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IDENTIFICATION OF SUPERIOR BREEDING POPULATION IN COWPEA FOR YIELD ATTRIBUTING TRAITS IN SEGREGATING POPULATION

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ABSTRACT

Identification of the best breeding population is prime requirement for breeders as it is impossible to select the best transgressant from forwarding all the crosses with their realistic limit of resources and time. Ten genotypes differing in seed size, seed coat colour and pod number were chosen and thirteen F_2 populations generated from these genotypes were used for the study. Genetic parameters *viz*., trait mean, standardized range, phenotypic coefficient of variability, genotypic coefficient of variability, heritability, transgressive segregation index, usefulness index and frequency of transgressants were estimated for pods plant^{-1}, pod length and seeds pod^{-1} for all the thirteen F_2 population to identify population with best breeding potential. Population derived from 520×200 ranked best for pod length, while population derived from 399×524 and 519×502 ranked for pods plant^{-1} and seed pod^{-1}. However, F_2 population from 520×200 cross ranked second for pod length and seeds pod^{-1} and population derived from cross 399×524 identified as potential breeding population to carry forward to recover majority of transgressants.

Key words : Transgressants, usefulness index, transgressive segregation index

Introduction

Cowpea (*Vigna unguiculata* L. Walp), a versatile crop and an excellent option for climate resilient agriculture cropping systems. Its inherent capacity to be productive in dry and marginal lands makes it ideal. Cowpea young leaves, immature pods, and immature seeds are used as vegetables (Umaharan *et al*., 1997), and plant leftovers can be composted and used as animal feed. Cowpea is cultivated from East to Southeast Asia, North America, Europe, Latin America and Northern Africa. Asia's leading cowpea-growing nations include China, Korea, Thailand, Bangladesh, Myanmar, Sri Lanka and India. In India, it is cultivated in Karnataka, Madhya Pradesh, Rajasthan, Maharashtra, Andhra Pradesh, Tamilnadu etc.

Cowpea has a genome size of about 640.6 Mbp. With roughly 25% protein and 64% carbohydrates (Bressani, 1985), cowpea grain has a great deal of potential to help farmers with limited resources, who suffer from malnutrition. In fact, there have been reports of certain cultivars with seed protein contents of roughly 30%, which is comparable to the levels found in soybeans (*Glycine max*) (Singh, 2007; Santos *et al*., 2012).

Lack of variation is one of the major problems observed in self-pollinated crops, which can be overcome by hybridization. Hybridization followed by selection in subsequent generation is highly recommended to create variation and select the superior segregants with better combination of genes controlling traits of economic relevance and to isolate a superior recombinant that could serve as a promising pure line variety.

In practice, large selection responses can be obtained with a wide range of combinations between number and size of breeding populations (Bernardo, 2003). Moreover, the existence of trade-off between breeding population and population size is always thought provoking to identify strategies to use resources and time in an efficient manner.

Achieving the yield potential is through contribution from significant genetic gains. Most of the time in breeding, 99% of the crosses and resources invested are not fruitful (Witcombe *et al*., 2013). High-volume crossing strategy is adopted by majority of breeders to increase inefficiency of having many failed crosses. Breeders can justify for fewer crosses using existing information on performance *per se* of parents. Forwarding all the crosses to select the best transgressant is impossible for breeder's with realistic limit of resources and time. Strategies are to be used to select the ideal breeding population to forward. First and second degree statistics based genetic parameters *viz*., trait mean, absolute range, standardized range, phenotypic coefficient of variability (PCV), genotypic coefficient of variability (GCV), heritability, transgressive segregation index (TSI), usefulness index (UI) and frequency of transgressants can be used as criteria. In this study, our objective is to identify breeding populations with higher estimates of above-mentioned genetic parameters to be forwarded to recover superior transgressants.

Materials and Methods

Ten genotypes (Table 1) differing in seed size, seed coat colour and pod number were chosen for the study. The crosses were made amongst the contrasting parents and F_1 was identified using significant male parent characteristic and variation in seed coat colours. The F_1's are forwarded to generate F_2. F_2 populations of thirteen crosses with respective F_1s and their parents constituted the experimental material and population size varied from 100 to 151. These populations were planted in contiguous blocks of 2m length by maintaining a spacing of 0.6m between rows and 0.2m between plants within row during summer 2023 at Department of Genetics and Plant Breeding (GPB), University of Agricultural Sciences (UAS), Bengaluru, India.

