

## BIOMETRIC AND PHYSIOLOGICAL INDICES AS EXPRESSION OF MAIZE GENOTYPES IN SPECIFIC AGROECOLOGICAL GROWTH CONDITIONS

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**Abstract.** The study used morpho-physiological indices and appropriate mathematical and statistical tools to evaluate 76 maize genotypes, cultivated in the specific agro-ecological conditions of ARDS Lovrin, Romania. For each genotype, the plant leaves number (Pln), the number of leaves up to ear (LnE), chlorophyll (Chl), leaf area (LA), panicle length (PL), the number of panicle branches (Pbn) were determined, and some ratios between indices were calculated (Pln/LnE, PL/Pln, PL/LnE, PL/Pbn, Pbn/Pln, Pbn/LnE). Low variability showed Pln ( $CV_{Pln} = 7.0382$ ), and moderate variability presented Pbn ( $CV_{Pbn} = 20.3474$ ). From the statistical analysis of the data series, some genotypes were placed in the upper quartile for several indices (e.g. L52, L62 for five indices each). According to PCA, PC1 explained 35.487% of variance, and PC2 explained 20.344% (based on indices); PC1 explained 45.47% of variance, and PC2 explained 36.195% of variance (based on calculated ratios). Based on the values of the Pln/LnE ratio, two groups of genotypes resulted, group G1 with  $Pln/LnE \geq 2.0$  (50 genotypes) and group G2, with  $Pln/LnE < 2.0$  (26 genotypes). The statistical analysis (t Test, Wilcoxon test) confirmed the differences between the two groups of genotypes, for each morpho-physiological index and calculated ratio, and facilitated the identification of valuable genotypes for maize breeding programs.

**Keywords:** *climatic changes, ecological adaptability, germoplasm, maize, productivity parameters*

### Introduction

Maize is a crop plant and food resource of global interest, and the implementation of genotypes with high tolerance to climate changes (thermal and water stress), to soil conditions (acidity, salinity), and resistance to diseases and pests, associated with high-performance technologies, show high interest for food security (Bojtor et al., 2021; Prasanna et al., 2021; Mukaro et al., 2023; Széles et al., 2024).

The appropriate management of maize germplasm, the comparative analysis of genotypes in relation to the specific agro-ecologies of the culture areas, are current concerns all over the world (Matova et al., 2023). In order to obtain valuable maize genotypes, in sustainable time and cost conditions, different methods were studied within the maize breeding programs (Bernardo, 2021; Mukaro et al., 2023).

For better genomic selections, comparative crops, and monitoring the behavior of populations by phenotyping a well-defined number of lines, are important in the development and effective progress in the breeding programs (Atanda et al., 2021). Of very recent interest are the “multi-environment (MET)” studies, in which the phenotypic prediction of maize hybrids (hybrids not tested in the field) was analyzed based on genomic prediction (Barreto et al., 2024).

Leaf morphological traits (e.g. leaf width, leaf surface, LAI) and physiological characteristics (e.g. Chl, total pigments, total N) were used for the comparative analysis of some corn hybrids in order to assess the growth advantage (Ibraheem and El-Ghareeb, 2019).

The number of leaves was considered as an important parameter in different study models of comparative maize crops (Liu et al., 2020). Foliar parameters (e.g. leaf surface, length and width of leaves, etc.) were analyzed in relation to flowering time and environmental conditions in a representative number of maize genotypes (127 genotypes; crops locations in Europe, Kenya, Mexico) (Lacube et al., 2020). The correlation between biometric plant growth parameters, yield, and specific genetic parameters (e.g. genetic variation, heritability, genetic advance) were analyzed for different maize genotypes (Magar et al., 2021).

Morphological and physiological parameters of the plants (e.g. plant height, leaf surface, root dimensions, root dry matter, and aerial part of the plants, etc.), were evaluated in relation to the plants in water stress conditions and irrigation conditions, in an experiment with different maize hybrids (Castro-Acosta et al., 2021).

Biometric parameters, as plant traits, were analyzed to evaluate the percentage of heritability in the maize breeding process by using inbred lines (Habiba et al., 2022). The spatial distribution of the leaves on the maize plant was analyzed in relation to the light capture efficiency (Serouart et al., 2023). Based on the ALAEM algorithm, the authors of the study quantified with high precision (RMSE values,  $R^2$ ) the differences in leaf orientation in relation to genotype, sowing density and environmental factors.

Reproductive success and crop yield was explained and associated with the architecture of rice panicles (Lu et al., 2017). Ecological plasticity, maize inflorescence architecture and reproductive success were studied in order to understand the genetic regulatory networks involved in their determination (Eveland et al., 2014; Koppolu et al., 2022). Inflorescence branching in maize, contributes to crop productivity and has been studied compared to panicles branching, in relation to genetic regulation pathways (Du et al., 2022).

Climate change generates pressure on agricultural systems, and farmers need resistant and productive genotypes in the new conditions (Stagnati et al., 2022).

Associated with climate change and the stress to which crops are subjected, with possible effects on food security, studies on the behavior and stress tolerance of maize genotypes are of great interest (Balbaa et al., 2022). Various ecological (biotic and abiotic) and socio-economic constraints influence maize yield (Dossa et al., 2023). The creation of genotypes with high nitrogen use efficiency (NUE) is of high importance in maize breeding programs worldwide (Santos et al., 2023).

Productivity gains, as an expression of genetic gains, are of interest in maize breeding, and in this sense public-private partnerships for the evaluation of traits and behavior under production conditions of different maize genotypes (genotypes in the pre-commercial, and commercial phase) were very useful (Asea et al., 2023). The efforts of cultivation and comparative testing of maize genotypes, in order to effectively promote performing genotypes and appropriate technologies to farming communities, in different agro-ecological zones, are of great interest (Prasanna et al., 2021; Omar et al., 2023; Széles et al., 2024).

Based on the interest expressed in the scientific literature in the field, for the behavior of maize genotypes in different agro-ecological areas, in the context of climate changes, the present study used representative morphometric and physiological indices, and different mathematical tools, to characterize 76 maize genotypes in the agro-ecological conditions of ARDS Lovrin, Western Plain of Romania.

## Materials and methods

The study took place within ARDS Lovrin, Timis County, Romania. The experimental conditions are representative for the Western Plain of Romania. The study took place in the 2022–2023 agricultural year, with the climatic conditions recorded at ARDS Lovrin Weather Station (*Table 1*).

**Table 1.** Climatic conditions in the study area

Climatic parameters		Values of climatic parameters during the study period										
		I	II	III	IV	V	VI	VII	VIII	IX	X	Avg/Sum
Rainfall (mm)	Monthly value	59	43.8	34.2	42	109.2	51.4	52.8	63.2	79	23.2	557.80
	Multiannual monthly average	32.7	29.6	32.3	42.7	57.3	68.1	55.8	32.3	42.4	40.5	433.70
	Deviation	26.3	14.2	1.9	-0.7	51.9	-16.7	-3	30.9	36.6	-17.3	124.10
Temperature (mm)	Monthly average	4.45	2.66	7.77	9.86	16.69	20.43	24.26	23.95	21.07	15.28	14.64
	Multiannual monthly average	-1.1	0.9	5.25	10.7	16.3	19.8	22.2	21.7	16.8	11.1	12.37
	Deviation	5.55	1.76	2.52	-0.84	0.39	0.63	2.06	2.25	4.27	4.18	2.28

The soil within the crop plot, ARDS Lovrin, is of typical chernozem type, weakly glazed, medium clay-clay, with the following physico-chemical parameters of fertility in the Ap horizon (0 – 20 cm): soil reaction, pH = 6.7; humus content, H = 3.55%; nitrogen index, NI = 3.07%; phosphorus content, P = 75.5 ppm; potassium content, K = 205 ppm.

