

Phenotypic Characterization and Multivariate Analysis of Fifteen Maize Lines developed by Induced Mutation in Daloa (Côte d'Ivoire)

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Abstract

In Côte d'Ivoire, maize is an important staple food for a large part of the population. However, its culture faces many constraints related to soil degradation, climate change and genetic degeneration of cultivated varieties. In order to create new varieties adapted to these constraints, mutant maize Lines have been developed using the gamma radiation technique. The present study aims to phenotyping 15 mutant Lines from the sixth generation of self-pollination, in order to characterize them and to give information on their genetic diversity. For this, an experiment in a randomized complete block design with three replications was conducted. Thirteen traits were evaluated and they showed wide variation not only between the mutant Lines but also between them and the non-irradiated control. Multivariate analysis structured this variability into five distinct groups with specific traits. The interest of the revealed traits and the future use of these mutant Lines are discussed.

Keywords: maize, gamma radiation, induce mutation, phenotypic characterization

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1. Introduction

Ranked third after wheat and rice, maize (Zea mays L.) is an important crop worldwide (FAO, 2022). In Côte d'Ivoire, it is the major staple diet of rural populations, mainly in the northern savannah region. In this region, it is the most energetic cereal due to its nutritional value and it has important economic stakes (Charcosset & Gallais, 2009). Indeed, it plays a significant role in reducing poverty and improving the food security status for poor families. However, in this region where agriculture production is largely rain-fed, the effects of current changes in climatic conditions constitute a huge limiting factor in maize production. In addition, it is projected that by 2050, global warming trends may render 40 % of the current maize growing areas in Africa unsuitable for varieties available today (Schlenker & Lobell, 2010; Neat, 2013). Hence, it becomes urgent to guide maize breeding towards new high-performance varieties adapted to strong environmental constraints. It is in this context that the present study aims at developing new elite maize varieties adapted to the agro climatic conditions of northern savannah region of Côte d'Ivoire, using the gamma radiation induced mutation technique. The action of these rays induces modifications of the genetic material (DNA) of a living organism such as a crop, thus increasing its variability and generating genotypes capable of adapting to the various constraints linked to their culture (Prouillac, 2006; Katiyar *et al.*, 2022).

Thus, 15 Lines of maize have been obtained after six generation of self-pollination of a maize genotype obtained by gamma irradiation of an appreciated variety in the north of Côte d'Ivoire. Phenotyping of these new maize Lines is highly recommended as the first step of the improvement program, prior to more in depth studies (Amiteye *et al.* 2019; Temam *et al.* 2020). Hence, the specific objective of the present study was to characterize phenotypicaly these 15 maize lines and to assess their genetic diversity, in order to generate information that could potentially help breeders to improve the crop.

2. Materials and Methods

2.1 Experimental Site

The study was carried out at the experimental farm of the Crop Improvement Laboratory of Jean Lorougnon

Guédé University (UJLoG) located in the town of Daloa (Center-West of Côte d'Ivoire, 130 km from the capital Yamoussoukro). The area was under humid tropical conditions with 1317 mm of rainfall per year. The vegetation, which used to be dense forest, has now disappeared in favor of various cash crops (Sangaré *et al*., 2009). The experiment site was located 6 ° 90 N latitude and 6 ° 37 W with an altitude of 238 m.a.s.l. (above sea level). The soil of the plot was sandy loam texture with good fertility, properly leveled and well drained. The mean temperature and relative humidity during the experiment period ranged from 25 to 35 ° C and 69 to 83 %, respectively.

2.2 Plant Material

The plant material used for this study consisted of 15 lines of maize coming from sixth generation of self-pollination of a maize genotype obtained by gamma irradiation of the EV8728 variety at doses of 200 and 300 grays in Seibersdorf, Austria. The non-irradiated EV8728 variety was used as a control.

