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Secondary trait and index selection determination for maize genotype selection in acidic tidal swamp environment

SUWARTI[1,3,♥], MUNIF GHULAMAHDI[2,♥♥], DIDY SOPANDIE[2], TRIKOESOEMANINGTYAS[2], EKO SULISTYONO[2], MUHAMMAD AZRAI[3]

[1]Graduate School of Institut Pertanian Bogor. Jl. Raya Dramaga Kampus IPB Dramaga Bogor 16680, West Java, Indonesia. Tel./fax.: +62-251-8622642, ♥email: suwarti_agh@apps.ipb.ac.id

[2]Department of Agronomy and Horticulture, Faculty of Agriculture, Institut Pertanian Bogor. Jl. Meranti Kampus IPB Darmaga, Bogor 16680, West Java, Indonesia. Tel.: +62-251-8629354, ♥♥email: mghulamahdi@yahoo.com

[3]Indonesian Cereals Research Institute. Jl. Dr. Ratulangi No. 274, Maros 90514, South Sulawesi, Indonesia

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Abstract. *Suwarti, Ghulamahdi M, Sopandie D, Trikoesoemaningtyas, Sulistyono E, Azrai M. 2022. Secondary trait and index selection determination for maize genotype selection in acidic tidal swamp environment. Biodiversitas 23: 4169-4179.* Determining secondary traits as complementary selection in maize genotypes for abiotic stress environment is compulsory since yield-based selection usually provides biased result. This study aimed to obtain secondary traits for maize selection in an abiotic multi-stresses of Fe, Al, and temporary flooding environments of acid sulfate tidal swamp land using multivariate analysis. A total of 150 (un-repeated) maize inbred lines and five check genotypes in five replicates were arranged in a randomized augmented plot design in three environments to generate 525 single plots. Three types of environments based on irrigation regimes were applied on acid soil tidal swamp land to study the agronomic characteristics of maize genotypes in each environment. Saturated Cultivation Technology (SSC) was considered the optimal environment. Dryland treatment was the condition to let the irrigation only depend on rainwater. Temporary flooding treatment was carried out at five stages of different growth periods. Several steps of analysis were applied in this study by taking variance, and heritability was used as the basis for selections. The characters affected by treatments with medium to high heritability were then analyzed with correlation and pathway analysis. Seven selection indexes, SSI, GMP, YSI, YI, TOL, STI, and MP, that have a high correlation to yield in both optimum and stress environments, were applied simultaneity with the selected trait characters to screen the genotypes utilizing PCA biplot analysis. Venn analysis was used to group the genotypes based on the result of the analysis. Indices of STI, MP, GMP, and traits of rows number per ear and the weight of 6 ears were selected as secondary characters for the maize genotypes selection in Saturated Soil culture with temporary flooding stress. Meanwhile, STI, MP, and ear stand height were the selected characters in the dryland of acid sulfate tidal swamp land. Based on the combination of all secondary character selection, genotypes were grouped into five types of stress tolerance in SSC+TF and DL environments. Eight genotypes were tolerant to both SSC+TF and DL stress, and two genotypes were susceptible to both environments.

Keywords: Acid soil**,** tidal swamp, heritability, pathway analysis, pyrite, temporary flooding

Abbreviations: SSC: Saturated soil culture, TF: Temporary flooding, SSC+TF: Saturated soil culture+temporary flooding, DL: Dry land, SSI: Stress susceptibility index, GMP: Geometric mean productivity, YSI: Yield stability index, YI: Yield index, TOL: tolerance index, STI: Stress tolerance index, MP: Mean productivity

INTRODUCTION

Improvement of maize production through extending crop area has strict obstacles because the convertion of agricultural land to non-agricultural purposes (Maulana et al. 2019). The utilization of suboptimal land such as sulfate acid soil tidal swamp areas to increase national maize production in Indonesia is an opportunity since the land is widely available, about 4.3 million hectares of tidal swampy areas can be managed as crop cultivation area (Surahman et al. 2018). Pyrite oxidation in the soil layer was the main restriction to developing maize crops on the sulfate acid soil tidal swamp to manage the land. Pyrite oxidation would trigger Fe^{3+} release in the soil layer leading to a decreased pH level (Toyip et al. 2019; Annisa et al. 2020). A low pH level in the soil triggers aluminum ion solubility and increases acidity (Pujiwati et al. 2015; 2016). Insoluble iron in the form of Fe^{3+} is found at the aerated soil layer and alkaline pH. In contrast, when soil is flooded and has an anaerobic condition or soil pH is decreased, there is a reduction of Fe^{3+} to Fe^{2+}, which is soluble and dangerous to the plant (Vahedian et al. 2014; dos Santos et al. 2017).

The toxicity of aluminum and iron to plant tissue has been studied previously in several experiments. In addition to damaging the plant tissues, the main risk of heavy metal stresses of Al and Fe is decreased crop yields. Low pH soil in acid soils increases the concentrations of Fe and Al in the solution, leading to the precipitation of inorganic phosphorus as iron and aluminium phosphates. The excess condition of Fe becomes toxic to the plant as a highly reactive Fenton catalyst. The formation of ROS such as hydroxyl radicals (OH) and superoxide anion radicals (O_2^-) is easily generated under Fe toxicity in the plant tissue

(Nikolic and Pavlovic 2018). Easy observed Fe excess stress symptoms in plant tissue were the appearance of bronzing color, starting from tip to basal of the leaf (Nugraha et al. 2016). There are two types of Fe uptake in the plant; strategy I for dicot and strategy II for graminea. Maize plant Fe uptake followed chelation-based Fe uptake by the so-called phytosiderophores strategy. Those plant hormones are secreted to the rhizosphere to bind Fe to become the Fe-phytosiderophores complex that enters the cell via the root ZmYS1 (*Yellow stripe1*) and YSL *(Yellow Stripe Like)* transporter. Chelating compounds phytosiderophores belong to the mugeneic acids family, released as Fe deficiency response (Bartucca et al. 2018).

