

'Feniks', 'Anatoliyivka', 'Oksana', 'Berehynia' were noticeable for increased protein content. The average protein content was 41-42%, but in some years it reached 46%.

Contents of trypsin and lipoxygenase inhibitors in seeds were very variable. 'Yuvileina', 'Donka' and 'Znakhidka' had the lowest amounts of trypsin inhibitors (28.6-36.1 mg/kg), and 'Ustia', 'Eldorado', 'Romantyka', 'Kyivska 98', and 'Yug 30' had decreased activity of lipoxygenase (0.33-0.38 Units).

**Conclusions.** We demonstrated that natural hybridization based on pollen sterility genes increased the hybrid number by 6 times in comparison with manual crossing. Varieties and collection accessions with high productivity parameters, high protein content and other economically valuable features were selected in dry years.

**Key words:** *soybean, starting material, gene sterility, natural hybridization, drought resistance, productivity, protein content in seeds*

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### **AMMI (ADDITIVE MAIN EFFECT AND MULTIPLICATIVE INTERACTION) MODEL FOR ASSESSMENT OF YIELD STABILITY OF SPRING BARLEY GENOTYPES**

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The article presents results of AMMI analysis of yield capacity and phenotypic stability of 8 promising spring barley lines and 2 standard varieties. The study identified genotypes that consistently fulfill their genetic potentials under contrastive hydrothermal conditions: lines 06-652 (G4) and 09-837 (G8). Line 09-837 named as 'Lord' was submitted to the state variety trial. Benefits of AMMI analysis for assessment of breeding material at the final stage of breeding are shown.

**Key words:** *AMMI, ASV, spring barley, genotype-environment interaction, yield capacity, stability, line*

**Introduction.** Taking into account climatic changes in the world and Ukraine, it is important to develop varieties that combine high yields with resistance to unfavorable environmental conditions in the modern era of breeding. Development of varieties adapted to specific agro-ecological conditions is the most expedient, since every variety has unique compensatory effects. Adaptive capacity of a variety is specific; hence breeding of cereals, including spring barley, should be closely associated with environmental conditions of an area, where a variety is developed. Drought is the most severe abiotic stress in the production of cereals, including barley, and intensity, duration and prevalence of drought are the biggest risks of instability in grain production.

Climatic changes necessitate the creation of varieties with high environmental stability, which can ensure the production of barley grain in the amount sufficient for the food security of this country. To select genotypes combining drought tolerance with high performance, evaluation of breeding material for depression in yields during dry years versus moisture-sufficient years is rather informative.

Therefore, statistical methods of calculation of adaptive capacity and environmental stability indices are among key lines in investigations of 'genotype-environment' interactions. Evaluation of adaptive capacity of genotypes and identification of conditions suitable for the maxi-

imum fulfillment of their potentials are necessary components of knowledge upon the development of varieties and hybrids.

**Analysis of literature, problem statement.** ‘Genotype-environment’ ( $G \times E$ ) interaction is a determinant feature, when highly adapted spring barley forms are created, and promising lines are submitted to the state variety trials. There are several statistical methods, which can determine the effect of  $G \times E$  interactions on yields, distinguish varieties in which this effect is minimal and predict phenotypic response to environmental changes. Linear regression analysis, non-linear regression analysis, multivariate analysis and nonparametric statistics are the most common methods [1, 2, 3]. The method of principal components is one of the most effective methods for quantification of  $G \times E$  interaction and evaluation of yield stability [4, 5, 6, 7].

Analysis of variance (ANOVA) is a simple additive model, in which  $G \times E$  interaction is a source of variation, but its internal effects are not analyzed. In contrast, principal component analysis (PCA) is a multiplicative model and, hence, it does not represent additive main effects either of environment or of genotype [8]. AMMI (additive main effects and multiplicative interaction) model includes ANOVA and PCA in a single approach, therefore it can be applied to analyze data of environmental trials [9, 10, 11]. AMMI uses ANOVA to verify the main effects of genotype and environment; it uses PCA for analysis of the residual multiplicative interaction between genotypes and environment to determine the sum of squares of  $G \times E$  interaction with the minimum number of degrees of freedom. Since ANOVA and PCA are parts of AMMI model, this model is one of the most effective to characterize  $G \times E$  interaction [9].