2.3 Experimental Design

The experiment was established using a randomized complete block design (RCBD) with three replications. The spacing between adjacent replications was 2 m. Three seeds per hill were sown at a depth of 3 cm and thinned to two plants per hill after two weeks. Each Line was raised in a single-row plot with a row-to-row spacing of 80 cm and plant-to-plant spacing of 40 cm. A plant population of twenty-four plants per row, plot and Line was maintained. Thus, each Line was represented by 72 plants, i.e. 1080 plants for the overall 15 Lines on the experimental field.

2.4 Field Managements

Standard agronomic and management practices were adopted to raise a healthy crop. Daily watering using a Drip Irrigation System was applied uniformly to all plots. Manual weeding by hoeing and handpicking (Figure 1) were carried out when necessary to avoid any competition between the crop and the weeds, and thus allow better crop development.

Figure 1. Experimental Field of the Studied Maize Showing Manual Weeding by Hoeing and Handpicking, and Drip Irrigation System.

2.5 Data Collection

Data were collected on 15 plants of each Line in each replication. Visual observations and measurements were done for this collection. The following traits were evaluated: Days to emergence (DE), emergence rate (ER), plant height (PH), ear height (EH), stem diameter at base (SDB), days to anthesis (DA), days to silking (DS), anthesis-silking interval (ASI), ear length (EL), ear diameter (ED), ear weight (EW), 100 grains weight (GW100), number of rows per ear (NRE), number of grains per row (NGR), number of grains per ear (NSE, NSE = NRE×NGR).

2.6 Statistical Analysis of Data

Microsoft Excel Software (2016 edition) was used to compile the data and the SPSS v22.0 and R v3.6.3 softwares for statistical analyses.

With the SPSS software, the means and the standard errors were calculated and the data were subjected to an Analysis of Variance (ANOVA) to determine the presence of statistically significant differences among cultivars for the traits measured. A p-value of 0.05 or less was considered statistically significant. The Student-Newman-Keuls post-hoc test was used to separate significantly different means.

With the R software, the variables were subjected to two methods of multivariate analyses: Principal Component Analysis (PCA) and Cluster Analysis (CA). PCA was done to transform the original variables into a limited number of uncorrelated new variables and to allow the visualization of differences among the Lines, the identification of groups, and the identification of relationships among Lines and variables. The Eigen Values and Eigen Vectors were computed, which represent the variance and the loadings of the corresponding principal components (PCs). A biplot analysis was carried out based on the two most important PCs to visualize the pattern of total diversity within the germplasm studied. The degree of correlation between the traits and the percentage contribution of each trait to the total diversity were determined. CA was used to group the Lines into various clusters according to genetic distance. The clustering was performed using the genetic distances computed from traits measured. The distance matrix was used to construct a dendrogram based on Ward's aggregation method. The genetic relationships among the Lines and between and within the clusters were determined and analyzed.

3. Results

3.1 Variation in Crop Phenology (Emergence and Flowering)

For the maize Lines studied, the mean values of days to seedling emergence, emergence rate and days to flowering, are recorded in Table 1. There were significant differences in the mean number of days taken by seedlings of Lines to emerge. The mean values ranged from 4.33 to 6.17 days. The Line L5 and the control (T0) took the shortest time to emerge (4.33 days after sowing) while lines L1 and L13 recorded the longest time (6.17 days). The mean values of emergence rates showed significant differences too. The values were ranged from 43.98 % to 93.05 %. The Lines with the lowest rates were L7 and L13 (43.98 and 48.61 % respectively), while Line L4 with 97.22 % exhibited the highest emergence rate. The emergence rate of the control was 93.05 %. The mean values of days to flowering (days to anthesis and days to silking) revealed significant differences. The T0 control showed the shortest time to flowering. Indeed, all the Lines recorded average values significantly higher than those of the T0 control regarding days to anthesis and days to silking. The anthesis-silking intervals varied between 0 and 3.78 days. Seven Lines (L1, L3, L5, L6, L7, L14 and L15) and the control T0 had anthesis-silking intervals inferior to 1 day.