The maize genotypes (76 genotypes) were cultivated in a non-irrigated system, under specific technological conditions. Sowing was done in the first decade of April, and harvesting was done in the third decade of September. The distance between rows was 70 cm, and the distance between plants per row was 23 cm. Crop technology ensured optimal conditions for the plants, through specific soil tillage, sowing in the optimal season, and maintenance works. Fertilization was done with complex fertilizers (NPK, 15:15:15) in a dose of 300 kg ha<sup>-1</sup>. During the vegetation period, BBCH code 14–16, leaves unfolded (Meier, 2001), ammonium nitrate was applied, at a dose of 200 kg ha<sup>-1</sup> (mechanical weeding, with the fertilizer incorporation near the plants row).

Maize hybrids were cultivated in randomized experimental plots, in four repetitions. For the phenological characterization of the plants, the total number of leaves per plant (Pln), the number of leaves up to the ear (LnE), the chlorophyll content (Chl, SPAD) and the leaf area (LA, cm<sup>2</sup>) were determined. Panicle length (PL, cm) and the number of panicle branches (Pbn) were also measured. Determinations were made at flowering, BBCH code 6, Flowering anthesis (Meier, 2001). Based on the determined indices, a series of ratios between the indices were calculated, in order to quantify, with high finesse, certain aspects of proportionality at the plant level. Thus, the following ratios were determined: Pln/LnE, PL/Pln, PL/LnE, PL/Pbn, Pbn/Pln, and Pbn/LnE.

The experimental data were analyzed mathematically and statistically through appropriate methods and tools (Hammer et al., 2001; JASP, 2022). Descriptive statistics was used for the purpose of general characterization of data series, calculation of mean

and median values, establishment of thresholds for quartiles. Correlation analysis was applied to evaluate the interdependence between the considered indices and the calculated ratios. Multivariate analysis (PCA) was applied to obtain the distribution of genotypes in relation to considered parameters, as biplot, as well as to find out how the main components (PC1, PC2) explained the variance in the analyzed data. Cluster analysis (CA) was applied to find out the grouping of genotypes into clusters, in relation to the degree of similarity for the indices considered, and to identify the best genotypes for each index. The t-test and the Wilcoxon test were applied to find out the reliability of the differences between the groups of genotypes (G1, and G2), in the case of each index and the calculated ratios.

## Results and discussion

From the maize germplasm collection within ARDS Lovrin, 76 maize genotypes were considered in the present study. The recorded values for the considered parameters were analyzed for descriptive statistical characterization. Based on the values of the determined physiological parameters, a series of ratios were calculated, in order to capture with a higher level of finesse a certain expression of the plants, which is not very obvious based only on the initial parameters.

The statistical values of the data series, for the physiological indices and the calculated ratios, are presented in *Table 2*.

**Table 2.** Descriptive statistics of the parameters of the maize genotypes studied

Parameters	Pln	LnE	Chl	LA	PL	Pbn	Pln/LnE	PL/Pln	PL/LnE	PL/Pbn	Pbn/Pln	Pbn/LnE
Valid	76	76	76	76	76	76	76	76	76	76	76	76
Missing	0	0	0	0	0	0	0	0	0	0	0	0
Median	12.60	6.00	55.00	627.90	33.95	13.30	2.09	2.72	5.72	2.53	1.07	2.25
Mean	12.68	6.07	54.59	626.25	34.03	13.54	2.14	2.70	5.77	2.64	1.07	2.28
Std. error of mean	0.10	0.12	0.54	7.42	0.42	0.32	0.04	0.04	0.13	0.09	0.03	0.06
Std. Deviation	0.89	1.08	4.72	64.67	3.64	2.76	0.32	0.33	1.16	0.77	0.22	0.54
Coefficient of variation	0.07	0.18	0.09	0.10	0.11	0.20	0.15	0.12	0.20	0.29	0.20	0.24
Variance	0.80	1.17	22.24	4181.5	13.27	7.59	0.10	0.11	1.34	0.60	0.05	0.29
Shapiro-Wilk	0.97	0.96	0.98	0.97	0.97	0.96	0.95	0.99	0.99	0.75	0.98	0.99
P-value of Shapiro-Wilk	0.08	0.02	0.30	0.04	0.05	0.02	0.01	0.71	0.49	<.001	0.29	0.56
Minimum	11.00	4.30	40.53	423.24	25.00	6.00	1.54	1.75	3.01	1.56	0.46	1.00
Maximum	15.00	8.30	70.17	819.22	40.00	18.30	3.17	3.45	9.30	6.67	1.53	3.45
25th percentile	12.00	5.30	51.50	589.38	32.08	12.00	1.90	2.49	4.87	2.20	0.94	1.97
50th percentile	12.60	6.00	55.00	627.90	33.95	13.30	2.09	2.72	5.72	2.53	1.07	2.25
75th percentile	13.30	7.00	57.33	657.75	36.70	15.70	2.26	2.89	6.62	2.85	1.22	2.62

Pln – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn – panicle branches number; Pln/LnE, PL/Pln, PL/LnE, PL/Pbn, Pbn/Pln, Pbn/LnE – calculated ratios

