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Research Article

Multi trait genotype- ideotype distance index - A tool for identification of elite parental inbreds for developing heterotic hybrids of fodder maize (*Zea mays* L.)

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Abstract

Forage maize breeders face a significant challenge in identifying genetic make-up that are superior for green fodder yield. Because it is influenced by polygenic trait. Therefore, understanding the relationship between biomass yield and other contributing characters would be very useful in selecting better genotype for improving green fodder yield. Forty one fodder maize inbred lines were evaluated in a randomized complete block design during June –September, 2021 to design selection index through various genetic and multivariate analyses to identify elite fodder maize inbreds for hybridization. The genotypes were evaluated for fourteen biometrical and four fodder quality traits. The association analysis revealed that plant height, leaf length, leaf breadth, number of nodes, stem girth, L/S ratio had highly significant contribution towards green fodder yield. The D^2 analysis indicated that green fodder yield has contributed about 55.60 per cent to the total variation followed by dry matter yield for about 17.92%. The Multivariate Genotype- Ideotype Distance Index analysis identified six superior inbreds namely GETM 25, UMI 1210, UMI 1221, African Tall, GETM 69 and TNFM 139-1.

Keywords: Fodder maize, Diversity, Green fodder yield, Principal component.

INTRODUCTION

Maize (*Zea mays* L.) is an important cereal crop belongs to the family Poaceae, also known as grass family. In India, it is being produced as a dual purpose crop for grain as well as fodder (Mahdi *et al.,* 2010). It is being utilized as a fodder crop for silage making besides feed as green forage for animals. Due to larger expansion of dairy, poultry, and corn-based industries, its demand has been raised (Ahmed *et al*., 2010). Because of the size and dispersion of its foliage, maize is better than most other cereal crops at utilising sunlight being a C_4 plant and develops more quickly (Warman, 2003). Maize is regarded as an ideal forage crop because it grows quickly, produces higher yields, palatable, nutrient-rich, and aids in increasing cattle body weight and milk quality (Sattar *et al*., 1994). Green fodder provides sufficient energy and proteins for animal growth and milk production (Takawale *et al*., 2009). By 2050, demand for green fodder and dry fodder will be expected to be 1012 million tonnes and 631 million tonnes, respectively. If demand for forage resources continues at its current rate, there will be

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18.4% deficit in green fodder and a 13.2% deficit in dry fodder by 2050. To bridge the gap, green fodder supply has to increase necessarily at a rate of 1.69% per year (2050 IGFRI Vision).

The current study aims to ascertain existing variability of traits and their relationship among them. The selection of superior genotypes based on selection index proposed by Smith (1936) and Hazel (1943) is a tedious method because it requires to give weightage for different traits. Therefore, multivariate selection index called Multi-trait Genotype–Ideotype Distance Index (MGIDI) developed by Olivoto and Lúcio, (2020) is the latest and useful tool for selection of superior genotypes. So, this study is focused on formulation of selection index to identify the best inbreds to develop heterotic single cross hybrids with higher biomass and quality in fodder maize.

MATERIALS AND METHODS

A set of 41 inbred lines of fodder maize (**Table 1**) were used for characterizing their quantitative and fodder quality traits. The experiment was organized in randomized block design with two replications at Department of Forage Crops, Tamil Nadu Agricultural University, Coimbatore. Each genotype was raised in 4m length row with 60cm × 30cm spacing during June – September, 2021. The quantitative traits related to the fodder yield were recorded from five randomly selected plants in each replication of each genotype at fifty percent flowering stage. Plants were harvested and dried for fodder quality analysis. A total of 12 quantitative traits *viz.,* days to fifty percent flowering, plant height, cob placement height, leaf length and breadth, number of leaves and nodes, internode length, stem girth, leaf stem ratio, green fodder and dry matter yield were observed. Fodder quality traits *viz.,* crude protein (CP), crude fiber (CF), acid detergent (ADF) and neutral detergent fiber (NDF) were analysed as per the standard protocol prescribed by Association of Official Analytical Collaboration (1970). The trait association and effects of different traits on green fodder yield were analysed through correlation and path coefficient analyses. The diversity of these fodder maize inbreds were analysed through Principle component analysis (PCA) and clustering. The PCA (Kassambara and Mundt, 2020), Karl pearson's coefficient of correlation (Wei and Simko, 2021) and path analysis were done by using R software version 4.2.1. The Mahalanobis D^2 analysis and diversity analysis by clustering of genotypes were carried out with TNAUSTAT (Manivannan, 2014) and DARwin (ver: 6.0.021) (Perrier & Jacquemoud-Collet, 2006) respectively. The multi-trait genotype-ideotype distance index (MGIDI) is calculated as per the formula given by