3.2 Variation in Growth and Production Parameters

Table 2 shows the distribution of the mean values of growth and production parameters. Except stem diameter, the values of each parameter studied showed highly significant differences. Concerning plant height, the shortest plants were recorded in Line L2 with an average height of 158.46 cm. Line L7 had tallest plants with 206.33 cm height on average. The mean height of the plants of the T0 control was 200.5 cm.

For the ear height, the control plants gave the highest ear heights (108.43 cm on average). Line L2 presented the shortest ear heights (76.43 cm on average).

The longest ears were observed in the control T01 with an average length of 15.24 cm; while the shortest ears were obtained in Line L3 with a length of 8.41 cm. All other lines presented ears with a length intermediate to these two values. For ear diameters, the control had the largest ears (45.66 mm); followed by Lines L6, L12 and L2 with 42.61, 41.93 and 40.46 mm respectively. The Line L9 had the smallest ear diameters (31.66 mm on average). Like the ear diameters, the control had the highest mass (128.18 g) before Line L12 (100.13 g). The least heavy ears were produced by Line L15 (31.11 g).

The number of grains per ear also contributed to discriminate studied Lines. The control plants had he most grains per ear (487.04 grains on average), followed by Lines L5 and L12 (424.59 and 424.3 grains respectively). With a mean of 141.13 grains, Line L15 exhibited the lowest number of grains per ear.

Furthermore, the heaviest grains were found in Lines L11, L15 and the Control with means of 24.32, 23.65 and 24.16 g for 100 grains weight respectively. Lines L5 and L9 had the least heavy grains (15.44 and 14.35 g 100 grains weight respectively).

Table 1. Mean Values and Standard Errors of the Phenological Traits of the 15 Maize Lines

Lines	DE (days)	ER (%)	DS (days)	DA (days)	ASI (days)
L1	6.17±0.9^{d}	60.65±8.3^{ab}	58±1.8^{e}	58.5±1.5^{de}	0.56±1.7^{a}
L2	5.33±0.5^{bc}	67.13±14.4^{b}	55.91±1.1^{c}	58.8±1.2^{e}	2.95±1.6^{c}
L3	5.5±0.8^{c}	76.39±9.4^{c}	60.48±2.8^{h}	60.4±2.5^{h}	0±0.9^{a}
L4	4.67±0.5^{ab}	97.22±3.5^{f}	53.03±1.5^{ab}	55.9±0.8^{bc}	2.93±1.3^{c}
L5	4.33±0.8^{a}	89.35±11.5^{de}	56.53±1.2^{d}	56.8±1.4^{cd}	0.27±1.2^{a}
L6	5.17±0.4^{bc}	87.5±7.1^{de}	54.83±1.4^{bc}	55.5±1.1^{b}	0.7±1.2^{a}
L7	5.83±0.4^{cd}	43.98±9.5^{a}	58.69±1.4^{f}	59.6±1.1^{f}	0.96±0.8^{a}
L8	5.17±0.4^{bc}	76.39±9.0^{c}	55.9±1.0^{c}	58±0.6^{d}	2.17±0.7^{b}
L9	5±0.0^{b}	77.78±11.6^{cd}	56.62±1.5^{d}	57.7±1.1^{d}	1.13±1.2^{ab}
L10	5.17±0.4^{bc}	91.67±5.5^{e}	56.33±0.9^{d}	58.9±1.2^{e}	2.6±1.3^{c}
L11	5.33±0.8^{bc}	88.43±9.1d^{e}	55.77±1.6^{c}	58.4±1.1^{de}	2.7±1.2^{c}
L12	5.33±0.5^{bc}	74.07±8.5^{bc}	54.2±2.0^{b}	55.3±1.3^{ab}	1.1±1.7^{ab}
L13	6.17±0.4^{d}	48.61±6.7^{a}	58.81±1.1^{f}	62.5±1.7^{i}	3.78±1.8^{d}
L14	5.33±0.5^{bc}	74.07±9.4^{bc}	56.4±1.3^{d}	56.5±1.1^{c}	0.13±0.5^{a}
L15	5±0.6^{b}	81.02±13.7^{d}	59.21±1.4^{g}	59.9±4.1^{g}	0.71±4.7a
T0	4.33±0.5^{a}	93.05±3.8^{e}	51.77±2^{a}	52.3±1.9^{a}	0.6±0.8^{a}
P-Value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