Table 1 : Parental genotypes with accession numbers.

S. no.	Genotypes	Pod length (cm)	Seeds pod^{-1}	Pods plant^{-1}
1	502 - IC268262	11.1	11.2	36
2	524 - IC263015	10.94	10	29
3	520 - EC738216	13.65	7.7	18
4	505 - EC725102	12.75	11	15
5	514 - EC724826	11.85	13.2	42
6	518 - EC724805	14.49	12	17
7	519 - EC244046	13.65	9.25	22
8	200 - IC590843	17.15	9.2	21.5
9	399 - EC738260	14.17	9.6	29.8
10	510 - EC724784	13.6	14.5	30

The data was recorded on all the F_2 individuals in the crosses for pods plant^{-1}, pod length and seeds pod^{-1} at maturity. Total number of pods borne by the plant from all the pickings were counted and expressed as pods plant^{-1}. Sample of five pods were chosen for calculating pod length which is averaged and expressed in centimetres. Seeds inside all the five sample pods are counted, averaged and expressed as seeds pod^{-1}.

Data analysis

The data was recorded on all the F_2 individuals of all crosses for pods plant^{-1}, pod length and seeds pod^{-1}. The individual plant data on three traits were used to calculate descriptive statistics such as mean, standardized range, PCV, GCV, heritability, TSI, UI and frequency of transgressive segregants for each F_2 population.

Quantitative traits mean $(\bar{X})$

$$(\bar{X}) = \frac{\text{All plants in each of the } F_2 \text{ population}}{\text{Number of plants}}$$

Quantitative traits range

$$\text{Standardized range (SR)} = \frac{(\text{Highest trait mean} - \text{Lowest trait mean})}{\text{Trait mean}}$$

Transgressive segregation index (TSI)

TSI was estimated as the proportion of the difference between the parents and the corresponding range for a phenotype in the segregating (F_2) populations. Based on descriptive statistics, TSI was estimated (Koide *et al*., 2019) as

$$\text{TSI} = \frac{\text{Absolute range in segregating population}}{\text{Trait mean difference between parents}}$$

Phenotypic (PCV) and genotypic coefficient of variability (GCV)

Phenotypic variance (σ_p^2) was standardized as phenotypic coefficient of variation (PCV) (Burton and Devane, 1953) to facilitate comparison across QTs.

$$\text{PCV}(\%) = \frac{\sigma_p}{\bar{X}} \times 100$$

$$\text{GCV}(\%) = \frac{\sigma_g}{\bar{X}} \times 100$$

Where, σ_p = Phenotypic standard deviation,

σ_g = Genotypic standard deviation,

$\overline{X}$ = Quantitative traits mean

The 'σ_g' was estimated as square root of σ^2_g; The $\sigma^2 g$ was estimated as σ^2_p - σ^2_e, where, σ^2_e for F_2 populations was estimated as the average of phenotypic variance in non-segregating populations (F_1, P_1, P_2)

$$\sigma^2_e = \frac{2V_{F1} + V_{P1} + V_{P2}}{4}$$

Heritability

Heritability was estimated to measure the degree of variation in a phenotypic trait in a population that is due to genetic variation between individuals in that population. In F_2 populations, broad sense heritability (h^2_{bs}) estimates for five quantitative traits were estimated according to the formula suggested by Lush (1945).

$$h^2_{bs} = \frac{\sigma^2_g}{\sigma^2_p} \times 100$$

Where,

σ^2_g = Genotypic variance; σ^2_p = Phenotypic variance

Usefulness index (UI)/ expected genetic gain

In F_2 populations, the UI for each trait was estimated as

$$UI = Trait\ mean + \left(k \times \frac{\sigma^2_g}{\sigma_p} \right)$$

Where, k = standardized selection differential at different selection intensities; k = 2.67 and 2.06 at 1% and 5% selection intensities, respectively (Bernardo, 2020).

