Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

Performance of 30 Maize Double Haploid Lines Under Drought Stress and Soil Conditioners

[1] Sabra D.M, [2] Younis A.S.M, [3] Nassar S.M.A, [4] Fathallah F.B, [5] A.M.A. Metwally

[1] Field Crops Research Department, National Research Centre, Dokki, Giza,

[2] Field Crops Research Department, National Research Centre, Dokki, Giza,

[3] Departments of Genetic Resources, Desert Research Center, Cairo,

[4] Genetics and Cytology Department, National Research Centre, Dokki, Giza, Egypt,

[5] Field Crops Research Department, National Research Centre, Dokki, Giza, Egypt,

(Received: 07 January 2024 Revised: 12 February 2024 Accepted: 06 March 2024)

KEYWORDS

DH lines, GT-biplot, PCA, Soil Conditioners, Water stress, Zea mays

ABSTRACT:

Background: Doubled haploid (DH) lines are widely employed in maize breeding programs to investigate the genetic and phenotypic characteristics of the lines under drought-stressed soil and various conditions. Agronomic performance and enhanced haploid inducer ability are always sought after through genetic development.

Objective: The study aimed to investigate the impact of drought stress and soil conditioners on the agronomic performance of 30 doubled haploid (DH) maize lines (were developed by Haploid inducers using 8 populations) under different irrigation levels and soil conditions.

Materials and Method: Two drought stress levels and two hydrogel levels with three replicates were used in a factorial experiment. In this experiment, dripper was used to implement a drip irrigation system. The experiment was conducted using a split-split plot design with three replications. Main plots were devoted to irrigation levels, two soil conditions was located sub plot and 30 DH lines were located in sub-sub plots. The drought stress levels were the normal condition (100% of field capacity) and the drought stress (75%) of normal condition respectively were design with three replicates.

Results: The results discuss the identification of agronomic and yield traits in a 30 set of multiple DH populations, as well as under drought stress and non-stress conditions; shown that mean squares due to DHL were significant (P≤ 0.01) for all studied traits, suggesting existence of genetic differences among lines for all studied characters. This also indicates that lines will be differ in their cross combinations, i.e. in their hybrid ability. Also for the Mean squares due interaction between (lines (L) × irrigation regimes (I)) were significant (P ≤ 0.01) for all studied traits, except KPR, suggesting that lines behaved differently under different water irrigation for most studied traits and indicating the possibility of selecting lines for improved performance under a specific drought stress. Mean grain yield per plant (GYPP) was significantly (P ≤ 0.01) reduced due to water stress (75%) water irrigation by 5.01% (96.76g) compared with the normal irrigation which recorded 102.37 g . Under water stress, plant height and ear height were reduced by 29% and 7.61%, respectively, compared with normal irrigation, while kernel weight is reduced by 0.79% compared with 100% irrigation. Furthermore, DTA, DTS, and ASI exhibited shorter days to flowering under water stress.

Conclusion: There were significant genetic variations between the DH lines for every variable that was examined, suggesting that lines could be chosen for better performance under particular drought stress conditions. The work offers insightful information about the application of DH lines in maize research as well as the discovery of genomic areas linked to drought stress and agronomic features in various DH lines.

Journal of Chemical Health Risks
www.jchr.org
JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

1. Introduction

Egypt is a significant importer of maize (corn) despite its substantial domestic production. In 2017, Egypt produced 7.1 million tons of maize from 920,600 hectares, with an average yield of 7.71 tons per hectare. However, in 2021, the country imported maize worth about 41.93 billion Egyptian pounds, and in 2022, it imported 52.5 billion Egyptian pounds worth of maize, mainly from Argentina, Brazil, Ukraine, Romania, and the United States [1, 2]. The high import volume is attributed to the country's dependence on maize imports to meet domestic consumption needs, particularly for the local market and to augment indigenous production [3]. Despite its significant domestic production, Egypt's maize imports are expected to remain high, with an estimated 6.5 million metric tons of maize imported in the marketing year 2023/24 [4].

Maize breeders are always looking for new methods to enrich breeding material of better tolerance to drought stress. Using modern biotechnological techniques in plant breeding could contribute to a great extent in the induction of novel genetic variation, which are not existed in the gene pool, such as soma clonal and gametoclonal variation [5]. Doubled haploid (DH) technology has become an important tool in maize breeding, offering several advantages over traditional breeding methods. DH lines are completely homozygous, allowing the development of inbred lines in a single year, compared to three to four years with the conventional recurrent selfing method. This technology also leads to maximum genetic variance between lines for per se and testcross performance from the first generation, reduced breeding cycle length, and increased efficiency in marker-assisted selection, gene introgression, and stacking genes in lines. DH lines have been widely adopted by leading seed companies, and the technology has also found its way into research, albeit at a slower pace due to the need for experienced staff and appropriate experimental facilities [6]. The production of maize DH lines involves four main steps: (1) induction of haploids, (2) identification of haploids at the seed or seedling stage, (3) doubling the chromosomes in putative haploid seedlings, and (4) developing completely homozygous lines. The in vivo induction of maternal haploids is the most commonly used technique for DH line development, as it offers the fastest and most efficient route to produce completely homozygous lines. This technique has been widely adopted in commercial hybrid maize breeding programs and has proven to be highly effective [7, 8]. The advantages of DH lines in maize breeding include the rapid development of completely homozygous lines, increased genetic variance between lines, reduced breeding cycle length, and improved efficiency in selection and trait introgression. This technology has been particularly beneficial in developing improved maize hybrids with DH lines as parental lines, and it has gained a foothold in tropical breeding programs due to intensive efforts by various organizations. Overall, DH technology has significantly improved breeding efficiency and reduced costs in maize breeding programs [9]. The objectives of the present research were: (i) to screen a set of 30 maize DH line in the field in order to identify tolerant ones, (ii) to identify the relative importance of agronomic parameters associated with drought tolerance and (iii) to determine the extent of genetic diversity among 30 DH lines of maize, under stressed conditions, using morphological data based on PCA, and assess interrelationships between yield and yield-related traits using GT-biplot analysis, to identify the secondary traits for selection for improved yield under such conditions.

2. Material and Method

Material: On March 21, 2022 at the National Research Center's El-Nubria Research Station, thirty DHL maize seeds derived from eight populations using the DH lines technique were planted.