$$MGIDI_i = \sqrt{\sum_{j=1}^{f}(F_{ij} - F_j)^2}$$

where, F_{ij} is i^{th} genotype score for j^{th} factor *(i = 1, 2, ..., g; j = 1, 2, ..., f)*, *g* and *f* stated as total number of genotypes and their factors. The analysis were done in R studio 4.2.1. using the package of "metan" (Olivoto and Lúcio, 2020) with the 'gamem' and 'mgidi' functions.

RESULTS AND DISCUSSION

Mean performance: Analysis of variance (**Table 2**) showed existence of significant variation for all the studied variables**.** The overall mean performance (**Table 3**) of 41 fodder maize inbred lines revealed that highest green fodder and dry fodder yield were recorded by DM 74-2. This genotype also recorded better crude protein content of 11.16%. The genotype 52021 had the highest crude protein content of 15.48%. The crude fiber content in the inbreds ranged from 22.15 to 32.04 per cent in N-10-86 and GETM-29 respectively. The acid detergent fiber and neutral detergent fiber ranged from 28.00 to 43.00 per cent and 45.00 to 62.00 per cent respectively. The crude fiber content in the inbreds ranged from 22.15 % to 32.04 % in N-10-86 and GETM-29 respectively.

Relative contribution of traits for GFY: The results of correlation among different traits (**Fig. 1**) showed

Table 1. List of fodder maize inbred lines and their performance

S.No.	Inbreds	S.No.	Inbreds	S.No.	Inbreds	S.No.	Inbreds
1	UMI 61	11	N-10-86	21	DM 12-5	31	GETM 25
2	UMI 112	12	N 61	22	DM 12-6	32	GETM 26
3	UMI 1200	13	N 66	23	DM 12-27	33	GETM 27
4	UMI 1201	14	52021	24	DM 74-2	34	GETM 29
5	UMI 1205	15	52485	25	DM 82	35	GETM 39
6	UMI 1210	16	DM-12	26	DM 84	36	GETM 40
7	UMI 1220	17	DM 12-1	27	DM 86	37	GETM 64
8	UMI 1221	18	DM 12 -2	28	DM 94	38	GETM 67
9	UMI 1230	19	DM 12 -3	29	GETM 14	39	GETM 69
10	N-09-160-2	20	DM 12 -4	30	GETM 15	40	TNFM 139-1
						41	African Tall

Table 2. Analysis of variance of 16 forage traits in 41 fodder maize inbred lines

Trait	DFF	PH	CH	LL	LB	NL	SG	NN	IL	LSR	CP	CF	ADF	NDF	DMY	GFY
Replication	58.06	57.01	512.05	0.94	6.28	8.21	0.11	0.13	0.11	0.002	0.02	19.34	61.48	41.02	35.6	942
Genotype	21.78*	1036.89*	2241.72*	4.44*	5.14*	243.64*	1.34*	15.06*	1.22*	0.004*	4.47*	12.27*	23.19*	27.74*	6765.50*	99947.00*
Error	2.21	33.97	56.69	0.55	0.57	11.52	0.12	0.97	0.06	0.001	0.78	1.74	2.7	3.72	87.9	644

Table 3. Range of 12 biometrical and 4 quality traits of fodder maize inbred lines

Traits	DFF (days)	PH (cm)	CH (cm)	LL (cm)	LB (cm)	NL	SG (cm)	NN	IL (cm)	LSR	CP (%)	CF (%)	ADF (%)	NDF (%)	DMY (g)	GFY (g)
Mean	56	219.26	96.42	88.60	9.51	13.19	5.99	12.08	16.99	0.26	12.18	27.83	36.12	54.88	132.71	595.41
Maximum	66	326.75	152.75	116.63	10.95	16.50	7.95	17.00	22.38	0.36	15.48	32.04	43.00	62.00	290.00	1182.00
Minimum	50	143.10	60.00	68.60	7.75	10.00	4.25	9.25	10.93	0.20	8.97	22.15	28.00	45.00	65.00	306.00