Frequency of transgressive segregants

The number of plants in each of the F_2 population registering trait values lower than lower scoring parent and higher than higher scoring parent involved in the construction of the respective F_2 population were counted and designated as transgressive segregants.

Criteria for identification of better breeding population

The crosses with higher estimates of the above statistical parameters are considered for identifying the breeding population with better breeding potential to forward. Rank-sum method, wherein highest estimates are assigned with ranks in ascending order and summation of ranks of crosses for all above mentioned genetic parameter estimates and all the traits, highest scores are assigned with ranks in descending order which are expressed as overall ranks of the F_2 populations (Kang, 1988; Pathy *et al.*, 2018).

Results and Discussion

Potential breeding population maintenance is an efficient technique to overcome the constraints faced by the breeder to handle the large breeding population for specific objectives of research in an efficient manner. In our study, we are presenting our results individually and combination to identify population with good breeding potential, which are worth forwarding to derive maximum number of transgressants.

Mean of the trait which depicts the tendency of the individuals in a population to congregate in the distribution is the important first degree statistic. Population derived from 519×524 recorded highest mean of 39.09 for pods plant^{-1}. While, 510×505 had lengthier pods and more seeds per pod (Tables 2, 3, 4).

Absolute range being one of the measures of dispersion denotes variability present in the population. However, standardized range is helpful when traits with different units of measurement are to be compared within and among segregating populations. All the populations exhibited lower magnitude of standardized range for pods

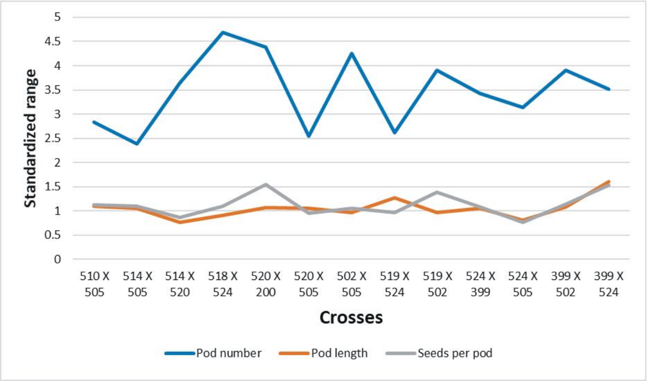

Fig. 1 : Estimates of standardized range for pods plant^{-1}, pod length, seeds pod^{-1} in thirteen crosses of cowpea.

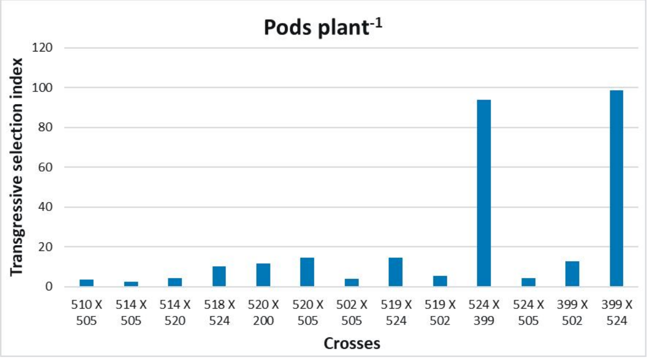

Fig. 2 : Estimates of transgressive segregation index for pods plant^{-1} in thirteen crosses of cowpea.

Table 2 : Genetic parameter estimates for pods plant^{-1} in thirteen crosses of cowpea.