Means followed by le same letter are statistically identical at the 5 % threshold. P-Value: Approximate Probability of Tests, DE: Days to emergence, ER: Emergence rate, DA: days to anthesis, DS: days to silking, ASI: anthesis silking interval.

Table 2. Mean Values and Standard Errors of Growth and Production Parameters of the 15 Maize Lines Studied

Lines	SDB (mm)	EH (cm)	PH (cm)	EL (cm)	ED (mm)	EW (g)	NSE	GW100 (g)
L1	23.4±2.3^{a}	83.5±9.7b	173.46±13.8bc	11.96±2.9bc	37.36±3.5c	51.52±21.5cd	262.22±99.8d	20.97±3.3b
L2	19.5±2.8^{a}	76.43±20.6a	158.46±10.8a	11.34±1.8b	40.46±2.6d	57.38±11.8d	271.73±41.4d	21.53±1.5b
L3	19.8±4.5^{a}	80.83±18.7ab	190.03±18.5de	8.41±2.0a	35.38±1.6b	38.68±7.0b	199.09±46.1b	18.9±3.2ab
L4	20.4±3.6^{a}	86.2±12.7bc	181.93±15.4d	10.5±2.1b	35.48±2.6b	47.16±14.0bc	183.12±51.6ab	24.57±3.0e
L5	22.1±2.4^{a}	97.7±35.2d	170.46±18.1b	12.32±1.9c	37.45±3.7c	60.64±22.7d	424.59±90.5h	15.44±3.3a
L6	21.1±3.5^{a}	99.1±14.4^{e}	196.24±12.3f	12.32±2.5c	42.61±3.2d	80.13±28.8g	293.48±86.3^{e}	24.69±2.9e
L7	30.2±10.8^{b}	98.37±14.0d	206.33±34.4h	12.3±1.5c	35.66±6.1b	52.35±14.7cd	297.92±54.2^{e}	19.04±2.4ab
L8	21.4±3.7^{a}	89.93±9.8c	179.1±13.0c	12.34±1.4c	36.37±2.0c	62.47±14.2d	329.14±57.9g	17.99±2.8ab
L9	18.4±2.1^{a}	94.37±8.4cd	180.23±11.8cd	10.94±2.3b	31.66±3.1a	36.16±14.0ab	269.57±109.9d	14.35±2.1a
L10	21.1±2.2^{a}	100.87±11.7^{e}	199.66±13.6g	13.99±1.3^{e}	38.17±2.8c	68.51±12.6f	313.68±39.2f	21.12±3.0b
L11	21.1±3.2^{a}	98.07±9.3d	197.4±19.5g	13.4±1.3d	35.85±1.9b	63.86±13.0e	245.83±35.0c	24.32±2.1d
L12	22.7±1.8^{a}	93.13±12.6cd	193.56±21.3e	13.87±2.4^{e}	41.93±7.1d	100.13±64.5h	424.3±75.4h	22.91±2.3c
L13	30.3±12^{b}	91.2±12.1c	188.23±33.0de	11.31±0.6b	37.03±2.6c	48.41±9.9c	211.92±41.7b	22.38±1.7bc
L14	20.7±2.8^{a}	80.23±8.2 ab	166.2±7.8ab	10.98±4.3b	36.44±2.8c	41.86±10.7b	231±50.3bc	19.05±2.6ab
L15	25.2±32.4^{ab}	97.63±10.8d	194.63±15.9ef	10.25±1.5ab	33.2±3.4ab	31.11±8.1a	141.13±32.7a	23.65±2.0d
T01	24±2.6^{ab}	108.43±17.5f	200.5±30.7g	15.24±2.0f	45.66±3.6e	128.18±30.2i	487.04±92.8i	24.16±2.8d
P-Value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

