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Estimation of genetic parameters and gene actions of sorghum [*Sorghum bicolor* (L.) Moench] tolerance to low p condition

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Abstract

A study of sorghum tolerance to low P condition was conducted in two experiments : (1) in nutrient solution and (2) in acid soil. The aim of this study was to obtain information on the genetic variability and heritability of agronomic characters that could be used as selection criteria in low P condition and to obtain information on gene action that controlled agronomic characters of sorghum. The plant materials used in this experiment were F_2 segregating population derived from the cross between B69 and Numbu. The first experiment was conducted in the green house of Bogor Agricultural University. B69, Numbu and F_2 population were grown hidroponically for 14 days. Composition of the nutrient solution used in the experiment followed the method suggested by Ohki with modification of P concentration. P is given in the form of KH_2PO4: 0.001 mM. The results showed that shoot dry weight (SDW) and total dry weight (TDW) had high estimate of heritability and moderate genotypic coefficient of variance (GCV), thus these characters can be used for criteria selection for low P tolerance at seedling stage. The significant value of skweness from the distribution shoot dry weight in F_2 segregants, indicate the presence of complementary epistasis gene action, whereas normal distribution of total dry weight showed additive gene action. The second experiment was conducted in acid soils of Bogor District Experimental Field Tenjo, Bogor. The results showed that stem diameter, panicle weight and grain yield per plant had high heritability and wide GCV value, indicating these characters can be used as the selection criteria in selecting sorghum genotypes for low P tolerance in acid soil. Based on the skewness values, all of the agronomy characters observed in this study were influenced by additive gene action.

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Introduction

Sorghum is the fifth major food crops in the world after rice, wheat, maize and barley (Reddy *et al.*, 2007a). Sorghum is a potential source of carbohydrates because it has a relatively high carbohydrate content of about 73 g/100 g of edible materials. Sorghum also contains protein, calcium and vitamins B_1 higher than rice and corn (Beti *et al.,* 1990). Another advantage of sorghum is gluten content and a low glycemic index so it is suitable for consumers with special nutritional needs (Schober *et al.*, 2007). In the Africa region, grain sorghum is consumed in the form of unleavened breads, porridge, beverages, popped grain and chips (Dicko *et al.*, 2006).

Sorghum plants known to be very efficient in water use because it has a deep and extensive root system (Dicko *et al.*, 2006). Sorghum requires less water that is 84% compared with the corn needs to produce an equivalent amount of dry matter (Reddy *et al.*, 2007b). Dry land area in Indonesia reached about 148 million ha, of which 102.8 million ha of land is an acid soil (Mulyani *et al.*, 2004). The use of acid dry land for agricultural will significantly increase food production.

Acid dry land is characterized by acidic pH and low soil fertility. At pH under 5.5 the solubility several elements, such as aluminum (Al), is very high and can be toxic to plants. In addition to Al toxicity, another limiting factor in crop production in acid soils is low soil fertility due to nutrient deficiency such as phosphorus (P), calcium (Ca) and magnesium (Mg) (Marschner, 1995). Binding of P by Al causes low availability of P in acid soils and causes P deficiency in plants. Deficiency of P compared to Al toxicity is the main cause of the low productivity of sorghum in acid soils of West Africa (Doumbia *et al.,* 1993).

Plants have developed numerous morphological, physiological, biochemical and molecular responses to cope with growth under Pi-limiting conditions, including changes in root morphology, improved Pi uptake efficiency, and changes in metabolism (Ragothama, 1999; Vance *et al.*, 2003). The development of adaptive varieties is an important factor in low input sustainable agriculture.

Studies on the mechanism and efficiency of nutrient uptake generally use nutrient solution (Blum, 1988), as an experiment in the nutrient solution is more accurate and controlled. Two experiment was done in this study, in nutrient solution and under acid soil condition. The purpose of this experiment was to obtain information on the estimation of genetic variability and heritability for characters of sorghum in F_2 segregating generations at seedling stage and in the field under low P condition. Also to obtain information on the gene actions of the traits related to sorghum adaptation to low P.

Materials and methods

This study consisted of two experiments: 1. study the inheritance of seedling characters of sorghum under low P condition in nutrient solution, 2. study the inheritance of agronomic characters under low P condition in acid soil. The first experiment was conducted in the green house of Bogor Agricultural University from January - Februari 2013 and the second experiment was conducted in acid soils of Bogor District Experimental Field Tenjo, Bogor from July to November 2014. The plant materials used in this experiment were F_2 segregating population derived from the cross between B69 and Numbu.

