

## Assessment of stability for some yield components in wheat under different fertilizations on saline soil

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### Abstract

Salt-affected soils constrain wheat production in many countries. It is estimated that more than 831 million hectares (Mha) are salt affected globally, with 10 Mha of cultivated land lost annually. Salinity adversely affects wheat grain yield and grain quality traits. Wheat genotypes show wide variation for salinity stress tolerance. The main objective of this study was to evaluate the performance and stability of spike traits in 14 winter wheat varieties with different origin, in order to identify the varieties that can exploit the growing conditions of saline soils. The wheat material was tested in nine environments as a combination of three years and three fertilization treatments (urea, calcium nitrate, ammonium nitrate), using a split-plot design with three replications. At maturity plant height, spike length, grains number per spike, and spike yield were determined. The significant effect of genotype × environment interaction on the phenotypic expression of different traits indicates differential response of the genotypes across the testing environments, thus offering the possibility of identifying some genotypes of interest. The Romanian varieties 'Alex' and 'Glosa' present the highest productivity of spike associated with an average stability, being specifically adapted to favourable rainfall conditions, achieving above-average yields in less favourable conditions. The varieties 'Cerere', 'Genesi', 'Alex' and 'Glosa' can be selected for crossing and included in wheat breeding programs for the development of stable cultivars adapted to saline soils.

**Keywords:** genotype × environment interaction; salinity stress; spike traits; winter wheat

### Introduction

Salinity stress causes osmotic stress and ion toxicity, through increasing the assimilation of Na<sup>+</sup> ion and decreasing the Na<sup>+</sup>/K<sup>+</sup> ratio due to lower osmotic potential within the plant roots. Further, these ionic imbalances affect the uptake and transport of other important essential ions in target cells and hamper the crucial plant processes and functions (Arif *et al.*, 2020). Salinity stress significantly reduces agricultural crop

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yields by affecting vegetative growth as well as fertility parameters (Roy *et al.*, 2014). The negative impact of salinity stress on plant growth is also associated with a considerable decrease of the nutritional composition of grains.

The tolerance of plants to salinity stress is a polygenic trait that is controlled by many genetic factors (Arzani and Ashraf, 2016). In order to optimize the growth and yield under saline environments the wheat plants use a wide range of physiological, biochemical and molecular mechanisms to adapt to salinity stress at the level of cell, tissue and whole plant (El Sabagh *et al.*, 2021). The salt-tolerant genotypes show an increase of enzymes activity, soluble sugars and proline amounts, associated with a decrease of osmotic potential and the lipid peroxidation, compared to the sensitive genotypes (Sen *et al.*, 2022).

Wheat is moderately tolerant to salt with threshold without yield loss at 6 dS m<sup>-1</sup> and with yield 50% loss at 13 dS m<sup>-1</sup> (Rasouli *et al.*, 2013). Salt stress causes retarded growth of the wheat plants as a consequence of the low uptake of water and nutrients and decreased pollen viability, reduced seed sets and finally reduces the yield and its components (Sen *et al.*, 2022). The wheat plants are very sensitive to salinity stress during anthesis and grain filling period (El Sabagh *et al.*, 2021). Identifying stable and high yielding genotypes can promote the sustainable use of saline soils. Therefore, selecting for salt stress tolerant wheat genotypes (Munns and Tester, 2008) associated with a more accurate agronomical practices are two complementary ways to a more stable yield in contrasting and limiting growth conditions (Yousfi *et al.*, 2010). The increasing of nitrogen supplementation in wheat regulates the salinity tolerance by modulations in the metabolism of different antioxidants, osmolytes and metabolites (Ahanger *et al.*, 2019) and improves water content of plant as well as the photosynthetic efficiency (Ahanger *et al.*, 2017).

Wheat tolerance to salt stress is influenced by genotype (G), environment (E), and the genotype × environment interaction (G × E). High and significant G × E have been defined as the failure of genotypes to achieve the same relative performance in different environments (Rharrabti *et al.*, 2003; Araus *et al.*, 2008). Identification of yield contributing traits and understanding the G × E interactions and yield stability are important for breeding new cultivars with improved adaptation to the environmental constraints (Rharrabti *et al.*, 2003).

Despite the considerable progress made during the last several decades in increasing yield, soil and climatic conditions are still one of the most important factors affecting the grain yield of wheat. Different genotypes may show different responses to a given environment as a consequence of G × E (Yan and Kang, 2003; Thungo *et al.*, 2019). The assessment of the relative contributions of the genotype, the G × E to cultivar performance is essential to determine the adaptation capacity which presents the cultivar's ability to reach its full potential in a specific environment in spite of the constraints imposed on the crop (Subira *et al.*, 2015). The analysis of G × E interaction effects is useful for the selection of genotypes with appropriate and stable performances under different unfavourable environmental conditions (Reynolds and Langridge, 2016; Vaezi *et al.*, 2019). In order to select and recommend wheat varieties suitable for cultivation in a particular environment, it is necessary to evaluate the effect of G × E interaction on the yield and its components (Leon *et al.*, 2016; Omrani *et al.*, 2022).

Among multivariate methods, the additive main effect and the multiplicative interaction analysis (AMMI) are widely used for G × E investigation (Tarakanovas and Ruzgas, 2006). AMMI model is distinguished because it decomposes main sources of variance (G, E and G × E) and at the same time it decomposes systemic variation and noise variation as well as other models (Branković-Radojčić *et al.*, 2018).

The main objective of this study was to evaluate the performance and stability of spike traits in 14 winter wheat varieties with different origin, in order to identify the varieties that can exploit the growing conditions of saline soils. The wheat material was tested in nine environments as a combination of three years and three fertilization (urea, calcium nitrate, ammonium nitrate) treatments.

## Materials and Methods

### *Biological material and experimental design*

The experimental material consisted of 14 winter wheat varieties with different genetic and ecologic origin: 'Alex', 'Glosa' (Romania); 'Apache', 'Soissons' (France); 'Capo', 'Josef' (Austria); 'Cerere', 'Genesi', 'Esperia' (Italy); 'Calisol', 'Cubus', 'Exotic', 'Solehio', 'Zephyr' (Germany). The plot was composed of 8 rows, 2 meter long and 12.5 cm rows spacing, using a density of 500 seeds per m<sup>2</sup>. The wheat material was tested in nine environments as a combination of three years (2021-2023) and three fertilization treatments (urea, calcium nitrate, ammonium nitrate), using a split-plot design with three replications.

In autumn (before sowing) 130 kg diammonium phosphate (18:46:0) was applied. The three studied fertilization treatments were applied in two stages: beginning of March and mid-April, using 50 kg N for each application. During the experiment, classical practices for wheat crop were applied. The research was carried out in the field of Genagricola Romania located near the town of Sannicolau Mare, on a solonetz soil with the following characteristics: pH, 8.64; humus, 2.1%; total nitrogen, 0.16%; mobile phosphorus, 16.13 ppm; mobile potassium, 179 ppm; zinc, 48.8 ppm; copper, 10.0 ppm; sodium, 45.04 mEq l<sup>-1</sup>; chlorine, 21.3 mEq l<sup>-1</sup>.

The temperatures and rainfall during the vegetation period of 2021-2023 are presented in Table 1. The average monthly temperatures from May-July showed variations of: 15.5-25.7 °C in 2021, 18.1-25.3 °C in 2022 and 9.9-24.5 °C in 2023. The amount of rainfall during the vegetation period was 63 mm in 2022, 106 mm in 2021 and 192 mm in 2023. Based on a lack of rainfall and higher temperatures, the year 2022 was considered the most unfavourable, while the year 2023 was the most favourable.