Maize crop productivity is negatively affected by aluminum presence in acid soil. The toxicity of aluminium primarily affects plant root growth, thus, reducing the ability to exploit the soil water and nutrients (Kopittke et al. 2015; Matonyei et al. 2020). Furthermore, the condition of low pH soil releases toxic forms of Al, such as $Al(OH)^{2+}$, $Al(OH)^{2+}$, and Al^{3+} into the soil solution (Xu et al. 2017). Plant mechanisms of aluminum tolerance could be summarized into two types: Al exclusion and detoxification. The exclusion mechanism is carried out to limit Al absorbance or reduce the root's uptake. The detoxification mechanisms were usually conducted by Al complexation, followed by transfer and storage of these complexes in vacuoles (Bian et al. 2018).

Development of maize variety tolerant to several abiotic stress troughs a breeding program for sulfate acid tidal swamp area environment is necessary along with the development of proper agronomic management (Sabagh et al. 2018). The breeding program for abiotic stress tolerant genotype usually starts with selecting tolerant genotypes for the target environment. Grain yield is usually chosen as the main character in the selection of maize tolerant to abiotic stress. However, using grain yield as solely character usually generate a biased result. Secondary traits and index selection could complement the yielding character as the plant tolerance to environmental stress measurement (Fadhli et al. 2020). Multivariate analysis to obtain the selection index value will simplify the characters in selecting genotypes tolerant to specific environments. Furthermore, a selection index based on the calculation of several formulas simultaneity can be used to strengthen the applied characters used in a selection in the abiotic stress environment (Anshori et al. 2019). This study aimed to obtain secondary characteristics for maize selection genotypes in acid sulfate tidal soil under Fe, Al, and temporary flooding stress on the Saturated Soil Culture crops technique and dryland environment.

MATERIALS AND METHODS

Study area

The study was carried out in the sulfate acid tidal swamp area of Karya Bhakti Village, Rantau Rasau District, Tanjung Jabung Timur Jambi Province Indonesia, from May to September 2019. The coordinates location of SSC treatment was -1°10'50"S 104°09'44"E, SSC+TF was -1°10'51"S 104°09'44"E, and DL was -1°10'50"S 104°09'37"E. The results of soil analysis at SSC, SSC+TF, and DL locations, respectively, showed pyrite levels of 400 ppm; 100 ppm, and 100 ppm, Fe levels were 45200 ppm; 35600 ppm, and 37300 ppm (very high), Sulphur levels of 200 ppm, 500 ppm and 100 ppm, Al^{3+} levels 52900 ppm, 47600 ppm and 46200 ppm (very high), soil pH H_2O 5.10, 5.20 and 5.2 (acidic), pH KCl 3.6, 3.7 and 3.6. Organic matter C was 7.39 and 11.23 (very high), and 1.28 (low). Total N 0.21%, 0.31% (moderate), and 0.09%. C/N ratio 35, 36 (very high) and 14 (medium). Available P_2O was 12 (medium), 8, and 8 (low). Cation exchange capacities were 17.2 $cmol.g^{-1}$ and 23.72 $cmol.g^{-1}$ (medium) and 8.97 $cmol.g^{-1}$ (low). Climatic data from January to June 2019 obtained from the Meteorology, Climatology and Geophysics Agency, Muaro Jambi Climatology Station, shows an average monthly temperature was 22.71°C - 33.65°C, relative humidity 77.22-85.77%, rainfall 42.70-191.70 mm per month.

Procedures

Design of field experiment

The experiment was arranged in an augmented randomized block design. Genetic materials consisted of 150 maize lines collected by the Indonesian Cereals Research Institute and five check genotypes. Check genotypes consisted of one commercial composite variety (Sukmaraga) as check number 1, two elite lines (MR-14 and Nei 9008) as check number 2 and check number 3, one commercial hybrid variety (P27) as check number 4, and one commercial three-way cross-hybrid variety (Bima 20) as check number 5. Maize seeds were planted at a depth of ±5 cm, the distance between rows was 70 cm, and between plants spacing was 20 cm; to gain a population per hectare of 71428 plants. Each plot is comprised of three rows of plants, with the row length being 4.2 m. Each block consisted of 30 test plots of genotypes and five plots of check genotypes.

Arrangement of soil irrigation in each treatment

There were three soil irrigation system treatments applied in this experiment. The treatments were Saturated Soil Culture (SSC), SSC+Temporary Flooding, and Dry Land (DL) environment. The saturated Soil Culture technique manages the water level to adjust 5 cm above the base of the 30 cm wide and 25 cm depth trench, the length of the trench following the length of the land perpendicular to the primary trench. This SSC technic purpose is to manage soil consistently in redox conditions. Floodgate was a tube applied to flow and retain water in the field at low tide and discharge water out of the field during high tides so that it also functions to clean the pyrite layer at the bottom of the trench.

The SSC+Temporary Flooding (SSC+TF) treatment was the SSC technique. Still, it allowed the water to overflow until it reached 10 cm above the root area, arranged periodically to allow roots in the anoxia condition. Temporary flooding in SSC+TF period occurred after planting (0 to 4 DAP); TF at 14-17 DAP; TF at 28-31 DAP; TF at 43-46 DAP (anthesis and tassel periods), and

TF at 58-61 DAP (R1/grain filling periods) (Figure 1). The water pump was used conditionally to regulate water flow if swamp water overflow was inadequate artificially. In addition, a 50 cm high embankment covered with tarpaulin was made around the land to prevent water leakage inside and outside the experimental area. The third environment treatment was dry land (DL), where soil watering in this experiment was dependent on rainwater.

Tolerance index formulas

The estimated heritability value (h^2) was calculated using the heritability equation in the broad sense, which is derived from the variance in the broad sense $h^2 = \frac{\sigma^2 g}{\sigma^2 p} x100\%$. In a broad sense, heritability values are expressed by decimal numbers ranging from 0 to 1 or percentages; the criteria for heritability values are classified by category; high $(h^2 > 50\%)$, medium $(20\% < h^2 < 50\%$ and low $h^2 < 20\%$. Finally, stepwise regression was applied to determine the most important character with a high correlation with the yield.