Estimation of genetic variability for sorghum seedling performance under low P condition

The experimental material comprised of 120 F_2 progenies and both of the parents each are 20 individu. Composition of the nutrient solution used in these experiments followed the method suggested by Ohki (1987), it was 0.24 mM NH_4NO_3; 0.03 mM $(NH_4)2SO_4$; 0.088 mM K_2SO_4; 0.38 mM KNO_3; 1.27 mM $Ca(NO_3)_2.4H_2O$; 0.27 mM $Mg(NO_3)_2$ .4H2O; 0.14 mM NaCl; 6.6 µM H_3BO_3; 5.1 µM $MnSO_4.4H_2O$; 0.61 µM $ZnSO_4.7H_2O$; 0.16 µM $CuSO_4.5H_2O$; 0.1 µM $Na_2Mo_7O_9.7H_2O$; 45 µM $FeSO_4.7H_2O$-EDTA. The concentration of P is given in the form of KH_2PO4: 0.001 mM.

Each pot filled with 2 liters of medium. Water is lost by transpiration is replaced by the addition of distilled water. The pH of the solution was daily monitored and maintained at 4.0±0.1 using HCl or NaOH. One week old seedling was transferred to a nutrient culture medium. Seedling wrapped with soft foam and then inserted into the holes of styrofoam and floated on nutrient solution. Each pot was planted with one seedling. Harvesting is done on day 14 when the plant reaches five-leaf growth stage. Plants were harvested and dried in an oven at a temperature of 60°C for 72 hours and then weighted. The observations were made for shoot length, root length, shoot dry matter (SDM), root dry matter (RDM), total dry matter (TDM), and shoot/root dry weight ratio.

Estimation of genetic variability for agronomy characters sorghum in F_2 population under low P condition in acid soil

The soil in the field is classified as acid soil with pH = 5.3 and P_2O_5 = 7.5 ppm. The parental lines B69 (P_1), Numbu (P_2) and F_2 were evaluated under acid soil conditions. Plant materials for this study consist of 400 individu F_2 generation and both of the parents each are 20 individu. All the agronomic practices such as land preparations, fertilizer application, weeding were done based on recommended practices. Fertilizers were applied as follows : 69 kg/ha of N, 36 kg/ha of P_2O_5 and 60 kg/ha of K_2O. Inter and intra row spacing was 70 cm and 15 cm. The seeds were sown and after emergence thinned to one plant per hole. Data were collected for the following traits: Plant height (cm), Stem diameter (mm), Panicle length (cm), Panicle weight (g), Grain weight (g), and 100 seed weight (g).

Statistical analysis

Analysis of variance and heritability in broad sense was determined according to the formula given by Allard (1960).

Variance $(\sigma^2) = \left(\frac{\sum(x_i - \bar{x})^2}{\mathrm{n}-1}\right)$

Phenotypic variance $(\sigma^2 p) = \sigma^2 F_2$

Environment variance $(\sigma^2 e) = \frac{\sigma^2 P_1 + \sigma^2 P_2}{2}$

Genotypic variance $(\sigma^2 g) = \sigma^2 p - \sigma^2 e$

The estimates of broad-sense heritability (h^2bs) were calculated for each trait using the formula:

$$\mathrm{h}^2 bs = \frac{\sigma^2 g}{\sigma^2 p}$$

Stanfield (1983) classified heritability value as high= $h^2 \geq 0.50$, moderate = $0.20 \geq h^2 \geq 0.50$, and low= $h^2 < 0.20$.

The genotypic coefficients of variation (GCV) was computed according to formula suggested by Knight (1979).

$$\mathrm{GCV} = \frac{\sqrt{\sigma^2 g}}{\bar{\mathrm{x}}} \times 100\ \%$$

$\sigma^2 g$ = genotypic variance and x = mean value of population.

Criteria: Low (0-10%), moderate (10-20%) and high (> 20%).

Skweness was estimated using formula :

Skweness = $\frac{\sum_{i=1}^{N}(Yi-Y)^3}{(N-1)s^3}$

Kurtosis was estimated using formula :

Kurtosis = $\frac{\sum_{i=1}^{N}(Yi-Y)^4}{(N-1)s^4}$

Yi = value of genotype-i, s = standart deviation, N = Number of data collected

The stastitical tests of significance of skweness and kurtosis was perform by comparing the skweness and kurtosis with their standard error. The value of skweness, kurtosis and standard error was computed using SPSS 15.0.