**Table 1.** Mean air temperature and rainfall of the three experimental years during the vegetation period

Month	Temperature (°C)			Rainfall (mm)		
	2021	2022	2023	2021	2022	2023
March	5.3	5.6	7.6	26	3	11
April	9.2	10.2	9.9	17	21	19
May	15.5	18.1	16.7	29	24	60
June	22.8	24.1	20.6	34	15	102
July	25.7	25.3	24.5	48	22	38

At the stage of full maturity, ten representative plants from each replication of each plot were selected for measurements. Plant height (cm) was measured from the stem base to the top of spikes (excluding awns). Spike length was measured from the first rachis node to the top of the upper spikelet (excluding awns). Grains number and weight per spike was determined for each plot by counting and weighting the spike grains.

### *Statistical analysis*

The data collected for plant height, spike length, grains number/spike and grain weight/spike were statistically processed by combined ANOVA and AMMI analysis using MATMODEL Version 3. The means for each trait were compared using Multiple Range Test (Ciulca, 2006). The significance of means was expressed with letters, being considered as significant the differences between means marked with different letters. The clustering of wheat varieties was performed using the UPGMA procedure, with the NEIGHBOR program of PHYLIP package, version 3.5c. (Felsenstein, 1993).

The AMMI stability value (ASV) was calculated as previously described by Purchase *et al.* (2000). It represents the distance to the origin of each variety in a two-dimensional space based on interaction principal component axis (IPCA1 and IPCA2) coordinates, considering that lower ASV indicate higher stability.

**Results**

*Analysis of plant height*

The AMMI analysis of variance for plant height indicated highly significant effects of genotype, environment, and their interaction (Table 2). The environment factors accounted for larger proportion (56.48%) of the total sum of squares, while 21.32% and 18.91% of the plant height variation were attributable to the genotype and the G×E interaction effects. The first, second, third and fourth principal components (IPCA) are significant, and explain a total of 99.64% of the interaction sum of squares. The IPCA1 accounted for 66.7% of the G×E interaction, while IPCA2 accounted for 22.02%.

**Table 2.** AMMI analysis of variance for plant height of 14 wheat varieties in nine environments

Source of variation	SS	DF	MS	F	SS % <sup>1</sup>
Genotype	13	10334	794.92	127.58**	21.32
Environment	8	27383	3422.88	444.53**	56.48
Genotype × Environment	104	9167	88.14	14.15**	18.91 (100)
IPCA 1	20	6114	305.70	49.06**	66.70
IPCA 2	18	2019	112.17	18.00**	22.02
IPCA 3	16	659	41.19	6.61**	7.19
IPCA 4	14	342	24.43	3.92**	3.73
IPCA residuals	36	33	0.92	0.15	0.36
Error	234	1458	6.23		

1- % of model sum of squares for genotype, environment and genotype × environment; \*\* Significant at  $p \leq 0.01$

The plants of ‘Solehio’ variety use the most efficiently the treatment with urea in 2021, registering a height of 80.27 cm, significantly higher by more than 10 cm compared to the other varieties, which generally recorded values of 60-70 cm (Table 3). The varieties ‘Alex’, ‘Capo’ and ‘Cerere’ presented the highest values (77.47-80.67 cm) of plant height under the treatment with calcium nitrate, while in ‘Soissons’ and ‘Zephyr’ the values of this trait were significantly lower (62.89-64.67 cm). Under the effect of ammonium nitrate fertilization, ‘Capo’ variety recorded a plant height of 83.2 cm, significantly higher than the rest of the varieties, followed by ‘Josef’ and ‘Alex’ varieties with 70.13-72.69 cm.

Regarding the results of plant height from 2022, the varieties recorded amplitudes between 18.60 cm for fertilization with calcium nitrate and 22.46 cm under the fertilization with urea. ‘Cubus’, ‘Capo’ and ‘Alex’ varieties utilized the fertilization with urea at a higher level achieving a plant height of 58.33-62.73 cm, while 57% of the varieties recorded values of 40-50 cm. The plants of the ‘Capo’ variety presented the highest values (63.67-64.47 cm) under the treatment with calcium nitrate and ammonium nitrate, registering significant increases compared to the rest of the varieties.

**Table 3.** Means of plant height (cm) for 14 wheat varieties in nine environments

Genotype	Environment									Mean G
	2021			2022			2023			
	U	CN	AN	U	CN	AN	U	CN	AN	
Alex	65.28 cde	80.67 a	72.69 b	50.07 d	52.40 bcd	59.93 a	78.53 b	85.65 b	81.11 b	69.59 B
Glosa	61.29 efg	72.78 cde	66.47 cd	45.27 ef	54.93 b	47.07 cd	71.83 de	79.69 c	70.87 b	63.36 C
Esperia	57.54 gh	69.01 cf	65.94 d	40.27 g	46.50 f	49.57 c	57.73 g	68.03 efgh	64.63 c	57.69 EF
Capo	61.49 efg	79.17 ab	83.20 a	62.73 a	64.47 a	63.67 a	86.80 a	92.15 a	94.20 a	76.43 A
Josef	60.00 fgh	69.47 def	70.13 bc	56.47 c	52.87 bcd	54.80 b	71.60 de	71.02 de	72.53 b	64.32 C
Cerere	56.13 h	77.47 ab	64.61 d	56.20 c	52.07 bcd	44.87 d	68.53 ef	72.24 d	61.20 cde	61.48 D
Genesi	60.68 f	69.47 def	58.31 ef	48.73 de	47.6 ef	44.07 d	74.13 cd	68.54 defg	59.96 de	59.05 E
Apache	63.09 def	75.61 bc	54.56 fg	58.33 bc	50.8 cde	49.67 c	61.07 g	66.23 fgh	52.03 g	59.04 E

Soissons	67.03 bcd	62.89 g	56.75 ef	44.93 ef	48.87 def	47.80 cd	65.20 f	64.56 gh	60.49 de	57.61 F
Exotic	67.93 bc	68.45 fg	55.31 ef	49.67 d	47.8 ef	54.60 b	71.67 de	67.55 efgh	63.86 cd	60.76 D
Solehio	80.27 a	75.33 bc	59.31 e	44.70 f	53.07 bc	49.40 c	66.33 f	67.58 efgh	57.20 ef	61.47 D
'Zephyr'	61.38 efg	64.67 g	59.03 e	44.06 fg	45.87 f	46.47 cd	71.60 de	66.44 fgh	60.28 de	57.75 EF
'Cubus'	70.90 b	73.26 cd	67.17 cd	61.67 ab	50.67 cde	49.57 c	75.87 bc	69.46 def	62.32 cd	64.54 C
Calisol	63.14 def	61.50 g	51.92 g	45.20 ef	53.27 bc	44.83 d	71.73 de	64.48 h	54.35 fg	56.71 F
Mean E	64.01 XY	71.41 V	63.24 Y	50.59 Z	51.51 Z	50.45 Z	70.90 V	71.69 V	65.36 X	62.13

U-Urea; CN-Calcium nitrate; AN-Ammonium nitrate; G-Genotype; E-Environment.

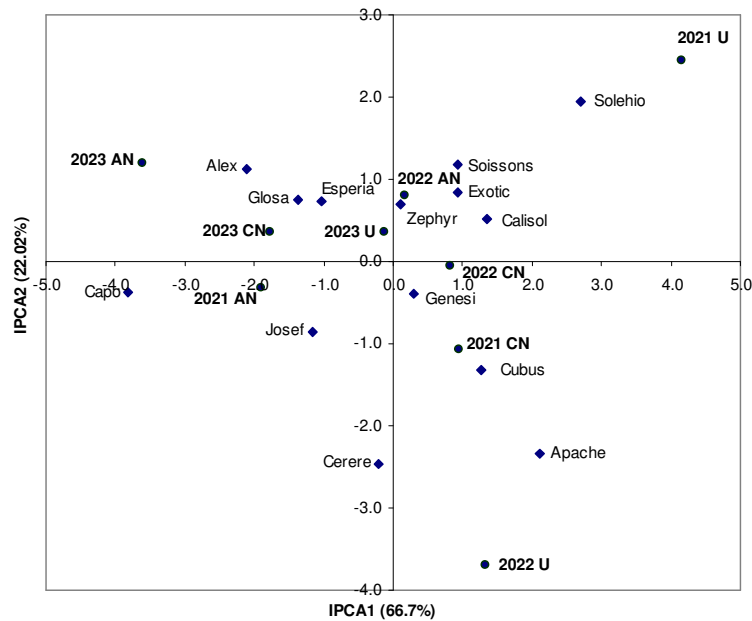
Environment LSD5%=1.36; Genotype LSD5%=1.34; Genotype × Environment LSD5%=4.02

Different letters (a-h) in the columns indicate significant differences (p<0.05) between genotypes.

