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Original Research Article

Genotypic Selection for Stability and Yield Adaptability of Short-Duration Pigeon Pea in Mozambique using REML/BLUP Mixed Models

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Abstract: The objective of the research was to select short-duration pigeon pea genotypes with high yield, stability and broad genotypic adaptability for northern region of Mozambique using REML/BLUP mixed model. Twelve short-duration genotypes were evaluated in twelve environments from combinations of locations and years: Nampula (2016, 2017 and 2018), Ribáuè (2016), Mogovolas (2016, 2017 and 2018), Namapa (2016, 2017 and 2018) and Montepuéz (2017 and 2018), in a randomized block design with three replications. The genetic parameters were estimated by the mixed model of restricted maximum likelihood (REML) and best linear unbiased prediction (BLUP). The significance of the deviance analysis effects was tested by the likelihood ratio test, considering the $\chi 2$ distribution. The selection genotypic strategies were based on the predicted genetic values free of interaction ($\mu + g$), with local interaction ($\mu + g + ge$), mean of environments ($\mu + g + gem$), and harmonic mean of relative performance (MHPRVG). Different genotypic responses were observed for genotypes x locations and genotypes x locations x years interactions. The REML/BLUP mixed model allowed the selection of the short-duration pigeon pea genotypes ICEAP 01107/5, ICEAP 01107/8 and ICEAP 01284, simultaneously for yield, stability and broad genotypic adaptability for northern region of Mozambique.

Keywords: *Cajanus cajan*, G x E interaction, MHPRVG.

INTRODUCTION

Pigeon pea [*Cajanus cajan* (L.) Millspaugh] is an important source of protein (20 - 23%) for human consumption and atmospheric nitrogen for soil fertility [1]. This legume is grown in semi-arid regions of Asia, Africa, Latin America and the Caribbean [2]. It is considered to be native to India [3] and is currently cultivated in many parts of the world, including southern Africa, particularly the region spanning Kenya, Mozambique, Malawi and southern Tanzania [4]. In Mozambique, it plays a greater role in feeding poor families, as fresh and dry grains are sold to generate income for small farmers. The increase in the number of households and the area per household justifies the increase in pigeon pea production, indicating that yield was not identified as a cause [5]. For a long time, only late-maturing local varieties, including medium and long duration improved varieties are dominated the agricultural landscape in Mozambique, however, according to [5] the yield of these cultivars at the level of small farmers varies from 300 – 400 kg ha^{-1}, which is comparatively low to the world mean yield (kg ha^{-1}): 910, from India: 730 [6] and from the neighboring country Malawi: 1102.8 [7]. The low yield can be attributed to the lack of genetically superior cultivars to the effects of biotic and abiotic factors in different environments. The use of short-duration pigeon pea varieties, which are, according to [8], photo-insensitive, productive, adapted to a wide range of altitudes, terminal drought stresses, can be an alternative for increasing yield, however, according to

[9, 10] when the yield is evaluated in a series of environments, it is influenced by genetic and environmental effects, and an additional effect of the interaction of both, meaning that the best genotype in one environment is not in another. To minimize the effects of GE interactions and to achieve higher performance predictability, the identification of the most stable genotypes and adapted to environments is necessary. Several methods have been described for examining stability and adaptability, as based on the analysis of variance, linear regression, nonlinear regression, multivariate analysis and nonparametric statistics.

In the context of REML/BLUP (restricted maximum likelihood/best unbiased linear predictor) mixed models method, the use of harmonic mean values of genotypic values (MHVG) simultaneously implies selection for yield and stability, the relative performance of genotypic values (PRVG) implies selection for adaptability and the harmonic mean of relative performance (MHPRVG) allows: i) simultaneous selection of yield, adaptability and stability; ii) considering the genotypic effects as random and, therefore, providing estimates of stability and adaptability for predicted and non-predicted genotypic values; iii) allowing the use of unbalanced data, non-orthogonal designs and heterogeneity of variance; iv) allowing the consideration of correlated errors within the locations in addition to stability and adaptability in the selection of individuals within progeny; v) providing genetic values already discounted (penalized) with regard to instability and allowing the application of any number of environments; and vi) generating results at the unit or scale of the evaluated trait, which can be directly interpreted as genetic values [11]. This methodology has been used to study stability and genotypic adaptability of various grain legumes such as common bean [12], soybean [13], cowpea [14] among others, however, its application in pigeon pea is still scarce. Thus, the objective of this work was to select short-duration pigeon pea genotypes with high yield, stability, and broad genotypic adaptability for the northern region of Mozambique using REML/BLUP mixed models.