Results and discussion

Genetic variability for sorghum seedling performance under low P condition

Mean value of F2, B69 and Numbu genotypes at seedling stage under low P condition are presented at Table 1.

Table 1. Mean performance and genetic parameters of seedling characters in F_2 population under low P condition in nutrient culture.

Characters	B69	Numbu	F_2 mean	F_2 range	h^2bs (%)	GCV (%)
Shoot length (cm)	38.67	42.10	49.22	32.00 – 65.00	23.16	5.80
Root length (cm)	31.63	32.97	44.13	20.00 – 65.00	30.77	10.70
Shoot dry weight (g)	0.080	0.097	0.125	0.068 – 0.192	58.33	14.97
Root dry weight (g)	0.029	0.040	0.053	0.033 – 0.079	50	11.93
Total dry weight (g)	0.109	0.138	0.178	0.101 – 0.252	60	13.76
Shoot/root ratio	2.779	2.441	2.342	1.448 – 3.245	0	0

The results showed that mean value of Numbu higher than B69 for shoot dry weight, root dry weight, total dry weight and shoot/root dry weight ratio, but not statistically significant for shoot length and root length. Tasliah *et al.* (2011) reported that phosphorus experiment in upland rice gave small effect on root length, but Al toxicity significantly reduced root length. Size and root system can be used as criteria for phosphorus deficiency tolerance (Wissuwa, 2003), and high root volume or root dry weight indicate P deficiency tolerant (Tasliah *et. al.*, 2011). Yaseen and Hussain (2000) reported that most of the genotypes adapted to P stress conditions increases their root growth, which in turn increase the above ground plant growth.

In this study, mean value of F_2 population was higher than the parental lines for most of the characters except shoot /root dry weight ratio. It indicates that there was possibility to obtain individu with better performance which indicated by higher mean value compared to parental lines.

The heritability and genetic coefficient of variance (GCV) are presented at Table 1. The heritability values were high for shoot dry weight and total dry weight. Moderate values of heritability were observed for shoot length, root length and root dry weight. It indicates that the selection for adaptation to low P condition will be effective for shoot dry weight and total dry weight.

Moderate GCV values were recorded for root length, shoot dry weight, root dry weight and total dry weight. Low GCV values were recorded for shoot length and shoot/root dry weight ratio. A high estimate of heritability together with moderate GCV was recorded for shoot dry weight and total dry weight. It indicates that shoot dry weight and total dry weight can be used for criteria selection in low P tolerant at seedling stage.

Frequency distribution and gene actions of seedling characters under low P condition

The skweness analysis of F_2 distribution provides information about nature of gene action (Fisher *et al.*, 1932), whereas kurtosis provides information on the number of genes controlling the traits (Robson, 1956). For a normal distribution, skweness is equal to zero in the absence of gene interaction. The positive skweness is assosiated with complementary gene interactions while negative skweness is assosiated with duplicate gene interactions (Roy, 2000).

Skweness describes the degree of departure of a distribution from symmetry and kurtosis characterizes the peakedness of a distribution. The traits with leptokurtic distribution are usually under the control of few segregating genes and the traits with platykurtic distribution is controlled by many genes. The positive value of kurtosis indicated leptokurtic curve and negative kurtosis indicated platykurtic curve (Jayaramachandran *et al.*, 2010).

Frequency distribution for sorghum characters in seedling stage are presented in Fig. 1. The characters observed showed continue distribution, indicating they are poligenic traits. The skweness and kurtosis values are presented in Table 2.

Table 2. Skewness and kurtosis value of seedling characters in F_2 population under low P condition in nutrient culture.

Character	Skewness	Gene action	Kurtosis	No. Of genes
Shoot length (cm)	0.200 ^{ns}	Aditif	-0.09 ^{ns}	Many
Root length (cm)	-0.312 ^{ns}	Aditif	0.30 ^{ns}	Many
Shoot dry weight (g)	0.452 *	Aditif + CE	-0.23 ^{ns}	Many
Root dry weight (g)	0.21 ^{ns}	Aditif	-0.29 ^{ns}	Many
Total dry weight (g)	0.26 ^{ns}	Aditif	0.57 ^{ns}	Many
Shoot/root ratio	0.38 ^{ns}	Aditif	-0.54 ^{ns}	Many

Note: * = significant at P>0.05 and ns = not statistically significant.
CE = complementary epistasis.

