Assessment of ecological stability in yield for breeding of spring barley cultivars with increased adaptive potential

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Introduction

One of the most significant global problems among those facing agricultural production and science is the unprecedented growth of the world’s population, which requires increasing production of the main crops by almost 100% by 2050 (Godfray et al., 2010; Tilman et al., 2011).

The current dynamics in crop yield increase do not correspond to the necessary rates to ensure the parity between production and consumption (Ray et al., 2013). The solution of the problem of providing food to humanity is complicated by global climate changes (Smith & Gregory, 2013; Moore & Lobell, 2015). Barley (Hordeum vulgare L.) is one of the five major crops in world agriculture (Ullrich, 2011). Therefore, barley grain production augmentation, as a result of increasing the genetic potential of productivity along with its stability, is one of the priority breeding tasks. Ukraine is one of the largest producers and exporters of barley grain. However, the territory of Ukraine is characterized by significant differences between agroclimatic zones and subzones, which require special attention to be paid to increasing adaptability in new cultivars. As an example, under conditions of the Ukrainian Forest-Steppe, modern spring barley cultivars should be characterized with high yield potential, as well as drought tolerance, lodging resistance, and resistance to a number of the most widespread pathogens (Hudzenko et al., 2017).

Grain yield, as the main integral economic characteristic of any commercial cultivar is formed under the influence of numerous different environmental factors. Genotypes differ in the efficiency of assimilation and commercial cultivar is formed under the influence of numerous different environmental factors. Genotypes differ in the efficiency of assimilation and commercial production and science is the unprecedented growth of the world’s population, which requires increasing production of the main crops by almost 100% by 2050 (Godfray et al., 2010; Tilman et al., 2011).
It rarely happens that the ranges of vital factors of growing conditions coincide with those optimal for plants. At least one factor is limiting. Its effect on gene products (proteins – enzymes) leads to the epigenetic regulation of genes and modifies the phenotypic manifestation of quantitative traits. The presence of the epigenetic regulation of gene expression causes elementary adaptive reactions, which are stages in the chain of genotype by environment realization and are ultimately expressed in quantitative and qualitative traits specific to it. Genotypes providing a relatively stable level phenotypic manifestation of traits in different environments are characterized by wide adaptation and high ecological stability (Zhabchenko, 1988). Genotypes prevailing only in a certain environment are specifically adapted and have low ecological stability. Specific adaptation is closely related to the genotype by environment interaction. This phenomenon is one of the central problems of plant breeding theory and practice (Hill, 1975; Zhao & Xu, 2012). There are different situations when genotype by environment interaction occurs: divergence, convergence, and cross-over interaction (Malosetti et al., 2013). Cross-over genotype by environment interaction is the most important for breeders as it implies that the choice of the best genotype is determined by the environment. Therefore, the selection of genotypes in some conditions does not guarantee their advantage in other conditions.

Thus, the aim of the present study is to theoretically substantiate and practically test the scheme of multi-environment trials, as well as interpret experimental data using modern statistical tools for evaluation of the genotype by environment interaction, and highlight the best genotypes for combining yield performance and ecological stability at the final stage of the spring barley breeding process.

