna Unguiculata L. Walp)

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ABSTRACT

Cowpea with large seed sizes is one of Ghana's preferred traits for producers, consumers and other value chain actors. Limited information on gene action controlling the inheritance of large seed size confounds the choice of appropriate breeding methods. Generation mean analysis was conducted to identify the gene action controlling the inheritance of cowpea seed size. Data on the seed length, seed width, and seed thickness of five generations (P_1, P_2, F_1, F_2, and F_3) of a cross between Kansa (large seed-sized variety) and Hewale (small seed-sized variety) were analyzed. The scaling test analysis showed additive-additive epistasis, and the additive-dominance model was inadequate in explaining the gene action controlling cowpea seed size. Significant additive, dominance, additive-additive, and dominance-dominance gene actions were found to control seed size. Also, allelic and non-allelic gene action were found to control seed size, hence recurrent selection could be used to improve the trait. From this study we conclude that we cannot improve on Cowpea seed size by making between large-seeded lines and small-seeded lines and recommend that breeders should make crosses among large-seeded lines if larger seeds are the desired trait as is the case in Ghana.

Keywords: Gene action, Seed size, Cowpea, Generation mean

1.0 Introduction

Cowpea is a nutritious grain legume that is eaten in many parts of the world. It is widely cultivated in the tropical and subtropical areas of Africa, Asia, and the United State of America. Particularly in Africa, the crop is cultivated by rural farmers who have limited resources and low-quality seeds (Martey *et al.*, 2022). Cowpea provides for the protein needs of people who may not afford animal protein and those who prefer plant protein. There are reasons to direct research focus on the improvement of cowpea now that there is a call to improve and sustain access to nutritious food in derived countries. In Ghana a larger number of cowpea consumers purchase their cowpea from the open market. A consumer's choice for a particular variety is influenced by several factors including seed quality traits such as seed coat colour, seed size, and duration of cooking (Quaye *et al.*, 2011). Most consumers prefer varieties with a white seed coat, short cooking time, and large seed size (Egbadzor, 2013). The large seed size of cowpea, aside been a quality choice for consumers, influences many biological and physiological processes such as germination, and vigorous seedling growth (Kandasamy *et al.,* 2020). The large seed-size cowpea available on the Ghanaian local market attracts a 30% extra premium (Ira *et al.,* 2019). However, these high-priced varieties are imported from Niger and Burkina Faso and they cost the country a huge amount of foreign exchanges to import these varieties (USAID, 2016). Unfortunately, the introduced varieties are poorly adapted to the local cropping systems and environment in Ghana. Additionally, large seed size is correlated with high yield since large seeds weigh heavier than small seeds. The improved germination, enhanced seedling vigor and high market premium which are benefits derived from producing cowpea seeds with large seed sizes make it essential to develop local cowpea varieties with large seed sizes. The choice of a breeding method depends on the amount of variability that exists in the breeding population, the number of genes controlling the trait, and the gene effect. Variations exist in seed size for cowpea and this variability can be harnessed in a breeding program that targets seed size. Several authors have reported that several genes control seed size (Lo *et al.,* 2019; Egbadzor, 2013) but information on the gene effect is limited and this makes the choice of a suitable breeding method for improving the seed size complicated. There is a need to determine the gene effect of the controlling genes to design an appropriate approach for developing cowpea varieties with large seed sizes. The present study seeks to determine the gene action controlling the seed size trait in cowpea.

2.0 Materials and Methods

Planting materials and Experimental design

The genetic materials comprise five generations of a cross between Kansa (large seed) and Hewale (small seed). The five generations are P_1, P_2, F_1, F_2, and F_3. The plants were raised in augmented design at the West Africa Centre for Crop Improvement (WACCI) research field at the University of Ghana.