In addition, AMMI simultaneously quantifies the contribution of each genotype and environment to the sum of  $G \times E$  squares and provides easy graphical interpretation of data by constructing a biplot [9, 12]. Therefore, it is easy to select productive varieties with wide adaptability, to perform agronomic zonation of varieties with certain adaptability and to determine optimal for trials conditions [9, 12, 13].

**The aim and tasks of the study.** The purpose of this study was to evaluate the adaptability and yield stability of spring barley lines using AMMI analysis and supportive nonparametric statistics in order to select genotypes with high performance and phenotypic stability due to decreased effects of  $G \times E$  interaction.

**Material and methods.** The study was conducted in the Laboratory of Barley Breeding and Genetics of the Plant Production Institute and a VYa Yuriev NAAS in 2012-2015. Eight original promising lines and 2 two standard varieties (‘Vzirets’ and ‘Komandor’) of spring barley were taken as starting material for the study.

Adaptability and stability were analyzed by the AMMI method described in Zobel R. W. et al. [9], using the following statistical model (1):

$$= \mu + g_i + e_j + \sum_{k=1}^n \lambda_k \alpha_{ik} y_{jk} + r_{ij} + \varepsilon_{ij} \quad (1)$$

where  $Y_{ij}$  - average response of genotype  $i$  in environment  $j$ ;  $\mu$  - grand average;  $g_i$  - fixed effect of genotype  $i$  ( $i = 1, 2, \dots, g$ );  $e_j$  - random effect of environment  $j$  ( $j = 1, 2, \dots, f$ );  $\varepsilon_{ij}$  - average error of the experiment;  $\lambda_k$  - unique value of the  $k^{\text{th}}$  analysis of a principal component of interaction (IPCA) ( $k = 1, 2, \dots, p$ , where  $p$  is the maximum number of estimated principal components);  $\alpha_{ik}$  - singular value for the  $i^{\text{th}}$  genotype in the  $k^{\text{th}}$  IPCA;  $y_{jk}$  - unique value of the  $j^{\text{th}}$  environment in the  $k^{\text{th}}$  IPCA;  $r_{ij}$  - error of  $G \times E$  interaction or AMMI residue;  $k$  - characteristic non-zero roots,  $k = (1, 2, \dots, \min(G - 1, E - 1))$ .

The level of phenotypic stability of genotypes was determined by ASV (AMMI stability value), which was calculated according to formula (2), as described by Purchase JL [14]:

$$ASV = \sqrt{\left[ \frac{SSIPC1}{SSIPC2} (IPC1score)^2 \right] + (IPC2score)^2} \quad (2)$$

where  $SSIPC1$  and  $SSIPC2$  are the sums of squares of the 1<sup>st</sup> and 2<sup>nd</sup> principal components of interaction;  $IPC1score$  and  $IPC2score$  are values of the 1<sup>st</sup> and 2<sup>nd</sup> principal components of interaction of genotype.

The yield stability index (YSI) was calculated using formula (3):

$$\sqrt{YSI} = RASV + RY \quad (3)$$

where *RASV* - rank of genotype based on ASV level; *RY* - rank based on yield (*Y*).

The stability index (SI) was calculated according to formula (4), as described by Barbarmanzoor A. et al. [15]:

$$Sul = \left( \frac{Y - \sigma n}{YM} \right) \times 100 \quad (4)$$

where *Y* - average performance of genotype;  $\sigma n$  - standard deviation, *YM* – maximum performance of genotype in any environment.

The data on yields of spring barley genotypes were mathematically processed using the program CropStat 7.2.

**Results and discussion.** Contrastive hydrothermal conditions in the study years allowed reliable estimating genotypes in terms of stability and adaptability. For example, 2013 and 2015 were not entirely favorable for growth and development of barley plants. Typically, plants suffered from drought (0-66% of normal rainfall) at high air temperature (by 2.3-6.0° higher than the norm). In 2013, unfavorable conditions (34-39% of normal rainfall; temperature by 1.1-5.4° higher than the norm) were during the tillering and ear initiation periods as well as during the filling stage, which also negatively affected the yields. It rained (140-163% of normal rainfall) during anthesis, but rain showers were local and accompanied by squalls. 2014 was extremely favorable for barley, which made it possible to evaluate the potential of genotypes for yield capacity.