Materials and methods

Plant material and experimental design. At the first stage in competitive strain testing at the V. M. Remeslo Myronivka Institute of Wheat (MIW) of National Academy of Agrarian Sciences of Ukraine (NAAS) (Central part of the Forest-Steppe of Ukraine) in 2016 we selected nine spring barley breeding lines Nutans 4066 (GRB/Luchezarnyi), Nutans 4982 (Sebastyan/Yukatan), Nutans 5069 (Bojos/Obolon), Deficiens 5145 (Celinka/Yakub), Nutans 5130 (Vivaldi/Elbon), Nutans 5152 (Vivaldi/Elbon), Nutans 5157 (Vivaldi/Elbon), Nutans 5032 (Sorozinsky/Celinka), Nutans 5093 (Seleni/Katrin) with a combination of valuable traits. These traits were yield performance, lodging resistance, tolerance to drought and resistance to widespread pathogens (Blumeria (Erysiphe) graminis (DC.) Golovin ex Speer & sp. hordei Em. Marchal, Pyrenophora teres Drechs., Bipolaris sorokiniana (Sacc.) Shoem., Pyrenophora graminis & A. K. Kurth. and Puccinia hordei Oth.). During the second stage in 2017 and 2018, we investigated the promising spring barley lines in three agroclimatic zones (Central part of the Forest-Steppe, Polissia and Northern Steppe of Ukraine). For a more reliable assessment, the breeding lines were compared not only with standard cultivar Vzirets, but also with the most widespread cultivars in production conditions of Ukraine which were developed in different Ukrainian institutions: Virazh, Talisman Myronivskyi, MIP Myrnyi, MIP Sotnyk (MIW); Sviatohir, Vakula (Plant Breeding and Genetics Institute – National Centre of Seed and Cultivar Investigation of NAAS), Dobuz, Aleho, Modern and Pan (The Plant Production Institute n.d. a. V. Y. Yuriev of NAAS). The spring barley cultivars were sown annually in the same block of competitive testing with the breeding lines. The trial was laid out with randomized complete block factors can be visualized simultaneously. Mathematically, a biplot is regarded as a graphical display of matrix multiplication (Yan & Tinker, 2006; Hongyu et al., 2014). Comparisons of AMMI and GGE biplot, their similarities, peculiarities and advantages have been discussed in several papers (Yan et al., 2007; Gauch et al., 2008). AMMI and GGE biplot analyses were performed when using non-commercial software GEA-R, version 4.1. Software review is provided in the publication (Fruntos et al., 2014). Analysis of variance of the AMMI conducted accordingly to approach described by Collob (1968).

Results

Spring barley genotypes yield performance. Grain yield of the spring barley genotypes significantly varied, depending on agroclimatic zones and meteorological conditions of the year (Table 2). We noted significant differences in yield performance of the genotypes throughout all environments (zones and years combinations). The maximum yield in the environments K16 (8.25 t/ha) and M17 (5.35 t/ha) was observed for the breeding line G10, in the M18 for the breeding line G7 (3.95 t/ha), in the N17 for the breeding line G3 (7.09 t/ha), in the N18 for the cultivar G15 (6.45 t/ha), in the K17 for the cultivar G13 (4.89 t/ha), in the K18 for the breeding line G5 (4.06 t/ha). The highest mean yield was in the breeding line G7 (5.46 t/ha), the lowest mean yield was in the cultivar G17 (4.26 t/ha). Breeding lines G3, G4, G5, G7, and G10, as well as cultivar G13 reliably predominated in yield over the standard cultivar G1. The breeding lines G2, G6, G8, G9 and the cultivars G11, G12, G18 and G19 had a slightly higher yield than the standard cultivar G1. The cultivars G14 and G15 had yield near to the standard cultivar G1. The cultivars G16, G17 and G20 were significantly inferior in yield compared to the standard. Thus, we found significant variability of the mean yield for all tested genotypes in different environments, as well as a changing in individual geno-
type yield ranks throughout the environments. It clearly indicates the presence of a cross-over genotype by environment interaction.

**Additive main effects and multiplicative interaction.** Analysis of variance of the AMMI (Gollob, 1968) showed a significant predomination of the contribution of environmental conditions to the total variation (82.8%) (Table 3). The genotype by environment interaction percentage was 12.5%, and the genotype contribution it was 4.7%. Despite the last two parameters having low numerical values, they were reliable. The first two principal components of the AMMI (Factor 1, Factor 2) explained 81.1% of the genotype by environment interaction. AMMI1 biplot (Fig. 1) allows graphic analysis of the variance of genotypes and test environments and the interaction between them. The variation of the main additive effects (mean yield) of genotypes (G1–20) and environments (M16–K18) is located on the horizontal axis (YLD). The first principal component of the multiplicative effects variation of the genotype by environment interaction is located on the vertical axis (Factor 1). The vertical line, which passes through origin of the AMMI1 biplot represents the grand mean yield in the trial (mean for all genotypes). The lines with the arrows at the end indicate the environment vectors. The vector length allows visualization of the remoteness of a certain environment from the origin of the AMMI1 biplot along the mean yield axis or the first principal component values axis.