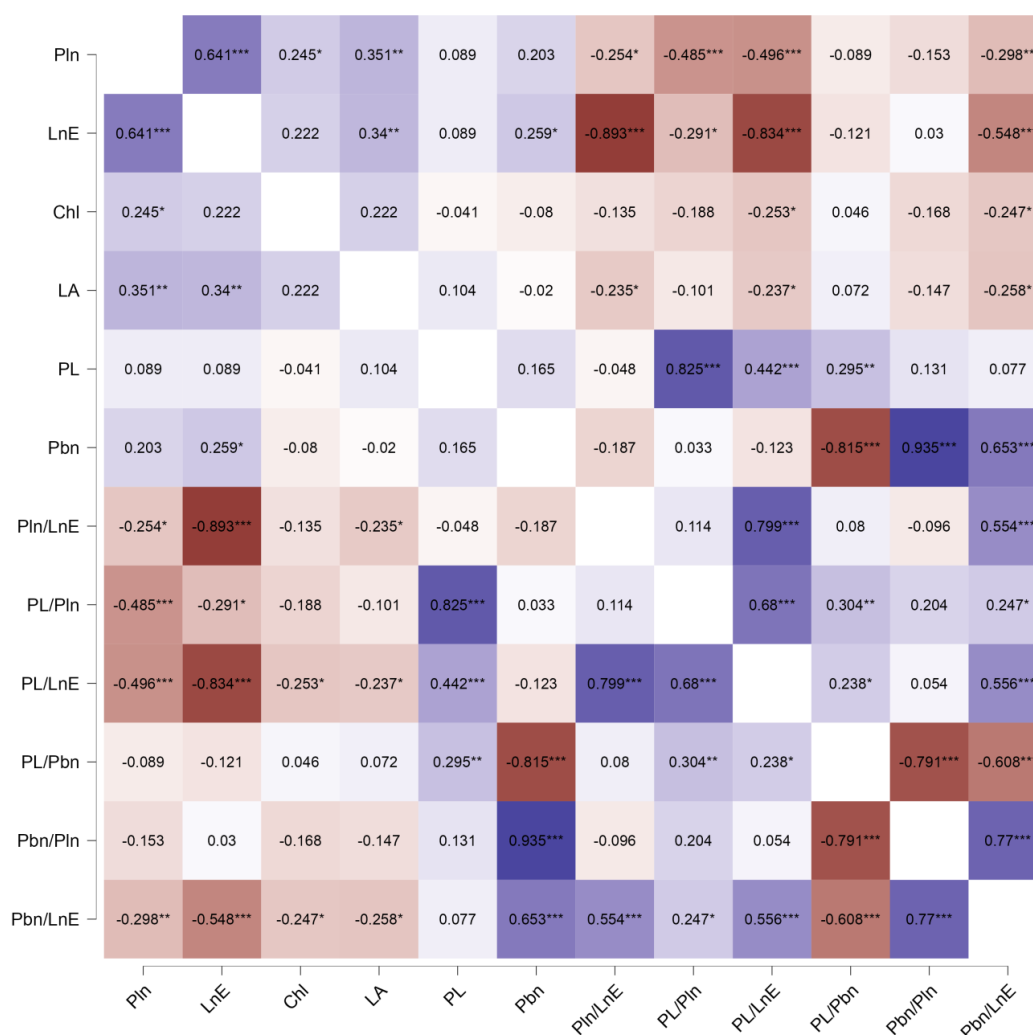
Based on the coefficient of variation (CV), the variability within each index and calculated ratio was analyzed. In the case of physiological indices, low variability was recorded in the case of Pln ( $CV_{Pln} = 7.0382$ ), followed by Chl ( $CV_{Chl} = 8.6389$ ), LA ( $CV_{LA} = 10.3257$ ), PL ( $CV_{PL} = 10.7057$ ), and LnE ( $CV_{LnE} = 17.8073$ ).

Moderate variability showed Pbn ( $CV_{Pbn} = 20.3474$ ). The LnE index showed higher variability compared to Pln ( $17.8073 > 7.0382$ ). Within the physiological indices, Pbn

presented moderate variability, the highest caloricity compared to the other indices ( $CV_{Pbn} = 20.3474$ ), which shows that it is the index with the highest sensitivity in the study conditions (genotype, environmental conditions).

In the case of the calculated ratios, low variability was recorded in the case of the  $PL/Pln$  ( $CV_{PL/Pln} = 12.0857$ ), and  $Pln/LnE$  ( $CV_{Pln/LnE} = 14.9059$ ) ratios. Moderate variability was recorded in the case of  $PL/LnE$  ratios ( $CV_{PL/LnE} = 20.0654$ ), followed by  $Pbn/Pln$  ( $CV_{Pbn/Pln} = 20.1212$ ), followed by  $Pbn/LnE$  ( $CV_{Pbn/LnE} = 23.5671$ ), respectively followed by  $PL/Pbn$  ( $CV_{PL/Pbn} = 29.3160$ ).

The correlation analysis was done to find out the interdependence between the physiological indices, the calculated ratios, respectively physiological indices and ratios. Resulted the correlation coefficient values, represented in map format (Fig. 1; \*  $p < 0.05$ ; \*\*  $p < 0.01$ ; \*\*\*  $p < 0.001$ ).



**Figure 1.** Correlation coefficient values in map format. *Pln* – plant leaf number; *LnE* – number of leaves up to the ear; *Chl* – chlorophyll content; *LA* – leaf area; *PL* – panicle length; *Pbn* – panicle branches number; *Pln/LnE*, *PL/Pln*, *PL/LnE*, *PL/Pbn*, *Pbn/Pln*, *Pbn/LnE* – calculated ratios

From the analysis of the correlation values (Fig. 1), various levels of correlation were found. Weak correlation was registered between *LnE* and *Pln* ( $r = 0.641^{***}$ ).

The Pln/LnE ratio in relation to Pln showed a correlation at the level of  $r = -0.254^*$ , and in relation to LnE it showed a correlation at the level of  $r = -0.893^{***}$ . This shows that the variation of the Pln/LnE ratio is much more dependent on the LnE value than on Pln, and highlights the importance of the LnE parameter in corn.

The PL/Pln ratio showed a negative correlation with Pln ( $r = -0.485^{**}$ ) and a positive correlation with PL ( $r = 0.825^{***}$ ). Taking into account the fact that PL in direct relation with Pln presented a correlation at the  $r = 0.089$  level, it shows the strong independence of this plant parameter with Pln.

The PL/LnE ratio showed a negative correlation with Pln ( $r = -0.496^{***}$ ) and with LnE ( $r = -0.834^{***}$ ) and a positive correlation with PL ( $r = 0.442^{***}$ ).

The PL/Pbn ratio showed a positive correlation with PL ( $r = 0.295^{**}$ ) and a negative correlation with Pbn ( $r = -0.815^{***}$ ). Considering the low correlation between Pbn and PL ( $r = 0.165$ ), and the values of the PL/Pbn ratio presented previously, it can be considered that the Pbn parameter is independent of PL.

The Pbn/Pln ratio showed a correlation with Pln at the level of  $r = -0.153$ , and with Pbn it showed a correlation at the level of  $r = 0.935^{***}$ , values that may suggest the independence of the Pbn parameter from the Pln parameter, or a very reduced interdependence with this parameter.

The Pbn/LnE ratio showed a correlation with Pln at the level of  $r = -0.298^{**}$ , with LnE it showed a correlation at the level of  $r = -0.548^{***}$ , and with Pbn it showed a correlation at the level of  $r = 0.653^{***}$ . These values confirm the low interdependence of the Pbn parameter with Pln, a moderate dependence on LnE.

Starting from the data obtained through the descriptive statistics analysis (*Table 2*), the maize genotypes placed in the best quartile, in relation to each determined index, were identified.

The obtained results are presented, with 19 maize genotypes, ranked descending according to the values for each index (*Table 3*; *Fig. 2*). In relation to the objectives of the maize breeding program, parental forms can be selected for directed crosses, at different stages of the breeding process. Some genotypes are present in this quartile for most of the determined indices (e.g. L52, L62 for five indices each), and other genotypes for a smaller number of indices (*Table 3*).

In relation to the values of the determined indices, the multivariate analysis (PCA) led to the specific diagram (*Fig. 3*). PC1 explained 35.487% of variance, and PC2 explained 20.344% of variance, and the maize genotypes were distributed according to the considered indices, as biplot. The graphic distribution for the Eigenvalue in relation to the main components is presented in *Figure 4*.

