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RESEARCH ARTICLE

NEW APPROACHES FOR EVALUATION THE GRAIN YIELD OF WINTER WHEAT IN CONTRASTING ENVIRONMENTS

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ABSTRACT

Purpose is to verify the new approach for correct separating of wheat varieties in groups matching particular standards of their level of grain yield during the years and towards the different environmental conditions.
Materials and Methods: 76 breeding lines were researched for two consecutive years. The first one was with favorable conditions for growing (2006) and the second one in 2007 was characterized by well-defined prolonged drought and unusually high temperatures during the active growing season of crop. Real values of grain yield of each variety were used for the calculation of breeding indexes, which establishes the level of tolerance to abiotic stress. It was made classification based on indices and parameters derived from combinations between them. The separation into groups according to their level of grain yield was founded on direct comparison using the correlations established these indices and combinations.
Main Results: By applying the score assessment of the values of the studied indices and parameters is quite possible grain yield genotype be evaluated during contrasting environmental conditions. It was very clear the importance of derivatives indices by newly formed parameters because they were used to value the other cultivars in the certain group.
Conclusions: The suitability of the new integrated approach to practical grouping of varieties, according to grain yield in contrast growing conditions was confirmed. An efficient method is proposed for grain yield assessment through all season which dissimilar weather conditions influences on wheat.

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INTRODUCTION

In the last few years global climate changes has been affecting the most severe grade food crops because they occupy large areas (Semenov *et al*., 2014, Hellin *et al*., 2012). This makes the breeding of each of these grain crops a bigger challenge. The assessment of yield grain variety in different environmental conditions has always been the focus of numerous wheat studies (Tsenov and authors, 2009, Anderson *et al*. 2016, Van Ittersum *et al*., 2013). The reason for this is that this crop is cultivated all around the world, regardless of the conditions for its cultivation (Carver *et al*., 2009). At least one third of the wheat grow in areas where abiotic stress occurs annually in different periods of its vegetation (Reynolds *et al*., 2016). In winter wheat is influenced on abiotic stress of high temperatures, drought and cold during the winter period (Gusta and Wisniewski, 2013, Fowler *et al*., 2014).If we add to stress and its other dimensions - high temperature, salinity, lack or excess of micro and / or macro-elements (mineral deficiency, toxicity), waterlogging), stress becomes very tangible part of wheat studies. (*www.plantstress.com*, Blum 2016). Since winter wheat is grown in such large areas it is essential to collect information about the behavior of the variety in the specific conditions of the region in which it is grown (Tsenov *et al*., 2011, Van Wart *et al*., 2013). Scientists have long sought a way to measure the response of genotype in contrasting environmental conditions (Finlay and Wilkinson, 1963) trying fundamentally different methods and approaches (Weikai, and Tinker 2002, Crossa 2004, (Mohammadi and Amri 2008). In the last 10-15 years scientists search for real ways to assess genotypes not only in times of stress (Moosavi *et al*., 2008), but in contrasting environmental conditions to identify the most adaptable genotypes, the yield of grain that is stable to wide range of weather conditions (Thiry *et al*., 2016). This is a new step forward in studying the behavior of the variety in conditions of stress and favorable conditions of different seasons, which are unpredictable and changing as a

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result of global warming (Kazandjiev and auto. 2011). The indices used for a long time to evaluate the tolerance / sensitivity abiotic stress are different depending on the approach. Their number varies from 5 (Thiry *et al*., 2016) to 16 types, (Brdar-Jokanovic *et al*., 2014), especially when two modes of cultivation of wheat are examined - without irrigation and watering. Between the values of these indices and grain yield stress has very high correlation (El-Rawy & Hassan 2014). Therefore the set of indices (different number) is widely used to assess the response of genotype by its yield, except for wheat and other major crops: Sunflower (Abdi *et al*., 2013); Corn (Naghavi *et al*., 2013); Sesame (Amani *et al*., 2012); Sorghum (Menezes *et al*., 2014); Rape (Curie-skłodowska and Polonia 2012); Barley (Benmahammed *et al*. 2010); Rice (Khan and Dhurve, 2016) and Soybeans (Rocha *et al*., 2014) and many others. This kind of test group of indices used to assess the behavior of groups of varieties in almost all types of stress, even biotic one (diseases).