MATERIAL AND METHODS

Study area

The experiments were carried out in field conditions from northern region of Mozambique, in 12 environments from combinations of locations and years: Nampula (2016, 2017 and 2018): 15°09'17.9"S 39°18'18.1"E, Ribáuè (2016): 15°03'27.4"S 38°13'46.1"E, Mogovolas (2016, 2017 and 2018): 15°44'05.3"S 39°17'01.9"E, Namapa (2016, 2017 and 2018): 13°43'32.0"S 39°46'18.1"E and Montepuéz (2017 and 2018): 13°13'04.2"S 38°52'11.6"E (Fig 1).

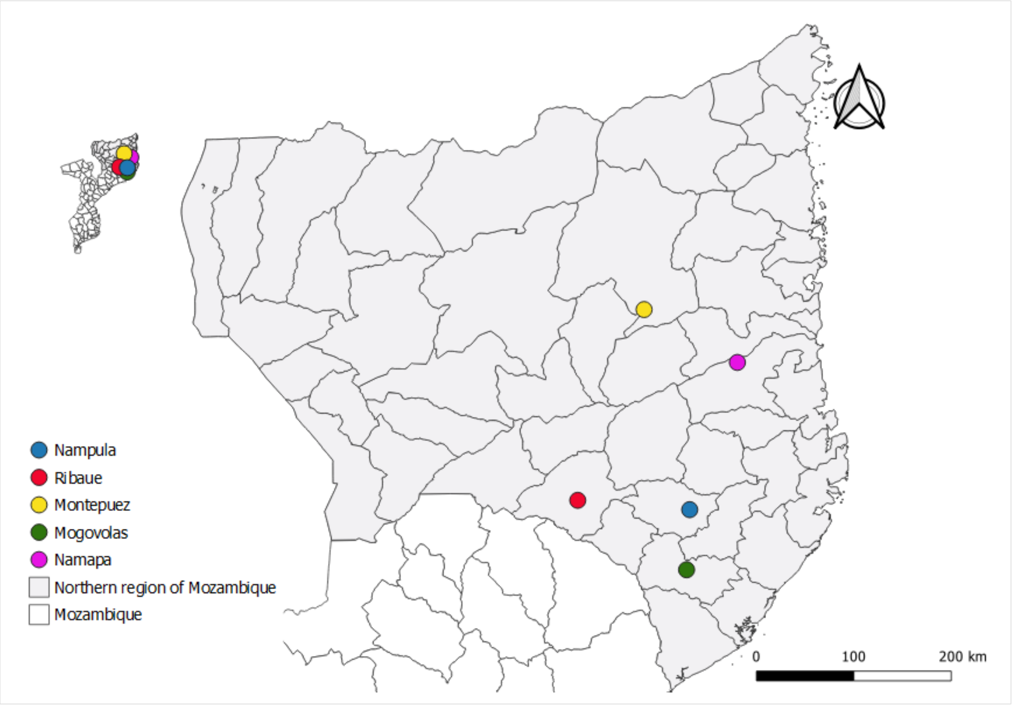

Fig-1: Experiment locations of the evaluated genotypes in Mozambique

Genetic material and experimental design

Were tested twelve ICRISAT (International Crops Research Institute for the Semi-Arid Tropics) pigeon pea genotypes (ICEAP 00604, ICEAP 00612, ICEAP 00652/2, ICEAP 01101/1, ICEAP 01101/2, ICEAP 01106/1, ICEAP 01107/5, ICEAP 01107/6, ICEAP 01107/8, ICEAP 01284, ICPL 86012 and ICPL 87091) of short-duration and determined growth, in a randomized block design with three replications.