DFF-days to fifty per cent flowering, PH- plant height, CH- Cob placement height, LL- Leaf length, LB –Leaf breadth, NL- Number of leaves, NN- Number of nodes, SG- Stem girth, IL- Internode length, LSR- Leaf stem ratio, CP- Crude protein, CF- Crude fiber, ADF- Acid detergent fiber, NDF- Neutral detergent fiber, DMY- Dry matter yield, GFY- Green fodder yield.

that the trait plant height proved to be an important trait in enhancing total green fodder yield. Plant height had positive relationship with most of traits like leaf length, leaf numbers, internode length and stem girth. Infant *et al.* (2018) reported that the plant height is ultimately affected by spacing of the plants and positively correlate with total biomass production. The leaf length also had positive association with internode length. Robertson (1994) also reported that the continued elongation of internode increases the leaf length at fully expanded stage.

The total green fodder yield is not a sole trait. It depends on many component traits like flowering, plant height, leaf length, breadth, number of nodes and leaves. In the present study, all these traits expressed positive relationship with green fodder yield whereas few fodder quality traits such as CP, CF and NDF does not display any association with fodder yield (**Fig. 1**). The ADF and NDF were positively correlated with total crude fiber because it contains the cellulose and lignin which forms the part of detergent fiber contents. Moreover, increase in the status of fibrous portion would decrease crude protein which is attributed to its indigestible portions of feed stuff, which subsequently reduce energy level of feed (Saha *et al.,*2010). The crude protein content is negatively associated with ADF, hence to reduce the indigestible carbohydrate fractions like ADF, the selection of genotypes with higher crude protein content would be rewarding to increase the intake of feed. This result was in accordance with the findings of Saiyad and Kumar, (2018).

Direct and indirect influence of traits for green fodder yield: The selection of genotypes based on association between two variables may give deceptive results because quantitative inheritance is influenced by more than two variables. So, the path coefficient analysis (**Table 4**) is a breeding tool which helps in understanding relationship and to split correlation into direct and indirect effects. As per the classification of path coefficient suggested by Lenka and Mishra (1973), in this study leaf length (0.439), number of leaves (0.143), internode length (0.335) and dry matter yield (0.617) had high positive direct effect on fodder yield. The yield contributing trait plant height (-0.356) had recorded negative direct effect on fodder yield. In case of indirect effect plant height had positive indirect effect on green fodder yield through dry matter yield (0.381) followed by leaf length (0.334). Also the leaf length had observed positive indirect effect on GFY through DMY (0.439) and internode length (0.224). Hence, traits *viz.,* plant height, leaf length, number of leaves, internode length, crude fat and dry matter yield may be considered as selection indices for higher green fodder yield.

Genetic diversity determined through Principle Component Analysis: The principle component analysis proposed by Karl Pearson in 1901 is the technique used for dimension reduction of datasets into few components (Venujayakanth *et al.,* 2017). Kaiser (1960) stated that the eigen value of PCs having greater than one will give more information. In the present study, first four PCs recorded the eigen value >1. These first four PCs (**Table 5**) have contributed 73.11 per cent of cumulative variance and the first two PCs alone reported to account for 53.58 per cent of variation. Saleh *et al.*(2022) also utilized PCA for grouping the genotypes and observed first three components accounting for 77% of total variation. Similarly, 76% of variation was accounted with first four components by the study of Sinha *et al.* (2019).
The eigen vectors are ranged from -1 to +1. The values