In 2015, the weather conditions were not favorable, though sowing was started early on optimum dates. However, the temperature fell drastically (to -6°C), it snowed, and sowing was only continued 10 days later. During the sprouting-tillering period, there was insufficient rainfall (37-56%), and there was drought at the end of filling (2% of normal rainfall). The average air temperature was close to the norm, but it reached 32.0°C in some decades (June, barley anthesis) and 33.8-35.9°C (July, filling-ripening). This negatively affected the tillering capacity and grain, plumpness, reducing yields.

The average yield of the test varieties and lines ranged from 2.79 t/ha in 2013 to 6.55 t/ha in 2014 (Table 1).

Table 1

Yield Capacity of Spring Barley Lines in 2012-2015, t/ha

Genotype code	Variety/line	2012, E1	2013, E2	2014, E3	2015, E4	Average
G1	‘Vzirets’	4.60	2.23	6.71	4.73	4.57
G2	‘Komandor’	4.60	2.10	6.24	4.42	4.34
G3	05-393	4.31	3.81	6.58	4.36	4.77
G4	06-652	4.48	2.65	6.73	5.20	4.77
G5	08-2455	5.12	2.59	6.40	5.06	4.79
G6	09-791a	3.43	2.60	6.52	4.47	4.26
G7	09-2162	3.69	2.78	5.89	3.43	3.95
G8	09-837	4.75	3.16	7.16	5.04	5.03
G9	08-1385	5.08	3.27	6.90	5.18	5.11
G10	09-409	4.63	2.70	6.34	4.37	4.37
	Average	4.47	2.79	6.55	4.63	4.61
	IPCA 1	-0.559	0.104	0.594	-0.540	
	IPCA 2	-0.747	-0.187	0.470	0.465	

Analysis of variance divided the sum of squares of the yield capacity into effects of genotype, environment and genotype-environment ( $G \times E$ ) interaction (Table 2). The environment effect contributed most of all to the variability (89.0%); the contributions of genotype and genotype-environment interaction were much weaker, accounting for 5.8% and 5.2% of the variance, respectively. The strong effect of the environment factor in the total variance of yield capacity is associated with significant fluctuations in hydrothermal conditions during the study.  $G \times E$  interaction was additionally divided using the method of principal components. IPCA 1 and IPCA 2 account for 95% of the interaction variability, enabling us to evaluate the stability of genotypes for these two components (Table 2).

Table 2

AMMI Model of Analysis of Variance of Yield Capacity of Spring Barley Genotypes					
Source of variability	df	SS	ms	Percentage of variability, %	Percentage of variability of interaction, %
Genotype(G)	9	4.611	0.512	5.8	
Environment (E)	3	70.881	23.627	89.0	
G×E interaction	27	4.131	0.153	5.2	
IPCA 1	11	2.858	0.260		69.2
IPCA 2	9	1.064	0.118		25.8
IPCA3	7	0.210	0.300		5.0
Total	39	79.624			

The AMMI model does not include quantification of the stability degree, that is why Purchase et al. (2000) [14] proposed ASV to evaluate it and to rank genotypes by yield stability. ASV is the distance from the center (zero) in a two-dimensional scattergram with IPCA1 (axis of principal component of interaction 1) and IPCA2 (axis of principal component of interaction 2). Since IPCA1 makes a greater contribution to the sum of squares of genotype-environment interaction (Table 2), it should proportionally contribute as compared to IPCA2 to compensate for a share of relative contribution of IPCA1 and IPCA2 to the total  $G \times E$  interaction. The distance from zero is determined using the Pythagorean theorem [14]. In the ASV method, the lower ASV is, the more stable genotypes are. Accordingly, lines G4 and G8 were the most stable across the test accessions, and lines G3, G7 and G9 showed significant variations in yields (Table 3).