Data Collection and Analysis

The seed length, seed thickness, and seed width of the five generations; P_1, P_2, F_1, F_2, and F_3 were recorded using a vernier caliper. A boxplot was constructed to visualize the distribution of traits across the five generations using R Software. The five-parameter model proposed by Hayman (1958) was used to perform the generation mean analysis. The scaling test C and D suggested by Mather (1949) was estimated to test the presence of epistasis. The scales C and D, and the five parameters which are mean effect (m), additive effect (d), dominance effect (h), additive-additive interaction (i) and dominance-dominance interaction (l) were estimated using the formula(e) below:

Scale test;

$$C = 4\overline{F_2} - \overline{2F_1} - \overline{P_1} - \overline{P_2}$$

$$D = 4(\overline{F_3} - 2\overline{F_2} - \overline{P_1} - \overline{P_2})$$

$$V_C = 16V\overline{F_2} + 4V\overline{F_1} + V\overline{P_1} + V\overline{P_2}$$

$$V_D = 16V\overline{F_3} + 4V\overline{F_2} + V\overline{P_1} + V\overline{P_2}$$

Genetic parameters;

m = mean of F_2

$$d = \frac{1}{2}\overline{P_1} - \frac{1}{2}\overline{P_2}\text{-}$$

$$h = \frac{1}{6}(4\overline{F_1} - 12\overline{F_2} - 16\overline{F_3})$$

$$I = \overline{P_1} - \overline{F_2} + \frac{1}{2}\overline{(P_1} - \overline{P_2}+\text{h})\text{-}\ \frac{1}{4}l$$

$$L = \frac{1}{3}(16\overline{F_3} - 24\overline{F_2} + 8\overline{F_1})$$

The variances of the gene effects were estimated as follows:

$$Vm = VF_2$$

$$Vd = \frac{1}{4}(V\overline{P_1} - V\overline{P_2})$$

$$Vh = \frac{1}{36}(16V\overline{F_1} + 144\overline{F_2} + 256\overline{F_3})$$

$$Vi = V\overline{P_1} + V\overline{P_2} + \frac{1}{4}\overline{(VP_1} + V\overline{P_2}+\text{Vh})\ \text{-}\ \frac{1}{16}Vl$$

$$Vl = \frac{1}{9}(256V\overline{F_3} + 576V\overline{F_2} + 64V\overline{F_1})$$

The standard errors for the gene effects were obtained by taking the square root of their corresponding variances. The ratio of the genetic effects to their respective standard errors was used to calculate a "t" to test for the significance of the genetic effects and their variances.

3.0 Results

3.1. Scaling test for seed length, seed width and seed thickness

The results of the scaling tests indicated that the scaling test C was not significant for all traits. However, scaling test D deviated significantly from zero for all traits (Table 1). The significant values of D suggest that the additive-dominance model was not adequate due to the presence of additive-additive epistasis.

Table 1: Scaling test for the adequacy of additive-dominance model for seed size in cowpea

	Scaling test		Variance		Calculated t value	
Traits	C	D	C	D	C	D
Seed length	**-0.66ns**	**-102.97****	5.9137±2.43	8.5497±2.92	-0.27	-35.21
Seed Width	**1.16ns**	**-73.66****	8.328±2.89	5.184±2.28	0.40	-32.35
Seed thickness	**0.76ns**	**-60.26****	3.4204±1.84	10.06±3.17	0.41	-18.99

ns= non-significant, **significant at 0.01.

3.2. Generation mean analysis for seed length, seed width, and seed thickness in five cowpea generation (P_1, P_2, F_1, F_2 and F_3)

The generation means, means of genetic parameters and the type of epistasis are presented in Table 2. The values of genetic components, m (mean effect), d(additive), and h(dominance) were significant for seed length, seed width and seed thickness. The i (additive*additive) interaction effect was significant for seed length, but it was not significant for seed width and seed thickness. The dominance-dominance (l) interaction was not significant for all three traits. The h and l gene effect were both negative for seed length indicating a complementary type of epistasis. The h and l gene effects had opposite signs for seed width and seed thickness. This indicates a duplicate type of epistasis for the two traits.