Observed yield variables were used to calculate the tolerance indices. The tolerance index used in this experiment was; *Stress susceptibility index* $(SSI) = \frac{1-(Yp/Yo)}{1-(Xp/Xo)}$, with tolerant criteria if S<0,5; moderate if the value is 0.5<S<1; Sensitive if the value of S>1, *Geometric mean productivity* $(GMP) = \sqrt{YpxYo}$; *Yield stability index* $(YSI) = \frac{Yp}{Yo}$; *Yield index* $YI = \frac{Yp}{Xp}$; *Tolerance Index* $(TOL) = Yo - Yp$; *Stress Tolerance Index* $STI = Yox\frac{Yp}{Yo^2}$; *Mean Productivity* $(MP) = \frac{(Yo+Yp)}{2}$. Description: Yp: Value of certain variables in varieties under stress; Yo: The value of a certain variable in the varieties that were not stressed; Xp: the average value of certain variables in all varieties under stress; Xo: The average value of a certain variable in all varieties that are not stressed.

Data analysis

All the collected data were analyzed over several stages. First, data were analyzed to obtain variance with a standard error of 5% at the initial process. Characters significantly affected by genetic variance and have high heritability values (>50%) were then reselected through correlation analysis and path analysis to determine the most important characters on the yielding character and directly affect the yield. Seven tolerance indices were connected to yield character by correlation analysis. The selected characters were then analyzed using PCA (Principal Component Analysis). Finally, Venn Diagram by Heberle et al. (2015) was applied to separate all the tested genotypes into tolerance groups in environmental stress trials.

RESULTS AND DISCUSSION

Genotypic variance, phenotypic variance, and heritability

The mean square of agronomic character data is presented in Table 1. Genotype factor was highly affected by yield character both in SSC+TF and SSC environments, while the DL environment showed significantly affected. Due to high variance, some data were being transformed at log (x+1) or at $(0.5+x)^{0.5}$. Most of the characters on SSC treatment were highly affected by genotypic factors. Senescence, leaf length, leaf width, anthesis days, silking days, and weight of six stems were the characteristics that were not affected by genotypic factors among all 23 observed traits. In the SSC+TF environment, genotypic factors did not affect the number of leaves and leaf length. The dry land treatment generated most characters that were not affected by genotypic factors compared to the other treatments. Those characters in dry land were a weight of 6 cobs, the weight of 1 cob, number of harvested plants, number of harvested cobs, plant aspect, senescence, ear diameter, stem diameter, and weight of 6 dry stems.

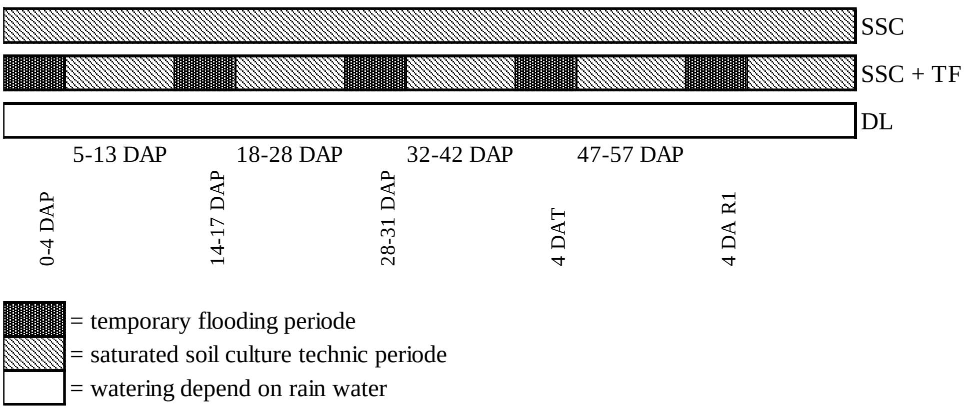


Figure 1. Irrigation treatment scheme on SSC technique and the temporary flooding period in the experimental environment

Table 1. Mean square of genotype factor at each character on environment treatments of SSC, SSC+TF, and DL

Source of variance	MSg SSC	CV	MSg SSC+TF	CV	MSg DL	CV
Yield	0.15**	30.59t2	0.03**	15.68t2	0.10*	22.01t2
Weight 6 cobs	0.07**	8.88t2	0.99*	11.73t2	0.01ns	12.71t2
Weight of 1 cob	0.10**	22.85	0.12**	22.19	0.02ns	2.71t2
Number of harvested plants	0.07**	10.59t1	0.99*	32.78t2	0.01ns	11.95t1
Number of harvested cobs	0.10**	13.55t1	0.12**	17.76t1	0.02ns	16.29t1
Kernels/Cob weight Ratio	7.18**	13.07	21.06**	7.89	0.04**	12.80t2
Weight of 1000 seed	2920.57*	23.37	0.02*	5.36t1	0.01**	3.97t1
Plant aspect	0.43**	16.38	0.02**	14.96t1	0.00ns	14.85t1
Senescence	0.02ns	22.98	0.03**	11.75t1	0.01ns	12.86t1
Ear length	9.03**	16.82	6.37**	21.22	0.01*	8.81t1
Ear diameter	0.43**	10.69	0.40*	21.06	0.25ns	17.92
Number of rows per cob	5.03**	10.69	6.14**	21.06	2.74**	17.92
Number of seeds per row	40.86**	23.07	0.07**	8.07t1	0.02**	8.77t1
Number of leaves	0.01*	11.93t1	1.82ns	24.68	2.57**	13.92
Height of cob	125.10**	19.59	0.03*	11.62t1	237.59**	20.01
Plant height	524.82**	14.47	468.22**	19.93	600.18**	13.16
Leaf length	101.09ns	21.31	0.01ns	6.86	84.13*	11.04
Leaf width	0.01ns	11.36t1	0.01*	12.50t1	1.11*	13.18
Stem diameter	6.51*	18.21	6.28*	22.87	0.01ns	9.91t1
SPAD	39.80**	12.29	43.49*	20.14	23.20*	11.04
Anthesis day	11.54ns	7.21	13.76**	4.91	9.21**	2.50
Silking day	16.03ns	6.58	19.12**	4.99	9.88**	2.54
Weight of 6 dry stem	0.01ns	10.91t2	0.01**	5.65t2	0.01ns	10.18t2