Methods: Over a three-year (2019, 2020, and 2021), pure lines of maize were developed using Doubled Haploid (DH) methods. In the first year of 2019, second-generation plants from hybrids were established. Subsequently, in 2021, the populations' seeds were produced, and the lines were developed in cooperation with the CIMMYT organization in Kenya to create the seed stock. The DH lines were then subjected to self-pollination in off-season in Toshka region. During the main season, the DH lines were evaluated as part of this breeding experiment. The current study was carried out in 2022 at the National Research Center's El-Noubaria Research Station in the El-Behaira Governorate to assess thirty DHL of maize, two drought stress levels, and two hydrogel levels with three replicates were used in a factorial experiment. In this experiment, drippers

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

will be used to implement a drip irrigation system. Three replications of a split split plot design in an RCBD arrangement were employed. Thirty DH lines were located in sub-sub plots, two soil conditions were located in sub-plots, and the main plots were used for irrigation levels. Three replicates were used in the design of the drought stress levels, which were (100% water irrigation) 8330 m3 ha−1 during the maize growing season was best than 6247 m3 ha−1 75% from water irrigation, and for hydrogel level which was zero adding (without for ha) and 30 kg ha−1 (with), respectively. There was one plant per hill in each experimental plot, with a row of 6 m in length and 70 cm in breadth and 30 cm in between hills.

Biometrical Analysis:

All the data were subjected to analysis of variance (ANOVA) of split-split plot experiment using SAS software. Comparisons of means were made using least significant difference (LSD) test at P< 0.05 and 0.01 levels of confidence according to Steel and Torrie [10]

3. Results and Discussion

Tables (1) display the comprehensive analysis of variance for the split split-plot design, the analysis covers several two water irrigation, two level of hydrogel and 30 DH lines and their interactions. The mean squares for water irrigations were not statistically significant for all features except for DTA, DTS, PH and 100 kernel weight which were significant and highly significant. This suggests that the level of water irrigation must be more sever in the next evaluations. On the other hand, soil conditioners were found to be statistically significant (P ≤ 0.05 or 0.01) for the traits except DTA, ASI and LANG, Additionally, DH lines were found to have a clear and significant effect on all studied traits. The mean squares resulting from the first-order interaction, specifically the interaction between the factors I (irrigation) and SC (soil conditioners) (I×SC), were not significant for the most traits. In the other hand the mean squares for the interaction between I×L and SC×L was found to be statistically significant (P ≤ 0.01) for all traits except for, RPE with I×L, and KPR (kernel per row) for the SC×L. Furthermore, the mean squares resulting from the second-order interaction, specifically the interaction between DHL, water irrigation, and soil conditioners (L×I×SC), were found to be statistically significant (P ≤ 0.01) for all traits except for KPR. These findings suggest that the performance of maize doubled haploids lines varies depending on the combination of irrigation and soil conditioners, and there is potential for selecting lines that perform well under specific water conditions, as suggested by previous studies conducted by [11, 12, 13, 14, and 15].

Table 1. Analysis of variance for 30 DH lines of maize under two water irrigation and soil conditioners in sandy soil

Source	df	DTA	DTS	ASI	PH	EH	LANG	EPP
Replication	2	30.25	74.2	10.41	563	35.93	3.4	35.88*
Irrigation (I)	1	1976*	2229*	7.58	104474**	471.23	2.5	7.8
Error(a)	2	119.5	63.33	10.41	373.4	52.14	27.71	1.34
Soil Con (SC)	1	447.78	203.63*	47.49	252.1**	1236**	33.61	12.47*
I×SC	1	3.5	7.88	21.88	1952**	418.45	202.5**	2.34
Error(b)	4	69.5	23.33	53.97	7.8	80.77	13.89	0.94
LINE (L)	29	296.2**	293.98**	3.49**	1301**	199.88**	246.83**	2.59**
I×L	29	24.00**	23.80**	2.29**	1749**	270.01**	162.56**	1.49**
SC×L	29	13.73**	11.15**	2.73**	365.6*	212.75**	94.53**	1.32**
I×SC×L	29	16.45**	13.50**	1.45**	649.3**	321.72**	101.64**	1.91**
Error (c)	232	0.03	0.22	0.28	227.6	60.27	18.24	0.31

Source	df	100KW	RPE	KPR	KPP	GYPP	GYPH(ton)
Replication	2	462.2**	3.22	384.9	150676	11087	20.35
Irrigation (I)	1	2.39**	0.71	2.06	9775	3209	5.89
Error(a)	2	0.02	0.9	122	90040	1723	3.16
Soil Con (SC)	1	4.85**	40.0**	210.5**	1163957**	63176**	115.9**
I×SC	1	0.06	9.34	1.99	2497	11102**	20.37**
Error(b)	4	0.1	2.61	5.29	49165	274	0.5
LINE (L)	29	299.81**	16.96**	188.7**	133805**	10957**	20.11**
I×L	29	16.44**	6.90**	0.26	61640**	2676**	4.91**
SC×L	29	80.67**	3.13	96.17**	78546**	5420**	9.95**

Journal of Chemical Health Risks
www.jchr.org
JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

I×SC×L	29	34.04**	7.07**	11.06	38593**	3555**	6.53**
Error (c)	232	0.1	3.06	21.35	13690	417	0.77

*and ** indicate significance at 0.05 and 0.01 probability level, respectively. DTA= Days to anthesis, DTS= Days to silking, ASI= Anthesis silking interval, PH= Plant height, EH= Ear height, LANG= Leaf angle, EPP= Ears plant^{-1}, 100KW= hundred Kernel weight, RPE= rows/ear, KPR= Kernels row^{-1}, KPP= Kernels plant^{-1}, GYPP= Grain yield plant^{-1}, GYPT= Grain yield hectar^{-1}.