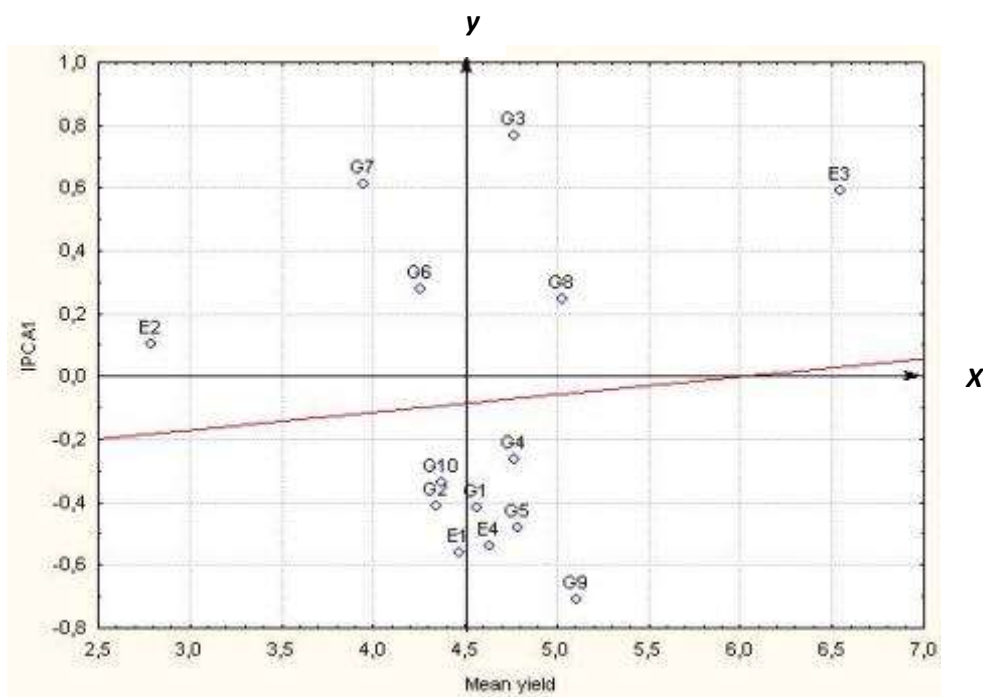
Table 3

Yield and Statistical Indices of Stability of Spring Barley Genotypes									
Genotype code	Variety/Line	Y, t/ha	Rank	IPCA1	IPCA2	ASV	Rank	YSI	SI,%
G1	‘Vzirets’	4.57	5	-0.415	0.128	0.693	4	9	40.8
G2	‘Komandor’	4.34	7	-0.412	-0.202	0.705	5	12	42.3
G3	05-393	4.77	4	0.767	-0.175	1.269	10	14	53.6
G4	06-652	4.77	4	-0.266	0.359	0.565	2	6	45.8
G5	08-2455	4.79	3	-0.482	-0.307	0.848	7	10	50.0
G6	09-791a	4.26	8	0.277	0.705	0.838	6	14	39.4
G7	09-2162	3.95	9	0.611	-0.273	1.039	8	17	44.1
G8	09-837	5.03	2	0.246	0.195	0.448	1	3	47.3
G9	08-1385	5.11	1	-0.708	-0.120	1.167	9	10	52.6
G10	09-409	4.37	6	-0.336	-0.310	0.632	3	9	43.9

Stability in itself, however, is not the only parameter of genotype evaluation, because the most stable genotypes do not necessarily have high performance [16, 17]. Hence, there is a need for approaches that integrally assess the average yield and stability as a single index, which prompted researchers to propose different criteria for simultaneous selection for yield and its stability [18, 19, 20, 21, 22, 23]. Since ASV is based on IPCA1 and IPCA2, which incorporate the greatest share of variations in  $G \times E$  interaction, ASV rank reliably assesses stability of genotypes. The sum of ASV ranks and yield comprehensively evaluate genotypes for stability and yield, giving so-called "yield stability index» (YSI). Genotypes with the lowest YSI are deemed the most stable and high-yielding. In our studies, such genotypes are lines 06-652 (G4) and 09-837 (G8).

The stability index SI is another integral parameter for simultaneous estimation of yield and stability. A lot of authors used it to select stable genotypes [24, 25, 26]. The stability index values were conveniently divided into 5 groups, namely: very low ( $\leq 20\%$ ), low (21-40%), medium (41-60%), high (61-80%), and very high (80%) [15] (Table 3). Most genotypes had a medium SI, and genotypes G1 and G6 only had slightly lower SI levels (40.8% and 39.4%, respectively). These results prove that in our case SI was not an informative index for ranking genotypes by yield and stability.