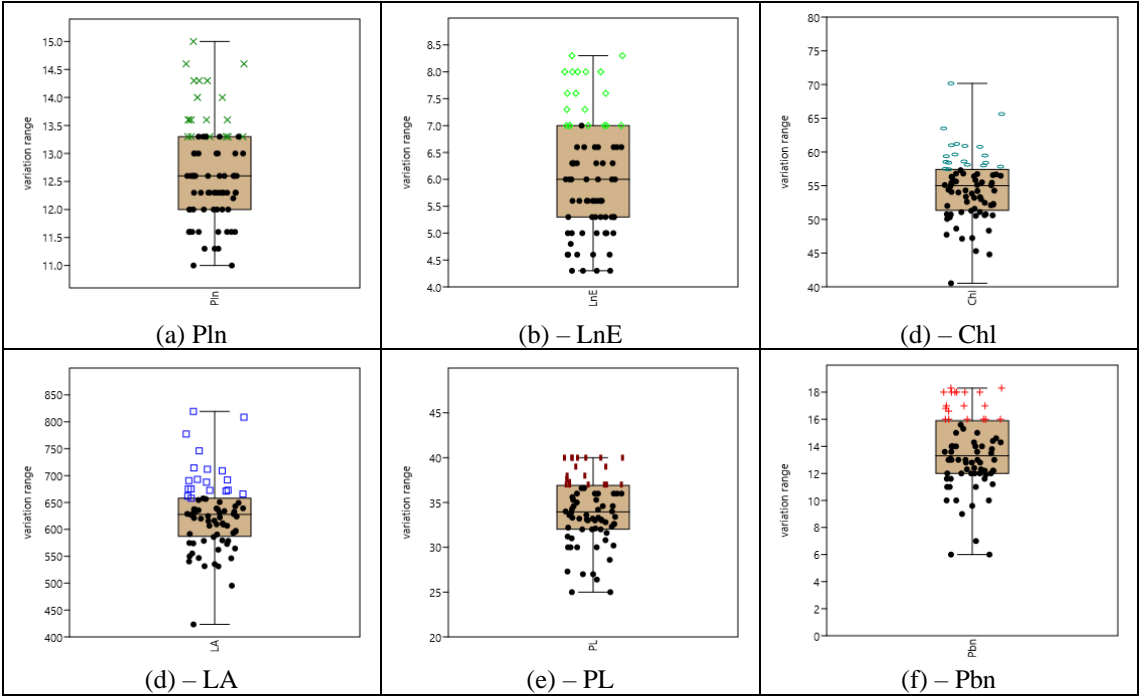
In relation to the values of the calculated ratios, the multivariate analysis (PCA) led to the diagram in *Figure 5*. PC1 explained 45.47% of variance, and PC2 explained 36.195% of variance. The maize genotypes were distributed according to the calculated ratios, as biplot. The graphic distribution for the Eigenvalue in relation to the main components is presented in *Figure 6*.

Cluster analysis (CA) facilitated the classification of maize genotypes based on similarity in relation to the studied indices, in statistical safety conditions (*Table 4*). From the resulting dendrograms, maize genotypes was associated in different sub-clusters, and were selected based on similarity, related to each index (*Table 4*). The statistical confidence values (Coph.corr.) for the dendrogram related to each index are presented, as well as the number of genotypes in the cluster, with different subclusters, as well as the code of the genotypes.

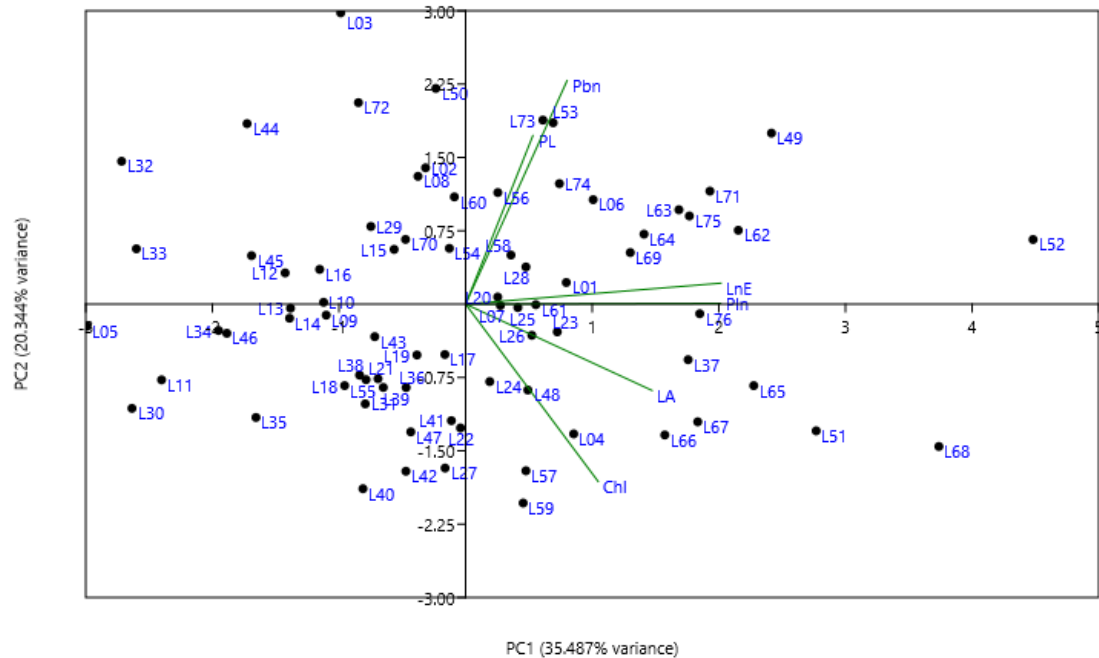
**Table 3.** The maize genotypes positioned in the upper quartile, within each parameter considered in the analysis

Pln		LnE		Chl		LA		PL		Pbn	
Code	Value	Code	Value	Code	Value	Code	Value	Code	Value	Code	Value
L52	15.00	L52	8.30	L68	70.17	L68	819.22	L41	40.00	L02	18.30
L04	14.60	L76	8.30	L27	65.63	L52	808.65	L44	40.00	L03	18.30
L06	14.60	L49	8.00	L04	63.50	L65	777.31	L68	40.00	L49	18.00
L49	14.30	L51	8.00	L51	61.20	L67	746.16	L70	40.00	L52	18.00
L51	14.30	L64	8.00	L24	61.00	L17	714.35	L71	40.00	L53	18.00
L76	14.30	L69	8.00	L67	60.90	L19	711.76	L73	40.00	L56	18.00
L26	14.00	L71	8.00	L47	60.77	L01	708.93	L75	40.00	L64	18.00
L75	14.00	L62	7.60	L38	59.63	L15	692.93	L60	39.00	L74	18.00
L23	13.60	L66	7.60	L62	59.47	L51	691.98	L61	39.00	L58	17.00
L37	13.60	L68	7.60	L66	59.37	L37	690.47	L15	38.00	L62	17.00
L62	13.60	L37	7.30	L64	58.60	L57	687.79	L72	38.00	L63	17.00
L63	13.60	L65	7.30	L22	58.53	L40	675.22	L03	37.60	L50	16.80
L66	13.60	L20	7.00	L39	58.40	L27	675.20	L16	37.30	L06	16.60
L07	13.30	L23	7.00	L75	58.40	L28	672.75	L43	37.00	L31	16.00
L20	13.30	L25	7.00	L61	58.10	L59	672.58	L49	37.00	L54	16.00
L24	13.30	L58	7.00	L54	58.00	L61	671.22	L52	37.00	L65	16.00
L28	13.30	L63	7.00	L65	57.83	L13	665.78	L62	37.00	L71	16.00
L53	13.30	L67	7.00	L36	57.50	L09	661.69	L69	37.00	L72	16.00
L57	13.30	L73	7.00	L31	57.43	L03	657.95	L74	37.00	L76	16.00