Mean Performance

1. Impact of Water Irrigations:

Investigate the effect of different water irrigations on the average values of various characteristics across all the DHLs in the years 2022 found in Table (2). Water amounts will be referred to as the control environment, representing a relative change of 100% compared to a non-stressed environment. A stressed water irrigation stress will be represented by 75% water quantities. The data shown that non-significant for most studied traits except for DTA and DTS were significant and plant height and 100 kernel weight were highly significant that main we must be used more water stress in next evaluation for these DH lines The DTA and DTS had a substantial and statistically significant decline of 6.43% and 6.62% when the water levels were reduced to 75%, respectively. And plant height and 100 kernel weight (100-KW) had a substantial and statistically highly significant decline of 29% and 0.79% when the water levels were reduced to 75%, respectively. Based on these findings, other researchers have documented decreases in 100 kernel weight and flowering traits as a result of drought stress [16, 17, 18, 19, 20]. Denmead and Shaw [21] observed that water scarcity during the vegetative phase of maize cultivation resulted in a 25% decrease in grain output.

Table 2. Means of studied traits under two water quantities across 30 DH lines of maize and two soil conditioners in 2022 season.

Traits	Irrigation (75%)	Irrigation (100%)	Change%	LSD0.05	LSD0.01
DTA (day)	68.18	72.87	6.43	4.96	11.44
DTS (day)	70.20	75.18	6.62*	3.61	8.33
ASI (day)	2.02	2.31	12.58	1.46	3.38
PH (cm)	83.43	117.50	29.00**	8.76	20.22
EH (cm)	27.78	30.07	7.61	3.27	7.55
LANG (^{o})	27.39	27.56	0.60	2.39	5.51
EPP	2.02	2.31	12.74	0.53	1.21
100KW (g)	20.44	20.61	0.79**	0.06	0.15
RPE	11.25	11.34	0.78	0.43	0.99
KPR	19.32	19.47	0.78	5.01	11.56
KPP	472.95	462.53	-2.25	136.09	313.92
GYPP (g)	96.76	102.73	5.81	18.83	43.43
GYPH (ton)	4.15	4.40	5.81	0.81	1.86

*****and ****** indicate significance at 0.05 and 0.01 probability level, respectively**.** **DTA**= Days to anthesis, **DTS**= Days to silking, **ASI**= Anthesis silking interval, **PH**= Plant height, **EH**= Ear height, **LANG**= Leaf angle, **EPP**= Ears plant^{-1}, **100KW**= hundred Kernel weight, **RPE**= rows/ear, **KPR**= Kernels row^{-1}, **KPP**= Kernels plant^{-1}, **GYPP**= Grain yield plant^{-1}, **GYPT**= Grain yield hectar^{-1}. Negative Ch (-) refers to increase

2. Impact of Soil Conditioners :

The impact of soil conditioners on DHLs maize growth and yield characteristics in all the DHLs maize (Doubled Haploid Lines) during the year 2022 as presented in Table (3). The data reveals that the use of soil conditioners had a significant impact on all the traits under study. Specifically, the addition of hydrogel resulted in a reduction of 40.4% in the number of days between the ASI (Anthesis-Silking Interval), leading to

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

enhanced pollination and fertility in plants. This ultimately translated into an increase in yield and its component, in which the increase in grain yield per plant and hectare was 23.45% for both traits. several researchers have documented the same result according to soil conditioners, Khan and Jan **[22]**, reported the use of farmyard manure as a soil conditioner has been found to significantly affect the growth characteristics and quality of maize, leading to higher crop growth rate, leaf area, plant height, biological yield, and grain yield, and Tanure et al **[23]** found , the incorporation of biochar as a soil conditioner has been shown to improve water retention, reduce bulk density, and enhance fertility in the soil, which can positively impact maize growth and physiology, Furthermore, recorded the reuse of sediment as a soil conditioner has been found to improve water retention capacity and increase organic carbon content, potentially reducing irrigation needs and preventing nutrient deficiencies in maize crops **[24]**.

Table 3. Means of studied traits under soil conditioners across 30 DH lines of maize and two water quantities in 2022 season.

Traits	Without (SC)	With (SC)	Ch %	LSD0.05	LSD0.01
DTA (day)	69.41	71.64	3.11	2.44	4.05
DTS (day)	71.94	73.44	2.05*	1.41	2.34
ASI (day)	2.53	1.80	-40.39	2.15	3.57
PH (cm)	99.63	101.30	1.65**	0.82	1.36
EH (cm)	30.78	27.07	-13.69**	2.63	4.36
LANG (°)	27.17	27.78	2.20	1.09	1.81
EPP	1.98	2.35	15.84*	0.28	0.47
100KW (g)	20.64	20.41	-1.14**	0.09	0.15
RPE	10.96	11.63	5.73**	0.47	0.78
KPR	18.63	20.16	7.59**	0.67	1.12
KPP	410.88	524.60	21.68**	64.89	107.61
GYPP (g)	86.50	112.99	23.45**	4.84	8.03
GYPH(ton)	3.71	4.84	23.45**	0.21	0.34

*****and ****** indicate significance at 0.05 and 0.01 probability level, respectively**.** **DTA**= Days to anthesis, **DTS**= Days to silking, **ASI**= Anthesis silking interval, **PH**= Plant height, **EH**= Ear height, **LANG**= Leaf angle, **EPP**= Ears plant^{-1}, **100KW**= hundred Kernel weight, **RPE**= rows/ear, **KPR**= Kernels row^{-1}, **KPP**= Kernels plant^{-1}, **GYPP**= Grain yield plant^{-1}, **GYPT**= Grain yield hectar^{-1}, Negative Ch (-) refers to increase.

3. Mean performance of Doubled Haploid Lines (DHLs)

The table (4) compared the performance of different DH lines of maize under sandy soil conditions in terms of grain yield. By comparing the GYPP and GYPH values, the Five lines (L94,L380,L260,L39,L525) have higher yields per plant and per hectare; Where is the line 94 recorded the highest value by 167.60 g for GYPP and 7.18 ton for GYPH, on the other side, the lowest DH line were L618and L66 by recorded 51.91 g and 47.59 g for GYPP and 2.22 ton and 2.04 ton for GYPH, respectively. These values provide insights into the highly different between the performance of the DH maize lines and that may be useful for maize breeder to find differences in mating design and hybrid vigor in the future.