Capital letters were used for environment means (V-Z) and genotype means (A-F) comparisons.

In the conditions from 2023, the 14 varieties achieved values of plant height with higher amplitudes than previous years, ranging between 27.67 cm for the treatment with calcium nitrate and 42.17 cm for ammonium nitrate. 'Capo' and 'Alex' varieties use more efficiently the treatments with the three fertilizers, registering values of plant height by 78.53-94.20 cm.

Considering the conditions of the combined environments, under the treatments with urea and calcium nitrate in 2023 as well as under the effect of calcium nitrate in 2021, the highest values of plant height were recorded. The conditions from 2022 have had a significant negative effect on the plant height for all varieties. Over all nine environments 'Capo' variety recorded a value of plant height significantly higher to other varieties with 9.8-34.8%, followed by the plants of 'Alex' variety.



**Figure 1.** Biplot of principal component axis (IPCA1 and IPCA2) for plant height of 14 wheat varieties in nine environments

According to the biplot from Figure 1, the first two principal components explained a total of 88.7% of the G×E interaction effects on plant height variability. The varieties 'Zephyr' and 'Genesi' located close to the origin are considered to be most stable regarding the plant height in the studied environments. The varieties 'Capo', 'Solehio', 'Apache' and 'Alex' appeared unstable considering their significant distance from the biplot's origin. The environments represented by the treatments with urea in 2021 and with ammonium nitrate in 2023 have had the highest contribution to the G×E interaction, since they were positioned far from the origin.

Under the treatments with urea in 2023 and with ammonium nitrate in 2022, the plant height exhibited the lowest variability, indicating that these two environments were the main contributors to the phenotypic stability of this trait for the studied varieties. In the biplot graph the varieties located in close proximity of an environment expressed a strong association with this one. Thus, 'Solehio' variety showed a specific adaptation to the treatment with urea in 2021, recording the highest value of this trait for all environments. Other specific associations can be observed for varieties 'Apache', 'Cubus' and 'Genesi' for the treatment with urea in 2021, and 'Capo' under the effect of calcium nitrate in 2021.

#### *Analysis of spike length*

The AMMI analysis of variance for wheat varieties over nine environments (Table 4) indicates that both varieties and environments, respectively their interaction had significant effects on spike length. The environment showed the highest influence (53.33%) on the variability of this trait, followed by G×E interaction (24.45%), amid a lower influence (9.44%) of genotype. The high contribution of the environment to the variation of the spike length indicates the existence of major differences between the effects of fertilization treatments during the three years. This model based on the first four principal components expresses almost entirely the effect of G×E interaction on the spike length.

**Table 4.** AMMI analysis of variance for spike length of 14 wheat varieties in nine environments

Source of variation	SS	DF	MS	F	SS % <sup>1</sup>
Genotype	13	53.3	4.10	15.18**	9.44
Environment	8	302.2	37.78	86.24**	53.53
Genotype × Environment	104	138	1.33	4.91**	24.45 (100)
IPCA 1	20	64.88	3.24	12.01**	47.01
IPCA 2	18	37.38	2.08	7.69**	27.09
IPCA 3	16	19.8	1.24	4.58**	14.35
IPCA 4	14	14.3	1.02	3.78**	10.36
IPCA residuals	36	1.64	0.05	0.17	1.19
Error	234	63.2	0.27		

1- % of model sum of squares for genotype, environment and genotype × environment; \*\* Significant at  $p \leq 0.01$

Under the conditions of 2021, the wheat varieties recorded spike length amplitude between 1.9 cm under the treatment with urea and 2.22 cm for calcium nitrate treatment (Table 5). The varieties 'Glosa' and 'Capo' use at a higher level the urea fertilization achieving a spike length significantly higher by 1.42-1.90 cm compared to the varieties: 'Esperia', 'Genesi', 'Solehio', 'Apache' and 'Soissons'. Under the fertilization with calcium nitrate, the varieties 'Glosa', 'Cerere' and 'Solehio' showed the highest values of this trait, corresponding to significant increases of 19.10-36.63% compared to 'Soissons', 'Josef' and 'Calisol'. The plants of the 'Alex' variety use more effective the fertilization with ammonium nitrate, achieving spikes significantly higher by 1.39-1.98 cm compared to the varieties: 'Genesi', 'Josef', 'Exotic', 'Zephyr' and 'Solehio'.

**Table 5.** Means of spike length (cm) for 14 wheat varieties in nine environments

Genotype	Environment									Mean G
	2021			2022			2023			
	U	CN	AN	U	CN	AN	U	CN	AN	
Alex	7.07 abc	7.75 abc	7.95 a	6.57 ab	6.13 bc	5.05 defg	9.10 ab	8.13 a	8.38 ab	7.34 A
Glosa	7.83 a	8.28 a	7.21 ab	5.53 def	6.03 bc	5.67 cde	7.90 cde	6.77 cde	7.67 bcdef	6.99 BC
Esperia	6.41 cde	7.27 bcd	6.71 bcd	5.53 def	5.10 de	4.53 g	8.13 cde	7.40 abc	7.29 cdefg	6.49 EF
Capo	7.48 ab	7.58 abcd	6.83 bc	5.70 cdef	5.87 bcd	5.60 cde	9.93 a	7.90 ab	8.11 bc	7.22 AB
Josef	6.93 bcd	6.17 e	6.37 cd	6.03 bcd	6.17 bc	5.07 defg	9.67 a	7.83 ab	7.46 cdefg	6.85 CD
Cerere	6.80 bcd	8.27 a	7.39 ab	5.07 f	6.27 b	7.20 a	9.53 a	6.53 def	9.02 a	7.34 A
Genesi	6.27 cde	7.20 cd	6.56 bcd	5.47 def	5.40 cde	5.60 cde	7.47 e	6.27 efg	6.99 efg	6.36 F
Apache	6.15 de	8.07 ab	6.77 bcd	5.93 bcde	5.67 bcd	5.60 cde	9.47 a	7.23 bcd	7.95 bc	6.98 BC
Soissons	5.93 e	6.86 de	7.26 abc	5.83 bcdef	4.73 cf	5.77 bcd	7.50 de	5.77 fgh	7.83 bcd	6.39 F
Exotic	6.56 cde	7.65 abcd	6.22 cd	5.13 ef	4.07 f	4.67 fg	8.23 cde	6.33 efg	7.11 defg	6.22 F
Solehio	6.33 cde	8.17 a	5.97 d	6.53 abc	6.01 bc	6.60 ab	7.77 cde	6.00 efgh	6.75 g	6.68 DE
Zephyr	6.82 bcd	7.67 abcd	6.27 cd	5.53 def	6.27 b	4.87 efg	7.47 e	5.37 h	5.78 h	6.23 F
Cubus	6.73 bcde	7.51 abcd	7.88 a	6.07 bcd	7.32 a	5.40 cdef	8.40 bc	5.53 gh	6.96 fg	6.87 CD
Calisol	6.85 bcd	6.06 e	7.21 ab	7.07 a	6.47 b	6.02 bc	8.33 bcd	5.73 fgh	6.97 fg	6.74 CDE
Mean E	6.73 Y	7.46 X	6.90 Y	5.86 Z	5.82 Z	5.54 W	8.49 V	6.63 Y	7.45 X	6.76

U-Urea; CN-Calcium nitrate; AN-Ammonium nitrate; G-Genotype; E-Environment.