The results showed that most of the characters observed in this study had non significant of skweness and non significant of kurtosis, indicating the absence of gene interaction. These traits were controlled by additive gene action and suggested the involvement of many number of segregating genes. On the other hand, shoot dry weight had positive significant of skweness, indicating the present of complementary epistasis. It means the trait is controlled not only by several genes with individual effects (additive effects) but also by interactions between genes (epistasis). Complementary gene interaction can occur when two or more genes code for enzymes that function at different point in the same pathway, so that functioning products from all genes in the set are needed to produce the final product (Holland, 2001).

Genetic variability for agronomy characters sorghum in F_2 population under low P condition in acid soil

The mean values recorded for agronomic characters in F_2, B69 (P_1) and Numbu (P_2) genotypes were presented in Table 3. Mean performance of F_2 for plant height, panicle weight, grain yield per plant and 100 seed weight were found to be intermediate between parental values. The F_2 showed a lower mean value for stem diameter when compared to parents, but it showed a wide range from 4.64 to 24.56. The mean value for panicle length in F_2 was higher than parents. Based on the range of F_2 population, indicating there was transgressive segregants in F_2 population. According to Rieseberg *et al.* (1999), transgressive segregation is the production of F_2 or later generation hybrid progeny with phenotypes that can fall outside the phenotypic range of the parental populations from which they were derived.

The criteria to be considered while evaluating a population is the magnitude of genetic variation and heritability of the characters. The estimates of heritability and genotypic coefficient of variation (GCV) are presented in Table 3.

Table 3. The value of mean, range, broad sense heritability (h^2_{bs}) and genotypic coefficient of variation (GCV) of agronomic characters of F_2 population under low P condition in acid soil.

Character	B69	Numbu	F_2 mean	F_2 range	h^2bs (%)	GCV (%)
Plant height (cm)	163.33	234.53	206.09	124.0 – 287.0	58.82	9.64
Stem diameter (mm)	14.83	17.07	13.45	4.64 – 24.56	75.33	22.30
Panicle lenght (cm)	19.00	18.07	19.87	10.50 – 27.00	88.49	13.56
Panicle weight (g)	39.31	76.54	60.64	10.28 – 143.07	82.10	36.07
Grain yield / plant (g)	31.20	62.75	49.47	8.18 – 120.23	79.61	35.67
100 seed weight (g)	2.54	3.17	2.72	1.50 – 3.64	65.16	11.68

The value of GCV were high for stem diameter, panicle weight and grain yield per plant. Moderate values of GCV were observed for panicle length and 100 seed weight. Low GCV observed for plant height. The extent of genetic variability gives the better chance in plant breeding through selection (Yunianti *et al.*, 2010).

High heritability estimates were observed for most of the traits except plant height that showed moderate value. The high heritability indicating the possibility of positive response to selection. It likelihood of transferring the heritable components from parents to offspring during breeding process. The effectiveness of selections for any character depends on the extent of genetic variability and the extent to which the characters can be transferable from one generation to the next (Sami *et al.*, 2013).

The results from this study showed that stem diameter, panicle weight and grain yield per plant under low P condition in acid soil had high heritability and wide GCV value. Therefore these characters can be used as the selection criteria in selecting sorghum genotypes for low P tolerant in acid soil.

Frequency distributions and gene actions in F_2 population under low P condition in acid soil

Frequency distribution for agronomic characters under low P in F_2 population showed continues distribution (Fig. 2). It indicates that these characters were controlled by many genes (polygenic). The information on frequency distributions can be used to identify transgressive segregants and to identify gene actions for the quantitative traits. The gene action can be estimated through skewness and kurtosis. Roy (2000) indicated that by observing the skweness of frequency distribution of F_2 generation plants, one can determined the gene action.

The skewness and kurtosis values for agronomic characters are presented in Table 4.

Table 4. Skewness and kurtosis values of agronomic characters of F_2 population under low P condition in acid soil.

Character	Skewness	Gene action	Kurtosis	No. of genes
Plant height (cm)	-0.047 ^{ns}	Aditif	0.004 ^{ns}	Many
Stem diameter (mm)	-0.132 ^{ns}	Aditif	-0.418 ^{ns}	Many
Panicle lenght (cm)	-0.215 ^{ns}	Aditif	-0.073 ^{ns}	Many
Panicle weight (g)	0.202 ^{ns}	Aditif	-0.234 ^{ns}	Many
Grain yield per plant (g)	0.221 ^{ns}	Aditif	-0.231 ^{ns}	Many
100 seed weight (g)	-0.184 ^{ns}	Aditif	-0.058 ^{ns}	Many

Note: ns = not statistically significant.