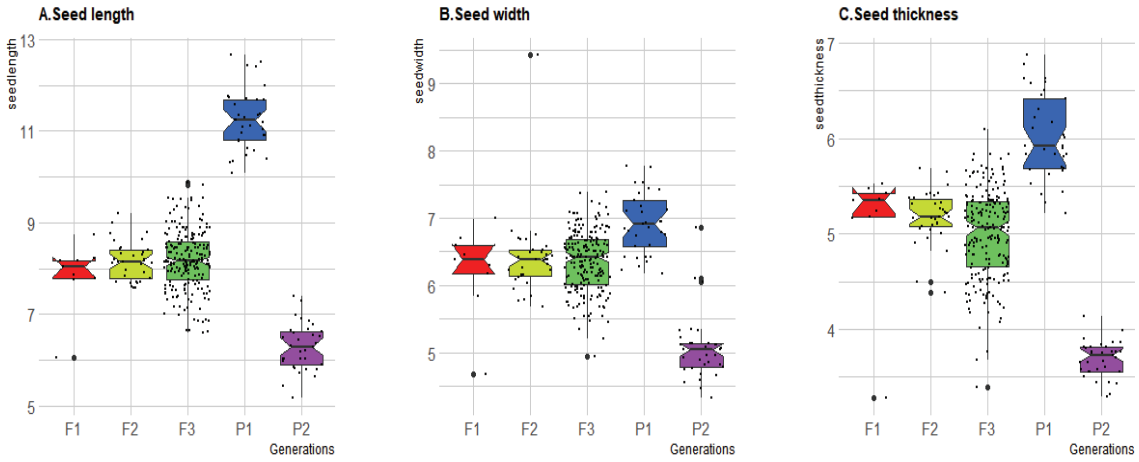
3.3. Distribution seed length, width, and thickness in five cowpea generation (P1, P2, F1, F2 and F3).

The five generations exhibited different levels of seed length (Figure 1A). The lower extreme, first quartile, median, third quartile, and upper extreme values of F_1 (7.75, 7.77, 8.03, 8.17, and 8.75), F_2 (7.57, 7.79, 8.15, 8.40, and 9.21), and F_3 (6.59,7.75, 8.16, 8.59, and 9. 82) was higher than their corresponding values in the P_2 (10.07, 10.81, 11.23, 11.68, and 12.65) and lower than their corresponding values in the P_1 (5.17, 5.91, 6.29, 6.62, and 7.39). The upper extreme value of the F_3 (9.82) was lower than the lower extreme of the P_1 but higher than the upper extreme of the P_2 (7.39). The outlier observed for seed length for the F_3 generation (9.878) was lower than the minimum value (10.07) for the higher parent (P_1) and greater than the maximum value (7.39) for the lower parent (P_2). The F_1 generation had an outlier (6.05) which was lower than the minimum seed length value (7.75).

For seed width, the lower extreme, first quartile, median, third quartile, and upper extreme values of the F_1 (5.83, 6.18, 6.39, 6.59, and 6.99), F_2 (5.68, 6.13, 6.38, 6.52, 6.81, and 6.81), and F_3 (5.21, 6.02, 6.42, 6.69, and 7.39) were lower than the corresponding values for P_1 (6.17, 6.57, 6.91, 7.26, and 7.78) and higher than their corresponding values for P_2 (4.32, 4.78, 5.04, 5.13, and 5.35) (Figure 1B). The value of the upper quartile for the

F_3 (7.39) was greater than the third quartile value (7.26) of the P_1. The value of the outlier for seed width in the F_2 generation (9.42) was greater than the maximum value (7.78) for the higher parent (P_1). The value of the outlier in the F_3 generation (4.98) was lower than the minimum value (6.17) in the higher parent and greater than the first quarter value of the lower parent (4.78). The outlier value observed in the F_1 (4.07) was lower than the minimum value (4.32) of the lower parent. The value of outliers (6.86, 6.04, and 6.1) observed for the F_2 population was below the median value of the higher parent (6.91).