Note: **significant effect on 1% level, *significant effect on 5% level, ns non-significant, t1 transformed at $\log(1+x)$; t2 transformed at $\sqrt{(0.5+x}$; CV: Coefficient of Variance; MS: Mean square of genotypes

Table 2. Agronomic characters of 150 maize lines under three environments types on acid soil tidal swamp, Jambi 2019

Agronomic Traits	SSC			SSC+TF			DL		
	σ^2p	σ^2g	h^2	σ^2p	σ^2g	h^2	σ^2p	σ^2g	h^2
Yield	0.21	0.35	59.81	-0.12	0.61	-19.23	0.10	0.49	20.40
Weight 6 cobs	14.64	14.65	99.95	8.51	8.52	99.96	2.52	2.54	99.18
Weight of 1 cob	17.62	17.62	100.00	6.98	6.98	100.00	5.57	5.57	99.99
Number of harvested plants	9.32	35.95	25.92	3.86	27.14	14.21	0.19	11.85	1.58
Number of harvested cobs	10.57	45.83	23.06	4.92	15.26	32.22	-0.92	31.52	-2.92
Kernels/Cob Weight Ratio	-34.04	150.50	-22.62	4.21	4.23	99.52	0.07	0.12	61.57
Weight of 1000 seed	218.23	2168.13	10.07	603.25	3458.62	17.44	228.26	2074.85	11.00
Plant aspect	0.05	0.21	24.80	0.07	0.40	18.33	0.02	0.22	8.57
Senescence	0.45	2.51	17.95	0.55	2.07	26.39	0.01	2.30	0.43
Ear length	0.87	5.20	16.76	0.96	2.54	37.80	0.41	3.37	12.03
Ear diameter	0.07	0.13	56.83	0.05	0.20	23.86	-0.48	2.40	-20.04
Number of Rows per ear	0.67	2.37	28.37	1.12	1.64	68.55	0.59	2.33	25.26
Seeds number per Row	4.64	22.43	20.70	6.34	14.14	44.83	2.68	12.87	20.83
Number of Leaves	0.46	2.03	22.83	0.18	1.11	16.14	0.37	1.15	31.94
Height of Cobs	20.86	41.66	50.08	10.23	52.65	19.42	0.22	0.41	52.49
Plant height	86.56	178.59	48.46	50.78	130.67	38.87	93.22	227.32	41.01
Leaf length	6.71	74.26	9.03	0.00	0.01	4.12	9.11	53.10	17.16
Leaf width	0.11	1.28	8.74	0.10	0.85	11.27	0.14	0.57	24.13
Stem diameter	0.79	3.34	23.80	0.70	3.47	20.28	-0.22	9.21	-2.41
SPAD	5.62	17.31	32.49	5.02	23.39	21.48	2.56	12.88	19.87
Anthesis day	0.54	9.39	5.73	1.85	6.37	28.98	1.65	2.61	63.08
Silking day	1.60	9.61	16.69	2.78	8.02	34.62	1.76	2.84	62.09
Weight of 6 dry stems	0.00	0.00	33.50	0.00	0.01	47.01	0.00	0.01	7.81

Notes: σ^2p: Phenotypic variance, σ^2g: Genotypic variance, h^2: Heritability, SSC: Saturated soil culture, DL: Dry land, SSC+TF: Saturated soil culture+temporary flooding

Other characters that were highly significantly affected by genotype and had moderate to high heritability values in the SSC+TF environment were the weight of 6 ears, the weight of 1 ear, number of harvested cobs, the ratio of shelled grain to cob, senescence score, ear diameter, ear length, rows number of kernels, number of seeds per row, plant height at 10 WAP, stem diameter at 8 WAP, SPAD value, anthesis day, silking days, and weight of 6 dry stems (Table 1 and Table 2).

A summary of the phenotypic variance, genotypic variance, and heritability observed characters are shown in Table 2. Characters with high heritability values can be used as selection criteria to increase the effectiveness of selection. Maize plants have different responses to the environment, so the characters utilized to increase the effectiveness of selection also vary, depending on the selection environment. The highest heritability on yield character was obtained in the SSC environment (scored 59.81%), followed by the DL environment, which scored 20.40% and was categorized as a medium category, and at the lowest was the SSC+TF environment, it had a negative heritability value (-19.23) which means there was no genetic progress and can be considered as 0 (zero). Low heritability indicates that the character is more affected by the environment rather than genotypic nature (Wening et al. 2020).

A high broad sense heritability value in the SSC environment (>50%) was obtained on the weight of 6 ears, the weight of 1 ear, ear diameter, and ear height position. Medium heritability values (between 20%-50%) were obtained on the number of harvested plants, the number of harvested cobs, plant aspect, seeds number per row, seeds rows number, plant height, number of leaves, stem diameter, SPAD value, and weight of 6 dry stems. High heritability in the SSC+TF environment was shown at characters of the weight of 6 cobs, the weight of 1 ear kernels/cob ratio, and the number of rows per ear. The number of harvested cobs, senescence, ear length, ear diameter, seeds number per row, plant height, stem diameter, SPAD value, anthesis day, silking day, and weight of 6 dry stems were categorized as a medium heritability. The high-category heritability values of agronomic characters in DL environment treatment were obtained on the weight of 6 cobs, the ratio of the kernel to cob, anthesis days, and silking days. Medium heritability values were obtained on the number of rows per ear, seeds number per row, number of leaves, plant height, and leaf width. The heritability of each maize character is influenced by additive or non-additive gene action and expressed at the phenotype performance (Sesay et al. 2016).