Journal of Chemical Health Risks
www.jchr.org
JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

Table 4. Means of grain yield per plant (GYPP) (g) and grain yield per hectare (GYPH) (ton) traits of all DHL of maize under sandy soil across 2022

Line	GYPP (g)			GYPH (ton)			
L23	69.43	L313	100.70	L23	2.97	L313	4.31
L39	137.57	L334	110.48	L39	5.89	L334	4.73
L54	126.35	L380	139.55	L54	5.41	L380	5.98
L67	126.85	L404	116.93	L67	5.43	L404	5.01
L91	86.45	L440	66.21	L91	3.70	L440	2.84
L94	167.59	L462	112.35	L94	7.18	L462	4.81
L104	85.13	L479	85.16	L104	3.65	L479	3.65
L106	67.58	L508	115.22	L106	2.89	L508	4.94
L142	97.71	L520	117.13	L142	4.19	L520	5.02
L166	47.59	L525	127.92	L166	2.04	L525	5.48
L226	71.49	L526	93.01	L226	3.06	L526	3.98
L243	119.16	L597	63.61	L243	5.10	L597	2.73
L246	98.19	L615	54.22	L246	4.21	L615	2.32
L260	138.73	L617	117.72	L260	5.94	L617	5.04
L294	80.39	L618	51.91	L294	3.44	L618	2.22
LSD 0.05	16.43			LSD 0.05	0.71		
LSD 0.01	21.65			LSD 0.01	0.93		

4. **Performance of interaction between DHL and water irrigation and soil conditioning for grain yield per plant**

In table (5) shown the effect of water irrigation (I) level (100 and 75%) and hydrogel (SC) (with and without) renamed as E1 for (I1 and SC0) (100 water irrigation + without hydrogel adding), E2 for 100% irrigation + with hydrogel), E3 for (75 irrigation + with hydrogel) and E4 for (75% irrigation + without hydrogel). The result show the best performance lines for grain yield per plant under E1 were L462 (176.4g), L94 (169.29 g) and L440 (136.49 g), for E2 the lines L380 (204.4 g) L54 (160.49g) and L525 (156.27g), for E3 the best value were recorded for L94 (205.16g), L525 (203.22g) and L260 (202.9g) and finally for E4 the L94 (158.27g), L39 (119.87g) and L617 (113.2g). These results provide insights into the relative performance of the different maize lines in terms of grain yield under the specified environments

Table 5. Means of grain yield per plant (GYPP) (g) traits of all DHL of maize under sandy soil across 2022

Lines	E1	E2	E3	E4
L23	63.32	79.08	103.40	31.92
L39	132.98	128.95	168.49	119.87

L54	93.34	160.49	198.43	53.12
L67	105.86	126.43	171.11	103.99
L91	68.73	67.43	150.56	59.07
L94	169.29	137.66	205.16	158.27
L104	128.59	50.02	88.67	73.23
L106	40.90	53.01	146.28	30.12
L142	108.80	114.90	74.84	92.31
L166	68.20	36.02	52.19	33.96
L226	116.82	70.34	58.88	39.94
L243	132.20	94.83	136.08	113.53
L246	75.38	134.26	141.95	41.15
L260	150.08	139.31	202.90	62.65
L294	70.15	86.19	114.84	50.37
L313	109.49	112.95	96.22	84.13
L334	109.69	86.19	187.64	58.38
L380	107.39	204.40	171.39	75.04
L404	160.94	86.09	161.06	59.63
L440	136.49	38.29	40.51	49.57
L462	176.40	126.11	114.78	32.11
L479	62.40	144.35	82.90	51.01
L508	108.81	115.18	160.02	76.88
L520	107.27	98.53	159.39	103.34
L525	81.52	156.27	203.22	70.67
L526	97.01	109.36	78.51	87.18
L597	61.08	73.47	61.69	58.20
L615	37.91	80.18	68.73	30.05
L617	123.72	141.70	92.27	113.22
L618	48.03	81.61	36.51	41.49

Principal Component Analysis

To display the genetic variability among maize DHLs, a principal component analysis (PCA) conducted on the data from all environments of DHLs for maize of standardized data was applied to display maize trait relationships, and its application in genotype 5characterization and comparison (Table 5) during the 2022. The table shows the loadings of each trait on the first two principal components (PC1 and PC2). Additionally, the eigenvalues, variability percentages, and cumulative percentages are provided. The PCA results indicate the relationships and patterns among the different traits. Traits with higher loadings on PC1 include DTA, DTS, ASI, EH, and EPP, while traits PH, LANG, 100KW, RPE, KPR, KPP, GYPP, and GYPH have higher loadings on PC2. These loadings represent

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

the contribution of each trait to the overall variability captured by the principal components.

Furthermore, the eigenvalues represent the amount of variability explained by each principal component. In this case, PC1 explains 34.46% of the total variability, while PC2 explains 17.61%. The cumulative percentages indicate the cumulative amount of variability explained by each principal component, with PC1 and PC2 accounting for 34.46% and 52.07% of the total variability, respectively. Principal component analysis (PCA), a multivariate statistical technique, has been used in a number of studies **[25, 26, 27]** to assess the degree of genetic diversity within crop germplasm and to condense a large number of observed traits into a smaller set of traits that have the greatest potential to separate genotypes.

Table 6. Principal component analysis (PCA) for all data across all environments of DHLs of maize in season 2022**.**

Traits	PC1	PC2	PC3	PC4	PC5	PC6	PC7
DTA	-0.39	-0.55	0.69	0.20	-0.11	-0.01	-0.07
DTS	-0.40	-0.50	0.73	0.17	-0.07	0.06	-0.07
ASI	-0.13	0.47	0.32	-0.36	0.33	0.63	-0.03
PH	0.06	0.49	0.30	0.51	0.39	-0.16	0.46
EH	-0.06	0.47	-0.09	0.67	-0.32	0.33	-0.19
LANG	0.36	-0.40	0.03	0.16	0.72	0.02	-0.24
EPP	-0.12	0.76	0.37	-0.06	0.00	-0.30	-0.37
100KW	0.72	-0.37	-0.33	0.29	-0.01	0.14	-0.10
RPE	0.74	-0.18	0.08	-0.18	0.05	-0.13	-0.07
KPR	0.73	-0.17	0.32	-0.17	-0.29	0.21	0.27
KPP	0.74	0.32	0.48	-0.19	-0.12	-0.14	-0.01
GYPP	0.96	0.08	0.10	0.12	-0.03	0.02	-0.07
GYPH	0.96	0.08	0.10	0.12	-0.03	0.02	-0.07
Eigenvalue	4.48	2.29	1.82	1.14	1.01	0.72	0.55
Variability (%)	34.46	17.61	14.03	8.80	7.75	5.57	4.21
Cumulative %	34.46	52.07	66.11	74.91	82.66	88.23	92.44

PC1-PC7= Principal component, **DTA**= Days to anthesis, **DTS**= Days to silking, **ASI**= Anthesis silking interval, **PH**= Plant height, **EH**= Ear height, **LANG**= Leaf angle, **EPP**= Ears plant^{-1}, **100KW**= hundred Kernel weight, **RPE**= rows/ear, **KPR**= Kernels row^{-1}, **KPP**= Kernels plant^{-1}, **GYPP**= Grain yield plant^{-1}, **GYPT**= Grain yield hectar^{-1},

The GT-biplot, visualizes the relationship between PC1 and PC2 for the 30 DH lines and 13 combined traits across four environments (Fig.1). This plot provides a graphical representation of how the different traits and DH lines are positioned about the principal components.