To illustrate the effects of each genotype and environment, we constructed AMMI1 (Fig. 1) and AMMI2 (Fig. 2) biplots, which are usually quite informative [11]. In Fig. 1, coordinate  $x$  shows basic effects (average yield), and coordinate  $y$  indicates the effects of genotype-environment interaction (IPCA1). The closer genotype markers to the start of axis  $y$  are, the smaller contribution to  $G \times E$  interaction they make. In this case, lines G4, G6 and G8 were the most stable. Across these genotypes, line G8 had the highest performance, which makes it the most desirable genotype. Line G4 is also a valuable genotype, since it is superior to standard varieties in terms both of the average yield and of stability.

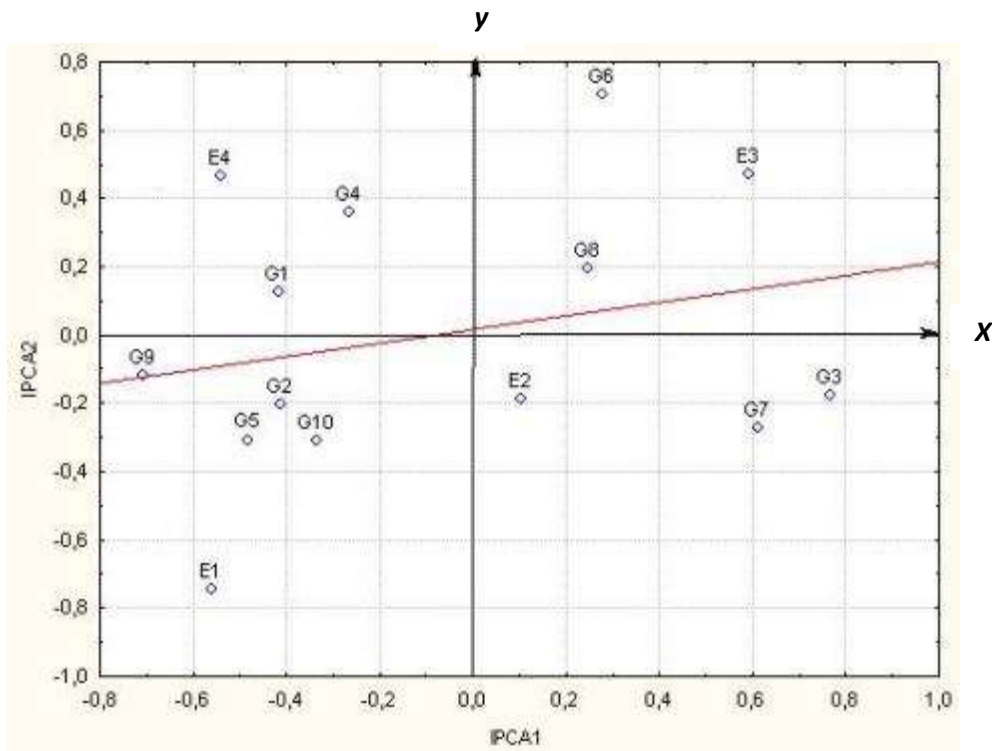


**Fig. 1.** AMMI1 Biplot (IPCA1 vs. average yield) for 10 spring barley genotypes in 4 environments

Environment E2 was characterized by a low level of interaction and low performance, while E3 had high levels of interaction and performance.

AMMI2 biplot based on IPCA1 and IPCA2 also distinguished lines G4 and G8 as the most stable genotypes, since they were closest to the biplot center. On the other hand, G3 and G9

genotypes were the most unstable, i.e. their performance depends on specific cultivation conditions. According to the AMMI2 model, unlike the AMMI1 model, genotype G6 with high IPCA2 is not stable.



**Fig. 2.** AMMI2 Biplot (IPCA1 vs. IPCA2) for 10 spring barley genotypes in 4 environments

The results of graphical analysis using AMMI1 and AMMI2 confirm the data obtained by computing ASV and YSI, making it a robust alternative to mathematical calculations.

**Conclusions.** The Plant Production Institute nd. a VYa Yuriev NAAS investigates spring barley response to changing weather conditions not only in Kharkiv region but also in other natural zones of Ukraine. Breeding material and varieties that can provide sufficient yields even under the harshest droughts have been developed, which will contribute to solving the food challenge is not only in our country but also worldwide.

The research using the AMMI model both with ASV and YSI computation and with construction of AMMI1 and AMMI2 graphs highlighted lines 06-652 (G4) and 09-837 (G8) as the most promising material. Line 09-837 named as ‘Lord’ was submitted to the state variety trial. The stability index SI turned out to be uninformative in our study.