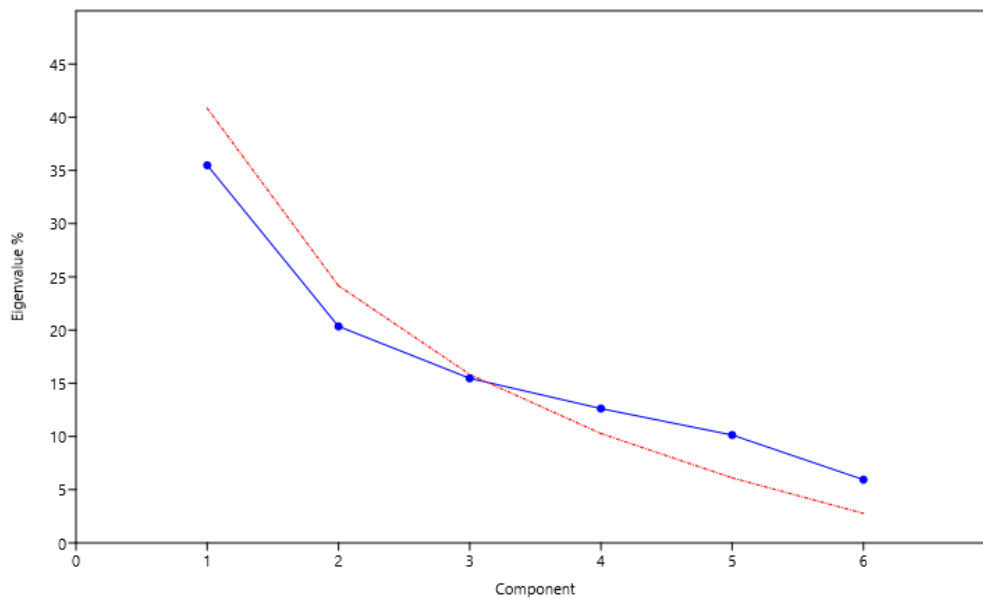
Pln – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn –panicle branches number



**Figure 2.** Box and jitter format distribution of maize genotypes, highlighting the “symbol & color” of those placed in the best quartile. Pln – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn –panicle branches number



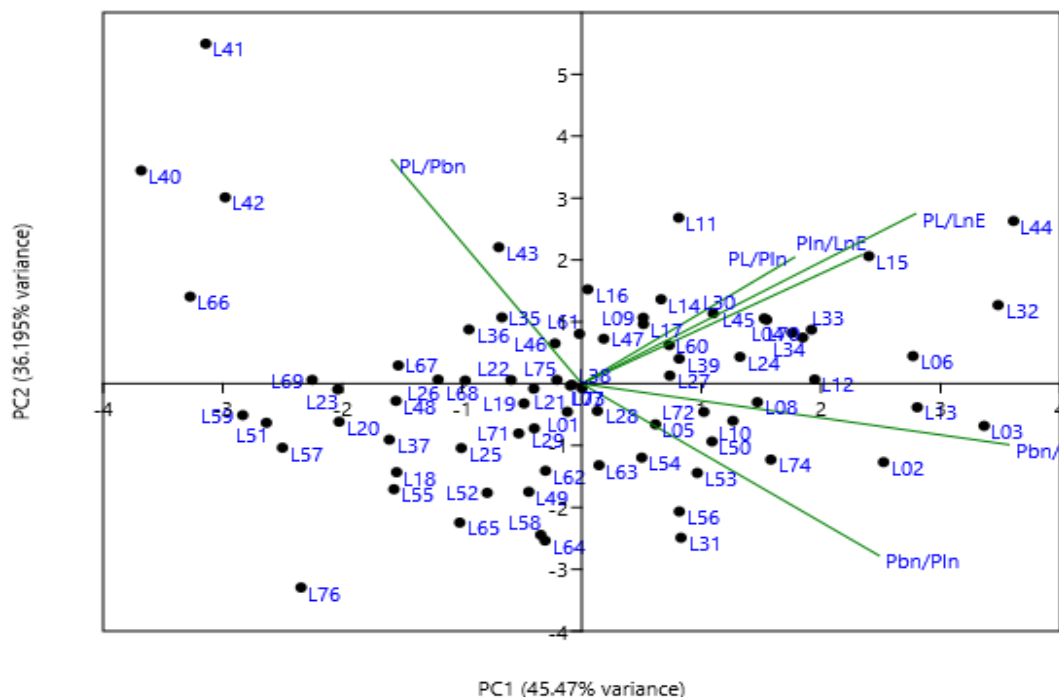
**Figure 3.** PCA diagram regarding the distribution of maize genotypes in relation to considered parameters and indices. Pbn – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn – panicle branches number



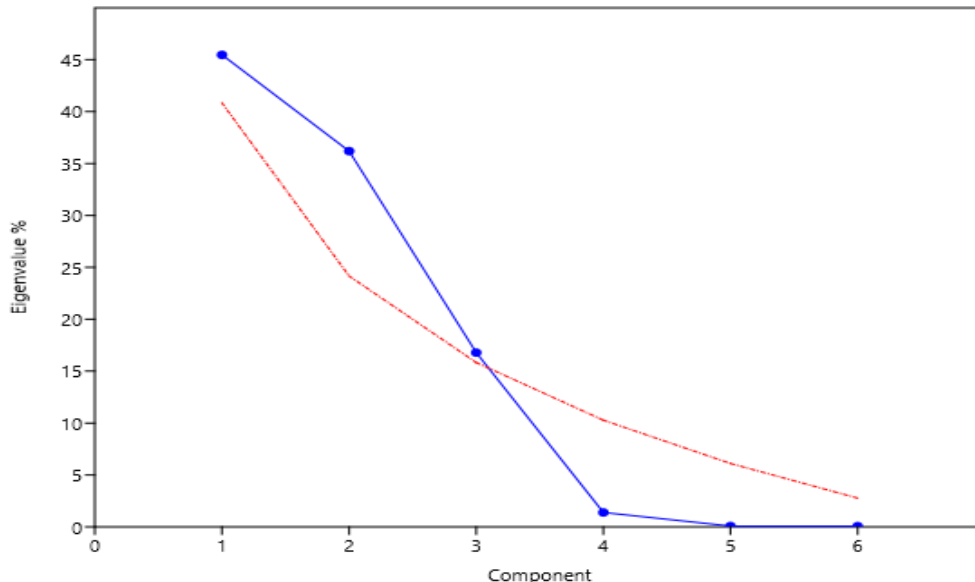
**Figure 4.** Eigenvalue in relation to the main components, under conditions of studied parameters and indices

The leaves of maize plants show high importance in relation to the plants productivity, with tolerance to environmental factors, through the total number of leaves per plant (Pbn), the positioning on the stem compared to the ear (LnE), through the dimensions of the leaves (LA), the content of photosynthetic pigments (Chl), but also other morpho-anatomical characteristics.