Correlation of important character to the yield

A stepwise regression analysis was conducted to determine the essential/dominant characters related to the main character (Ayvat and Omeroglu 2022). The generated model by stepwise analysis was considered more precise in predicting the results based on the resulting model than the correlation involving all observation variables (Andayani et al. 2016). This study found that, in the SSC environment weight of 6 ears (X6e), the weight of 1 ear (X1e), and the height of the ear stand (XeSt) were the dominant characteristics that affect maize grain yield (Table 3). Therefore, the SSC treatment in acid soil tidal swamp is considered an optimum crop technic in the tidal swamp.

The stepwise regression correlation analysis on all maize characters observed in the SSC+TF environment showed that four traits had a significant effect, with an R^2 value of 70.00% and a model F value of 87.52 significantly different. These characters were the weight of 6 ears (X6E), the weight of 1 ear (X1E), kernels per cob weight ratio (XKpC), and the number of rows per ear (XNR) (Table 4). The equation model formed from this analysis were Y(SSC+TF)= -0.09 + 1.63 X6E + 0.71 X1E - 0.01 XKpC + 0.01 XNR. Meanwhile, the stepwise regression correlation results on dry land treatment showed three characters (weight of 6 ears, the height of ear stand, and anthesis day) that were dominant to affected yield. Equation model formed in dry land treatment was Yield DL= 4.26 - 0.30 XKpC + 0.02 XESt - 0.07 XAD. The R^2 value explained from this model is 40.81%, with a model F value was 34.7, which is highly significant (Table 5).

Table 3. The most important trait correlation based on stepwise regression analysis of SSC tidal swamp treatment

Characters	YieldSSC	(X6e)	(X1e)
YieldSSC	1		
Weight of 6 ears (X6e)	0.75**	1	
Weight of 1 ear (X1e)	0.44**	0.34**	1
Height of ear stand (XeSt)	0.51**	0.53**	0.26**

Notes: **significant at 1% level, *significant at 5% level

Table 4. The most important trait correlation based on stepwise regression analysis on SSC+GS tidal swamp treatment

Characters	YieldSSC+GS	(X6e)	(X1e)	(XKpC)
YieldSSC+GS	1			
Weight of 6 ears (X6E)	0.82**	1		
Weight of 1 ear (X1E)	0.1ns	0.05ns	1	
Kernels/cob weight Ratio (XKpC)	-0.23**	-0.17*	0.31**	1
Number of rows per ear (XNR)	0.52**	0.47**	0.17*	-0.2*

Notes: **significant at 1% level, *significant at 5% level

Table 5. The most important trait correlation based on stepwise regression analysis on DL tidal swamp treatment

Characters	YieldDL	(XKpC)	(XeSt)
YieldDL	1		
Kernels/cob weight Ratio (XKpC)	-0.31**	1	
Height of ear stand (XeSt)	0.53**	-0.12ns	1
Anthesis day (XAD)	-0.32**	0.03ns	-0.11Ns

Notes: **significant at 1% level, *significant at 5% level

Each environment shows a different important selected character with a high correlation to yield. Two characters selected in optimum treatment (SSC) were similar to those found in SSC+TF. The result was understandable though both environments lay in the same area. Height ear stands as the dominant character that affected grain yield in stress DL environment was found in SSC optimum environment likewise. Kernels per cob ratio in SSC+TF is also shown as the dominant character in the DL environment. The high correlation between variables shows a close relationship and can be an indirect selection character for quantitative traits (Hamawaki et al. 2017).

Direct and indirect effects of agronomic traits on the yield characters

The pathway analysis results on the Saturated Water Cultivation (SSC) treatment showed a residual value of 0.39, meaning that the path model could explain 61% of the variance. The most considerable direct effect was obtained on the weight of 6 ears, which has a direct effect coefficient of 0.61, weight of 1 ear get 0.20, and the height of ear stand get 0.13 score the direct effect. The pathway analysis of characters in the SSC+TF environment showed a residue of 0.30, meaning that 70% of the yield characters could be explained by the four characters used in the test. The character weight of 6 ears (0.73), the weight of 1 ear (0.7), and the number of rows per ear (0.14) showed a positive direct effect coefficient on the yield. However, the kernels per cob ratio have a negative direct effect (-10). Characters of kernels per cob ratio and anthesis day had a negative direct effect on grain yield, with values of -0.25 and -0.26, respectively.

In contrast, the height of the ear stand has a positive direct effect (0.47) in the DL environment. The residual effect in this environment stress treatment was 0.59, meaning that 41% of the path model could explain the yield character. The positive direct characters to yield in path analysis indicated that increasing maize grain yield was supported by increasing the character value in those environments. A negative direct effect demonstrates that the yield response is more influenced by the indirect effect (Putri and Ashari 2019). Path analysis enables breeders to improve understanding of the relationships between several traits by revealing the correlation coefficients into direct and indirect effects (Machado et al. 2017).

Correlation of selection index to yield characters

A total of seven stress tolerance indices (GMP, YSI, SSI, YI, TOL, STI, and MP) were used to determine the appropriate index for maize genotype selection in tidal areas under the stress of Al, Fe, and temporary flooding (Table 9). The correlation coefficient of tolerance indices with productivity in SSC+TF treatment shows a highly significant and high value based on the Pearson correlation on the GMP (r=0.73), STI (r=0.63), and MP (r=0.86) indices; all those indices had an exact high correlation to yield on the optimum treatment (SSC) that has correlation indices of r=0.89, r=0.85, and r=0.82, respectively.

Index selection that has a high correlation with grain yield on DL environment stress was STI (r=0.81) and MP (r=0.77), the correlation of optimum SSC treatment on those selection indices was STI (r=0.76) and MP (r=0.98). The correlation coefficient score is useful to explain the relationship within traits in terms of degree, characteristic, and direction of selection to apply (Sayo et al. 2017).