Environment LSD5%=0.28; Genotype LSD5%=0.28; Genotype × Environment LSD5%=0.84

Different letters (a-h) in the columns indicate significant differences (p<0.05) between genotypes.

Capital letters were used for environment means (V-W) and genotype means (A-F) comparisons.

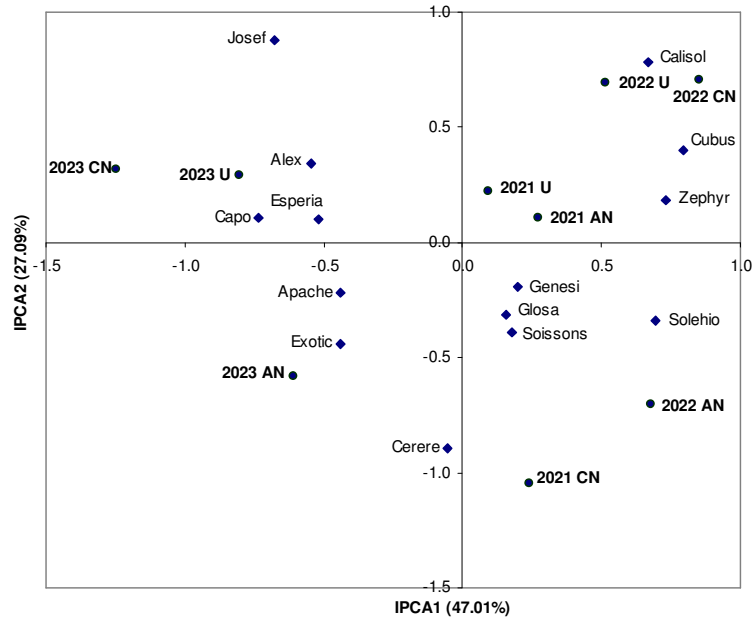
The effect of the genotype on the expression of the spike length in 2022 was higher than the previous year, with variations from 2 cm for the treatment with urea and 3.25 cm for the fertilization with calcium nitrate, respectively. ‘Calisol’ variety showed the most efficient use of urea treatment, achieving significantly higher values of this trait by 1.37-2 cm compared to 50% of the other varieties. Under the fertilization with calcium nitrate, the plants of ‘Cubus’ variety recorded a spike length significantly higher by 22-79% compared to other seven varieties. The treatment with ammonium nitrate was exploited at the highest level by the ‘Cerere’ variety, which showed significant increases of 1.43-2.67 cm compared to spike length of most other varieties.

The amplitude of the spike length in 2023 ranged between 1.30 cm for the treatment with urea and 2.23 cm for the fertilization with ammonium nitrate. Under the effect of urea treatment, the wheat varieties recorded the highest values of this trait, ranging from 7.47 cm for ‘Zephyr’ to 9.93 cm for ‘Capo’. Also, values over 9 cm of spike length were recorded at ‘Alex’, ‘Josef’, ‘Cerere’ and ‘Apache’ varieties. On the soil fertilized with calcium nitrate, the plants of the ‘Alex’ variety showed the highest spike length values, significantly higher than 75% of the other varieties. Under the effect of ammonium nitrate treatment, ‘Cerere’ variety achieved a spike length significantly higher by 15.2-40.3 % than 12 varieties.

The average values of spike length achieved by the 14 varieties showed amplitude of 1.02 cm, ranging from 6.49 cm in ‘Zephyr’ variety to 7.51 cm in ‘Cerere’ variety. Thus, ‘Cerere’ variety achieved significantly higher values of spike length than most of the other varieties, associated with increases between 6% to ‘Cubus’ and 15% to ‘Zephyr’ variety, respectively. The treatment with urea in 2023 showed the highest positive effect on spike length, followed by the treatments with calcium nitrate in 2021 and ammonium nitrate in 2023. Under the conditions from 2022, the fertilization had a low effect on the spike length in all varieties, with significantly lower values to other environments.

Considering that IPCA1 and IPCA2 expresses 77.1% of the G×E interaction from Figure 2 it is observed that the effects of fertilization treatments in 2022 was associated with low values of spike length and high IPCA1 scores, while under the fertilization in 2023 the high values of this trait were associated with low IPCA1 scores. Also, according to the coordinates of each environment it turns out that the treatment with calcium nitrate in 2022-2023 has had the highest interaction with the spike length of wheat varieties, while the

treatments with urea and ammonium nitrate in 2021 showed lower contribution to the genotype × environment interaction.



**Figure 2.** Biplot of principal component axis (IPCA1 and IPCA2) for spike length of 14 wheat varieties in nine environments

The close position of ‘Calisol’ varieties to the treatment with urea in 2022 and also the position of ‘Cubus’ variety to the calcium nitrate treatment in 2022, showed a specific adaptation to these environments, were these varieties registered the highest spike length. In the case of ‘Genesi’, ‘Glosa’ and ‘Soissons’ varieties, their close position to the origin indicates a high stability of spike length associated with low values. Given their distance from the origin, ‘Josef’, ‘Cubus’ and ‘Calisol’ varieties expresses a low stability associated with spike length values close to the overall mean.

*Analysis of grains number/spike*

In the case of grains number/spike, the AMMI analysis of variance indicated highly significant effects of all three sources of variation (Table 6).

**Table 6.** AMMI analysis of variance for grains number/spike of 14 wheat varieties in nine environments

Source of variation	SS	DF	MS	F	SS % <sup>1</sup>
Genotype	13	3022	232.46	11.33**	7.27
Environment	8	22900	2862.50	128.36**	55.12
Genotype × Environment	104	10417	100.16	4.88**	25.07 (100)
IPCA 1	20	5049	252.45	12.30**	48.47
IPCA 2	18	2711	150.61	7.34**	26.02
IPCA 3	16	1239	77.44	3.77**	11.89
IPCA 4	14	942	67.29	3.28**	9.04
IPCA residuals	36	476	13.22	0.64	4.57
Error	234	4803	20.53		

1- % of model sum of squares for genotype, environment and genotype × environment; \*\* Significant at  $p \leq 0.01$

The environment factors accounted the largest contribution (55.12%) to the total sum of squares, while 25.07% and 7.27% of the grain's number/spike variation were attributable to the G×E interaction and genotype effects. The first, second, third and fourth principal components (IPCA) are significant, and explain a total of 95.43% of the interaction sum of squares. The IPCA1 accounted for 48.47% of the G×E interaction, while IPCA2 accounted for 26.02%.

Taking into account the combined effect of climatic conditions and fertilization on the grains number/spike, in 2021 the varieties registered amplitudes from 17.92 for the treatment with calcium nitrate to 24.25 under the treatment with ammonium nitrate (Table 7). 'Glosa' variety achieved the highest value of this trait under the fertilization with urea, associated with significant increases of 10.76-18.84 compared to 50% of the other varieties. 'Alex', 'Solehio', 'Glosa' and 'Exotic' varieties capitalized at a high level the treatment with calcium nitrate, reordering approximately 40.5-42.5 grains/spike, significantly higher than the values associated to 'Cubus', 'Josef', 'Soissons', 'Capo' and 'Calisol' varieties. Under the effect of ammonium nitrate treatment, the plants of 'Calisol' variety achieved 44.22 grains/spike, significantly higher than 10 of the other varieties, followed by 'Apache' and 'Cerere' varieties with 38.36-38.5 grains/spike.