The median value of the seed thickness in F_2 (5.18) was below the lower quartile (5.22) of the P_1 (Figure1C). The third quartile value for the seed thickness of the F_2 (5.69) was equal to the first quartile value of the P_1 and greater than the third quartile value of the P_2 (4.14). The median value of the seed thickness for the F_3 (5.063) was found to be lower than the minimum value of the P_1 (5.22) and higher than the third quartile value of the P_2 (4.14). The third quartile value of the F_3 (6.098) was greater than the median value of the P_1 (5.92) and lower than the third quartile value of the P_1 (6.42). The outlier observed within the F_1 population (3.28) was below the minimum value of the lower parent (3.30). Transgressive segregants for seed thickness were identified within the F_2 and F_3 generations. The outliers for the F_2 generation (4.38 and 4.50) were below the minimum value (4.82) of the F_2 population. The outliers for the F_2 population (4.38, 4.50) had higher seed thickness values compared to the maximum seed thickness value of the lower parent (P_2) but these outliers were below the minimum value (5.22) of the higher parent (P_1). The value for the outlier in the F_3 population was below the first quartile value of the lower parent (3.56)


A.Seed length

B.Seed width

C.Seed thickness

Figure1: The seed length, seed thickness, and seed width distribution in five generations of cowpea. P_1=Paternal parent (large seed); P_2,= maternal parent(small seed); F_1=first filial generation; F_2=second filial generation; F_3=third filial generation; black dots are outliers; points are data points

4.0 Discussions

The types and magnitude of gene action influencing the inheritance of a trait are important in determining the breeding methodology for crop improvement. The scales C and D were used to test the adequacy of additive-dominance interaction of genes. The results from this study indicated that scale C was insignificant for seed length, width, and thickness. However, scale D was significant for seed length, seed width, and seed thickness. This could be a result of non-allelic interaction Panigrahi et al., (2020) obtained similar results in blackgram cowpea.

The results from the generation mean analysis estimated using five generations (P_1, P_2, F_1, F_2 and F_3), we deduce that the mean effect *(m),* additive effect *(d),* dominance effect *(h)* and additive-additive effect (*i*) were significant for seed length. Therefore, the additive, dominance and the epistatic component of additive-additive gene actions control seed length. This result is consistent with reports by Vadive *et al.,* (2019).

However, the dominance-dominance effect (*l*) was not significant for seed length. Also, the gene effects, m, d and h were significant for seed width and seed thickness but the gene effects *(i)* and *(l)* were not significant for seed width and seed thickness. This shows that additive and dominance gene action controls seed width and seed thickness, and that epistatic effect did not play major role in the control of the traits. This result agrees with reports from Thamdhara *et al.,* (2017). The value of the dominance (h) gene effect was relatively greater than that of the value of the additive gene effect signifying the prevalence of dominance allelic gene action on the inheritance of the traits. Consequently, the overall observation on the generation mean analysis confirms earlier reports by Egbadzor, (2013) that seed size was controlled by both additive and non-additive gene action.

Recurrent selection should be used for improvement on the trait. The seed width, seed thickness, and seed length distribution indicated that the progeny generations showed higher values over the lower parent (P_2) but lower values relative to the higher parent (P_1) and this shows that there is progress in the improvement of small seed size in cowpea however slow. Among the progeny generations, the F_3 generation had the highest values over the lower parent (P_2) indicating that selection for large seed size can begin from the F_3 generation. Also, the seed length, seed width, and seed thickness observed in the F_1, F_2, and F_3 generations were lower than the higher parent (P_1). This implies repeated backcrossing to the higher parent will be required to enhance selection for large seed size.

5.0 Conclusions

Seed size trait in cowpea is heritable and regulated by dominance, additive-additive, and dominance-dominance gene actions. Breeding programmes can make progress by using recurrent selections and backcrosses to make quick genetic gains when breeding for larger seeds. From this study we conclude that we cannot improve on Cowpea seed size by making between large-seeded lines and small-seeded lines and recommend that breeders should make crosses among large-seeded lines if larger seeds are the desired trait as is the case in Ghana.

Competing Interest: The authors declare none.

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