Means followed by same letter are significantly identical at the 5 % threshold. P-Value: Approximate Probability of Tests. EL: ear length. ED: ear diameter. EW: ear weight. GW100: 100 grains weight. NSE: number of seeds per ear. PH: plant height. EH: ear height. SDB: stem diameter at base.

3.3 Correlation between the Studied Traits

Table 3 presents the correlation matrix between the different traits studied. The analysis of this matrix indicates a significant correlation (≥ ± 0.50) between some pairs of traits. Thus, strong positive correlations were found between ear size (length and diameter) and ear weight and number of ear grains; between Plant height and ear height; and between days to anthesis and days to silking. On the other hand, strong negative correlations were observed between ear weight and days to flowering (anthesis and silking) and between emergence rate and days to emergence.

Table 3. Correlation Matrix between Studied Traits

	DE	ER	SDB	EH	PH	EL	ED	EW	NSE	GW100	DA	DS	ASI
DE	-												
ER	**-0.82**	-											
SDB	0.441	**-0.65**	-										
EH	-0.42	0.324	0.319	-									
PH	0.021	0.065	0.441	**0.74**	-								
EL	-0.24	0.214	0.154	**0.666**	0.368	-							
ED	-0.21	0.184	0.035	0.269	0.139	**0.653**	-						
EW	-0.36	0.308	0.041	0.54	0.357	**0.839**	**0.896**	-					
NSE	-0.43	0.199	0.002	0.495	0.118	**0.781**	**0.689**	**0.837**	-				
GW100	0.037	0.217	0.186	0.186	0.406	0.27	0.478	0.399	-0.13	-			
DA	**0.679**	**-0.6**	0.365	-0.34	-0.03	-0.54	-0.61	**-0.72**	**-0.65**	-0.2	-		
DS	**0.612**	**-0.59**	0.321	-0.31	-0.02	-0.63	-0.62	**-0.73**	**-0.57**	-0.38	**0.872**	-	
ASI	0.203	-0.08	0.124	-0.08	-0.02	0.118	-0.05	-0.06	-0.23	0.321	0.352	-0.15	-

DE: Days to emergence. ER: emergence rate. EH: ear height. DA: days to anthesis. DS: days to silking. ASI: anthesis silking interval. EL: ear length. ED: ear diameter. EW: ear weight. GW: 100 grains weight. NSE: number of seeds per ear. PH: plant height. EH: ear height. SDB: stem diameter at base

3.4 Principal Component Analysis (PCA)

The principal component analysis (PCA) transformed the 13 raw set of data into 13 factors loadings or principal components, with the first principal component (PC1) contributing the most variability (43.38 %) and the last principal component (PC13) contributing the lowest variability (7.85 x 10^{-7} %). Eigen values are often used to determine how many principal components to retain. Usually, components with Eigen values less than 1 are excluded (Shah *et al*. 2018). The first four PC (PC1, PC2, PC3 and PC4) had Eigen values greater than 1 (Table 4), and showed therefore high significant variability compared to the rest of the PCs which had Eigen values less than 1. These latter PCs had not been considered, as they were not significantly influencing the variability among the maize Lines studied. The percentage of variation explained by the first four PCs, their Eigen value and the factor scores for the 13 traits studied are presented in Table 4. These four PCs cumulatively explained 86.589 % variation for the maize Lines studied. The first two PCs together explained 63.52 % of variation. This value is higher than the reference value of 45.15 %. PC1 (43.38 % variation) had a greater weightage on DE, RE, DA, DS, EH, EL, ED and EW; and PC2 (20.14 % variation) had a greater weightage on SDB and PH. The remaining variation (22.53 %) was contributed by the other two PCs (PC3 and PC4) with greater weightage on the rest of the traits. Thus, PC1 was mainly attributed to phenologic and production parameters and PC2 to growth parameters.