Overall, PCA helps to identify patterns, associations, and the relative importance of different traits in the dataset, providing valuable insights for further analysis and decision-making in maize breeding programs.

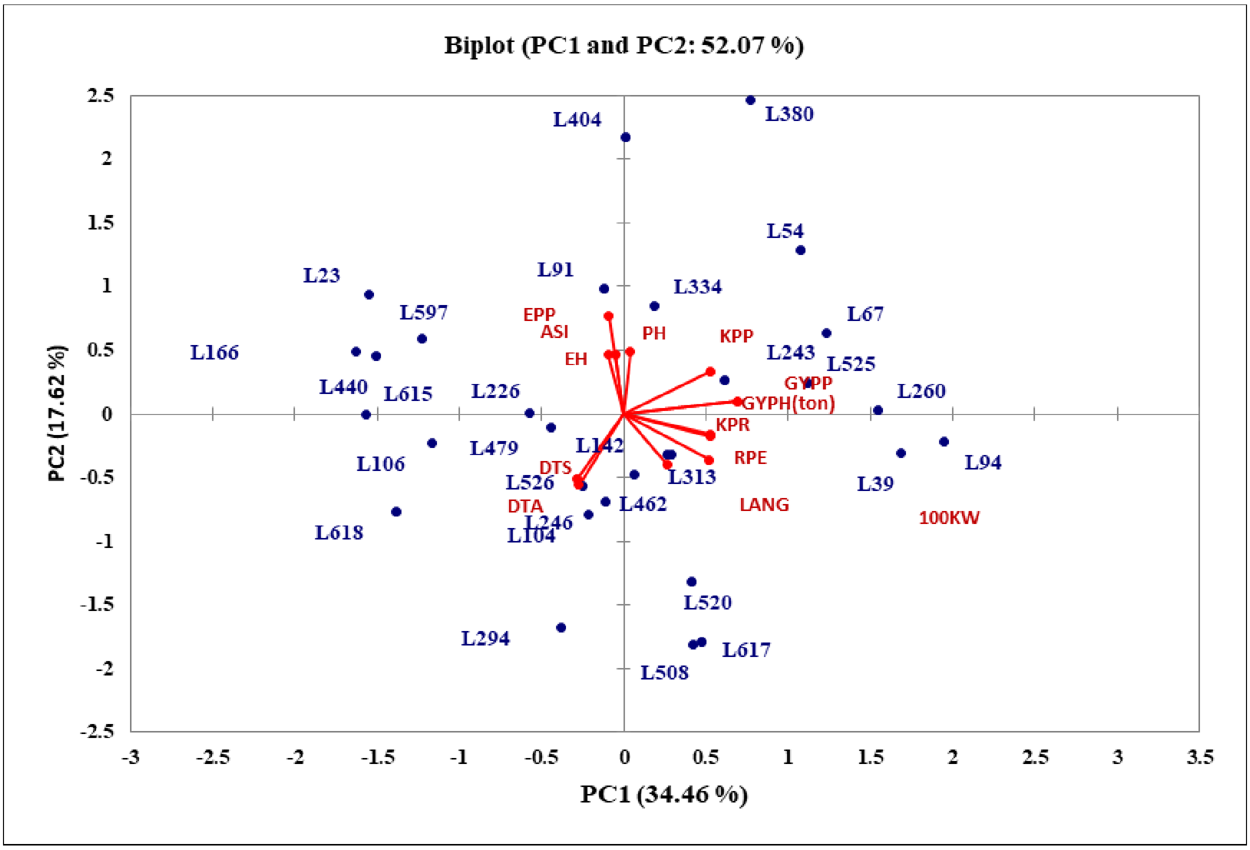

Fig 1. GT-biplot showing the relationship between PC1 and PC2 for 30 DH lines and 13 traits combined across 4 environments.

Figure (1) showed four groups of traits versus four groups of genotypes, namely GYPP, GYPH, KPP, and PH for the DHLs; L54, L67, L243, L260, L380 and L334 in the first group, EPP, EH and ASI for the DHLs; L23, L91, L166, L226, L404, L440, L597 and L615 in the second group, RPE, GFC, ASI and, DTA and DTS for the DHLs; L 104, L106 , L142, L246, L294, L479, L526 and L618 in the third group, KPR, RPE, LANG and 100-KW for the DHLs; L39, L94 , L313 , L462 ,L508 , L520 and L617 in the 4th group. **Yan [28]** was indeed one of the early advocates of the biplot method, which has since been widely used in various fields, including agriculture. The biplot approach has been particularly effective in evaluating the relationships between different characteristics, such as plant traits and grain production in maize, as demonstrated in several studies. In a study published in Scientific Reports **[29]** used the GGE biplot analysis to evaluate the grain yield of drought-tolerant maize hybrids in Inner Mongolia. The GGE biplot was employed to assess the performance of maize in drought-tolerant hybrid selection trials, providing insights into genotype-environment interactions and stability. Another study, published in Agronomy, utilized the GGE biplot graphical technique to assess the stability of maize hybrids. The study focused on evaluating the grain yield and stability of hybrids using the biplot of average environment, demonstrating the practical application of the GGE biplot in assessing the performance of maize hybrids across different environments. The biplot method, including the GGE biplot, has proven to be a valuable tool for evaluating the relationships between different characteristics, such as plant traits and grain production in maize, and assessing the performance and stability of maize lines across various environments.