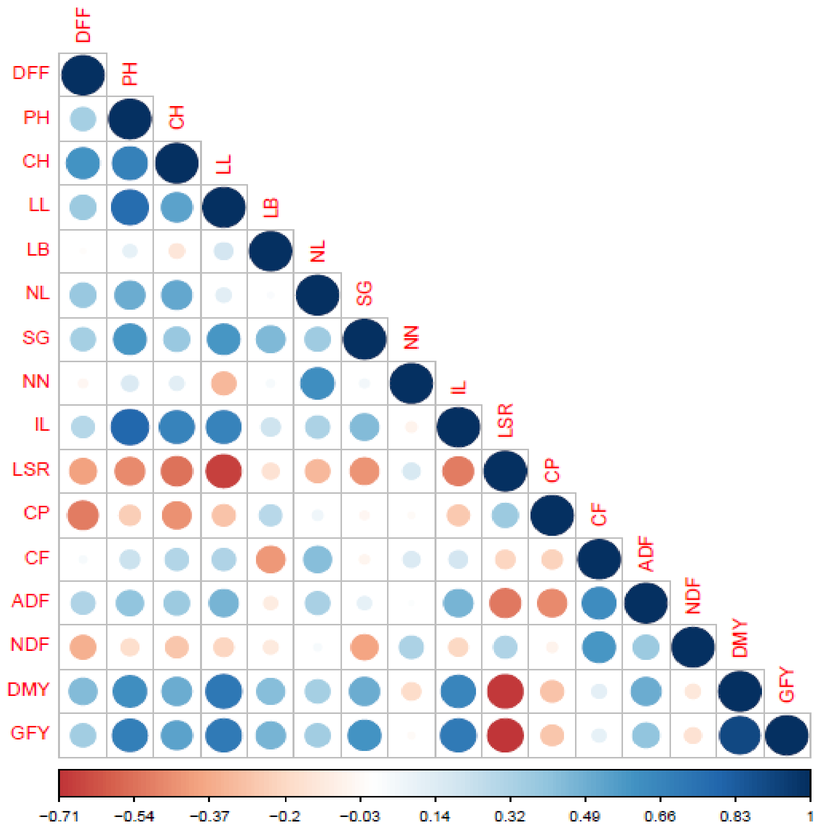

Fig. 1. Correlation between 12 biometrical and 4 fodder quality traits

Table 4. Path coefficient analysis of various traits on green fodder yield

Traits	DFF	PH	CH	LL	LB	NL	SG	NN	IL	LSR	CP	CF	ADF	NDF	DMY	Genotypic correlation with GFY
DFF	**-0.123**	-0.120	0.009	0.162	0.002	0.083	0.045	-0.007	0.094	0.064	0.057	-0.011	-0.070	-0.105	0.269	0.348
PH	-0.042	**-0.356**	0.010	0.334	-0.011	0.111	0.079	0.022	0.264	0.076	0.027	-0.073	-0.090	-0.052	0.381	0.681
CH	-0.074	-0.242	**0.014**	0.236	0.014	0.115	0.051	0.017	0.223	0.086	0.050	-0.101	-0.082	-0.082	0.306	0.532
LL	-0.046	-0.271	0.008	**0.439**	-0.020	0.028	0.079	-0.047	0.224	0.109	0.032	-0.102	-0.104	-0.064	0.439	0.703
LB	0.002	-0.036	-0.002	0.082	**-0.108**	0.005	0.061	0.005	0.068	0.025	-0.030	0.148	0.024	-0.035	0.260	0.468
NL	-0.046	-0.176	0.007	0.055	-0.002	**0.225**	0.048	0.088	0.105	0.052	-0.007	-0.145	-0.074	0.010	0.209	0.349
SG	-0.041	-0.208	0.005	0.256	-0.048	0.080	**0.135**	0.007	0.147	0.071	0.005	0.018	-0.024	-0.117	0.307	0.593
NN	0.006	-0.054	0.002	-0.142	-0.004	0.137	0.007	**0.144**	-0.023	-0.026	0.003	-0.053	-0.003	0.094	-0.116	-0.030
IL	-0.035	-0.281	0.010	0.293	-0.022	0.070	0.060	-0.010	**0.335**	0.082	0.029	-0.064	-0.106	-0.060	0.402	0.703
LSR	0.049	0.170	-0.008	-0.300	0.017	-0.074	-0.061	0.023	-0.173	**-0.159**	-0.040	0.072	0.118	0.089	-0.435	-0.711
CP	0.064	0.089	-0.007	-0.126	-0.030	0.015	-0.006	-0.004	-0.090	-0.058	**-0.110**	0.076	0.107	-0.020	-0.177	-0.277
CF	-0.004	-0.077	0.004	0.133	0.047	0.096	-0.007	0.022	0.063	0.034	0.025	**-0.338**	-0.143	0.175	0.074	0.105
ADF	-0.038	-0.141	0.005	0.203	0.011	0.074	0.015	0.002	0.157	0.083	0.052	-0.213	**-0.226**	0.109	0.303	0.394
NDF	0.044	0.062	-0.004	-0.095	0.013	0.008	-0.053	0.045	-0.068	-0.048	0.007	-0.199	-0.083	**0.296**	-0.079	-0.153
DMY	-0.054	-0.220	0.007	0.312	-0.046	0.076	0.067	-0.027	0.218	0.112	0.032	-0.040	-0.111	-0.038	**0.617**	0.906