Appropriate secondary trait and index selection for genotype selection in tidal swamp land using PCA analysis

PCA biplot analysis was employed to explain the relationship between tolerance index and productivity of stress environment in this experiment. PCA analysis is widely used as it simplifies large amounts of data into easy-to-understand visualizations (Metsalu and Vilo 2015). Furthermore, the data analyzed was standardized beforehand—data closer to the biplot axis indicates more stable characters than those more distant from the axis. In the previous study, Principal Component Analysis is useful to predict the relationship between germplasm to the environment and cassava sample according to several of its enzymatic activity. The analysis work with the approach of multiple correlations (Wang et al. 2014; Mursyidin and Khairullah 2020).

Table 6. Agronomic characters with direct effect and indirect effect on maize grain yield in SSC treatment in acid soil tidal swamp

	Direct effect	Indirect effect			
		(X6e)	(X1e)	(XeSt)	Residual
Weight of 6 ears (X6e)	0.61		0.07	0.07	0.39
Weight of 1 ear (X1e)	0.20	0.21		0.03	0.39
Height of ear stand (XESt)	0.13	0.32	0.05		0.39

Table 7. Agronomic characters with direct effect and indirect effect to maize grain yield of SSC+TF treatment in acid soil tidal swamp

	Direct effect	Indirect effect				
		(X6E)	**(X1E)**	**(XKpC)**	**(XNR)**	**Residual**
Weight of 6 ears (X6e)	0.73		0.00	0.02	0.07	0.30
Weight of 1 ear (X1e)	0.07	0.04		-0.03	0.02	0.30
Kernels/cob weight Ratio (XKpC)	-0.10	-0.12	0.02		-0.03	0.30
Number of rows per ear (XNR)	0.14	0.34	0.01	0.02		0.30

Table 8. Agronomic characters with direct effect and indirect effect on maize grain yield of DL treatment in acid soil tidal swamp

	Direct effect	Indirect effect			
		(XKpC)	**(XESt)**	**(XAD)**	**Residual**
Kernels/cob weight ratio (XKpC)	-0.25		-0.06	-0.01	0.59
Height of ear stand (XESt)	0.47	0.03		0.03	0.59
Anthesis day (XAD)	-0.26	-0.01	-0.05		0.59

Table 9. Pearson's correlation of tolerance indices and grain yield characters on the SSC + TF treatment acid soil tidal swamp

Characters	Yp		Ys_gs		GMP		YSI		SSI		YI		TOL	STI		MP
Yp	1															
Ys_dl	0.42	**	1													
GMP	0.89	**	0.73	**	1											
YSI	-0.2	**	0.07		-0.2	**	1									
SSI	-0.2	**	0.07		-0.2	**	1	**	1							
YI	0.42	**	1	**	0.73	**	0.07		0.07		1					
TOL	0.46	**	-0.6	**	0.06		-0.3	**	-0.3	**	-0.6	**	1			
STI	0.85	**	0.63	**	0.9	**	-0.1		-0.1		0.63	**	0.13	1		
MP	0.82	**	0.86	**	0.96	**	-0.1		-0.1		0.86	**	-0.1	0.87	**	1

Notes: ** significant at 1% level, Yp: The yield on SSC treatment, Ys_gs: The yield on SSC+GS treatment; GMP: Geometric mean productivity; YSI: Yield stability index; SSI: Susceptibility index; YI: Yield index; TOL: Tolerance index; STI: Stress tolerance index; MP: Mean productivity

Table 10. Pearson's correlation of tolerance indices and grain yield characters on the DL treatment acid soil tidal swamp

Characters	Yp	Ys_dl	GMP	YSI	SSI	YI	TOL	STI	MP
Yp	1								
Ys_gs	0.62**	1							
GMP	0.87**	0.9**	1						
YSI	-0.2*	0.16	-0.1	1					
SSI	0.18*	-0.2*	0.07	-1**	1				
YI	0.62**	1**	0.9**	0.16	-0.2	1			
TOL	0.95**	0.33**	0.67**	-0.3**	0.28**	0.33**	1		
STI	0.76**	0.81**	0.89**	-0	0.04	0.81**	0.58**	1	
MP	0.98**	0.77**	0.95**	-0.1	0.1	0.77**	0.85**	0.84**	1

Notes: ** significant at 1% level, Yp: The yield on SSC treatment, Ys_dl: Yield in dry land treatment; GMP: Geometric mean productivity; YSI: Yield stability index; SSI: Susceptibility index; YI: Yield index; TOL: Tolerance index; STI: Stress tolerance index; MP: Mean productivity.

The results of the biplot analysis show that the STI, MP, and XESt indices vectors coincide with the yield character in the SSC+TF environments (Figure 2). In the DL environment, the STI, GMP, MP, XNR, and X6E were at PC 1. The value of PC1 on SSC+TF was able to explain 52.40% of the variance and on PC2 explained 16.30% of the variance. PC1 in the DL environment explained 57.10% variance on PC1 and 16.60% variance on PC2. Data in Table 9 show that the highest PC coefficient was obtained on PC 1 in SSC+TF and DL environment treatments. As the main character, yield is given 3 points in the model to optimize the selection. PC1 was the fit model to explain the character factor based on the eigenvalue. Hence models to explain index selection in both SSC and DL environments were:

I_{SSC+TF} = 3*0.94 Yield+0.98GMP+0.89STI+0.90MP+ 0.83X6E+ 0.59XNR

I_{DL} = 3*0.90Yield+0.84STI+0.96MP+0.65XESt

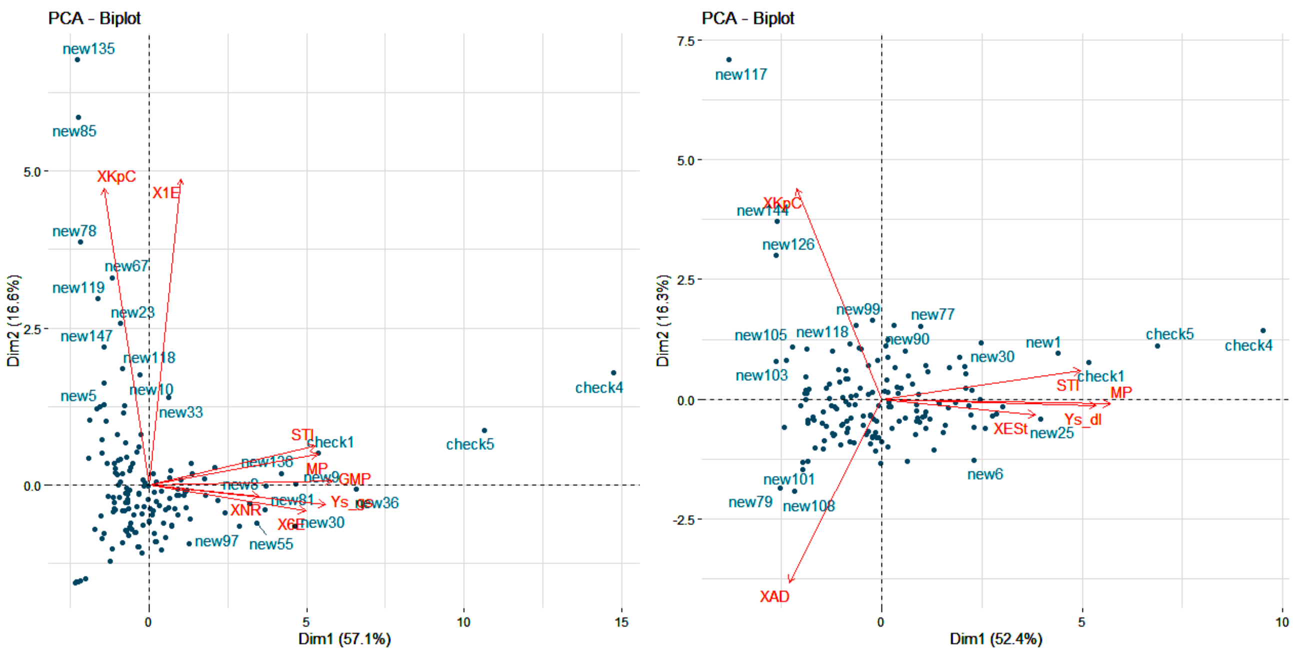


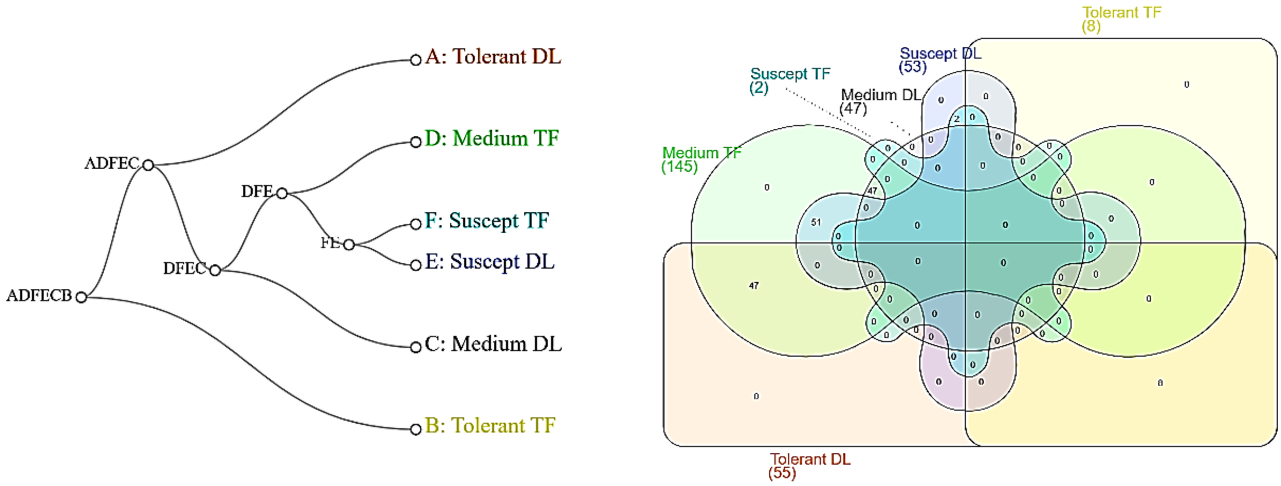
Figure 1. Biplot principal components 1 and 2 for all tolerance indices with yield in dryland (DL) and saturated water+temporary flooding (SS+TF) cultivation treatments

Table 11. PCA eigenvalues for each dimension in SSC + TF treatment and treatment (DL) dry land in tidal land

	PC 1	PC 2	PC 3	PC 4	PC 5
SSC+TF treatment					
Yield SSC+TF	0.94	-0.05	-0.02	0.07	-0.13
Geometric Mean Productivity (GMP)	0.98	0.01	-0.11	-0.04	0.12
Stress Tolerance Index (STI)	0.89	0.10	-0.29	-0.13	0.07
Mean Productivity (MP)	0.90	0.08	-0.19	-0.12	0.25
Weight of 6 Ear (X6E)	0.83	-0.07	0.02	0.29	-0.43
Number of Rows Per Ear (XNR)	0.59	-0.03	0.73	0.25	0.23
Variance percent	57.07	16.57	10.44	7.35	4.80
Cumulative variance percent	57.07	73.64	84.08	91.44	96.24
Eigenvalue	4.57	1.33	0.84	0.59	0.38
DL treatment					
Yield DL	0.90	-0.02	-0.02	-0.03	0.42
Stress Tolerance Index (STI)	0.84	0.10	0.13	0.43	-0.28
Mean Productivity (MP)	0.96	-0.02	0.09	0.19	0.03
Height of Ear Stand (XESt)	0.65	-0.05	0.41	-0.62	-0.16
Variance percent	52.35	16.33	14.50	11.07	4.98
Cumulative variance percent	52.35	68.69	83.19	94.26	99.24
Eigenvalue	3.14	0.98	0.87	0.66	0.30