Experiment size, crop management and data collection

Sowing was carried out manually at 0.5 m spacing between rows, in plots of two rows of 5 m in length and with 10 seeds per linear meter. Two weedings were carried out throughout the crop cycle. Pest control was carried out with synthetic insecticides (Cypermethrina and Zakanaca) every 15 days, from flowering to physiological maturation of the pods. The Harvest was carried out manually in all plots when about 90% of the pods were dry and golden. It was followed by drying in the sun, manual threshing to obtain the grain production per plot and subsequent determination of grain yield in kg ha^{-1}.

Statistical data analysis

The data regarding the grain yield (kg ha^{-1}) were evaluated by the mixed model methodology, in which the genetic parameters were estimated by the Restricted Maximum Likelihood (REML) method [11] and the predicted genotypic values by the method of the Best Linear Unbiased Prediction (BLUP) [15], using model:

$$y = Xb + Zg + Qa + Ti + Wt + e,$$

Where y is the data vector, b is the vector of the effects of replication-location-year combinations (fixed) added to overall mean, g is the vector of the genotypic effects (random), a is the vector of the effects of genotypes x years interaction (random), i is the vector of the effects of genotypes x locations interaction (random), t is the vector of the effects of genotypes x locations x years interaction (random) and e is the vector of residuals (random). Capital letters represent the incidence matrices for these purposes.

Vector b contains the replication effects group with locations and years, locations, years and locations-years interaction (adjusting the replication-locations-years combinations). The structure of means and variances were given by [16] and, for the adopted model, the equations for the mixed model were as follows:

$$\begin{bmatrix} X'X & X'Z & X'Q & X'T & X'W \\ Z'X & Z'Z + I\lambda_1 & Z'Q & Z'T & Z'W \\ Q'X & Q'Z & Q'Q + I\lambda_2 & Q'T & Q'W \\ T'X & T'Z & T'Q & T'T + I\lambda_3 & T'W \\ W'X & W'Z & W'Q & W'T & W'W + I\lambda_4 \end{bmatrix} \begin{bmatrix} \hat{b} \\ \tilde{g} \\ \tilde{a} \\ \tilde{\imath} \\ \tilde{t} \end{bmatrix} = \begin{bmatrix} X'y \\ Z'y \\ Q'y \\ T'y \\ W'y \end{bmatrix}, \text{ Where:}$$

$$\lambda = \frac{{\sigma_e}^2}{\sigma_g^2} = \frac{1-h^2-a^2-i^2-t^2}{h^2}, \lambda_2 = \frac{{\sigma_e}^2}{\sigma_{gy}^2} = \frac{1-h^2-a^2-i^2-t^2}{a^2} \lambda_3 = \frac{{\sigma_e}^2}{\sigma_{gl}^2} = \frac{1-h^2-a^2-i^2-t^2}{i^2} \text{ and }, \lambda_4 = \frac{{\sigma_e}^2}{\sigma_{gly}^2} = \frac{1-h^2-a^2-i^2-t^2}{t^2},$$

Where: ${\sigma_e}^2$ is the residual variance, σ_g^2 is the genotype variance among elite genotypes, σ_{gy}^2 is the variance of the g x y interaction, σ_{gl}^2 is the variance of the g x l interaction, σ_{gly}^2 is the variance of the g x l x y interaction, h_g^2 is the individual heritability in the broad sense, at the block level, equal to: $\frac{\sigma_g^2}{\sigma_g^2+\sigma_{gl}^2+\sigma_{gy}^2+\sigma_{gly}^2+{\sigma_e}^2}$, and a^2, i^2 and t^2 are the coefficients of determination of the effects c_{gy}^2 , c_{gl}^2 e c_{gly}^2 , respectively.

Genotypic correlations across locations and years were determined using the equations: $r_{gl} = \frac{\sigma_g^2}{{\sigma_g}^2+\sigma_{gl}^2}$ is the genotypic correlation across locations for any year, $r_{gy} = \frac{\sigma_g^2}{{\sigma_g}^2+\sigma_{gy}^2}$ is the genotypic correlation across years for any local, $r_{gl\_y} = \frac{\sigma_g^2+\sigma_{gy}^2}{(\sigma_g^2+\sigma_{gy}^2)+\sigma_{gl}^2}$ is the genotypic correlation between locations across the years, $r_{gy\_l} = \frac{\sigma_g^2+{\sigma_{gl}}^2}{({\sigma_g}^2+\sigma_{gl}^2)+\sigma_{gy}^2}$ is the genotypic correlation between years across the locations, and $r_{gly} = \frac{\sigma_g^2}{\sigma_g^2+\sigma_{gy}^2+\sigma_{gl}^2+\sigma_{gly}^2}$ is the genotypic correlation between locations and years [16].