Residual effect = 0.0981

Table 5. Variance and different principal components of 16 traits of fodder maize

PCs	EV	VP	CV	DFF	PH	CH	LL	LB	NL	SG	NN	IL	LSR	CP	CF	ADF	NDF	DMY	GFY
PC1	6.10	38.15	38.15	0.07	0.88	0.64	0.69	0.08	0.20	0.44	0.01	0.89	-0.32	-0.21	0.15	0.29	-0.17	0.53	0.63
PC2	2.47	15.43	53.58	-0.19	0.07	-0.09	0.17	-0.15	0.13	-0.13	0.14	0.03	-0.04	-0.19	0.78	0.71	0.84	0.12	0.08
PC3	1.65	10.33	63.91	0.16	0.20	0.29	-0.30	0.02	0.81	0.12	0.88	0.03	0.04	0.09	0.13	0.03	0.16	-0.04	0.06
PC4	1.47	9.20	73.12	-0.06	0.02	-0.30	0.06	0.90	-0.12	0.32	0.10	0.06	-0.04	0.05	-0.45	-0.08	0.13	0.38	0.42
PC5	0.97	6.05	79.17	0.73	0.09	0.38	0.12	-0.13	-0.01	0.03	0.03	0.10	-0.12	-0.89	-0.02	0.26	-0.07	0.19	0.14
PC6	0.75	4.68	83.85	0.34	0.11	0.32	0.32	0.13	0.33	0.11	-0.26	0.22	-0.84	-0.01	0.13	0.36	-0.21	0.64	0.53

EV-Eigen value, VP- Variance percentage, CV- Cumulative variance

with higher points in both the direction contributes more to the total variance (Anthony, 2014). Based on this approach internode length (0.89), plant height (0.88), leaf length (0.69), cob placement height (0.64), green fodder yield (0.63) and dry matter yield (0.53) had higher vector coefficients in the positive direction. Hence, these are all important variables which contribute more towards the 38.14 per cent of variation of first PC. In the case of second PC, three fodder quality traits *viz.,* crude fiber, ADF and NDF possessing higher vector coefficients in the positive direction to the 15.43 per cent of variation. Pavithra *et al.* (2022) also reported that fodder yield, number of leaves, plant height and stem girth displayed positive eigen vector coefficients for higher variation of first two components.

The biplot showed in the **Fig. 2** would exhibit more about information accompanying with trait association and clustering of genotypes. The acute angle between two variables revealed their positive association and from the biplot, the GFY had very high positive relationship with most of the traits like Dry matter yield, leaf length, inter node length and plant height. But the obtuse angle between GFY and leaf stem ratio, NDF and CP indicate that these traits do not have any positive contribution towards fodder yield. The variables which plotted close to the genotypes marked with plus symbols revealed that these genotypes had higher fodder yield as compared with others and those genotypes are very similar in response. Those genotypes which are grouped at opposite to GFY symbol with triangle recorded lower fodder yield indicating that these two groups are opposite to each other. The vector length of CF, ADF, NDF and