**Table 7.** Means of grains number/spike for 14 wheat varieties in nine environments

Genotype	Environment									Mean G
	2021			2022			2023			
	U	CN	AN	U	CN	AN	U	CN	AN	
Alex	37.24 ab	42.28 a	32.47 bc	35.87 ab	30.40bcde	26.93 bcde	59.00 ab	46.80 a	44.86 cd	39.54 A
Glosa	42.67 a	42.11 a	32.39 bc	27.87bcdef	30.80bcde	30.13 abc	55.07 bcd	31.40 cd	41.43 cde	37.10 B
Esperia	33.54 bcd	36.20abcd	34.39 bc	30.47bcdef	25.47 de	22.40 defg	52.80 bcd	39.33 ab	45.41 bcd	35.56 BCDE
Capo	23.83 e	26.10 f	32.40 bc	28.87bcdef	36.27 ab	36.73 a	56.87abcd	39.07 b	52.42 ab	36.95 B
Josef	29.07 cde	29.40 def	33.47 bc	23.73 f	27.60 cde	18.73 g	58.33 abc	34.47 bc	44.80 cd	33.29 E
Cerere	37.40 ab	35.87abcd	38.36 ab	24.13 ef	42.47 a	32.87 ab	59.87 ab	31.33 cd	55.07 a	39.71 A
Genesi	26.87 de	34.47bcde	28.45 c	31.40bcde	32.13 bcd	26.60bcdef	51.20 cde	34.27 bc	40.16 de	33.95 CDE
Apache	31.91 bcd	33.60bcde	38.50 ab	32.13 bcd	27.13 cde	23.93cdefg	59.47 ab	34.93 bc	47.58 bc	36.58 B
Soissons	28.80 cde	28.17 ef	37.03 ab	31.67 bcd	30.40bcde	32.53 ab	42.67 f	28.53 cd	43.98 cde	33.75 DE
Exotic	28.00 cde	40.53 ab	28.11 c	27.73 cdef	23.93 e	29.67 abcd	44.93 ef	28.60 cd	37.28 ef	32.09 E
Solehio	35.13 bc	42.47 a	19.97 d	24.87 def	31.07bcde	30.53 abc	50.93 de	26.87 d	31.30 fg	32.57 E
Zephyr	31.80 bcd	37.50 abc	20.61 d	25.73 def	34.27 bc	22.33 efg	45.07 ef	18.13 e	25.52 g	29.00 F
Cubus	34.33 bc	31.20 cdef	44.22 a	33.53 bc	24.47 e	19.60 fg	63.67 a	28.33 cd	47.76 bc	36.35 BC
Calisol	34.86 bc	24.56 f	28.03 c	42.47 a	41.67 a	27.13 bcde	58.20abcd	28.53 cd	37.04 ef	35.83 BCD
Mean E	32.53 XY	34.60 X	32.03 YZ	30.03 Z	31.29 YZ	27.15 W	54.15 T	32.19 YZ	42.47 V	35.16

U-Urea; AN-Ammonium nitrate; CN-Calcium nitrate; G-Genotype; E-Environment.

Environment LSD5%=2.31; Genotype LSD5%=2.43; Genotype × Environment LSD5%=7.29

Different letters (a-g) in the columns indicate significant differences (p<0.05) between genotypes.

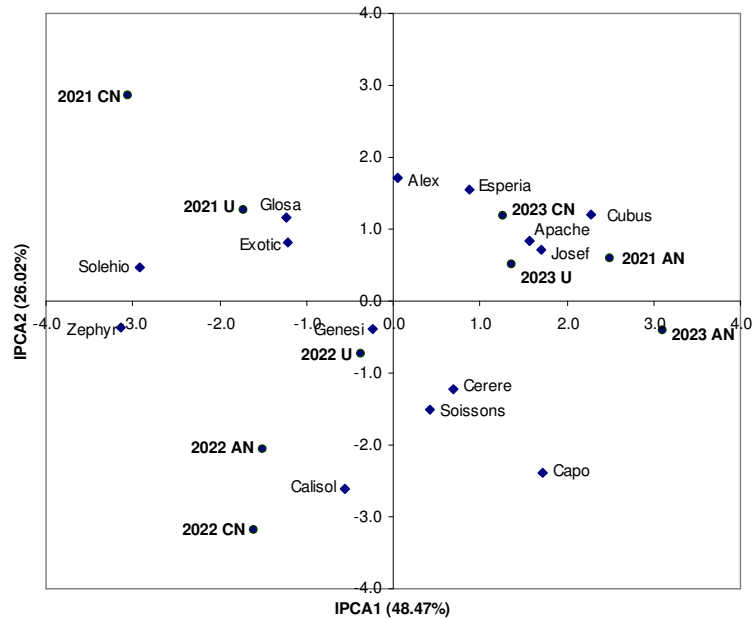
Capital letters were used for environment means (T-W) and genotype means (A-F) comparisons.

Regarding the results from 2022, very close amplitudes of grains number/spike were observed, ranging from 18 for ammonium nitrate treatment and 18.74 grains for the treatment with urea. 'Calisol' variety stood out in particular under the effect of urea, registering a significantly higher grains number/spike compared to 85% of the varieties. The plants of 'Cerere' and 'Calisol' varieties use the most efficient the treatment with calcium nitrate achieving 41.67-42.47 grains/spike, associated with significant increases compared with ten varieties. Under the fertilization with ammonium nitrate, the spikes of 'Capo' variety presented the highest grains number (36.73) registering significant increases of over 35% compared to other eight varieties.

Under the conditions from 2023, the 14 varieties achieved values of grains number/spike with higher amplitudes than in previous years, ranging from 21 for urea treatment and 29.55 for ammonium nitrate treatment. 'Apache' variety achieved under the treatment with urea the highest value (63.67) of this trait over all environments, followed by 'Cerere', 'Apache' and 'Alex' varieties with 59-60 grains/spike. Under the effect

of calcium nitrate treatment, ‘Alex’ variety showed significantly higher value of this trait than 85% of the other varieties. ‘Cerere’ and ‘Capo’ varieties use more efficient the ammonium nitrate fertilization, recording values of 52.42-55.07 grains/spike, associated to significant increases by over 9.75%.

Given the conditions of the combined environments, under the treatments with urea in 2023 the highest values of grains number/spike were recorded followed by the results obtained in 2023 under the effect of calcium nitrate. Generally, the fertilization in 2022 was associated with a low effect on grains number/spike, while under the conditions from 2021 the treatment with calcium nitrate was associated with an increase of this trait. Over all nine environments ‘Cerere’ and ‘Alex’ variety were highlighted by significantly increases of grains number/spike with 6.6-19.28% to other varieties.



**Figure 3.** Biplot of principal component axis (IPCA1 and IPCA2) for grains number/spike of 14 wheat varieties in nine environments

According to the biplot from Figure 3, the first two principal components explained a total of 74.49% of the G×E interaction effects on grains number variability. The variety ‘Genesi’ located close to the origin are considered to be the most stable regarding the variation of this trait in the studied environments, but its stability was associated with a low grain’s number/spike. The varieties ‘Solehio’, ‘Zephyr’ and ‘Cubus’ appeared unstable considering their significant distance from the biplot’s origin. The environments represented by the treatments with calcium nitrate in 2021 and with ammonium nitrate in 2023 have had the highest contribution to the G×E interaction, since they were positioned far from the origin. Under the treatments with urea in 2022 and 2023, the grains number/spike exhibited the lowest variability, indicating that these two environments were the main contributors to the phenotypic stability of this trait for the studied varieties. ‘Cubus’, ‘Apache’ and ‘Josef’ varieties showed a specific adaptation to the treatment with urea in 2023, recording the highest value of this trait for all environments.

*Analysis of grains weight/spike*

The results of the analysis of variance for grains weight/spike show that the genotype, environment and interaction effects are significant (Table 8). The environment showed the highest influence (60.94%) on the variability of this trait, followed by G×E interaction (17.27%), amid a lower influence (8.29%) of genotype.