The significance of the effects of the model was estimated by the joint analysis of deviance through the likelihood ratio test (LTR), without considering the effects of g, gy, gl and gly. The deviance without the effect was subtracted by the deviance of the complete model followed by the comparison with the chi-square value (χ2), with a degree of freedom, at 1% probability according to [17].

Through the model used, the BLUP predictors of the free genotypic values of the interaction were obtained, given by $\hat{\mu} + \hat{g}_i$, where $\hat{\mu}$ is the mean of all environments and $\hat{g}_i$ is the free genotypic effect of the G x E interaction. For each environment j, the genotypic values (Vg) were predicted by $\hat{\mu}_j + \hat{g}_i + (\hat{g}e)_{ij}$, where $\hat{\mu}_j$ is the mean of environment j, $\hat{g}_i$ is the genotypic effect of genotype i, in the environment j, and $(\hat{g}e)_{ij}$, is the effect of the G × E interaction relative to genotype i. The prediction of genotypic values capitalizing the mean interaction in the different environments is given by $\hat{\mu}_j + \hat{g}_i + \hat{g}e_m$, being calculated by $\hat{\mu} + (\hat{\sigma}_g^2 + \hat{\sigma}_c^2/n)/\hat{\sigma}_g^2\ x\hat{g}_i)$, where $\hat{\mu}$ is the overall mean of all environments, n is the number of environments and $\hat{g}_i$ the genotypic effect of genotype i [11].

The harmonic mean of genetic values related to genotype i (MHVG) considered to assess the stability and yield of the genotypes and the relative performance of the predicted genotypic values related to genotype i (PRVG) considered to assess the adaptability were obtained, respectively, by:

$$\mathrm{MHVG_i} = \frac{\mathrm{n}}{\left[\sum_{\mathrm{j=1}}^{\mathrm{n}}\left(\frac{1}{\mathrm{Vg_{ij}}}\right)\right]}$$

$$\mathrm{PRVG_i} = \frac{1}{\mathrm{n}}\left[\sum_{\mathrm{j=1}}^{\mathrm{n}}\left(\frac{\mathrm{Vg_{ij}}}{\mathrm{u_i}}\right)\right]$$

And the model which was considered simultaneously for yield, adaptability and stability, obtained by the harmonic mean of the relative performance of genetic values related genotype i (MHPRVG), was calculated by:

$$\mathrm{MHPRVG_i} = \frac{\mathrm{n}}{\left[\sum_{\mathrm{j=1}}^{\mathrm{n}}\left(\frac{1}{\mathrm{PRVG}}\right)\right]}$$

Where, n is the number of environments where the genotype I was evaluated; $\mathrm{Vg_{ij}}$ is the genotypic value of genotype i in environment j, expressed as the proportion of the mean of this environment; and μ_j is the overall mean of each environment j. The mean genotypic value, capitalizing on adaptability, is obtained by PRVG multiplied by the general mean of all environments (PRVG*μ), and the average genotypic value penalized by instability and capitalized by stability is calculated by the MHPRVG multiplied by the general mean of all environments (MHPRVG*μ). All analyses were carried out in the software Selegen-REML/BLUP [11].

RESULTS AND DISCUSSION

Deviance analysis and variance components (Individual REML)