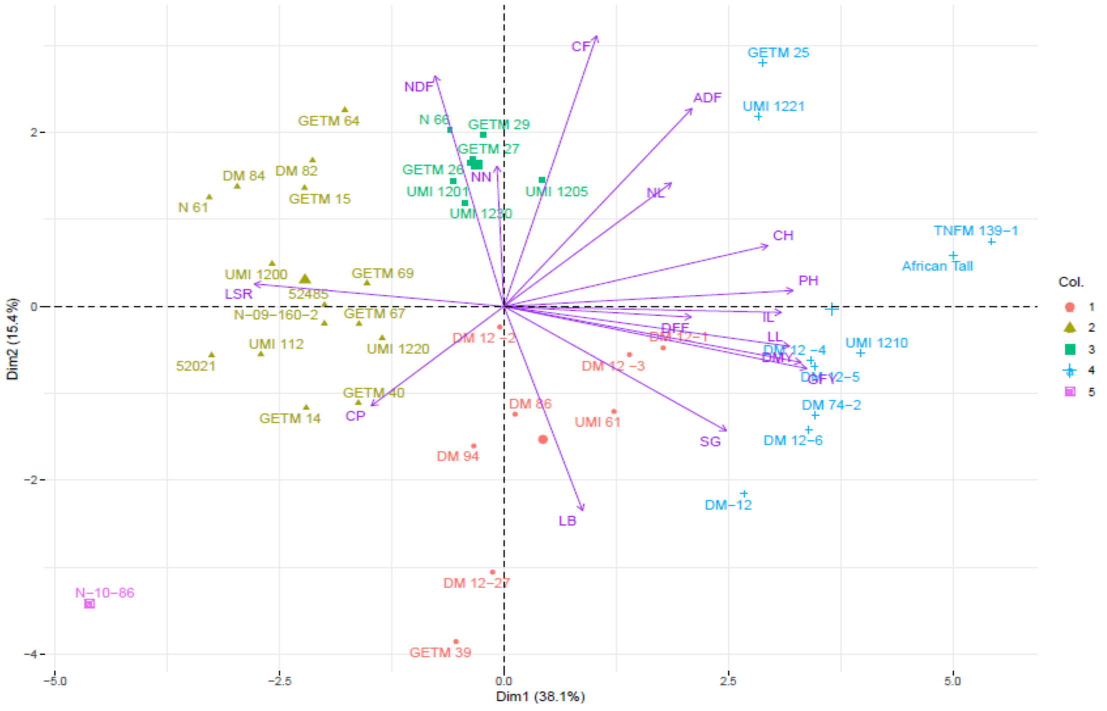

Fig. 2. PCA biplot showing relationship of traits and grouping of genotypes

GFY are high and indicates that these are most variable traits whereas the CP and NN are the least variable traits under the study. Kifayat *et al.* (2022) also reported similar findings on green fodder yield related traits.

Dissection of genetic diversity through D^2 analysis: The genetic divergence of genotypes from Mahalanobis D^2 analysis resulted in total of eight clusters including four solitary clusters (**Table 6**). The maximum of 18 inbred lines were grouped in a Cluster II and eight in Cluster I. Cluster III & VIII (**Table 7**) had very high inter cluster distance (2152.27) followed by fourth cluster (1784.30) revealing the existence of greater divergence among genotypes within the group. The solitarily clustered genotype African Tall had greater inter cluster distance with many other clusters. Hence, it can be concluded that this is a highly diverse genotypes than others. These type of facts was also observed in the study of Rathod *et al.* (2021) in 54 forage maize inbred lines.

The cluster mean values revealed that the Cluster III had recorded lowest GFY of 381.60 and Cluster IV had highest of 971.33 suggesting that the genotypes present in this cluster can be used as a parent to develop highly heterotic single cross fodder maize hybrids or composites. Comparatively, the genotypes present in the Clusters I & IV had better mean for plant height, leaf length and better crude protein content, so these could be exploited to develop outstanding hybrids through heterosis breeding. The results showed that DFY and DMY contributed maximum of 55.6 and 17.92 correspondingly for divergence. Prasad *et al.* (2021) reported that DMY had contributed 49.52 percent of variation in a fodder maize evaluation study.

Unweighted Pair Group Method with Arithmetic Mean (UPGMA) of hierarchical clustering based on the Euclidean distance between each genotype registered total of seven groups **(Fig. 3)** with the maximum of eleven in the first group and solitary in seventh group (N-10-86). The dissimilarity index was calculated by using Euclidean standardized method for all genotypes and displayed the maximum of 11.21 between the genotype TNFM 139-1 and N-10-86 indicating that these genotypes are highly dissimilar and crossing of these inbred lines would yield heterotic fodder hybrids. These genotypes were found to be grouped separately, whereas the minimum dissimilarity of 1.65 is observed between DM 12-3 and DM 12-1which were clustered in the same group.