Agglomerative Hierarchical Clustering (AHC) analysis

The dendrogram (Fig.2) of the DH lines produced from the entire linkage method's standardized phenotypic data across all conditions. The 30 DH lines were divided into three categories by the analysis. Group 1

Journal of Chemical Health Risks
www.jchr.org
JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

consisted of seven lines: L91, L104, L142, L226, L246, L313, L334, L404, L462, L479, L526 and L597 in one subgroup and the second group were L23, L106, L166, L294, L440, L617, and L618 in another. Eleven DHLs were in the third group; they are very closely linked and are identified as L39, L54, L67, L94, L243, L260, L380, L508, L520, L525 and L617. Every subgroup's DHLs are closely interrelated.

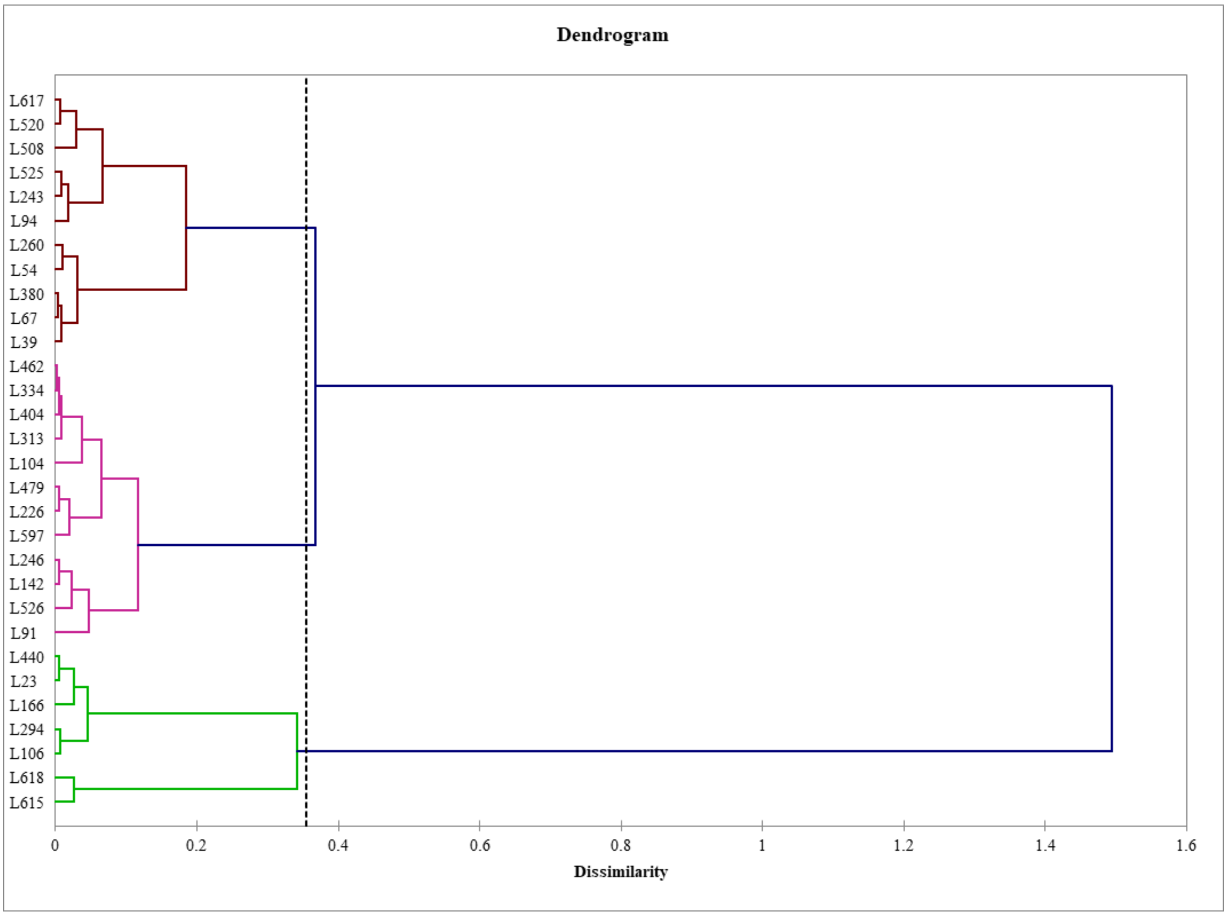

Fig. 2. Dendrogram of 30 DHLs of maize based on 13 traits measured across all environments using the average method of clustering.

Dissimilarity Euclidean coefficients based on phenotypic traits

The table (7) provided represents the dissimilarity coefficients based on yield and its components analysis among 30 DHLs (Double Haploid Lines) across different environments. The dissimilarity coefficients are calculated using the Euclidean distance. Dissimilarity Euclidean coefficients indicated that the genotype L618 was the most dissimilar with each of L39 (0.98), L54 (0.96), L 67(0.91),L91 (0.62), L94 (0.94), L104 (0.89), L142 (0.73), L166 (0.50), L226 (0.67), L243 (0.76), L246 (0.67), L260 (1.04), L313 (0.82), L334 (0.83), L380 (0.94), L404 (0.88), L462 (0.80),L479 (0.68), L508 (0.77), L520 (0.80), L525 (0.88), L597 (0.71) and L617 (0.74) lines, since L618 exhibited the highest dissimilarity And L615 also was dissimilar with all DHLs Euclidean coefficients with these lines; so these pairs of lines are the most unrelated lines and that can help maize breeder to find variations in breeding programs.