**Figure 5.** PCA diagram of the distribution of maize genotypes based on the calculated ratios. Pln – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn – panicle branches number



**Figure 6.** Eigenvalue in relation to the main components, under the conditions of the calculated ratios

In this study, the variable values of the Pln/LnE ratio were especially taken into account. Thus, in the case of  $\text{Pln/LnE} \geq 2.0$ , the number of leaves up to the ear (LnE) is less than, or equal to the number of leaves above the ear, and in the case of  $\text{Pln/LnE} < 2.0$ , the number of leaves up to the ear (LnE) is higher.

**Table 4.** Positioning of maize genotypes based on cluster analysis, in relation to the studied parameters and indices

Pln	LnE	Chl	LA	PL	Pbn
Coph.corr.					
0.719	0.713	0.782	0.825	0.789	0.822
Number of genotypes in the best cluster					
8	7	3	4	9	8
Maize genotypes in the best cluster					
L52 (L76,L51,L49) (L06,L04) (L75,L26)	(L52,L76) (L71,L69,L64, L51,L49)	L68 (L27,L04)	(L68,L52) (L65,L67)	(L41,L44,L68, L70,L71,L73, L75) (L60,L61)	(L02,L03) (L49,L52,L53, L56,L64,L74)

Pln – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn –panicle branches number

Starting from this observation, the genotypes were grouped according to the value of the Pln/LnE ratio. Two groups resulted, group G1 which included 50 genotypes with Pln/LnE value  $\geq 2.0$ , and group G2 which included 26 genotypes with Pln/LnE value  $< 2.0$ .

A comparative analysis was made of the G1 group of genotypes, with the mean and median value for each index and calculated ratio of the G2 group of maize genotypes (significance of differences was applied only under these conditions).

In the case of the studied indices, the analysis led to the results in Table 5. According to the t test, significant differences were recorded, at the level of  $p < 0.001$  (\*\*\*) for Pln, LnE, Chl, LA and Pbn indices. In the case of the PL index, the differences were insignificant (ns). The Wilcoxon test confirmed the recorded results.

**Table 5.** Results of the comparative analysis of the indices between the G1 and G2 groups of maize genotypes

Statistical parameters	Morphological parameters and physiological indices					
	Pln	LnE	Chl	LA	PL	Pbn
t test						
Given mean:	13.185	7.242	55.705	642.54	34.215	14.485
Sample mean:	12.414	5.462	54.01	617.78	33.928	13.052
95% conf. interval:	(12.174 12.654)	(5.2664 5.6576)	(52.687 55.333)	(602.3 633.26)	(32.953 34.903)	(12.263 13.841)
Difference:	0.771	1.78	1.6952	24.759	0.287	1.433
95% conf. interval:	(0.53136 1.0106)	(1.5844 1.9756)	(0.37249 3.0179)	(9.2765 40.241)	(-0.68846 1.2625)	(0.64438 2.2216)
t:	-6.4654	-18.285	-2.5755	-3.2137	-0.59126	-3.6516
p (same mean):	4.47E-08	1.57E-23	0.013078	0.0023194	0.55706	0.00063295
Differences significance	***	***	***	***	ns	***
Wilcoxon test						
Given median:	13.3	7	56.15	634.89	34.1	14
Sample median:	12.3	5.45	54.235	622.99	33.75	13
W:	932	1225	962.5	856	664	782.5
Normal appr. z:	5.1144	6.1067	3.1374	2.1093	0.2559	2.3134
p (same median):	3.15E-07	1.02E-09	0.0017047	0.034923	0.79803	0.0207
Differences significance	***	***	***	***	ns	***

Caption: Pln – plant leaf number; LnE – number of leaves up to the ear; Chl – chlorophyll content; LA – leaf area; PL – panicle length; Pbn –panicle branches number

The comparative analysis was also done in the case of the calculated ratios, and the results are presented in *Table 6*. The results showed statistical certainty (\*\*\*) in the case of the Pln/LnE, PL/Pln, PL/LnE, PL/Pbn and Pbn/LnE.

Parameters considered in the study, for the evaluation of maize genotypes, were representative, presented statistical reliability, and were communicated in the specialized literature in similar studies on maize.

In a comparative study on maize (127 genotypes; crops locations in Europe, Kenya, Mexico), Lacube et al. (2020) recorded, based on some morphometric parameters (e.g. leaf surface, length, width of leaves, etc.), differentiated effects of flowering time and environmental factors on leaf surface (flowering time had a predominant effect).

**Table 6.** Results of the comparative analysis of the calculated ratios between the groups of genotypes G1 and G2

Statistical parameters	Calculated ratios					
	Pln/LnE	PL/Pln	PL/LnE	PL/Pbn	Pbn/Pln	Pbn/LnE
t test						
Given mean:	1.827	2.603	4.753	2.433	1.1	2.008
Sample mean:	2.3004	2.743	6.3056	2.7478	1.0544	2.4258
95% conf. interval:	(2.2243 2.3765)	(2.6531 2.8329)	(6.022 6.5892)	(2.5029 2.9927)	(0.98997 1.1188)	(2.2648 2.5868)
Difference:	0.4734	0.14	1.5526	0.3148	0.0456	0.4178
95% conf. interval:	(0.39727 0.54953)	(0.050119 0.22988)	(1.269 1.8362)	(0.069901 0.5597)	(-0.018832 0.11003)	(0.25682 0.57878)
t:	12.496	3.1302	11.001	2.5832	-1.4222	5.2154
p (same mean):	7.50E-17	0.0029426	7.71E-15	0.012826	0.1613	3.68E-06
Differences significance	***	***	***	***	ns	***
Wilcoxon test						
Given median:	1.84	2.635	4.72	2.41	1.115	2
Sample median:	2.22	2.78	6.3	2.555	1.045	2.435
W:	1275	896.5	1217	898.5	817	1050
Normal appr. z:	6.1563	2.5003	6.0133	2.8451	1.7331	4.3523
p (same median):	7.44E-10	0.012408	1.82E-09	0.00444	0.083077	1.35E-05
Differences significance	***	***	***	***	ns	***

Pln/LnE, PL/Pln, PL/LnE, PL/Pbn, Pbn/Pln, Pbn/LnE – calculated ratios

Based on morphological and physiological plant parameters of different maize hybrids (e.g. plant height, leaf surface, root size, root dry matter and aerial part of plants, etc.) under conditions of water stress and irrigation (Castro-Acosta et al., 2021) identified genotypes with high tolerance to drought, and recorded adaptations and specific responses of plants under water stress conditions, compared to normal vegetation conditions.