MGIDI selection index: The MGIDI results figured out that 16 traits were separated as four factors (FA) and the FA1 includes most of the biometrical traits which related to biomass (**Table 8**). In that fodder yield and their supportive traits were gathered as a single factor (FA1) and the fiber quality traits under FA2. The traits like GFY, DMY, CH and PH had higher selection gain percentage (37.00%, 30.10%, 26.70% and 19.40% respectively). All

Table 6. Mahalanobis D^2 analysis and clustering of genotypes

Cluster	Genotypes
I	UMI 61, DM 12-27, DM 12-1, UMI 1221,DM 12-3, GETM 39, UMI 1230, GETM 40
II	UMI 112, DM 86, GETM 67, GETM 64, GETM 26, 52485, GETM 14, DM 12-2, N66, N-09-160-2, GETM 15, UMI 1205, GETM 27, DM 94, GETM 29, UMI 1200, 52021, DM 82
III	UMI 1201, UMI 1220, DM 84, N 61, N-10-86
IV	UMI 1210, DM 12-6, DM 12-4, DM 12-5, DM 74-2, TNFM 139-1
V	DM 12
VI	GETM 25
VII	GETM 69
VIII	African Tall

Table 7. Average Intra (bold) and inter cluster distance of fodder maize genotypes

Clusters	I	II	III	IV	V	VI	VII	VIII
I	**202.006**	442.269	856.537	444.513	355.214	385.733	351.865	700.128
II		**242.188**	435.827	1104.793	566.963	828.061	501.410	1181.782
III			**277.875**	1784.304	858.776	1564.267	609.080	2152.278
IV				**246.783**	485.689	491.765	944.565	457.873
V					**0**	787.340	684.062	683.713
VI						**0**	660.860	520.886
VII							**0**	1573.566
VIII								**0**

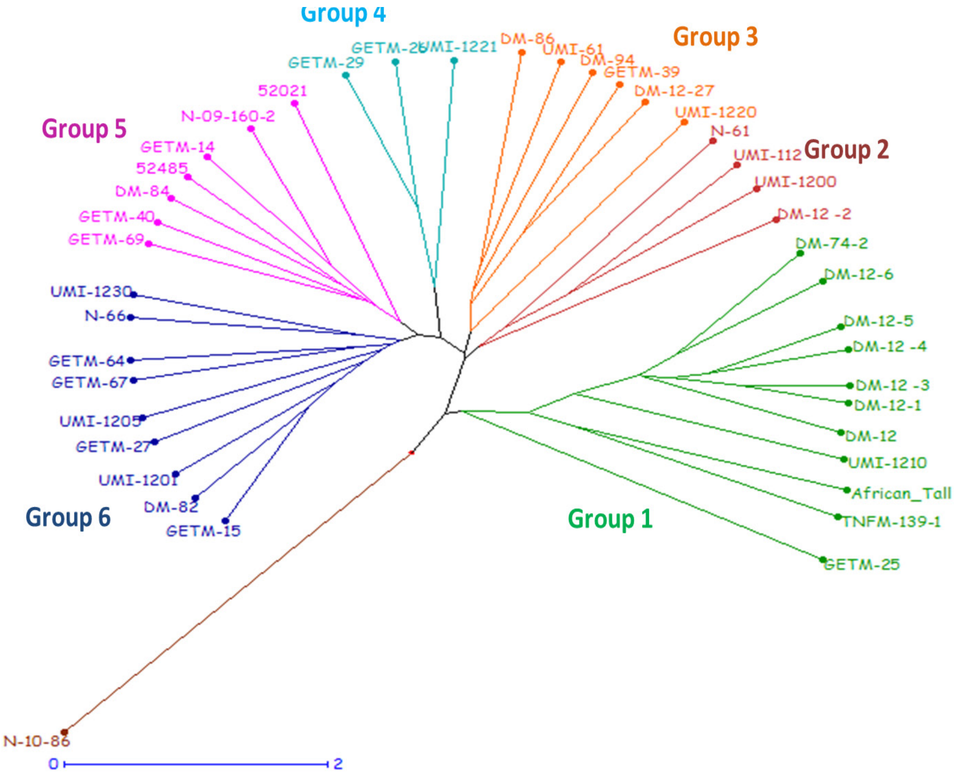

Fig. 3. UPGMA clustering of 41 fodder maize inbreds

Table 8. Genetic gain of different traits based on MGIDI index

S.No.	Traits	Factor	SG(%)
1	PH	FA1	19.40
2	LL	FA1	8.40
3	SG	FA1	12.00
4	IL	FA1	10.80
5	LSR	FA1	-10.00
6	DMY	FA1	30.10
7	GFY	FA1	37.00
8	CF	FA2	4.46
9	ADF	FA2	6.17
10	NDF	FA2	1.14
11	NL	FA3	11.00
12	NN	FA3	9.64
13	DFF	FA4	2.68
14	CH	FA4	26.70
15	LB	FA4	0.45
16	CP	FA4	-2.98

the studied traits exerted positive genetic gain except for LSR and crude protein content. LSR and crude protein accounted about 12.98% selection gain percentage. Maranna *et al.* (2021) also observed negative selection for plant height and days to flowering that negatively impact on grain yield on soybean. The selection pressure 15% (red line indicated that genotypes based on selection pressure) was used with aim of selection