We demonstrated a possibility of using AMMI analysis to assess adaptive features of promising breeding material at the final stage of breeding.

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**AMMI (ADDITIVE MAIN EFFECT AND MULTIPLICATIVE INTERACTION) МОДЕЛЬ ОЦІНКИ СТАБІЛЬНОСТІ ГЕНОТИПІВ ЯЧМЕНЮ ЯРОГО**

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**Мета і задачі дослідження.** Метою даних досліджень була оцінка адаптивності та стабільності врожайності ліній ячменю ярого за допомогою АММІ аналізу і допоміжних непараметричних статистик для добору генотипів з високою продуктивністю та фенотиповою стабільністю, завдяки мінімальним ефектам  $G \times E$ .

**Матеріал і методика.** Дослідження проведено протягом 2012–2015 рр. у лабораторії селекції та генетики ячменю Інституту рослинництва ім. В. Я. Юр'єва НААН. Вихідним матеріалом для дослідження були вісім перспективних ліній ячменю ярого та два сорти-стандарту Взірець і Командор. Аналіз адаптивності та і стабільності здійснювали за допомогою метода АММІ за Zobel et al. (1988).

**Обговорення результатів.** За допомогою дисперсійного аналізу варіабельність урожайності було розділено на ефекти генотипу, оточуючого середовища та взаємодію генотип-середовище. Найбільшу частку в загальній дисперсії склали ефекти оточуючого середовища – 89,0 %, ефекти генотипу та взаємодії генотип-середовище були значно меншими – 5,8 % і 5,2 % відповідно. Взаємодію генотип-середовище було додатково розподілено за допомогою метода головних компонент (РСА). Перші дві вісі РСА визначали 95 % від варіабельності взаємодії генотип-середовище, що дозволило оцінити стабільність генотипів за цими двома компонентами. Найнижчий рівень ASV серед досліджених генотипів і, відповідно, максимальну стабільність мали лінії 06-652 (G4) та 09-837 (G8), а лінії 05-393 (G3), 09-2162 (G7) та 08-1385 (G9) характеризувались високою варіабельністю врожайності. Лінії 06-652 і 09-837 також характеризувалися високим рівнем інтегрального показника YSI, який поєднує врожайність та її стабільність, що робить їх найціннішим селекційним матеріалом. Графічний аналіз із застосуванням АММІ1 та АММІ2 біплот показали на генотипи 06-652 (G4) і 09-837 (G8) як найцінніші.

**Висновки.** Результати дослідження як з використанням показників ASV і YSI, так і за допомогою побудови графіків АММІ1 і АММІ2, показали, що лінії 06-652 (G4) та 09-837 (G8) є найперспективнішим матеріалом. Лінію 09-837 було передано до Державного сортопробування України під назвою Пан. Індекс стійкості SuI виявився неінформативним у наших дослідженнях. Таким чином, показано можливість використання АММІ

аналізу для оцінки адаптивних особливостей перспективного селекційного матеріалу на завершальних етапах селекційного процесу.

**Ключові слова:** АММІ, ASV, ячмінь ярий, взаємодія генотип-середовище, урожайність, стабільність, лінія

### **АММІ (ADDITIVE MAIN EFFECT AND MULTIPLICATIVE INTERACTION) МОДЕЛЬ ОЦЕНКИ СТАБИЛЬНОСТИ ГЕНОТИПОВ ЯЧМЕНЯ ЯРОВОГО**

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**Цель и задачи исследований.** Целью данных исследований была оценка адаптивности и стабильности урожайности линий ячменя ярового с помощью АММІ анализа и вспомогательных непараметрических статистик для отбора генотипов с высокой продуктивностью и фенотипической стабильностью, благодаря минимальным эффектам  $G \times E$ .

**Материал и методика исследований.** Исследования проведены в течении 2012-2015 гг. в лаборатории селекции и генетики ячменя Института растениеводства им. В. Я. Юрьева НААН. Исходным материалом для исследований служили восемь перспективных линий ячменя ярового и два сорта-стандарта Взирец и Командор. Анализ адаптивности и стабильности осуществляли с помощью метода АММІ по Zobeletal. (1988).