Balba et al. (2022) recorded the increase in photosynthetic pigment content (chlorophyll), proline content, and transpiration rate under conditions of water stress in different maize genotypes. Based on the drought tolerance index and yield, the authors made a hierarchy and hierarchical grouping of the tested lines.

Testing maize genotypes in different agro-ecological conditions and recording the response at the level of some morpho-physiological parameters and indices, is of interest for the purpose of selecting genetic resources for breeding programs, but also for the promotion of valuable genotypes for agricultural production (Dossa et al., 2023).

The maintenance of genetic bases, the conservation, analysis and characterization of local genetic resources, based on morpho-physiological parameters and representative indices, together with germplasm collections, in order to identify sources of useful genes, are very important and necessary for future improvement programs (Stagnati et al., 2022).

The creation of genotypes with high nitrogen use efficiency (NUE) is of great importance in the maize breeding programs worldwide (Santos et al., 2023). In relation to nitrogen use efficiency, Santos et al. (2023) recorded variations in the root system (root architecture, many lateral roots, reduced branching), as well as a more efficient photosynthesis, as an adaptation of the plants in order to reduce the metabolic cost, and a better photosynthetic yield, in conditions of suboptimal nitrogen supply.

Qadeer et al. (2024) reported the variation of plant growth parameters, physiological indices, and elements of productivity, yield and quality in maize, in relation to the differentiated application of urea-phosphate fertilizers.

Results in the form of series of data on plant morpho-physiological parameters were analyzed by different mathematical and statistical methods and tools, in order to capture the homogeneity and heterogeneity of the data, ranges of variation, genotypes positioned in the upper quartiles, etc., or to understand answers adaptation of plants to environmental and technological factors (Supasri et al., 2020; Paul, 2021; Lopez-Cruz et al., 2023).

The number of leaves, in relation to the position of the ear (leaves below the ear, leaves above the ear), was significantly correlated with environmental factors, temperature and photoperiod and presented a relevant indicator for yield (Liu et al., 2020). The recorded results facilitated the authors to obtain, through adequate statistical analysis, ranges of variation and to rank the genotypes, which led to the identification of the valuable ones, in relation to the purpose of the study.

The hierarchical grouping analysis of some maize genotypes (45 lines) based on some quantified traits (18 traits), showed importance, and was also communicated in other studies with obtaining groups of genotypes, in relation to drought tolerance (Balbaa et al., 2022).

The results recorded for parameters and physiological indices studied in the collection of 76 maize genotypes, are in line with the trend of studies in the field, and have facilitated the evaluation of the response of each genotype to the agro-ecological crop conditions, and the identification of valuable genotypes for maize breeding programs.

## Conclusions

The considered morpho-physiological parameters and indices highlighted the differentiated behavior during the vegetation period of the 76 maize genotypes in the specific agro-ecological conditions of ARDS Lovrin.

Low variability showed the number of leaves on the plant,  $Pln$  ( $CV_{Pln} = 7.0382$ ), and moderate variability showed the number of ramifications of the panicle,  $Pbn$  ( $CV_{Pbn} = 20.3474$ ). In the case of the calculated ratios, low variability was recorded in the case of  $PL/Pln$  ( $CV_{PL/Pln} = 12.0857$ ) and  $Pln/LnE$  ( $CV_{Pln/LnE} = 14.9059$ ), and moderate variability in the case of the other ratios, with an example for  $PL/Pbn$  ( $CV_{PL/Pbn} = 29.3160$ ). Various levels of correlation were recorded between parameters and morpho-physiological indices and the calculated ratios, under conditions of statistical safety (at least  $p < 0.05$ ).

The descriptive statistical analysis facilitated the grouping of values by quartiles, and the identification of genotypes in the upper quartile, related to each parameter and index. Some genotypes were positioned in the upper quartile for most indices (e.g. L52, L62 for five indices each), and other genotypes for a smaller number of indices.

Multivariate analysis (PCA, CA) facilitated the distribution and grouping of genotypes based on similarity in relation to considered parameters and indices. Depending on the values of the Pln/LnE ratio ( $\text{Pln/LnE} \geq 2.0$ ;  $\text{Pln/LnE} < 2.0$ ), the maize genotypes were grouped into two groups, group G1 (50 genotypes) and group G2 (26 genotypes). The two groups of hybrids presented differences in terms of statistical safety ( $p < 0.001$ ), except for the PL parameter and the Pbn/Pln ratio ( $p > 0.05$ ).

Based on the morpho-physiological parameters and indices and the mathematical and statistical analysis tools, the study accurately described the behavior of the maize genotypes in the agroecological study conditions and facilitated the identification of valuable genotypes for maize breeding programs.

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## REFERENCES

- [1] Asea, G., Kwemoui, D. B., Sneller, C., Kasozi, C. L., Das, B., Musundire, L., Makumbi, D., Beyene, Y., Prasanna, B. M. (2023): Genetic trends for yield and key agronomic traits in pre-commercial and commercial maize varieties between 2008 and 2020 in Uganda. – *Frontiers in Plant Science* 14: 1020667.
- [2] Atanda, S. A., Olsen, M., Burgueño, J., Crossa, J., Dzidzienyo, D., Beyene, Y., Gowda, M., Dreher, K., Zhang, X., Prasanna, B. M., Tongoona, P., Danquah, E. Y., Olaoye, G., Robbins, K. R. (2021): Maximizing efficiency of genomic selection in CIMMYT's tropical maize breeding program. – *Theoretical and Applied Genetics* 134: 279-294.
- [3] Balbaa, M. G., Osman, H. T., Kandil, E. E., Javed, T., Lamlom, S. F., Ali, H. M., Kalaji, H. M., Wróbel, J., Telesiński, A., Brysiewicz, A., Ghareeb, R. Y., Abdelsalam, N. R., Abdelghany, A. M. (2022): Determination of morpho-physiological and yield traits of maize inbred lines (*Zea mays* L.) under optimal and drought stress conditions. – *Frontiers in Plant Science* 13: 959203.
- [4] Barreto, C. A. V., das Graças Dias, K. O., de Sousa, I. C., Azevedo, C. F., Nascimento, A. C. C., Guimarães, L. J. M., Guimarães, C. T., Pastina, M. M., Nascimento, M. (2024): Genomic prediction in multi-environment trials in maize using statistical and machine learning methods. – *Scientific Reports* 14: 1062.
- [5] Bernardo, R. (2021): Upgrading a maize breeding program via two-cycle genomewide selection: same cost, same or less time, and larger gains. – *Crop Science* 61(4): 2444-2455.
- [6] Bojtor, C., Mousavi, S. M. N., Illés, Á., Széles, A., Nagy, J., Marton, C. L. (2021): Stability and adaptability of maize hybrids for precision crop production in a long-term field experiment in Hungary. – *Agronomy* 11: 2167.
- [7] Castro-Acosta, M. L., Sánchez-Soto, B. H., RuelasIslas, J. R., Romero-Félix, C. S., Buelna-Tarín, S., Almada-Ruíz, V. G. (2021): Morpho-physiological characteristics of corn (*Zea mays* L.) affected by drought during its vegetative stage. – *Agro Productividad* 14(8): 71-77.
- [8] Dossa, E. N., Shimelis, H., Mrema, E., Shayanowako, A. T. I., Laing, M. (2023): Genetic resources and breeding of maize for *Striga* resistance: a review. – *Frontiers in Plant Science* 14: 1163785.