Crosses	Mean	PCV (%)	GCV (%)	Heritability (%)	Freq. >Higher parent	Freq. <Lower parent
510 × 505	17.95	67.03	65.64	95.90	0.15	0.41
514 × 505	26.31	58.65	57.35	95.62	0.16	0.27
514 × 520	27.21	68.33	66.42	94.50	0.20	0.34
518 × 524	25.78	73.34	71.98	96.32	0.29	0.34
520 × 200	9.37	88.77	82.71	86.82	0.05	0.82
520 × 505	17.31	61.44	60.80	97.91	0.33	0.42
502 × 505	19.77	77.16	74.76	93.88	0.12	0.32
519 × 524	39.09	63.34	62.52	97.40	0.49	0.16
519 × 502	18.92	88.82	86.94	95.82	0.15	0.68
524 × 399	21.89	66.43	64.73	94.93	0.30	0.68
524 × 505	18.46	65.03	63.14	94.27	0.15	0.25
399 × 502	19.93	80.66	78.66	95.09	0.16	0.72
399 × 524	22.47	81.29	80.00	96.86	0.27	0.71

Table 3 : Genetic parameter estimates for pod length in thirteen crosses of cowpea.

Crosses	Mean (cm)	PCV (%)	GCV (%)	Heritability (%)	Freq. >Higher parent	Freq. <Lower parent
510 × 505	13.00	20.03	15.03	56.26	0.45	0.39
514 × 505	11.17	20.79	13.12	39.85	0.25	0.56
514 × 520	11.39	16.59	4.92	8.81	0.14	0.01
518 × 524	10.08	19.11	7.22	14.27	0.00	0.64
520 × 200	12.38	27.71	21.88	62.35	0.03	0.59
520 × 505	12.63	23.23	17.16	54.60	0.51	0.49
502 × 505	9.17	22.69	11.99	27.93	0.02	0.80
519 × 524	10.74	21.60	15.18	49.43	0.02	0.56
519 × 502	10.93	22.34	14.63	42.90	0.09	0.44
524 × 399	9.32	20.98	10.50	25.06	0.00	0.78
524 × 505	10.85	19.16	11.61	36.70	0.17	0.46
399 × 502	9.70	22.47	12.85	32.70	0.00	0.69
399 × 524	8.06	27.42	18.32	44.64	0.01	0.91

plant^{-1}, pod length and seeds pod^{-1}. However, pods plant^{-1} recorded higher estimates in comparison with pod length and seeds pod^{-1} (Fig. 1). Furthermore, population derived from 518×524 recorded a greater number of pods per plant compared to other crosses (Fig. 1).

Dispersion observed amongst the population can be attributed to genotypic and environmental variability. These variabilities are expressed in the form of phenotypic coefficients and genotypic coefficient of variability, their heritable aspect is expressed in the form of heritability. Population derived from cross 519×502 had higher variability for number of pods borne by the plant while heritability estimates were higher in 520×505 (Table 2). Whereas, variation in length of pods as well as seeds per pod and heritability for the trait were found to be promising in the population from cross 520×200 (Tables 3 and 4).

Frequency of the transgressants with estimates higher than higher parent is most important. Population derived from 519×524 (Table 2) had more segregants for pod number while, Population from crosses 510´505 (Table 3) and 524×505 (Table 4) consisted of segregants with lengthier pods and a greater number of seeds per pod.

Transgressive segregation index (TSI) thus indicates occurrence of standardized and unit-independent extreme trait phenotypes. Higher the TSI, better would be the breeding potential of crosses. Transgressive segregation (TS) is the reason for the success of plant breeding. (Mackay *et al.*, 2020). TSI estimates for trait differed among the population and we could observe huge differences in the estimates amongst the population.

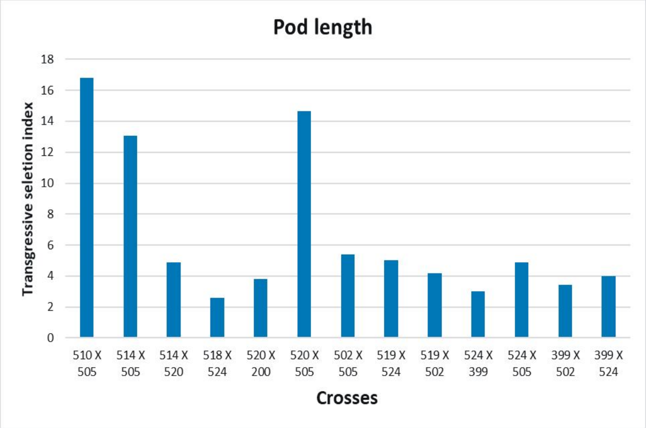

Fig. 3 : Estimates of transgressive segregation index for pod length in thirteen crosses of cowpea.