**Table 8.** AMMI analysis of variance for grains weight/spike (g) of 14 wheat varieties in nine environments

Source of variation	SS	DF	MS	F	SS % <sup>1</sup>
Genotype	13	8.1	0.62	12.49**	8.29
Environment	8	59.55	7.44	88.83**	60.94
Genotype × Environment	104	16.88	0.16	3.25**	17.27 (100)
IPCA 1	20	6.48	0.32	6.50**	38.39
IPCA 2	18	5.59	0.31	6.23**	33.12
IPCA 3	16	2.18	0.14	2.73**	12.91
IPCA 4	14	1.16	0.08	1.66	6.87
IPCA residuals	36	1.47	0.04	0.82	8.71
Error	234	11.67	0.05		

1- % of model sum of squares for genotype, environment and genotype × environment; \*\* Significant at  $p \leq 0.01$

The high contribution of the environment to the variation of the spike length indicates the existence of major differences between the effects of fertilization treatments during the three years. Given the decomposition of the G×E interaction through the AMMI model, it was observed that the first four principal components express 91.29 % of this interaction effect.

In the conditions of 2021, the wheat varieties recorded spike yield amplitudes between 0.59 g under the treatment with ammonium nitrate and 1.05 g for urea treatment (Table 9). ‘Solehio’ variety achieved the highest spike yield (1.84 g) under the treatment with urea, associated with significant increases compared to six other varieties. Under the effect of fertilization with calcium nitrate, ‘Exotic’ variety registered a grain weight/spike of 1.91 g significantly higher than the other six varieties, followed by the ‘Solehio’ and ‘Glosa’ varieties with 1.77-1.78 g. The plants of ‘Cubus’ variety use most effective the treatment with ammonium nitrate, recording a spike yield of 1.53 g, significantly higher to ‘Zephyr’ variety.

**Table 9.** Means of grains weight/spike (g) for 14 wheat varieties in nine environments

Genotype	Environment									Mean G
	2021			2022			2023			
	U	CN	AN	U	CN	AN	U	CN	AN	
Alex	1.58 abc	1.66 abc	1.13 bcd	1.64 a	1.24 cd	1.33 abc	3.02 a	2.36 a	2.11 a	1.78 A
Glosa	1.71 abc	1.77 ab	1.37 ab	1.25 bcde	1.42 abcd	1.38 ab	2.84 ab	1.60 b	2.14 a	1.72 A
Esperia	1.38 cde	1.39 cde	1.21 abcd	1.18 bcde	1.07 d	0.95 d	2.27 efg	1.56 bc	1.85 abc	1.43 CDE
Capo	1.11 cde	0.98 f	1.03 bcd	1.29 abcde	1.55 abc	1.51 a	1.92 gh	1.33 bcd	1.53 cde	1.36 E
Josef	1.53 fg	1.21 def	1.16 bcd	1.01 de	1.13 d	0.97 d	2.85 ab	1.24 cd	1.82 abcd	1.44 CDE
Cerere	1.48 bcd	1.49 bcd	1.32 abc	0.96 e	1.71 a	1.08 bcd	2.78 abc	1.27 bcd	1.98 ab	1.56 B
Genesi	1.03 abcd	1.56 abcd	1.19 abcd	1.33 abcd	1.28 bcd	1.25 abcd	2.53 bcde	1.31 bcd	1.95 ab	1.49 BCD
Apache	1.15 abc	1.26 def	1.20 abcd	1.30 abcde	1.07 d	0.98 cd	2.61 bcde	1.20 d	1.70 bcd	1.38 DE
Soissons	0.79 g	1.07 ef	1.37 ab	1.21 bcde	1.18 d	1.23 abcd	2.03 fgh	1.16 d	2.15 a	1.35 E
Exotic	1.15 def	1.91 a	1.28 abcd	1.25 bcde	1.17 d	1.42 ab	2.33 def	1.26 bcd	1.94 ab	1.52 BC
Solehio	1.84 a	1.78 ab	1.00 cd	1.10 cde	1.34 bcd	1.40 ab	2.47 cde	1.26 bcd	1.49 de	1.52 BC
Zephyr	1.16 ab	1.25 def	0.94 d	1.03 cde	1.41 abcd	0.91 d	1.79 h	1.03 d	1.22 e	1.19 F
Cubus	1.50 abcd	1.09 ef	1.53 a	1.37 abc	1.37 abcd	0.97 d	2.74 abc	1.26 bcd	2.02 ab	1.54 BC
Calisol	1.62 efg	1.62 abc	1.11 bcd	1.47 ab	1.63 ab	1.19 abcd	2.67 abcd	1.20 d	1.73 bcd	1.58 B
Mean E	1.36 XY	1.43 X	1.20 ZW	1.24 YZW	1.33 XYZ	1.18 W	2.49 T	1.36 XY	1.83 V	1.49

U-Urea; AN-Ammonium nitrate; CN-Calcium nitrate; G-Genotype; E-Environment.

Environment LSD5%=0.14; Genotype LSD5%=0.12; Genotype × Environment LSD5%=0.36

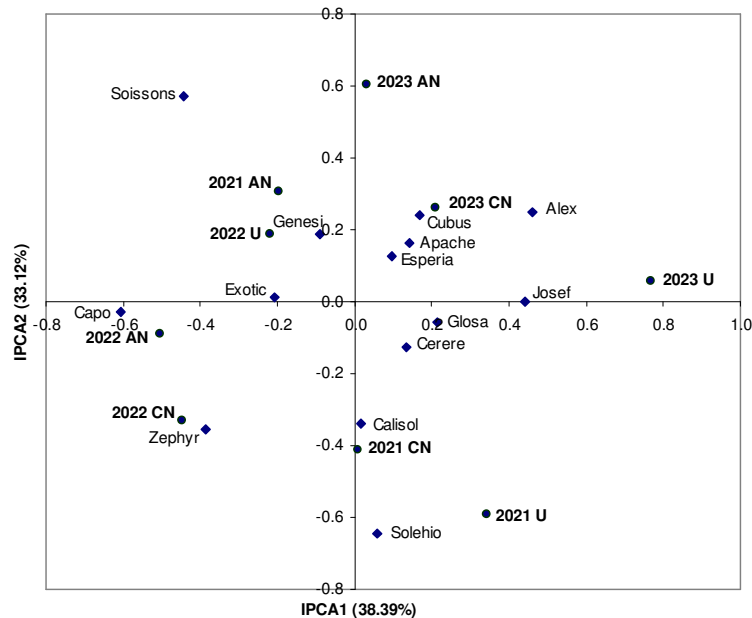
Different letters (a-g) in the columns indicate significant differences ( $p < 0.05$ ) between genotypes.

Capital letters were used for environment means (T-W) and genotype means (A-F) comparisons.

The effect of the genotype on the expression of the spike yield in 2022 was lower than the previous year, with variations from 0.6 g for the treatment with ammonium nitrate to 0.68 g for the fertilization with urea, respectively. Under the effect of fertilization with urea, the plants of ‘Alex’ variety achieved significantly higher spike yields than ‘Josef’, ‘Cerere’ and ‘Zephyr’ varieties, while the variations between the other varieties were small and non-significant. ‘Cerere’ variety was highlighted in the case of treatment with calcium nitrate, achieving significantly higher spike yield compared to ‘Esperia’, ‘Josef’ and ‘Apache’ varieties. Most varieties have valued at a similar level the fertilization with ammonium nitrate by recording spike yield of 0.95-1.42 g, except for the ‘Capo’ variety which has achieved a significantly higher value compared to ‘Zephyr’ variety.

The 14 varieties showed under the conditions from 2023 values spike yield with amplitudes superior to the previous years, from 0.93 g for the treatment with ammonium nitrate and 1.33 g for the treatment with calcium nitrate. ‘Alex’ variety exploited more effectively the fertilizations with urea and calcium nitrate, registering significant increases compared to the rest of the varieties on the calcium nitrate treatment and six of the varieties under the effect of urea treatment. In the case of ‘Soissons’, ‘Alex’ and Glosa varieties, the fertilization with ammonium nitrate has shown the highest efficiency on spike yield, offering the possibility of obtaining significantly higher values than ‘Capo’, ‘Solehio’ and ‘Zephyr’ varieties.