Tolerance grouping based on multivariate analysis

The grouping of genotype tolerances was based on the index value obtained from the calculation following the index selection model. The genotypes were then classified into three categories, i.e., tolerant, moderately tolerant, and susceptible. The tolerant category score was $X_i > X + SE*t_{table}$; the susceptible category score was $X_i < X - SE*t_{table}$, while the moderately-tolerant category score was between these two values. Finally, a Venn diagram was applied to describe the tolerance category in each environment. The results of the tolerance grouping in the two experimental environments classified the genotypes into five categories (Figure 3A). Eight genotypes were tolerant in both SSC+TF and DL stress environments, and 47 genotypes were classified tolerant in the DL environment and moderately tolerant in SSC+TF environments. Forty-seven genotypes were moderately tolerant in both SSC+TF and DL environments, and 51 genotypes were moderately tolerant at SSC+TF and susceptible in the DL environment. Two genotypes were susceptible to SSC+TF and DL environments (Figure 3B).



A B

Figure 2. A. Venn diagram summarizing adaptation of maize lines on SSC+TF and B. DL of acid soil tidal swamp environments based on selected indices

Table 12. Venn diagram of maize genotype grouping based on the tolerance in SSC+TF and DL stress acid soil tidal swamp environment

Group of tolerance	Genotypes	Mean yield		Number of genotypes
		SSC+TF	DL	
[Tolerant DL] and [Tolerant TF]	Sukmaraga, P27, BIMA20URI, M-35-1-B, MPop11_01, MPop18_02, MPop27_08, MPop28_01	0.93	2.14	8
[Tolerant DL] and [Moderate TF]	MPop27_01, MPop28_02, MPop28_03, C44-3-1, C27-4-B, MPop28_04, E57-B, MGOLD, MPop28_05, DKL9001, M1-3-B, M-10-1-B, DYW7-B, DYW34-B, MPop03_06, MPop02_02, MPop02_04, MPop02_06, MPop11_06, MPop18_01, MPop26_01, MPop26_03, MPop27_04, MPop05_01, MPop05_02, MPop05_04, MPop05_06, MPop24_01, MPop24_02, MPop24_03, MPop27_05, MPop24_06, MPop24_08, MPop24_09, MPop24_10, MPop21_03, MPop21_04, MPop21_08, MPop21_09, MPop27_06, PAC2244-2-3-1-3-9-B-B, PAC2244-2-3-1-4-11B-B, PAC9996-2-4-1-2-11B-B, PAC2247-1-1-1-1-8B-B, PAC2244-1-6-1-1-9B-B, PAC2245-3-1-1-2-8B-B, C17-1-B	0.20	1.90	47
[Medium DL] and [Moderate TF]	NEI9008, C39-1-B, C31-1-B, C25-1-B, E31B, E90B, M42-1-B, DYW15-B, DYW24-B, CLYN231, AL46, MAL03, MR15, B11209, G1026-12, G1044-14, MPop03_01, CY-7, MPop03_09, MPop03_10, MPop10_01, MPop10_07, MPop10_08, MPop02_03, MPop15_01, MPop11_02, MPop11_03, MPop11_08, MPop05_03, MPop24_04, MPop24_05, MPop24_07, PAC99912-2-4-1-4-10-B-B, PAC2247-1-1-1-4-12B-B, PAC2247-1-1-1-2-9BB, MPop27_07, PAC2244-1-6-1-3-7B-B, PAC2244-2-3-1-2-9B-B, PAC2246-2-4-1-2-10B-B, PAC2245-3-1-1-4-5B-B, PAC9999-1-2-1-4-9B-B, PAC2247-1-1-1-2-8B-B, C40-2-B, C61-1-B, C32-5-B, C58-1-B, C19-4-B	0.14	1.11	47
[Moderate TF] and [Susceptible DL]	MR-14, C93-1-B, C63-1-B, C48-5-B, C46-2-B, C72-1-B, C47-3-B, C73-2-B, C6-3-B, C62-2-B, C37-1-B, C11-1-B, C60B-B, C16B, C41-1-B, E100B, E93B, E52B, E102B, WYW27B, WYW18B, WYW11B, M-6-1-B, DYW-17-B, MPop28-06, MPop03_03, MPop27_02, MPop10_02, Pop10_03, MPop27_03, MPop26_02, MPop05_05, MPop21_10, G104414, G2013631, PAC2245-3-1-1-4-4B-B, PAC9999-1-2-1-1-10BB, PAC2243-1-5-1-1-9B-B, PAC99912-3-1-1-3-9B-B, PAC9997-1-1-1-3-8B-B, PAC99912-2-4-1-3-8B-B, PAC99912-2-4-1-3-7B-B, PAC2241-2-3-1-3-4B-B, PAC2246-2-4-1-1-7B-B, PAC99912-2-4-1-1-7B-B, C54-1-B, C43-1-B, C56-1-B, C12-1-B, C42-3-B, C10-2-B	0.08	0.71	51
[Susceptible DL] and [Susceptible TF]	AMB30, AMB20	0.00	0.78	2

Sukmaraga, P27, and BIMA20URI varieties belong to the tolerant group in two stress treatment environments of SSC+TF and DL. Those check varieties belonged to heterozygous genotypes of open-pollinated, hybrid, and three-way cross variety, respectively. Maize tolerance to abiotic stress was commonly controlled by additive genes, which is why heterozygotes are more desirable to survival in an abiotic stress environment. Genotypes of M-35-1-B, MPop11_01, MPop18_02, MPop27_08, MPop28_01. Genotypes of AMB30 and AMB20 were susceptible in both SSC+TF and DL treatment environments.

In conclusion, based on multivariate analysis, secondary characters/selection indices employed for selecting maize genotype in acid tidal swamp land using saturated soil culture technique with temporary flooding (SSC+TF) were STI, MP, GMP, rows number per ear, and weight of 6 ears. STI, MP, and ear stand height characters were the most appropriate for selecting maize genotype on the acidic tidal swamp land on dryland stress.

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