The deviance analysis to yield showed that the effects of the genotypes x locations and genotypes x locations x years interactions were significant ($p \leq 0.01$) indicating an inconsistency in the performance of genotypes in different locations and locations x years interactions. The effects of genotypes and genotypes x years interaction were not significant, this may be due to the consumption of genetic variability among pigeon pea genotypes, caused by the occurrence of high genotypes x locations interaction according to [9], however, the genotypes x years interaction did not change the performance of genotypes over the years (Table 1). The significant effects can be explained by the magnitude of the variance componentes (Table 2), which show low correlations of genotypes across locations in any year (*rgl*), through the years in any location (*rgy*), locations in a given year (*rgl_y*), locations for the mean of all years (*rgl_my*) and between locations and years (*rgly*) which confirms the existence of the complex interaction caused when the correlation between the performance of genotypes across environments is low, changing the relative position of genotypes due to different responses to environmental variations according [9]. However, the non-significance effects can be explained by the high genotypic correlations observed over the years in a given location (*rgy_l*) and over the years for the mean of all locations (*rgy_ml*) confirming that, in a given location and for the mean of all locations, the variability of unpredictable biotic and abiotic factors over the years was not sufficient to cause changes in the performance of the genotypes.

The low correlations between the G x E interactions are explained by the greater contribution of the σ_{gl}^2 (50.78%), followed by $\sigma_g^2 ly$ (24.88%) and σ_e^2 (20.21%) in phenotypic expression, while the high correlations are explained by the low influence of the σ_{gy}^2 (3.91%) and σ_g^2 (0.22%) in phenotypic expression. The low σ_g^2 was reflected in the reduction of individual genotypic heritability in the broad sense, this reduction may be associated with the quantitative nature of the trait that is highly influenced by the environment and the low genetic variability between genotypes, caused by the occurrence of high genotypes x locations interaction according to [18, 9].

Table-1: Deviance of grain yield (kg ha^{-1}) of short-duration pigeon pea (*Cajanus cajan*) genotypes evaluated in 12 environments in northern region of Mozambique

Effects	**$^{+}$Deviance**	**LRT (χ2)**
Genotypes	4841.43	-0.01
Genotypes x Years	4842.96	1.52
Genotypes x Locations	4849.12	7.68**
Genotypes x Locations x Years	4906.49	65.05**
Full Model	4841.44	-

$^{+}$: Model deviation adjusted without corresponding effects, LTR: likelihood ratio test. Chi-square: χ2. **: Significant by chi-square test at 1% (6.63) probability. -: absent.

Table-2: Variance components of grain yield (kg ha^{-1}) of short-duration pigeon pea (*Cajanus cajan*) genotypes evaluated in 12 environments in northern region of Mozambique

Variance Components (Individual REML)		
σ_g^2	Genotypic variance	304.4195
σ_{gy}^2	Variance of the genotypes x years interaction	5456.3994
σ_{gl}^2	Variance of genotypes x locations interaction	70900.9510
σ_{gly}^2	Variance of the genotypes x locations x years interaction.	34734.5904
σ_e^2	Residual variance	28217.3876
σ_f^2	Individual phenotypic variance	139613.7479
h_g^2	Individual genetic heritability in the broad sense	0.00218 ± 0.0064
c_{gy}^2	Coefficient of determination of genotypes x years interaction	0.0391
c_{gl}^2	Coefficient of determination of genotypes x locations interaction	0.5078
c_{gly}^2	Coefficient of determination of the genotypes x locations x years interaction	0.2488
rgl	Genotypic correlation across locations, for any year	0.0043
rgy	Genotypic correlation across years, for any location	0.0528
rgl_y	Genotypic correlation across locations, in a given year	0.0752
rgy_l	Genotypic correlation across years, in a given location	0.9288
rgl_my	Genotypic correlation across locations, for the mean of all years	0.1620
rgy_ml	Genotypic correlation across years, for the mean of all locations	0.7971
rgly	Genotypic correlation across locations and years	0.0027
μ	Overall mean	840.4203

The coefficients of determination c_{gy}^2, c_{gl}^2 *and* c_{gly}^2 indicate how much each component contributes to the total variance of the phenotype (Table 2). Thus, these coefficients explain that the genotypes x locations, genotypes x locations x years and genotypes x years interactions contributed 63.83, 31.29 and 4.88%, respectively to the total phenotype variance, with the genotypes x locations interaction being the main cause of the differentiated response of genotypes followed by the interaction genotypes x locations x years. The greater influence of locations for interaction can be explained by the great predominance of the variability of unpredictable biotic and abiotic factors in different locations. Similar results were observed by [19, 8, 20, 2] when studying the effect of genotypes x environments interaction on yield and physiology of 30 short-duration genotypes in 20 environments in India, the stability and yield adaptability of short-duration pigeon pea hybrids in 10 environments in India, the stability of 15 genotypes in Kharif, in five environments and the adaptability of 28 pigeon pea genotypes in 10 locations, under rainfed conditions in the states of India, respectively, using AMMI and GGE-Biplot methods.