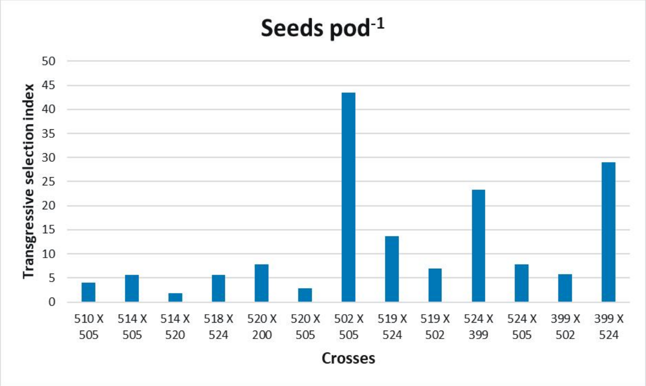

Fig. 4 : Estimates of transgressive segregation index for seeds pod^{-1} in thirteen crosses of cowpea.

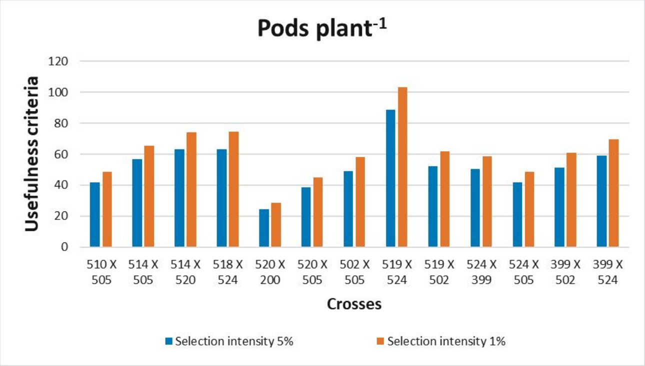

Fig. 5 : Estimates of usefulness index for pods plant^{-1} in thirteen crosses of cowpea.

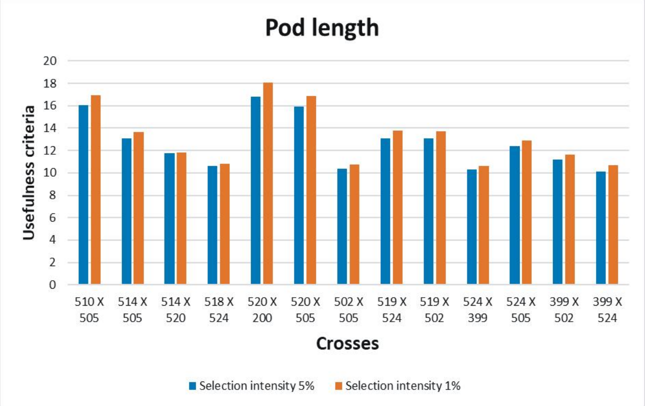

Fig. 6 : Estimates of usefulness index for pod length in thirteen crosses of cowpea.

Table 4 : Genetic parameter estimates for seeds pod^{-1} in thirteen crosses of cowpea.

Crosses	Mean	PCV (%)	GCV (%)	Heritability (%)	Freq. >Higher parent	Freq. <Lower parent
510 × 505	12.65	20.17	14.69	53.06	0.20	0.23
514 × 505	11.41	21.17	14.41	46.37	0.24	0.38
514 × 520	11.29	20.99	15.42	53.95	0.21	0.10
518 × 524	10.14	23.74	17.67	55.39	0.24	0.47
520 × 200	7.56	37.33	33.94	82.66	0.32	0.36
520 × 505	9.67	23.70	18.46	60.69	0.36	0.16
502 × 505	8.29	24.82	16.05	41.81	0.06	0.88
519 × 524	10.48	23.15	19.48	70.78	0.31	0.40
519 × 502	9.88	29.28	24.85	72.03	0.35	0.37
524 × 399	8.62	24.37	19.91	66.73	0.25	0.69
524 × 505	10.27	19.42	13.59	48.98	0.40	0.35
399 × 502	8.09	26.90	19.93	54.93	0.08	0.73
399 × 524	7.58	29.93	25.36	71.77	0.12	0.79

Population derived from 399×524 (Fig. 2), 510×505 (Fig. 3), 502×505 (Fig. 4) had a greater number of segregants with increased pod number, pod length and seeds per pod, respectively.