Journal of Chemical Health Risks
www.jchr.org
JCHR (2024) 14(2), 1199-1211 | ISSN:2251-6727

Table 7. Discuss the following table Dissimilarity Euclidean coefficients based on yield and its components analysis among 30 DHLs across environments

	L23	L39	L54	L67	L91	L94	L104	L106	L142	L166	L226	L243	L246	L260	L294	L313	L334	L380	L404	L440	L462	L479	L508	L520	L525	L526	L597	L615	L617
L39	0.64																												
L54	0.57	0.18																											
L67	0.56	0.12	0.17																										
L91	0.26	0.47	0.43	0.36																									
L94	0.64	0.25	0.37	0.21	0.43																								
L104	0.52	0.38	0.28	0.39	0.49	0.57																							
L106	0.16	0.57	0.52	0.50	0.29	0.60	0.43																						
L142	0.39	0.27	0.27	0.21	0.27	0.33	0.32	0.31																					
L166	0.21	0.76	0.67	0.69	0.41	0.80	0.56	0.25	0.52																				
L226	0.29	0.44	0.36	0.39	0.30	0.54	0.25	0.20	0.23	0.34																			
L243	0.44	0.26	0.30	0.18	0.24	0.22	0.43	0.39	0.15	0.59	0.34																		
L246	0.32	0.34	0.32	0.29	0.25	0.41	0.31	0.23	0.10	0.44	0.17	0.21																	
L260	0.67	0.17	0.14	0.23	0.54	0.41	0.29	0.60	0.34	0.76	0.43	0.38	0.39																
L294	0.23	0.54	0.52	0.48	0.29	0.55	0.46	0.12	0.28	0.34	0.25	0.35	0.23	0.59															
L313	0.44	0.30	0.20	0.26	0.36	0.45	0.20	0.36	0.19	0.51	0.19	0.30	0.20	0.26	0.37														
L334	0.44	0.25	0.15	0.20	0.32	0.39	0.23	0.38	0.16	0.53	0.22	0.24	0.18	0.25	0.38	0.10													
L380	0.58	0.11	0.14	0.08	0.40	0.24	0.38	0.53	0.24	0.71	0.41	0.21	0.31	0.21	0.51	0.27	0.21												
L404	0.48	0.22	0.10	0.18	0.36	0.37	0.25	0.43	0.20	0.58	0.27	0.25	0.23	0.21	0.44	0.14	0.07	0.17											
L440	0.11	0.65	0.60	0.58	0.30	0.66	0.53	0.12	0.40	0.20	0.30	0.45	0.32	0.69	0.20	0.46	0.46	0.60	0.51										
L462	0.42	0.25	0.18	0.21	0.31	0.38	0.23	0.35	0.13	0.52	0.20	0.22	0.15	0.26	0.35	0.10	0.05	0.22	0.09	0.44									
L479	0.28	0.38	0.30	0.32	0.24	0.46	0.27	0.22	0.16	0.38	0.09	0.26	0.11	0.40	0.25	0.17	0.16	0.34	0.22	0.31	0.14								
L508	0.59	0.40	0.51	0.39	0.47	0.31	0.58	0.51	0.32	0.75	0.53	0.30	0.38	0.53	0.43	0.50	0.47	0.42	0.48	0.58	0.44	0.47							
L520	0.52	0.25	0.35	0.25	0.39	0.25	0.43	0.45	0.19	0.67	0.41	0.18	0.26	0.38	0.40	0.36	0.31	0.27	0.32	0.52	0.28	0.34	0.19						
L525	0.55	0.18	0.25	0.10	0.35	0.14	0.45	0.50	0.22	0.69	0.42	0.13	0.30	0.31	0.46	0.32	0.26	0.14	0.25	0.57	0.25	0.34	0.32	0.21					
L526	0.34	0.40	0.41	0.36	0.29	0.42	0.41	0.29	0.20	0.50	0.31	0.24	0.18	0.49	0.27	0.35	0.31	0.37	0.35	0.34	0.28	0.25	0.32	0.23	0.35				
L597	0.34	0.47	0.39	0.45	0.39	0.61	0.22	0.28	0.30	0.39	0.16	0.42	0.24	0.45	0.34	0.25	0.26	0.45	0.31	0.35	0.26	0.19	0.57	0.44	0.49	0.31			
L615	0.53	1.11	1.08	1.02	0.70	1.04	1.02	0.60	0.85	0.56	0.79	0.87	0.80	1.17	0.61	0.94	0.95	1.05	0.99	0.51	0.92	0.80	0.91	0.93	0.99	0.75	0.84		
L617	0.44	0.26	0.32	0.23	0.32	0.30	0.38	0.36	0.10	0.59	0.32	0.15	0.17	0.37	0.31	0.28	0.24	0.26	0.27	0.44	0.22	0.26	0.24	0.11	0.22	0.18	0.36	0.87	
L618	0.45	0.98	0.96	0.91	0.62	0.94	0.89	0.48	0.73	0.50	0.67	0.76	0.67	1.04	0.47	0.82	0.83	0.94	0.88	0.43	0.80	0.68	0.77	0.80	0.88	0.62	0.71	0.23	0.74

In contrast, dissimilarity Euclidean coefficients indicated that the most closely related lines based on phenotypic traits; i.e. those exhibited the lowest dissimilarity Euclidean coefficients, the lines L 243, L 246 and L617 were similarity with most DHLs in this experiment.

The pair L39 and L23 is 0.64, indicating a relatively high dissimilarity between DHL L39 and DHL L23. Similarly, the pair of L67 and L54 was 0.17, suggesting a lower dissimilarity between DHL L67 and DHL L54. These dissimilarity coefficients provide a measure of the genetic distance or dissimilarity between the DHLs based on their yield and its components across different environments. The coefficients can be used to assess the diversity and similarity among the DHLs and to understand the relationships between them in terms of their performance in various environments and that relationship can be useful for maize breeder in making hybrids. Although phenotypic analysis for assessment of genetic diversity presents many limitations as low polymorphism and influence of environment on morphological expression **[30]**

4. Conclusion

There were significant genetic variations between the DH lines for every variable that was examined, suggesting that lines could be chosen for better performance under particular drought stress conditions. The work offers insightful information about the application of DH lines in maize research as well as the discovery of genomic areas linked to drought stress and agronomic features in various DH lines.

Funding

This research was funded by the "Science, Technology & Innovation Funding Authority in Egypt, (STDF) "Young Researchers Grant (STDF-YRG)", and "Call number (10), Project number (43264) during the project "Developing drought tolerant hybrids of maize by using conventional and biotechnological techniques".

Acknowledgments

The authors are grateful to the "Science, Technology & Innovation Funding Authority (STDF)" and its staff members for their support to carry out this work.