Mean BLUP components

The G x E interaction makes it difficult to widely select and recommend superior genotypes [9, 10]. Thus, the mean BLUP components associated with the predicted genotypic values free of interaction (μ + g) and the mean genotypic values predicted for various environments (μ + g + gem) were estimated (Table 3). Based on the μ + g strategy, it was found that the predicted genotypic values were close to each other and to the overall mean. This can be explained by the non-significant genotypic effect, previously shown in Table 1. Whereas, the strategy based on μ + g + gem, showed a variation from 702.94 to 921.58 kg ha-1, indicating the genotypes ICEAP 01107 /5 (921.58), ICEAP 00612 (919.89), ICEAP 01284 (917.69), ICPL 86012 (913.39) and ICEAP 01107/8 (841.48 and 890.81) more productive than the most genotypes evaluated. According to [11] genotypes with high values of μ + g + gem can be recommended for cultivation in other locations with the same pattern of interaction. These values were above 360 and below 2009 kg ha^{-1} observed by [20], below 1094.19 to 2300.36 kg ha^{-1} by [8, 20] and above 516 and below 2590 kg ha^{-1} by [2].

In general, the yield (kg ha^{-1}) per environment ranged from 348.93 to 2072.03. The highest performance of the genotypes was observed in the environments of Nampula and Ribáuè, while the lowest performance occurred in the environments of Mogovolas, Namapa and Montepuéz, respectively (Table 3). In these locations, a specific recommendation of genotypes capable of maximizing their yield potential is suggested. [11] recommends using predicted genotypic values (μ + g + ge) to select the best genotypes in each location. Based on this strategy, the yield performance (kg ha^{-1}) of the genotypes ICEAP 00612 and ICPL 87091 was better in Nampula with 1357.19 and 1313.95, respectively. The ICPL 86012 and ICEAP 00652/2 genotypes were better in Ribáuè with 2699.14 and 2531.32, respectively. The ICPL 87091 genotype was better in Montepuéz with 1050.54. These values were similar to those observed by [19, 8, 20, 2].

Table-3: Mean BLUP components associated with predicted genotypic values (µ + g) free of interaction, predicted genotypic values (µ + g + ge) capitalizing the interaction with the environment and mean genotypic value of environments (µ + g + gem) in 12 short-duration pigeon pea (*Cajanus cajan*) genotypes evaluated in 12 environments in northern region of Mozambique

Genotypes	µ + g +ge					µ + g	µ + g +gem
	Nampula	Ribáuè	Mogovolas	Namapa	Montepuéz		
ICEAP 01107/5	1136.24	2038.27	327.13	955.48	931.53	842.13	921.58
ICEAP 00612	1357.19	2423.71	332.22	557.47	709.68	842.09	919.89
ICEAP 01284	1182.67	2457.51	437.1	752.64	539.37	842.04	917.69
ICPL 86012	916.19	2699.14	309	716.43	707.17	841.95	913.39
ICEAP 01107/8	1134.05	2248.42	353.94	703.19	795.89	841.48	890.81
ICEAP 00652/2	968.94	2531.32	377.01	718.31	564.56	841.16	875.67
ICPL 87091	1313.95	1963.7	290.53	521.12	1050.54	841.08	871.59
ICEAP 00604	1073.84	2063.77	376	632.94	678.83	839.75	808.42
ICEAP 01107/6	1231.73	2111.97	328.21	555.7	584.88	839.69	805.82
ICEAP 01101/1	1280.22	1266.55	367.34	800.45	715.81	838.08	729.06
ICEAP 01101/2	1192.19	1795.09	326.71	480.91	631.16	838.06	728.19
ICEAP 01106/1	1158.09	1264.88	362	725.15	790.25	837.53	702.94
Mean	1162.11	2072.03	348.93	676.65	724.97	840.42	840.42