**Обсуждение результатов.** С помощью дисперсионного анализа вариабельность урожайности была разделена на эффекты генотипа, окружающей среды и взаимодействия генотип-среда. Наибольшую долю в общей дисперсии составляли эффекты окружающей среды – 89,0 %, эффекты генотипа и взаимодействия генотип-среда были значительно меньше – 5,8 % и 5,2 %, соответственно. Взаимодействие генотип-среда было дополнительно разделено с помощью метода главных компонент (РСА). Первые две оси РСА определяли 95 % от вариабельности взаимодействия генотип-среда, что позволило оценить стабильность генотипов по этим двум компонентам. Наименьший уровень ASV среди исследованных генотипов и, соответственно, максимальную стабильность имели линии 06-652 (G4) и 09-837 (G8), а линии 05-393 (G3), 09-2162 (G7) та 08-1385 (G9) характеризовались высокой вариабельностью урожайности. Линии 06-652 и 09-837 также характеризовались высоким уровнем интегрального показателя YSI, сочетающего урожайность и ее стабильность, что делает их наиболее ценным селекционным материалом. Графический анализ с использованием АММІ1 и АММІ2 биплот указали на генотипы 06-652 (G4) и 09-837 (G8), как наиболее ценные.

**Выводы.** Результаты исследований, как с использованием показателей ASV и YSI, так и с помощью построения графиков АММІ1 и АММІ2, показали, что линии 06-652 (G4) и 09-837 (G8) являются наиболее перспективным материалом. Линия 09-837 была передана в Государственное сортоиспытание Украины под названием Пан. Индекс устойчивости SuI оказался неинформативным в наших исследованиях. Таким образом, показана возможность использования АММІ анализа для оценки адаптивных особенностей перспективного селекционного материала на завершающих этапах селекционного процесса.

**Ключевые слова:** АММІ, ASV, ячмень яровой, взаимодействие генотип-среда, урожайность, стабильность, линия

## **AMMI (ADDITIVE MAIN EFFECT AND MULTIPLICATIVE INTERACTION) ANALYSIS OF STABILITY OF SPRING BARLEY GENOTYPES**

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**The aim and tasks of the study.** The purpose of this study was to evaluate the adaptability and stability of the yield capacity of spring barley lines using AMMI analysis and supplementary non-parametric statistics for selection of genotypes with high productivity and phenotypic stability due to the minimal  $G \times E$  effects.

**Materials and methods.** The research was conducted in the Laboratory of Barley Breeding and Genetics of the Plant Production Institute nd. a VYa Yuryev NAAS in 2012–2015. The starting material for the study was 8 promising lines of spring barley and two varieties-standards, ‘Vzirets’ and ‘Komandor’. Adaptability and stability were analyzed by the AMMI method according to Zobel et al. (1988).

**Results and discussion.** Analysis of variance divided variability of the yield capacity into the effects of genotype, environment and genotype-environment interaction. The environment effects accounted for the greatest share in the total variance accounted for environmental effects - 89.0%; the genotype and genotype-environment interaction were significantly lower – 5.8% and 5.2%, respectively. The genotype-environment interaction was further divided by principal component analysis (PCA). The first two PCA axes determined 95% of the genotype-environment interaction variability, which allowed us to estimate the stability of genotypes by these two components. Across the test genotypes, the lowest ASV and, accordingly, the maximum stability, were observed for lines 06-652 (G4) and 09-837 (G8), and lines 05-393 (G3), 09-2162 (G7) and 08-1385 (G9) were noticeable for high variability of the yield capacity. Lines 06-652 and 09-837 were also characterized by high values of the integral YSI index combining yield capacity and its stability, making them the most valuable breeding material. Graphical analysis using AMMI1 and AMMI2 biplot highlighted genotypes 06-652 (G4) and 09-837 (G8) as the most valuable ones.

**Conclusions.** The results both of using YSI and ASV parameters and of constructing AMMI1 and AMMI2 graphs showed that lines 06-652 (G4) and 09-837 (G8) were the most promising material. Line 09-837 named ‘Pan’ was submitted to the state variety trial of Ukraine. Stability index  $SuI$  proved uninformative in our research. Thus, we demonstrated a possibility of using AMMI analysis to assess adaptive features of promising breeding material at final stages of breeding.

**Key words:** *AMMI, ASV, spring barley, genotype-environment interaction, yield capacity, stability, line*