Usefulness index (UI) has been suggested as an unambiguous objective method of selecting early segregating generations to increase the frequency of recovering superior RILs for use as pure-line cultivars in self-pollinated crops (Mackay *et al.*, 2020; Melchinger, 1987). UI at selection intensity of 1% recorded higher

Table 5 : Rank-sum estimates of thirteen crosses to identify potential cross for pods plant^{-1}, pod length and seeds pod^{-1}.

Crosses	Pod length rank sum	Rank a	Pod number rank sum	Rank b	Seeds per pod rank sum	Rank c	Rank sum (a+b+c)	Overall rank
510 × 505	38	3	82	11	72	8	192	7
514 × 505	55	5	86	12	72	8	213	10
514 × 520	88	12	58	6	84	13	230	12
518 × 524	95	13	44	2	62	6	201	9
520 × 200	35	2	65	8	43	2	143	2
520 × 505	34	1	72	9	73	10	179	5
502 × 505	69	8	75	10	74	11	218	11
519 × 524	50	4	55	5	51	4	156	4
519 × 502	64	7	50	4	38	1	152	3
524 × 399	85	11	58	6	56	5	199	8
524 × 505	76	10	92	13	81	12	249	13
399 × 502	71	9	48	3	66	7	185	6
399 × 524	56	6	34	1	47	3	137	1

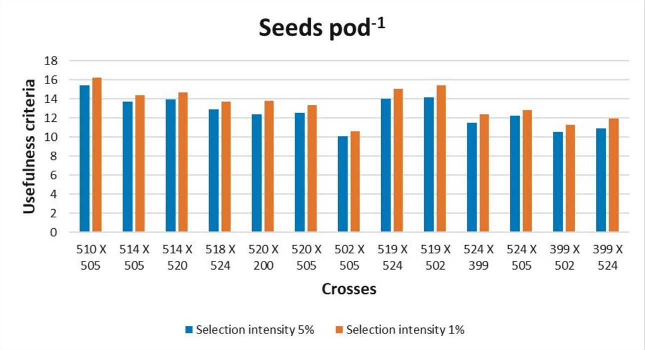

Fig. 7 : Estimates of usefulness index for seeds pod^{-1} in thirteen crosses of cowpea.

estimates than at 5%. Population derived from 519×524 (Fig. 5), 520×200 (Fig. 6), 510×505 (Fig. 7) had a greater number of pods, lengthier pods and increased number of seeds per pod, respectively.

According to the aforementioned results and discussion, the ranking of the thirteen crosses in terms of breeding potential is different from the statistic. The concept of rank sum method can be used in cowpea to identify best breeding populations for extracting superior RILs for use as parents and cultivars (Chandana and Ramesh, 2021). In our results, we considered all the genetic parameters with equal weightage for assigning the ranks, which later is used as tool to select the segregating population.

Genetic parameter estimates for F_2 populations for each trait was ranked and ranks of crosses for each trait are mentioned in Table 5. Population derived from 520×200 ranked first for pod length, while population derived from 399×524 and 519×502 for pods plant^{-1} and seed pod^{-1} ranked lowest. 520×200 cross ranked second for pod length and seeds pod^{-1} (Table 5). Apart from that none crosses had common lowest rank, hence rank-sum across the trait were pooled and overall rank was assigned to the crosses. The population derived from 399×524 were found to be promising and potential. Forwarding such multi trait derived superior populations can assist isolation of elite recombinants with multiple target traits.

Conclusion

It is not reliable to identify one best population based on the one genetic parameter or one trait. The best strategy like rank-sum method can be used to identify potential breeding population considering majority of the traits and efficient genetic parameters. From our study, it can be concluded that population derived from 399´524 was found to be potential to carry forward to recover majority of transgressants.

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