Based on the skewness values, all of the agronomy characters in this study were influenced by additive gene action. The skewness value was not significantly different from zero, indicating that epistasis appears to be absent for the characters. Similar result has been reported by Mary *et al.* (2015) which showed that heading date, number of leaves, plant height and 100 grain weight in sorghum are controlled by additive gene action. None of the kurtosis value in F_2 population was different from zero. It means that all of the characters are controlled by many genes. Eventhough it is controlled by many genes, but there is no epistasis effect. It indicates that loci affecting the characters are not interacting.

Jayaramachandran *et al.* (2010) reported that in segregating generations, skewness could result when certain combinations of genes are lethal, presence of incomplete linkage of certain genes, presence of epistasis and one gene has a much larger effect than others. Kurtosis will occur if either of few genes are contributing to the phenotypic distributions or there are inequalities in the additive genetic effect at different loci.

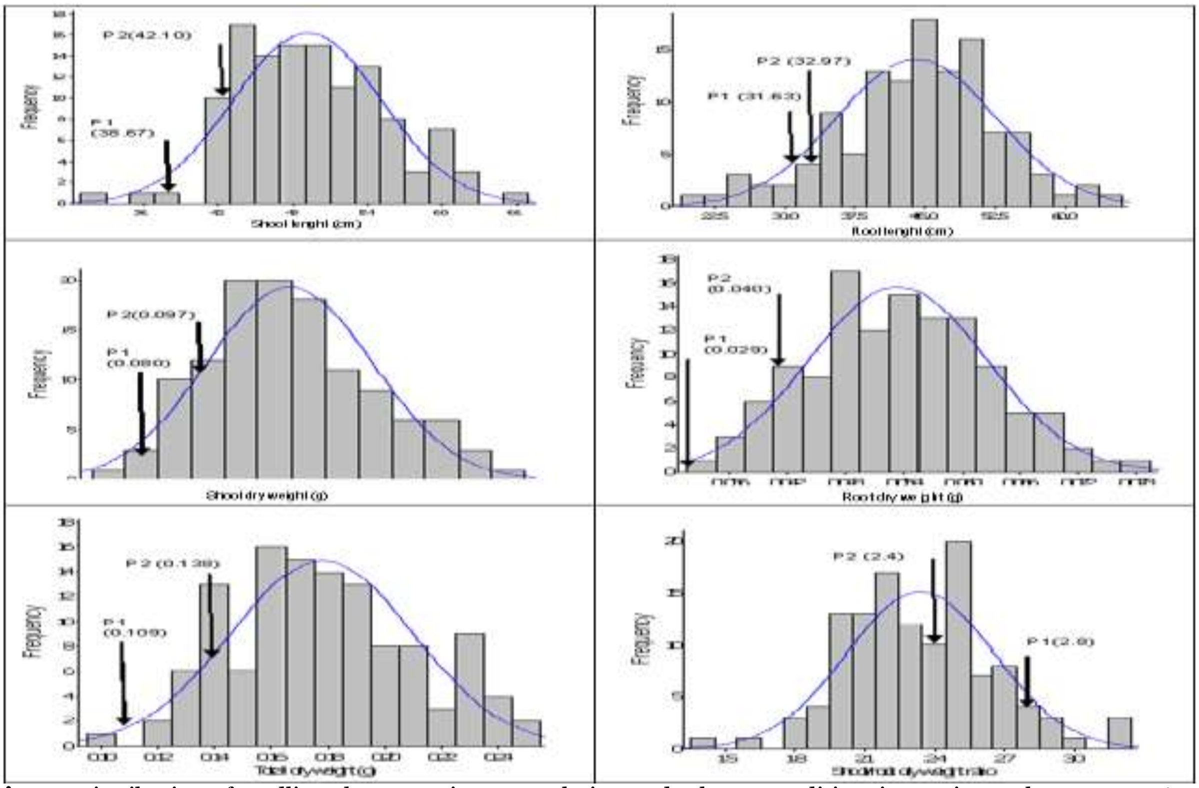

Fig. 1. Distribution of seedling characters in F_2 population under low P condition. in nutrient culture. P1=B69, P2=Numbu.