The average values of spike yield achieved by the 14 varieties showed amplitude of 0.59 g, ranging from 1.19 g in ‘Zephyr’ variety to 1.78 in ‘Alex’ variety. Thus, ‘Alex’ and ‘Glosa’ variety achieved significantly higher values of spike yield than the other varieties, associated with increases between 10.25% to ‘Cerere’ and 49.58% to ‘Zephyr’ variety, respectively. The treatment with urea in 2023 showed the highest positive effect on spike yield, followed by the treatments with ammonium nitrate in 2023 and calcium nitrate in 2021. Under the conditions from 2022, the fertilization with ammonium nitrate had a low effect on the spike yield in all varieties, with significantly lower values to other environments.



**Figure 4.** Biplot of principal component axis (IPCA1 and IPCA2) for grains weight/spike of 14 wheat varieties in nine environments

Considering that IPCA1 and IPCA2 expresses 71.51% of the G×E interaction from Figure 4 it is observed that the effects of urea treatment in 2022 was associated with highest values of spike yield and high positive IPCA1 scores, while under the fertilization in 2022 and with ammonium nitrate in 2021 the lowest spike yield was associated with low IPCA1 scores. Also, according to the coordinates of each environment it

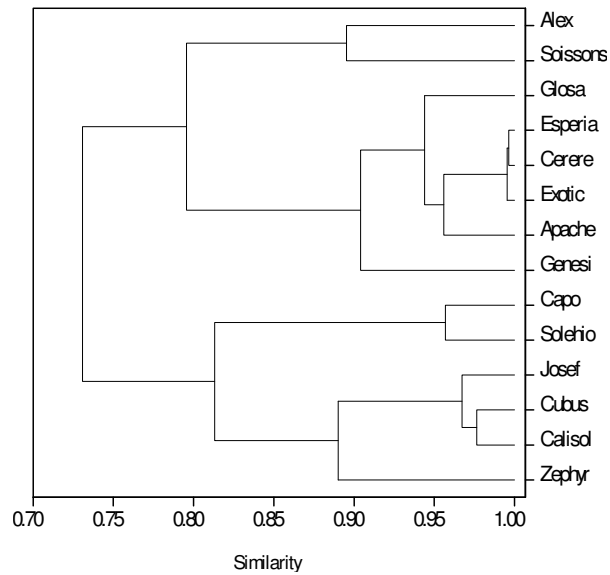
turns out that the treatment with urea in 2021 and 2023 has had the highest interaction with the spike yield of wheat varieties, while the treatments with urea in 2022 and with calcium nitrate in 2023 showed lower contribution to the genotype  $\times$  environment interaction. The close position of 'Capo' variety to the treatment with ammonium nitrate in 2022 and also the position of 'Calisol' variety to the calcium nitrate treatment in 2022 showed a specific adaptation to these environments, where these varieties registered the highest spike yield. In the case of 'Esperia', 'Cerere' and 'Genesi' varieties, their close position to the origin indicates a high stability of spike yield associated with values around the overall mean. Given their distance from the origin, 'Soissons', 'Capo' and 'Solehio' varieties express a low stability associated with spike yield below the overall mean.

#### *Evaluation of multitrait stability*

For a multitrait stability evaluation of wheat varieties the average ranks and its coefficient of variation associated to ASV were used (Table 10). Thus, it can be observed that 'Genesi' variety showed the highest stability of the analyzed traits. In the case of 'Cerere' variety the high overall stability was associated with a low  $G \times E$  for spike yield and plant height and average stability for spike length. 'Esperia' variety expressed the highest stability for spike yield and average stability for the other traits. The environmental conditions influenced to the same extent the phenotypic expression of the four traits in 'Exotic' variety, which showed an average stability. The performances of 'Capo' variety were highly influenced by the environmental conditions, on the background of low stability for all traits, especially for plant height and spike yield. Also, a low stability of all traits was observed at 'Solehio' variety, followed by 'Josef' and 'Cubus'.

**Table 10.** Multitrait stability of wheat varieties based on AMMI stability value (ASV)

Genotype	ASV				Ranks	
	Plant height	Spike length	Grains number/spike	Grains weight/spike	Average	Coefficient of variation
Alex	6.465	1.011	1.709	0.590	8.25	45.76
Glosa	4.208	0.419	2.582	0.255	6.25	52.86
Esperia	3.197	0.911	2.250	0.171	4.75	55.37
Capo	11.593	1.283	4.006	0.703	12.25	12.24
Josef	3.605	1.470	3.239	0.511	10.00	29.44
Cerere	2.535	0.898	1.766	0.199	3.75	45.54
Genesi	0.958	0.399	0.572	0.215	1.75	54.71
Apache	6.805	0.797	3.046	0.230	7.25	54.45
Soissons	3.059	0.495	1.700	0.766	6.00	91.29
Exotic	2.944	0.885	2.404	0.240	5.00	16.33
Solehio	8.418	1.253	5.473	0.649	11.75	16.11
Zephyr	0.759	1.281	5.853	0.569	8.75	62.86
Cubus	4.052	1.437	4.405	0.311	10.00	29.44
Calisol	4.106	1.397	2.816	0.340	9.25	20.46



**Figure 5.** UPGMA clustering of wheat varieties based on the GE interaction

Based on the stability of the studied traits in the nine environments, the genotypes were grouped into four clusters (Figure 5). The first cluster is composed of 'Alex' and 'Soissons' varieties that show a low stability for plants height and spike yield. From the second cluster, 'Esperia', 'Cerere' and 'Exotic' varieties which express a high stability of spike length and yield, are highlighted. Also, as a part of the second cluster, 'Genesi' variety showed the highest stability of all traits. The third cluster consists of 'Capo' and 'Solehio' varieties, in which the  $G \times E$  interaction has shown the greatest influence on the four traits. In the case of 'Josef', 'Cubus', 'Calisol' and 'Zephyr' varieties, the  $G \times E$  interaction had a high influence on the length and the number of grains/spikes.

## Discussion

The combined environmental conditions had the main influence on the phenotypic expression of the studied traits, with contribution to total variation from 53.33% in the case of spike length to 60.94% for spike yield. The high influence of environment to the variation of yield traits in wheat growth under stress conditions is consistent with previous studies (Al-Sayaydeh *et al.*, 2023; Omrani *et al.*, 2022). A higher contribution of environmental effects to the total variances than the genotype and  $G \times E$  effect was also reported by Tanin *et al.* (2022) for plant height (41.54%) and grains number/spike (81.21%), while Ljubicic *et al.* (2022) reported 48.8% for spike index. The high effect of the environment was mainly due to the fact that the variation of weather conditions during the study strongly influenced the effect of fertilization treatments. Similar findings were reported by another study on spike yield stability in wheat under saline stress (Petrovic *et al.*, 2012). The highly contribution of the environment effect to the total differences for all traits indicates that the response of studied wheat varieties to variation of the environmental factors was very different, implying that selection of environment-specific varieties is required (Enyew *et al.*, 2021).

The genotype contributed to the total variation of the four traits with 7.27-9.44% for grains number/spike, grains weight/spike and spike length, while the genotype effect on plant height was significantly higher (21.32%). The contribution of the genotype to the variation of plant height indicates that for these varieties this trait is more stable compared with the spike traits (Popovic *et al.*, 2020).

The results of our study have revealed a G×E interaction of 17.27-18.91% for grains weight per spike and plant height, and 24.45-25.07% for spike length and grain number/spike, respectively. This contribution of G×E interaction to the phenotypic expression of different traits resulted from genotypes with low potential that are unable to use the advantage of favourable condition (Padi, 2007). These interactions are of special interest when they produce inconsistencies in relative evaluation of genotypes measured by changes in the rank (of traits levels) of genotypes namely crossover interactions (Leon *et al.*, 2016). Given that the climate change increases the crossover interactions, it represents a critical indicator for changes of cultivar performances under different environmental conditions (Xiong *et al.*, 2021).