- [9] Du, Y., Wu, B., Xing, Y., Zhang, Z. (2022): Conservation and divergence: regulatory networks underlying reproductive branching in rice and maize. – *Journal of Advanced Research* 41: 179-190.
- [10] Eveland, A. L., Goldshmidt, A., Pautler, M., Morohashi, K., Liseron-Monfils, C., Lewis, M. W., Kumari, S., Hiraga, S., Yang, F., Unger-Wallace, E., Olson, A., Hake, S., Vollbrecht, E., Grotewold, E., Ware, D., Jackson, D. (2014): Regulatory modules controlling maize inflorescence architecture. – *Genome Research* 24(3): 431-43.
- [11] Habiba, R. M. M., El-Diasty, M. Z., Aly, R. S. H. (2022): Combining abilities and genetic parameters for grain yield and some agronomic traits in maize (*Zea mays* L.). – *Beni-Suef University Journal of Basic and Applied Science* 11: 108.
- [12] Hammer, Ø., Harper, D. A. T., Ryan, P. D. (2001): PAST: Paleontological Statistics software package for education and data analysis. – *Palaeontologia Electronica* 4(1): 1-9.
- [13] Ibraheem, F., El-Ghareeb, E. M. (2019): Assessment of natural variability in leaf morphological and physiological traits in maize inbreds and their related hybrids during early vegetative growth. – *Egyptian Journal of Basic and Applied Sciences* 6(1): 25-45.
- [14] JASP Team (2022): JASP (Version 0.16.2). – Computer Software.
- [15] Koppolu, R., Chen, S., Schnurbusch, T. (2022): Evolution of inflorescence branch modifications in cereal crops. – *Current Opinion in Plant Biology* 65: 102168.
- [16] Lacube, S., Manceau, L., Welcker, C., Millet, E. J., Gouesnard, B., Palaffre, C., Ribaut, J. M., Hammer, G., Parent, B., Tardieu, F. (2020): Simulating the effect of flowering time on maize individual leaf area in contrasting environmental scenarios. – *Journal of Experimental Botany* 71(18): 5577-5588.
- [17] Liu, W., Ming, B., Xie, R., Liu, G., Wang, K., Yang, Y., Guo, X., Hou, P., Li, S. (2020): Change in maize final leaf numbers and its effects on biomass and grain yield across China. – *Agriculture* 10(9): 411.
- [18] Lopez-Cruz, M., Aguade, F. M., Washburn, J. D., Leon, N., Kaeppler, S. M., Lima, D. C., Tan, R., Thompson, A., De La Bretonne, L. W., Campos, G. (2023): Leveraging data from the Genomes-to-Fields Initiative to investigate genotype-by-environment interactions in maize in North America. – *Nature Communications* 14: 6904.
- [19] Lu, H., Dai, Z., Li, L., Wang, J., Miao, X., Shi, Z. (2017): OsRAMOSA2 shapes panicle architecture through regulating pedicel length. – *Frontiers in Plant Science* 8: 1538.
- [20] Magar, B. T., Acharya, S., Gyawali, B., Timilsena, K., Upadhayaya, J., Shrestha, J. (2021): Genetic variability and trait association in maize (*Zea mays* L.) varieties for growth and yield traits. – *Heliyon* 7(9): e07939.
- [21] Matova, P. M., Kamutando, C. N., Warburton, M. L., Williams, W. P., Magorokosho, C., Shimelis, H., Labuschagne, M., Day, R., Gowda, M. (2023): New techniques for breeding maize (*Zea mays*) varieties with fall armyworm resistance and market-preferred traits for sub-Saharan Africa. – *Plant Breeding* 142(1): 1-11.
- [22] Meier, U. (2001): Growth Stages of Mono- and Dicotyledonous Plants. BBCH Monograph. 2. Ed. – Federal Biological Research Centre for Agriculture and Forestry, Berlin.
- [23] Mukaro, R., Kamutando, C. N., Magorokosho, C., Mutari, B., Zaidi, P. H., Kutwayo, D., Sibiyi, J. (2023): Genetic potential of tropically adapted exotic maize (*Zea mays* L.) heat-tolerant donor lines in sub-tropical breeding programs. – *Agronomy* 13: 2050.
- [24] Omar, S., Abd Ghani, R., Khalid, N., Jolánkai, M., Tarnawa, Á., Percze, A., Mikó, P. P., Kende, Z. (2023): Effects of seed quality and hybrid type on maize germination and yield in Hungary. – *Agriculture* 13: 1836.
- [25] Paul, L. A. (2021): Heterogeneous and conditional returns from DT maize for farmers in Southern Africa. – *European Review of Agricultural Economics* 48(5): 1224-1248.
- [26] Prasanna, B. M., Cairns, J. E., Zaidi, P. H., Beyene, Y., Makumbi, D., Gowda, M., Magorokosho, C., Zaman-Allah, M., Olsen, M., Das, A., Worku, M., Gethi, J., Vivek, B. S., Nair, S. K., Rashid, Z., Vinayan, M. T., Issa, A. B., San Vicente, F., Dhlwayo, T.,

- Zhang, X. (2021): Beat the stress: breeding for climate resilience in maize for the tropical rainfed environments. – *Theoretical and Applied Genetics* 134(6): 1729-1752.
- [27] Qadeer, A., Yaseem, M., Naveed, M., Shahbaz, M. (2024): Effect of urea-phosphate and its application methods on maize (*Zea mays* L.) growth, yield and nutrient use efficiency. – *Applied Ecology and Environmental Research* 22(1): 265-280.
- [28] Santos, T. d. O., Amaral Junior, A. T. d., Moulin, M. M. (2023): Maize breeding for low nitrogen inputs in agriculture: mechanisms underlying the tolerance to the abiotic stress. – *Stresses* 3: 136-152.
- [29] Serouart, M., Lopez-Lozano, R., Daubige, G., Baumont, M., Escalé, B., De Solan, B., Baret, F. (2023): Analyzing changes in maize leaves orientation due to GxExM using an automatic method from RGB images. – *Plant Phenomics* 9(5): 0046.
- [30] Stagnati, L., Soffritti, G., Desiderio, F., Lanubile, A., Zambianchi, S., Marocco, A., Rossi, G., Busconi, M. (2022): The rediscovery of traditional maize agrobiodiversity: a study case from Northern Italy. – *Sustainability* 14: 12110.
- [31] Supasri, T., Itsubo, N., Gheewala, S. H., Sampattagul, S. (2020): Life cycle assessment of maize cultivation and biomass utilization in northern Thailand. – *Scientific Reports* 10: 3516.
- [32] Széles, A., Huzsvai, L., Mohammed, S., Nyéki, A., Zagy, P., Horváth, É., Simon, K., Arshad, S., Tamás, A. (2024): Precision agricultural technology for advanced monitoring of maize yield under different fertilization and irrigation regimes: a case study in Eastern Hungary (Debrecen). – *Journal of Agriculture and Food Research* 15: 100967.