Table 2

| Code | Genotype (cultivar/breeding line) |
|------|----------------------------------|
| G1   | Voriz (standard)                 |
| G2   | Nutans-4966                      |
| G3   | Nutans-4982                      |
| G4   | Nutans-5069                      |
| G5   | Deficiesz 5145                   |
| G6   | Nutans-5150                      |
| G7   | Nutans-5157                      |
| G8   | Nutans-5032                      |
| G9   | Nutans-5033                      |
| G10  | Nutans-5039                      |
| G11  | Vinzh                            |
| G12  | Talismean Myronivskyi            |
| G13  | MIP Myronivsky                   |
| G14  | MIP Sotnyk                       |
| G15  | Svitatorh                        |
| G16  | Vakula                           |
| G17  | Dokaz                           |
| G18  | Akhle                           |
| G19  | Pan                              |
| G20  | Modern                          |

Note: * – The V. M. Remeslo Myronivka Institute of Wheat of NAAS, ** – Nosivka Plant Breeding and Experimental Station of the V. M. Remeslo MIW of NAAS, *** – Institute of Agriculture of Steppe of NAAS.

Table 3

| AMMI model analysis of variance (Golob’s test) of yield of the spring barley genotypes and sum of squares decomposition of the genotype by environment interaction |
|---------------------------------------------------------------|
| Source of variation | Sum of squares | Degree of freedom | Mean square | Percentage relative to the sum of squares |
|---------------------|----------------|------------------|-------------|----------------------------------------|
| Genotype (G)        | 46.00          | 19               | 2.42        | 4.78%                                  |
| Environment (E)     | 816.79         | 6                | 136.13      | 82.88%                                 |
| G x E interaction   | 123.08         | 114              | 1.08        | 12.58%                                 |
| Factor 1*           | 75.18          | 24               | 3.13        | 61.11%                                 |
| Factor 2            | 24.63          | 22               | 1.12        | 20.01%                                 |
| Factor 3            | 10.75          | 20               | 0.54        | 8.77%                                  |
| Factor 4            | 8.47           | 18               | 0.47        | 6.90%                                  |
| Factor 5            | 2.88           | 16               | 0.18        | 2.3%                                   |
| Factor 6            | 1.17           | 14               | 0.08        | 1.0%                                   |
| Factor 7            | 0.00           | 12               | 0.00        | 0.0%                                   |
| Residuals           | 9.55           | 280              | 0.03        | 0.0%                                   |

Note: * – Factor 1–7 – principal components, ** – significant at 1% probability level.

The environment M16 had maximum distance from the biplot origin. The environments M16 and N17 were the most productive, and the environments K18 and M18 were the least productive. The more desirable are genotypes with high productivity and located closer to zero along the vertical axis. In our case genotypes can be divided into several groups: 1) genotypes that had higher yield performance than mean yield in the trial and combined it with a high stability (G5, G13, G4, G3, G7 and G10); 2) genotypes with yield performance close to the mean yield in the trial and with relatively high stability (G12, G11, G14, G2, G8, G9, G6 and G19); 3) genotypes with yield performance close to the mean yield in the trial and with high variability (G18 and G15); 4) genotypes that had poor yield performance but had high relative stability (G17 and G16); 5) the genotype G20 that had yield lower than mean yield in trial and combined it with maximal variability.

The AMMI2 biplot (Fig. 2) displays the multiplicative effects of genotype by environment interaction in the coordinates of the first (Factor 1) and second (Factor 2) principal components. It is possible to visualize dispersion of the genotypes and environments in the principal component coordinates. Genotypes that shifted from the origin of the AMMI2 biplot towards the specific environment have a stronger positive reaction to it. The stable genotypes throughout environments should be located as close as possible to the origin of the AMMI2 biplot. Accordingly, the breeding line G5 had the highest stability, and the cultivars G15 and G17 were the most variable.
Genotype main effects plus genotype by environment interaction. Figure 3 shows the representativeness and discriminating power of environments according to the GGE biplot. The first two principal components (axis 1 and axis 2) explained 81.6% of the genotype by environment interaction. The line that intersects the biplot origin is the average environment axis (AEA). The average environment is represented on the AEA with a small circle at the end of the arrow. The dashed lines indicate the vectors of individual test environments. The length of the vector characterizes the discriminating power of an environment. Particularly, the environment M16 had the longest vector, and accordingly, it was characterized by the highest discriminating power. The lowest discriminating power was in the environment N17. The angle between an environment vector and the AEA shows its representativeness. A test environment that has smaller angle with AEA is more representative than other test environments. Thus, the highest representativeness was in the environment M18, the vector of which coincided with the AEA. The environments M16 and K17 were the most distant from each other, as is evidenced by the largest angle between their vectors. The environments N17 and M17, as well as K18 and N18 were more similar to each other, since they had the smallest angle between their vectors.