Taking into account the genotype × year interaction, it can be seen that ‘Esperia’, ‘Cerere’, ‘Cubus’, ‘Genesi’ and ‘Calisol’ varieties recorded a low variation under the climatic conditions from 2021 and 2023 and at the same time they achieved significantly higher values of plant height compared to the results from 2022. The other varieties express a low stability of plant height registering significant variations of this trait over the years, with the highest values in 2023 and the lowest under the low amount of rainfall from 2022.

The genotype × year interaction had a high effect on spike length in the case of ‘Alex’, ‘Esperia’, ‘Capo’, ‘Josef’, ‘Cerere’ and ‘Apache’ varieties. ‘Glosa’, ‘Genesi’, ‘Soissons’, ‘Exotic’, ‘Solehio’ and Zepyr varieties showed a stability of the spike length in 2021 and 2023 recording significantly higher values than in 2022.

In the case of grains number/spike, ‘Alex’, ‘Josef’, ‘Cerere’, ‘Genesi’, ‘Apache’, ‘Soissons’, ‘Exotic’ and ‘Solehio’ varieties exhibited low genotype × year interaction during 2021-2021, showed a significant decrease of grains per spike compared to the results from 2023. Based on significant variations of grains number/spike over the years, the other varieties had a low stability of this trait.

Most of the varieties showed a stability of the spike yield during 2021-2022 associated with significantly lower values compared to those from 2023. ‘Genesi’ exhibited a different genotype × year interaction, displayed great stability and performed well under the conditions from 2022-2023.

From the four studied traits, the spike yield and grain numbers per spike were more sensitive to drought stress in accordance with the findings of Bapela *et al.* (2022). These are consequences of the fact that drought at anthesis produces abortion of ovules and as such reduce the grains number per spike and grain weight (Pradhan *et al.*, 2012; Weldearegay *et al.*, 2012). Also, during grain filling, water deficit may disrupt the uptake of nutrient and photosynthesis, what leads to the production of shrivelled grains (Iqbal, 2019). The low level of rainfall from 2022 has led to a reduction in the values of the four traits: 19.25-27.98% for plant height; 15.28-26.56% for spike length; 4-40% for grains per spike; 7.05-51.76% for spike yield. These negative impacts of drought on different traits in wheat were also reported by several studies: 14.7-34.5% for plant height; 23.7% for spike length; 38.1-50% for grains per spike; grain yield 40-62.75% (Afzal *et al.*, 2017; Bayoumi *et al.*, 2008; Khokhar *et al.*, 2017; Ljubicic *et al.*, 2021; Mkhabela *et al.*, 2020; Mwadzingeni *et al.*, 2018; Prasad *et al.*, 2011; Zhang *et al.*, 2018).

Considering the genotype × fertilization interaction, the plants of ‘Josef’, ‘Soissons’ and ‘Zephyr’ varieties showed a stability of height, registering low and non-significant variations between treatments. In the case of other varieties, the fertilization had a significant effect of the plant height. At ‘Alex’, ‘Esperia’, ‘Cubus’, ‘Exotic’ and ‘Glosa’ varieties, the spike length showed low genotype × fertilization interaction under the effect of the three nitrogen treatments. ‘Alex’, ‘Esperia’, ‘Genesi’ and ‘Exotic’ varieties exhibited a stability of the grain’s number/spike associated with close values of this trait regardless the nitrogen treatments. The grains per spike in ‘Cubus’ variety was the most influenced by the fertilization. The spike yield of ‘Esperia’, ‘Capo’, ‘Cerere’ and ‘Genesi’ exhibited low genotype × fertilization interaction while in the case of ‘Alex’ and ‘Cubus’ varieties the grains weight per spike was highly influenced by the nitrogen treatments.

The treatment with urea showed a superior effect compared to the other treatments especially under the conditions from 2023, characterized by a higher level of rainfall. These results are consistent with the findings that the fertilization with urea is more suitable for environments with higher water availability because its rapid

solubilization and infiltration into the soil decreases the nitrogen losses due to ammonia volatilization (Ferreira *et al.*, 2021).

Under the treatment with calcium nitrate the plants height was superior to other treatments regardless the climatic conditions. Also, the fertilization with calcium nitrate caused an increase of grains number and weight per spike in 2021-2022, having a superior effect compared with urea and ammonium nitrate treatments. In a study conducted on eutriccambisol Cui *et al.* (2023) found that the treatments with urea and calcium nitrate had similar effects on grains number per spike of two wheat varieties, while the highest grains yield under the treatment with urea was affected by genotype  $\times$  year interaction. The ammonium nitrate showed a lower effect on grains number and weight per spike during 2021-2022 compared to the other treatments, while under the favourable conditions from 2023 the effect of ammonium nitrate was higher to calcium nitrate. The results can be due that a single supply with ammonium nitrogen accelerated the crop senescence and decrease the leaf area, resulting a slow photosynthetic rate associated with a short photosynthetic time, thus lower grain weight and yield (Luo *et al.*, 2020).

Against the background of the unfavourable weather conditions during the vegetation period of 2022 characterized by a low amount of rainfall, the salinity of the soil had a pronounced negative effect, causing a considerable decrease of the four traits regardless of the applied fertilization. These results are in accordance with the findings that salinity stress inhibited the division and elongation of cell, photosynthesis and assimilation of nutrients and finally reduced the plant height and spike length in wheat (Kalhor, 2016). These results indicate that biomass at flowering could be a reliable indicator of plant tolerance and performance under salinity stress conditions (Chamech, 2015). Under saline stress the organ's reproductive senescence is accelerated (Khataar *et al.*, 2018), with negative effect of spike yield components by decreasing the number of filled grains per spike and the grains weight (Darko *et al.*, 2017; Sen *et al.*, 2022). A stable canopy development of wheat plant during the vegetative phase provides enough pre-anthesis reserves of nitrogen for grain filling and thereby ensures yield stability (Wang, 2023).

Under the conditions analysed in this study, 'Alex' and 'Glosa' varieties with the best average spike yield did not underperform in relatively unfavourable conditions (Stella *et al.*, 2023). This was regarded as a result of good adaptation of these two Romanian varieties, able to achieve appropriate performances under both favourable environmental conditions as well as in marginal environments such as saline soils (Welcker *et al.*, 2022).

## Conclusions

The significant effect of G $\times$ E interaction on the phenotypic expression of different traits indicates differential response of the varieties across the testing environments, thus offering the possibility of identifying some varieties of interest. The variety 'Cerere' displays higher stability over the environments associated with a spike yield close to the overall mean. This variety showed a specific adaptation to the fertilization with calcium nitrate under rainfall deficit, and to the fertilization with urea and ammonium nitrate under appropriate rainfall conditions. The highest stability of 'Genesi' variety was associated with performances close to the mean under unfavourable rainfall and above the mean under the fertilization with urea and ammonium nitrate in favourable rainfall conditions. The Romanian varieties 'Alex' and 'Glosa' present the highest productivity of spike associated with an average stability, being specifically adapted to favourable rainfall conditions, achieving above-average yields in less favourable conditions. As such, these varieties can be recommended for cultivation on saline soils in environments with similar ecological conditions, especially under fertilization with urea, in order to obtain sustainable yields. The previously mentioned varieties are useful for further studies in order to dissect the genetic and physiological bases that ensure their performance under salinity conditions. Therefore,

they can be selected for crossing and included in wheat breeding programs for the development of stable cultivars adapted to saline soils.

### Authors' Contributions

Conceptualization: GV, EO, SC, and AC; Methodology: DC, MM, IR, EO; Data analysis and interpretation: SC, GV and AC; Writing- original draft: GV, TI, MM, and DC; Writing-review and editing: GV, SC, IR, TI and AC. All authors read and approved the final manuscript.

### Ethical approval (for researches involving animals or humans)

Not applicable.

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### Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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