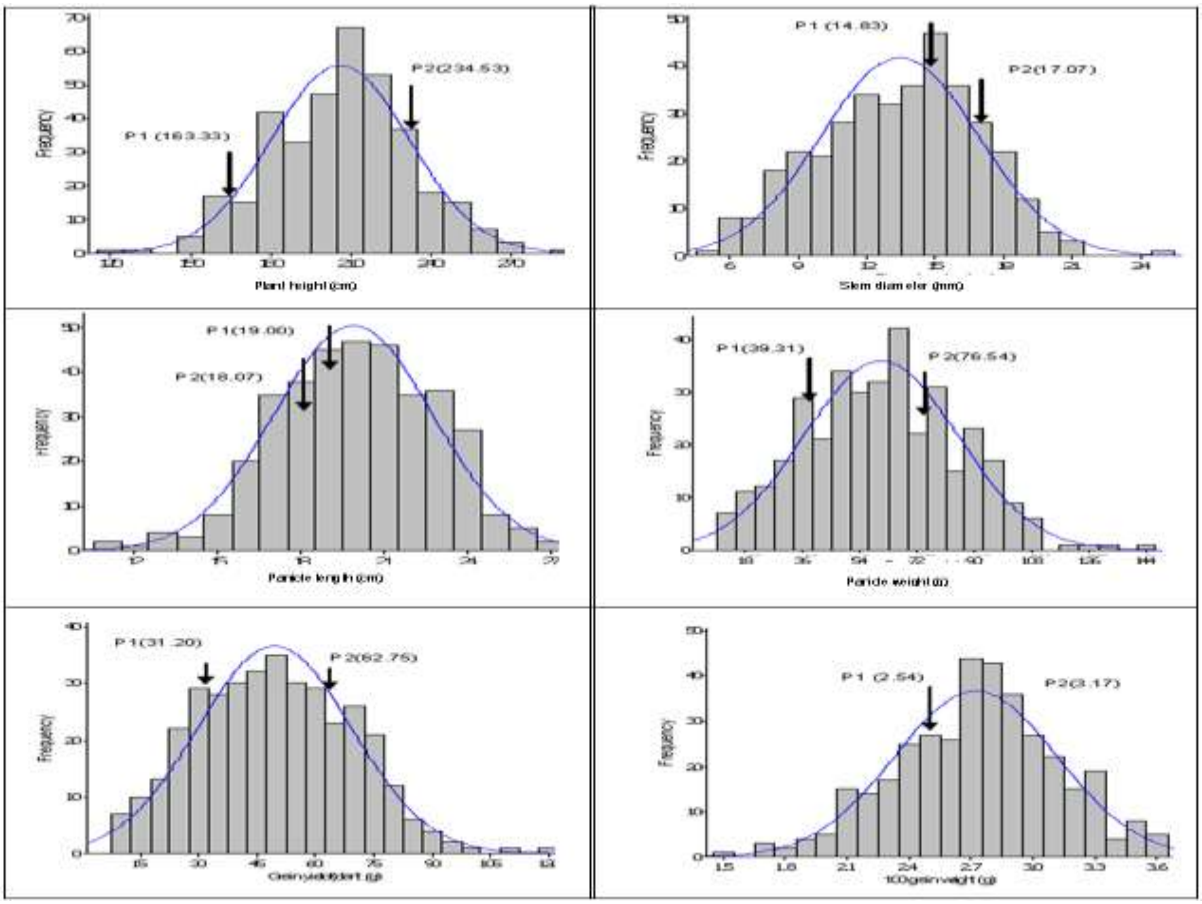

Fig. 2. Distribution of agronomic traits in F_2 population under low P condition in acidsoil. P1=B69, P2=Numbu.

Conclusion

Seedling characters of sorghum such as shoot length, root length, root dry weight, total dry weight and shoot/root dry weight ratio were controlled by additive gene action, whereas shoot dry weight is influenced by complementary epistasis gene action. Shoot dry weight and total dry weight can be used for selection criteria in low P condition at seedling stage because of high heritability and moderate GCV. Agronomy characters of sorghum in acid soil such as plant height, stem diameter, panicle length, panicle weight, grain yield per plant and 100 seed weight were controlled by additive gene action. Stem diameter, panicle weight and grain yield per plant had high broad sense heritability and wide GCV, thus they can be used as the selection criteria in selecting sorghum genotypes for low P tolerant in acid soil.

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