Biplot analysis was carried out based on the first two PCs. The traits and the maize Lines were shown on the biplots (Figure 2 and Figure 3A, respectively) to clearly visualize their associations and differences. The scatter plot of the maize Lines (Figure 3A) presented the diversity pattern of the Lines studied and classified them into five groups. The group 1 contained exclusively the Lines L1, L3 and L15. The group 2 comprised Lines L7 and L13. Lines L14, L9, L8, L5, L4 and L2 formed group 3. Group 4 contained Lines L12, L11, L10 and L6; and the latest group (5) contained exclusively the control T01.

Table 4. Principal Component Analysis of 13 Traits in the Maize Lines Studied Showing Eigen Vectors, Eigen Values, Total and Cumulative Percentage of Variance Explained by the First Four PC Axes

TRAITS	PC1	PC2	PC3	PC4
DE	**-0,635**	0,552	0,074	-0,37
ER	**0,586**	-0,57	0,24	0,47
SDB	-0,145	**0,872**	-0,176	-0,005
EH	**0,629**	0,415	-0,255	0,544
PH	0,31	**0,655**	-0,059	0,59
EL	**0,816**	0,366	-0,054	-0,133
ED	**0,775**	0,204	0,102	-0,445
EW	**0,925**	0,241	-0,018	-0,222
NSE	**0,799**	0,062	-0,438	-0,31
GW100	0,353	0,384	**0,717**	0,107
DA	**-0,865**	0,344	0,063	0,08
DS	**-0,857**	0,223	-0,317	0,124
ASI	-0,108	0,27	**0,734**	-0,077
Eigen value	5,64	2,618	1,526	1,403
Total variance (%)	43,382	20,137	11,74	10,793
Cumulative variance (%)	43,382	63,519	75,259	86,052

Bold values are the highest contribution for each trait to total variance in the respective axes. DE: Days to emergence. ER: emergence rate. EH: ear height. DA: days to anthesis. DS: days to silking. ASI: anthesis silking interval. EL: ear length. ED: ear diameter. EW: ear weight. GW: 100 grains weight. NSE: number of seeds per ear. PH: plant height. EH: ear height. SDB: stem diameter at base

3.5. Cluster Analysis (CA)

A hierarchical ascending classification of the studied Lines was performed. The dendrogram (Figure 3B) generated with Ward's aggregation method, at 50 % level of similarity, confirmed the grouping in five distinct groups as revealed by the PCA.

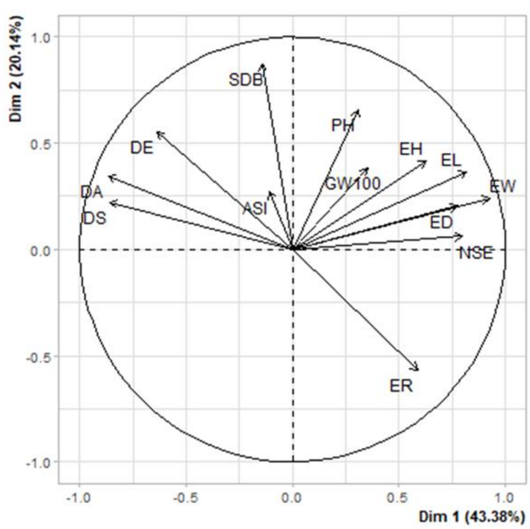

Figure 2. Plot of Components Weight of the 13 Traits of the Studied Maize

(A) (B)