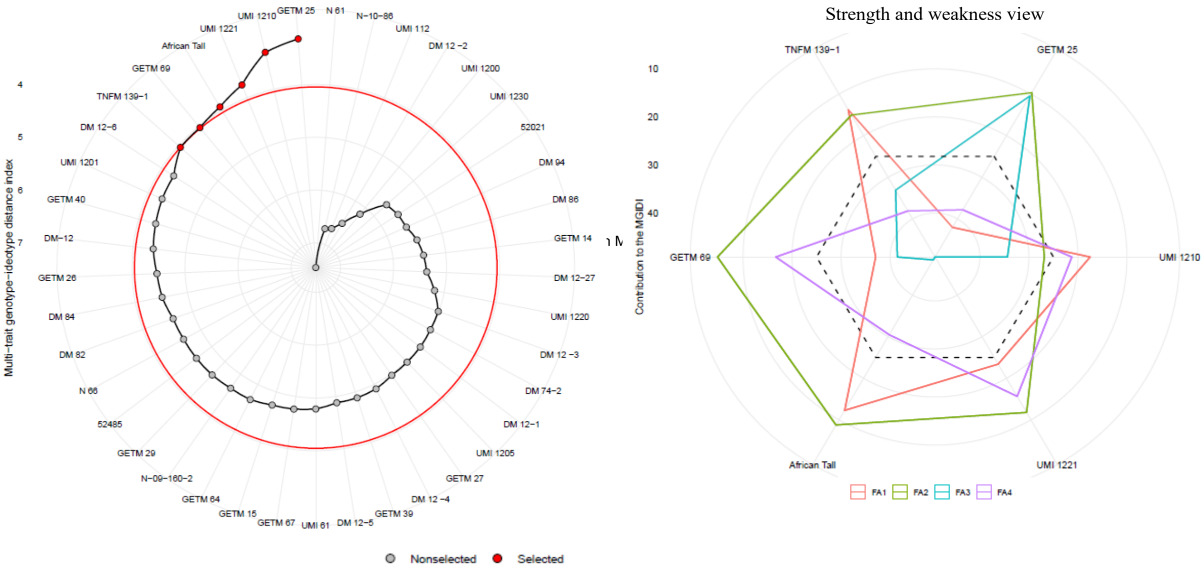

Fig. 4. Selection of genotypes based on MGIDI selection index

of superior genotypes. It resulted in the identification of six inbreds as best performer (**Fig. 4**). The strength and weakness plot showed that relative contribution of different traits on superior genotypes. It showed the traits which presented in the first two factors determine their superiority compared with others. The traits plant height, stem girth, internode length and both dry matter & green fodder yield are the strength for the genotypes TNFM 139-1, African Tall and UMI 1210. The fiber traits in the FA2 had very high contribution in the genotypes GETM 69, GETM 25 and African Tall. Whereas the traits number of leaves and number of nodes presented in the FA3 is the weakness of the selected genotypes but those are very solid strength for GETM 25. Uddin *et al.* (2021) also utilized this approach to select superior phosphorous tolerant genotypes in maize.

The results from fodder maize inbreds evaluation revealed that existence of significant variation among genotypes for all the studied variables. Since it is fodder crop emphasis given on fodder component traits which relatively enhance the total biomass. Considering the different genetic analysis and MGIDI, a selection index to select the genotypes with increased biomass could be achieved by selection of associated traits like plant height, leaf length, stem girth and internode length. The development of single cross fodder maize hybrids with higher heterosis is the breeding task of ours to increase the biomass and as a necessary first step, the valid parental selection could be done by using MGIDI. The MGIDI index facilitated the identification of elite inbreds exhibiting superior performance for fodder yield component traits like plant height, cob height, stem girth, internode length and dry matter yield. These traits were also found to be the valid selection indices as understood from the results of others genetic analyses performed in the present study.

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