Stability and genotypic adaptability

Other selection strategies were based on MHGV criteria for selecting genotypes with high yield and stability, PRGV*µ for selecting genotypes with high adaptability considering genotypic value multiplying adaptability and MHPRGV*µ for simultaneous selection of genotypes with high yield, stability and genotypic adaptability, considering genotypic value penalized by instability and multiplied by adaptability (Figure 2). Based on these criteria, there was a consensus for the selection of genotypes ICEAP 01107/5, ICEAP 01107/8, ICEAP 01284 with high yield, stability and genotypic adaptability, although the ICEAP 01107/8 and ICEAP 01284 genotypes have shown a change in classification for MHPRGV. However, the predicted mean values for genotypic stability (MHGV) of the selected genotypes were below the overall mean and the mean predicted values for adaptability (PRVG) and simultaneous selection for genotypic yield, stability and adaptability (MHPRVG) were above the overall mean. These last two criteria were more efficient in selecting superior genotypes in this study. Thus, according to [11], these genotypes can be recommended for wide adaptability and genotypic stability, that is, for cultivation in multiple locations with the same pattern and in multiple other locations with a varying pattern of g x e interaction. These values penalize the instability of genotypes and capitalize on their ability to respond to improved environments. The yield of the genotypes selected is lower than the genotypes selected by [19, 8, 20, 2]. However, they have yields that can be recommended for cultivation in the northern region of Mozambique, where it is necessary to popularize short-duration pigeon pea varieties at the small farmer's level.

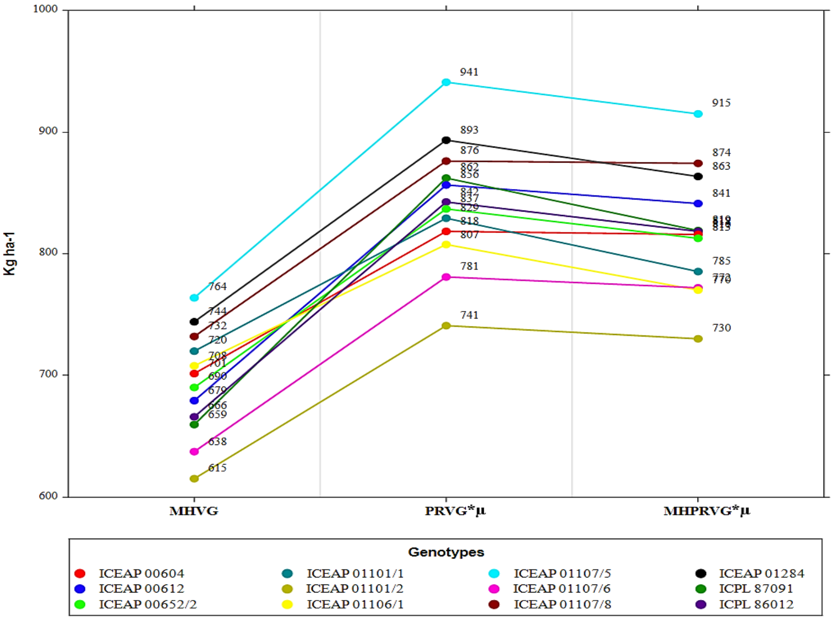

Fig-2: Genotypic stability (MHVG), genotypic value multiplying adaptability (PRVG*µ) and genotypic value penalized by instability and multiplied by adaptability (MHPRVG*µ) for yield (kg ha^{-1}) of 12 short-duration pigeon pea genotypes evaluated in 12 environments in northern region of Mozambique

The genotypes ICPL 87091, ICEAP 00612, ICPL 86012, ICEAP 00652/2, ICEAP 01101/1, ICEAP 00604 and ICEAP 01106/1 have lower performance and changes in classification regarding one of the three selection attributes considered. While the ICEAP 01107/6 and ICEAP 01101/2 genotypes, although they showed the worst performance, they did not show changes in the classification for the three attributes of the evaluation of yield, stability and adaptability.

CONCLUSION

The genotypes x locations and genotypes x locations x years interactions were the main reason for the classification of genotypes in different environments.

The short-duration pigeon pea genotypes ICEAP 01107/5, ICEAP 01107/8 and ICEAP 01284, simultaneously present high yield, stability and broad genotypic adaptability;

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