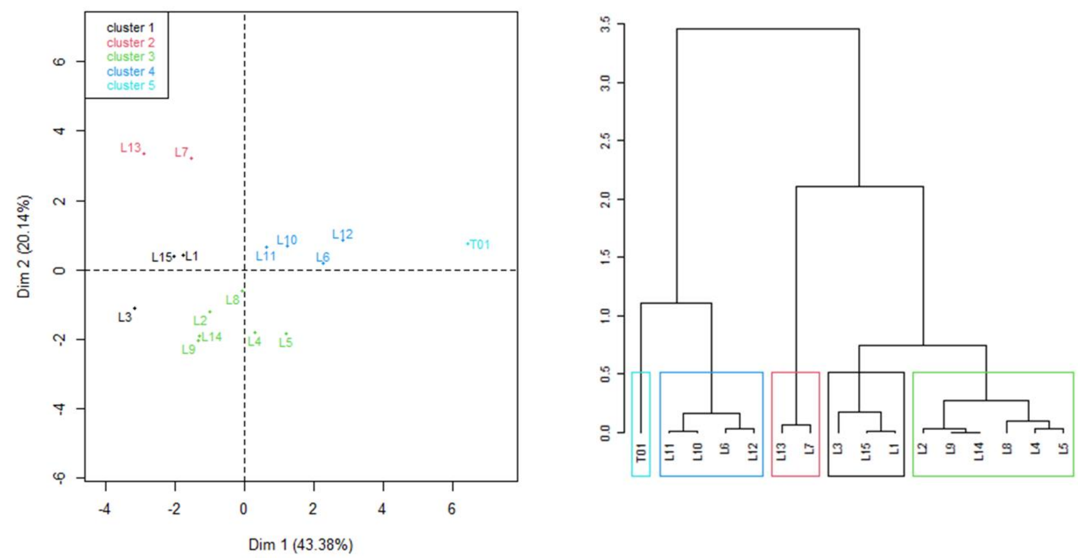

Figure 3. Principal Component (PCA) and Cluster Analysis (CA): A) Scatter Plot of the First and Second PC for the Maize Lines Studied, Showing Five Groups; B) CA Dendrogram Showing Five Clusters.

4. Discussion

Phenotypic characterization is the first essential step in genetic resources description (Kaur *et al.,* 2022; Sanchez *et al*., 2023. It provides breeders with important information necessary for their breeding program (Jung *et al*., 2021). The phenotyping of the 15 maize Lines used in the present study was done to characterize them and to give information on their genetic diversity. The traits evaluated showed wide variation not only between the maize Lines studied but also between these Lines and the control. First, these results indicate that the traits evaluated are suitable to discriminate the genotypes studied. According to Louette (1994), vegetative traits (plant height and ear insertion height), length of vegetative stage (days to flowering) and certain ear parameters, such as diameter, are the main criteria to identify maize varieties. Second, the significant differences observed could be explained by the effects of gamma radiation applied to the grains of the control variety which made it possible to obtain the Lines studied. Such exposure of grains to gamma radiation could have caused modifications to their genome, which could be the origin of the variability observed. Indeed, according to Feuk *et al* (2006) this practice leads to random modifications at several levels of the genome, which could allow obtaining several lines differing from each other at different levels.

Emergence parameters (days to emergence and emergence rate) are important criteria for assessing germination capacity of each Line. Successful germination alone does not guarantee successful emergence of a maize crop. Elongation of the mesocotyl must elevate the coleoptile to the soil surface before the inner true leaves emerge from the protective tissue of the coleoptiles (Nielsen, 1995). In our study, emergence varied between 4 and 6 days after planting. This is a narrower range than that reported by Nemergut *et al*. (2021), who evaluated in-field maize emergence in different soils and at different planting depths and reported emergence between 4 and 13 days after planting. Except for lines L7 and L13 which showed emergence rates of 43.98 and 48.61 % respectively, the other Lines recorded rates above 50 %. Overall, these results imply that the lines studied have a good germination capacity.