Thanks are also extended to “National Research Centre (NRC), Cairo, Egypt”

References:

1. Bhorat H., Ewinyu A., Lilenstein K., Rooney C., Steenkamp F. and Thornton A. (2019). Economic complexity and employment expansion: The case of South Africa.
2. Ibrahim H. A. and Baqutayan S. M. (2022). Sudan's Government Interventions: A Review Article On How To Overcome The Gap Between Wheat Production And Consumption. Journal of Science, Technology and Innovation Policy, 8(1), 58-68.
3. Ahmed Y. N. and Elasraag Y. H. (2023). The impact of population growth on food security in Egypt. Egyptian Journal of agricultural economics, 33(3), 1218-1231.
4. Kaur H., Kyum M., Sandhu S., Singh G. and Sharma P. (2023). Protocol optimization and assessment of genotypic response for inbred line development through doubled haploid production in maize. BMC Plant Biology, 23(1), 219.
5. Khan S. U., Zheng Y., Chachar Z., Zhang X., Zhou G. and Zong N. (2022). Dissection of maize drought tolerance at the flowering stage using genome-wide association studies. Genes 13, 564. doi:10.3390/genes13040564.
6. Chaikam V.V.R. (2016). Doubled Haploid (DH) Technology in Maize Breeding: Application and Technology for Production of DH Lines.
7. Chaikam V., Molenaar W., Melchinger A. E., and Boddupalli P. M. (2019). Doubled haploid technology for line development in maize: technical advances and prospects. Theoretical and Applied Genetics, 132, 3227-3243
8. Trindade R.D.S. (2022). The use of doubled haploid technology for lines development in maize breeding. Revista Brasileira de Milho e Sorgo, 21.
9. Chen Y.R., Lübberstedt T. and Frei U.K. (2023). Development of doubled haploid inducer lines facilitates selection of superior haploid inducers in maize. Frontiers in Plant Science, 14.
10. Steel R.G.D. and J.H. Torrie (1980). Principles and procedures of statistics. A biological apporoach. 2nd ed. McGraw Hill Inc., New York.
11. Tollenaar M. and Wu J. (1999). Yield Improvement in Temperate Maize is Attributable to Greater Stress Tolerance. Crop Science, 39, 1597-1604.
12. Al- Naggar A.M.M., R. Shabana and A.M. Rabie (2011). Per se performance and combining ability of 55 newly – developed maize inbred lines for tolerance to high plant density. Egyptian Journal of Plant Breeding. (15) 5: 59-82.
13. Al- Naggar A.M.M., R. Shabana, M.M.M. Atta and T.H. Al-Khalil (2014). Genetic parameters controlling some maize adaptive traits to elevated plant densities combined with reduced N-rates. World Research Journal of Agronomy 3 (1), 70-82.
14. Al-Naggar A.M.M., R.A. Shabana, M.M.M. Atta and T.H. Al-Khalil (2015). Maize response to elevated plant density combined with lowered N-fertilizer rate is genotype-dependent. The Crop Journal. 3 (2), 96-109.
15. Duvick D.N. (1984). Genetic contributions to yield gains of U.S. hybrid maize, 1930 to 1980. p. 1–47. In W.R. Fehr (ed.) Genetic contribu- tions to yield gains of five major crop plants. CSSA Spec. Publ. 7 ASA and CSSA, Madison, WI.
16. Al-Naggar A.M.M., A.E.I. Ragab, S.S Youssef and M.R. Al- Bakry (2004). New genetic variation in drought tolerance induced via irradiation and hybridization of Egyptian cultivars of bread wheat.Egyptian Journal of Plant Breeding 8, 353-370.
17. Al- Naggar A.M.M., W.A. El-Murshedy and M.M.M.Atta (2008 a). Genotypic variation in drought tolerance among fourteen Egyptian maize cultivars. Egyptian Journal of Applied Sciences 23 (2B), 527-542.
18. Al- Naggar A.M.M., R. Shabana, A.A. Mahmoud and S.A.M. Shaboon (2008 b). Genetic improvement of maize for low-soil nitrogen tolerance via S1 recurrent selection. Egyptian Journal of Plant Breeding 12 (2), 255-277.
19. Al- Naggar A.M.M., R. Shabana, A.A. Mahmoud, M.E.M. Abdel El-Azeem, S.A.M. Shaboon (2009). Recurrent selection for drought tolerance improves maize productivity under low-N conditions. Egyptian Journal of Plant Breeding. 13 : 53-70.
20. Al- Naggar A.M.M., S.M. Soliman and M.N. Hashimi (2011a). Tolerance to drought at

flowering stage of 28 maize hybrids and populations. Egyptian Journal of Plant Breeding 15 (1), 67-87.

21. Denmead O. T. and R. H. Shaw (1960). The Effect of Soil Moisture Stress at Different Stages of Growth on the Development and Yield of Corn. Agronomy Journal 52(5): 271-274.
22. Braga B. B., Costa C. A., Lima G. D., de Lacerda, C. F., Foerster, S., Brosinsky, A., and Medeiros, P. H. (2023). Reuse of sediment as a soil conditioner in a semiarid region dominated by subsistence farming: sediment characterization at the regional scale and effects on maize crop. Journal of Soils and Sediments, 1-17.
23. Amarasinge Y.P.J., Wijesinghe G. and Pushpakumara R.W., (2016). Multivariate Analysis, Genetic Diversity and Phenotypic Correlation of Nineteen Exotic Groundnut Accessions. J. Agrisearch, 3(1): 7-12.
24. Makinde S.C.O. and Ariyo, O.J., (2010). Multivariate analysis of genetic divergence in twenty-two genotypes of groundnut (Arachis hypogaea L.). J. Plant Breeding & Crop Sci., 2(7): 192-204.
25. Makinde S. and Ariyo O., (2013). Genetic divergence, character correlations and heritability study in 22 accessions of groundnut (Arachis hypogaea L.). J. Plant Studies, 2(1): 7-17.
26. Yan W., (2001). GGE biplot-a windows application for graphical analysis of multi-environment trial data and other types of two-way data. Agro. J., 93(5): 1111–1118.
27. Li Y., Bao H., Xu Z., Hu S., Sun J., Wang,Z., and Gao, J. (2023). AMMI an GGE biplot analysis of grain yield for drought-tolerant maize hybrid selection in Inner Mongolia. Scientific Reports, 13(1), 18800.
28. Badu-Apraku B, Akinwale R.O., Franco J. and Oyekunle M., (2012). Assessment of reliability of secondary traits in selecting for improved grain yield in drought and low-nitrogen environments. Crop Sci., 52(5): 2050-2062.