The GGE biplot “which-won-where” polygon view is an effective tool to visualize the interaction patterns between genotypes and environments (Fig. 4). The polygon is formed by connecting the genotypes that are farthest away from the origin of GGE biplot, such that all other genotypes are included within the polygon. A set of perpendiculars to each side of the polygon lines divide the GGE biplot into several sectors. The sectors at the vertex of the polygon contain genotypes that have an advantage in a single environment or in a set of environments (mega-environment).

In our study, three sectors contained both environments and genotypes. The first mega-environment included four environments N17, M17, M18 and K18. The breeding line G7 had advantage in it. Breeding lines G10, G3, G4, G5 and cultivar G13 were also located in the first mega-environment. The second sector included only the environment M16 and breeding lines G6, G9, G2 and G8. It should be noted that the breeding line G10, which was in the first mega-environment, also aspired towards the environment M16, since this breeding line had the highest productivity in this environment. However, the high yield performance in the individual environments that formed the first mega-environment determines the localization of the breeding line G10 exactly in it. The third mega-environment combined the conditions K17 and N18 environments. The cultivar G15 had an advantage in this mega-environment. Cultivars G18 and G19 were located in the third mega-environment also. The standard cultivar G1, as well as the cultivars G11, G12, G14, G16, G17 and G20 had poorer performance compared to the above mentioned genotypes and they were in sectors that did not contain environments.

Figure 5 shows the average environment coordination of breeding lines in terms of mean yield and stability. The line which intersects the origin of the GGE biplot from left to right is the AEA for the environments. In the direction marked with an arrow in the small circle on the abscissa, the genotypes are ranked accordingly to mean yield, however, not in tons per hectare, but in the principal components values.
The AEA intersects in the GGE biplot origin with the average ordinate. The intersection point represents the mean yield of the experiment (grand mean). In our research, the breeding line G7 had the highest yield performance, and the cultivar G17 had the poorest one. The breeding lines G4, G3, G10, G5, G8, G9, G2, G6 and the cultivar G13 exceeded the grand mean yield in trial also. The breeding line G5 and the cultivar G16 had the highest stability, as was evidenced by their minimal deviation from the absissa. However, the cultivar G16 was significantly inferior to most of the genotypes in yield performance. Cultivars G15, G18, G20 and G19 had strong variability. The breeding line G7 had optimal combination yield performance and stability throughout test environments. Accordingly, this breeding line was as close as possible to the “ideal” genotype, which should conditionally be in the centre of centric circles (Fig. 6).

Fig. 6. GGE biplot ranking spring barley genotypes based on both mean performance and stability with reference to the “ideal” genotype

BREEDING LINES AND CULTIVARS

Breeding lines G4, G3, G10 and the cultivar G13 were slightly inferior to the breeding line G7 in mean yield and stability. The breeding line G5 was inferior to the G7 only in yield performance. The breeding lines G8, G2, G9 and G6 formed a cluster of genotypes which had lower yield and stability in comparison with the above mentioned genotypes, but at the same time prevailed over the majority other genotypes.

Discussion

It should be mentioned that several other researchers have also reported high year to year, or site to site grain yield variation in spring barley genotypes in Eastern European conditions (Pržulj & Momčilović, 2012; Miroslavčević et al., 2014; Pržulj et al., 2015) and in particular in Ukraine (Solonechnyi et al., 2015; Manukhnyk, 2018). Thus, testing genotypes in various environmental conditions (multi-environment trial) is effective for assessment of genotype by environment interactions and selecting desirable ones. For interpretation of experimental data from multi-environment trials it is necessary to use the most appropriate statistical models (van Eeuwijk et al., 2016). AMMI (Mehari et al., 2014; Abtew et al., 2015; Bocianowski et al., 2019; Verma et al., 2019) and GGE biplot analysis (Bilgin et al., 2019; Al-Gheewali et al., 2019; Al-Sayyady et al., 2019; Gradzinsko, 2019) have been the most widely used in recent years in order to interpret the experimental data from genotype by environmental trials. A number of researchers combine both of these statistical tools (Vaezi et al., 2017; Fana et al., 2018; Solonechnyi et al., 2018; Kendal et al., 2019). It has been shown that AMMI as well as GGE biplot explain the significantly higher percentage of genotype by environment interaction compared to other statistical methods (Nannorato et al., 2009; Pereira et al., 2011). In our study, the first two principal components AMMI and GGE biplot explained more than 80% of the genotype by environment interaction. Thus, both graphical models used sufficiently contributed to the in-depth visual analysis of the experimental data from the multi-environmental trial and allowed us to provide effective differentiation of genotypes in terms of wide and specific adaptation. In addition, the GGE biplot has a number of different functions for comprehensive assessment of test environments and providing mega-environment analysis (Yan et al., 2007).