Flowering time (or vegetative stage length) is a complex trait that displays a large range of variability. More than 60 QTLs for flowering time with five to six major chromosomal clusters/regions have been detected in maize and each QTL contributes to a small part of the phenotypic variation (Durand, 2012). In the present study, the length of the vegetative stage was shorter for the control (51.77 DA and 52.3 days DS) than the studied Lines (53.03 - 60.48 DA and 55.3 - 62.5 DS). This result suggests that the mutations induced in the Lines by the gamma irradiation slightly lengthened their vegetative stage. In addition, the anthesis-silking interval was relatively small, less than 4 days. This indicates the synchronization of male and female flowering of these Lines.

Our results are in line with those of Mercer & Perales (2019), who reported that the time interval required between male and female flowering in maize is less than five days. Moreover, a strong positive correlation was found between these two parameters (male and female flowering). These results are consistent with those of Moussa *et al* (2018) who showed that duration of male and female sowing-flowering cycles are positively and significantly correlated.

Ear height and plant height are of great importance for the morphological characterization of maize variety. Both are important agronomic traits in maize that directly affect nutrient utilization efficiency and lodging resistance and ultimately relate to maize yield (Wang, 2023). Apart from Line L18, all studied Lines were shorter and had shorter ear insertion heights than the control. These results showed that gamma irradiation reduced the growth of the mutants. These results are in line with those of Singh *et al* (2009) who studied generation 1 to 3 mutants of wheat obtained by gamma irradiation, and found shorter mutants compared to the non-irradiated parent. This reduction can be beneficial and desirable breeding efforts as these mutants can be better adapted to high wind areas to limit lodging damage (Useni *et al,* 2012). Indeed, according to Zhao *et al*. (2022) growth reduction reduces the center of gravity, increases the mechanical strength of the basal stem and increases anchorage strength of brace roots in maize; which leads to increased resistance to lodging.

The heaviest, largest and longest ears were recorded in the control. These results indicate that irradiation had a deleterious effect on these traits. This is consistant with the results of Irfaq & Nawab (2001) who also observed a decrease in the number of grains in wheat under the effect of gamma radiation.

PCA grouped the 15 mutant Lines studied in five distinct group. This result was confirmed by CA with the dendrogram that grouped the Lines under five major clusters. Clustering is a technique that can conveniently show the pattern of genetic relationships or proximity among the genotypes studied such that each group is homogeneous with respect to certain characteristics and each group should be different from other groups with respect to the same characteristics (Gangopadhyay *et al.* 2017). Thus, the Lines in Group 1 are characterized by short ears and relative late female flowering. Group 2 is characterized by high stem diameter and relative late seedling emergence and male flowering. Group 3 is characterized by short plants and relative light grains mass. Group 4 is characterized by longest ears. Group 5, is characterized by plants with earliest emergence and flowering, and highest values for the ear parameters (mass, diameter, number of grains and ear length). This structuring of the diversity showed for each groups, specific interesting traits. This offers the possibility of improving the crop by pyramiding the useful genes of each cluster in a new genotype. For this, the use of the best Mutant from each cluster in a hybridization program could generate the expectation of obtaining hybrids with a greater heterotrophic effect.

5. Conclusion

This study constitutes a first step in the characterization of 15 maize Lines developed by induced mutation using gamma radiation. Major differences were recorded within the germoplasm studied. The significant differences revealed between the studied Lines and the non-irradiated parent prove the occurrence of mutations in these Lines. Multivariate analysis structured the variability into five distinct groups. These groups already constitute interesting reservoirs of genes for the crop improvement. Nevertheless, the characterization work must continue. For this, these mutant Lines need to be tested in the unsuitable environmental and edaphic conditions of the northern savannah region of Côte d'Ivoire to assess their response to various abiotic stresses like heat, salinity and drought.

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