Summarizing the environment characteristics in representativeness and discriminating power according to GGE biplot, we should note both the similarities between different agroclimatic conditions (N17 and M17, K18 and N18), as well as differences in the same agroclimatic conditions in different years (N17 and N18, M16 and M18). In addition, the considered environments had a different level of productivity. This indicates that not only does the environments’ mean productivity characterize their similarities or differences in terms of discriminating power and representativeness, but also the genotypes’ ranking in yield performance. In other words, the mode of reaction of the studied genotypes influenced the statistical characteristics of the test environments. The GGE biplot “which won-where” visualization also demonstrates that mega-environments were formed with a combination of different environmental conditions (agroclimatic zones) and years of the trial: I – N17, M17, M18 and K18; II – M16; III – K17 and N18. This is since even at the same ecological niche, the genotypes reacted differently to changes in weather conditions in different years. Experimental data are consistent with the prevalence of environmental conditions (82.8%) in the total dispersion and significant effect of the genotype by environment interaction (12.5%). The low share of the genotype contribution (4.7%) to the total dispersion, in our opinion is because only the best breeding lines and cultivars were involved in the experiment. At the same time, since the genotype contribution was reliable, this indicates the validity of the proposed combination of spatial (zones) and temporal (years) gradients for more efficient differentiation and selection of the best of the genotypes at the final stage of spring barley breeding process. It is demonstrated that even among of three lines G6 (Nutans 5152), G7 (Nutans 5152) and G8 (Nutans 5157) selected from one crossbreeding combination (Vivaldi/Ebson), the line G7 (Nutans 5152) was significantly superior the others. Difference in the adaptive reactions among these lines was expressed by different yield performance in different environments, which was clearly shown with AMMI and GGE biplot analysis. That is, due to genetic recombination, genotypes with significantly different reactions to environmental conditions could be formed even from one crossbreeding combination. Therefore, after a detailed study of the genetic source material and successful selection of parent components for crossing, the main aim of the breeder in the subsequent breeding process is to identify such genotypes.

In general, we determined that all studied promising breeding lines exceeded the widespread cultivars in production conditions in yield performance and, in most cases, in stability. Only the spring barley cultivar MIP Myrmty (G13) was the exception. The best spring barley breeding lines in yield performance and stability combination Nutans 5152 (G7), Nutans 5009 (G4), Nutans 4982 (G3) and Nutans 5093 (G10) were selected for further investigation in the State Variety Testing of Ukraine.

Conclusion

We developed an effective approach for organizing multi-environment trials at the final stage of breeding process which contribute to an in-depth assessment of the genotype by environment interaction and differentiation of genotypes in terms of yield performance and ecological stability. The main aspects of the proposed approach are: 1) Promising breeding lines highlighted by the results in the first year of the competitive test at the MIW in the next two years are also tested in two other research institutions (NPBES and IAS). Thus, for three years of competitive testing, the genotypes are evaluated in seven environments, which represent a combination of contrasting agroclimatic zones and different years (three years in MIW (Central part of the Forest-Steppe of Ukraine) + two years in NPBES (Polissia of Ukraine) + two years in IAS (Northern Steppe of Ukraine). Furthermore, MIW is an ecological niche where the breeding lines selected have multicrop representation in the total multi-environment trial. 2) Breeding lines are compared not only with the standard, but also with the other cultivars widespread in production conditions. It allows us to select only the best genotypes for growing in certain production conditions. 3) The experimental data from the genotype by environmental testing are analysed using AMMI and GGE biplot, which contribute to the comprehensive differentiation of genotypes in terms of wide and specific
adaptability. In addition, these statistical tools make possible the most qualitative characterization of test environments and provide mega-environment analysis. As a practical result of the multi-environment trial in 2016–2018 spring barley breeding lines with the optimal combination of yield performance and ecological stability, Naturas 5152, Naturas 5069, Natures 4982 and Natures 5009 as new cultivars MIP Sharm, MIP Tytul, MIP Deviz and MIP Zakhnyshyn respectively, have been submitted to the State Variety Testing of Ukraine.

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