In Bulgaria Tsenov *et al*. (2012) found that five of the eight indexes they examined, which are effective for the climatic conditions of the country. When working with these indexes a questions always arises if it is possible to identify genotypes whose yield remained relatively high in terms of stress and favorable growing conditions? A similar question is asked by (Fernandez 1993), which is actually the first one who gives a realistic idea that such an analysis is possible. In literature abounds predominate the hypothesis that variety which has a good expression under stress, have low yield in favorable conditions and is almost impossible to have a "universal" variety for all types of conditions (Păunescu and Boghici, 2008). In research we made after intense and prolonged drought in 2007, we found similar trenlines of our native varieties (Ivanova *et al*., 2011, Tsenov *et al*. (2012), Tsenov *et al*., 2015). Then we had no information on possible opportunities for making such analysis. It was impossible until this fall when Thiry *et al*. (2016) showed clearly established and realistic approaches for such clustering. The availability of data with the results of contrast growing years were not published, strengthen our desire to verify the effectiveness of these options. The aim of this study is to analyze the possibilities for effective evaluation and clustering of genotypes in contrasting environmental conditions using new indices such as systemic approach

MATERIALS AND METHODS

The reason for writing this paper is the publication of (Thiry *et al*., 2016), which makes a new interpretation of the role of indices for analyzing the behavior of wheat in contrast growing conditions. 76 breeding lines and varities were sdudied, using data for harvesting grain of CSR in the 2006-2007 and even then were calculated number 8 of indices to assess tolerance to drought like the publication of Tsenov *et al*. (2012). Assigning a score of each variety being index is made exactly according to their approach (Thiry *et al*., 2016). This approach lies in the introduction of grading (scoring, clustering) assessment for each variety group. For each of the 5 indices used there a 10-grading simple assessment is done. The extent of variation in the values of each trait (difference between maximum and minimum value) is divided by 10. This determines the range of values between which appropriate score will be made. For example, let see the situation with the index SSI (Table 1). In this case, score 5 g*et al*l sorts of variation row that have values between 0.74 and 0.83.

For more convenience in the analysis, indexes and consequently the parameters are grouped into two categories: 1 category (class) - related to stress conditions **(**index **SSI, TOL;** parameters **Ys, RSI, YSSI)** and 2 categorie (class) related to the favorable conditions of the season indexes **(MP, GMP, STI;** parameters **Yp, PSI, YPSI).** Details of each of the indexes and derived parameters can be seen in the following references: Fischer and Maurer (1978), Hossain *et al*. (1990), Fernandez, (1993), Thiry *et al*. (2016). All similarities and differences in approaches from the last group of authors will be discussed in the following sections. Due to the limited volume of standard scientific communication part of the intermediate results are not presented here, but are discussed in the context of the working out. Ranking (Scoring) and statistical data processing is undertaken primarily module XL Stat 2014 of MS Excel 2007. The main part of the analyzes take correlations, verify the authenticity of groups of varieties and graphical analysis of the scattering of points of each variety used according to indexes or formulas, so called scatter plot.

RESULTS

The separation of varieties based on their level of grain yield is graphically represented in Fig.1. In summary, this is the grouping according to the reaction of any variety relative to the average response of the group in different conditions (favorable conditions is the vertical line, - Yp) or conditions of abiotic stress is the horizontal line - Ys). This grouping seems a lot easier until you start using each index separately and you realize it is impossible to proceed.

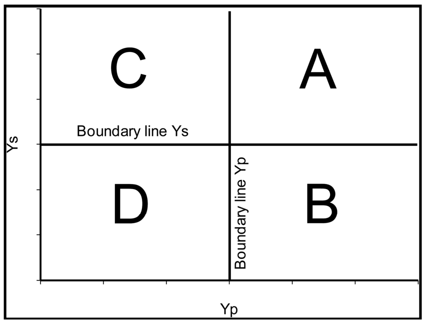

Fig.1. Principal Presentation of the different response group (A, B, C, and D), defined by Fernandez (1993), according to their grain yield under abiotic stress conditions (*Ys*) and grain yield under yield potential conditions (*Yp*)

After entering the assessment scores we did analyze whether this approach is correct by directly comparing the values of the indices groups with their "new" assessment values. The data in Tables 2 and 3 show the correlation interrelatioships between them. The rated assessments of the first group are presented in two versions, because there is a dispute about the way the scores are formed. One is in descending order (higher value of the index - a lower score and vice versa). The simplicity of these indices show tolerance (sensitivity) of genotype and therefore low value should be combined with a higher score. This is consistent with the way of placing the score in the indices of second grade. In this study the second way ❷ has been accepted for the "right" one. We accept scores evaluation for proper expression data because each index has fairly high correlation with their physical values. The thesis is strongly corroborated by low, respectively high values of the parameters *p-value* and $R^{2,}$ which clearly we are verifying entirely.

Table 1. Way of putting of scoring assessment on index SSI

Min	0,33	Score	1	2	3	4	5	6	7	8	9	10
Max	1,34	From	0,33	0,44	0,54	0,64	0,74	0,84	0,94	1,04	1,14	1,24
Difference	1,01	To	0,43	0,53	0,63	0,73	0,83	0,93	1,03	1,13	1,23	1,34

Table 2. Spearman correlation ships between the new score indexes of class 1 group and their original values

Indexes	SSI	*p-value*	R^2	TOL	*p-value*	R^2
Score SSI ❶	0,9774	0,0000	0,9553	0,9446	0,0000	0,8923
Score SSI ❷	-0,9774	< 0,0001	0,9553	-0,9446	< 0,0001	0,8923
Score TOL❶	0,9442	< 0,0001	0,8916	0,9765	< 0,0001	0,9536
Score TOL ❷	-0,9442	< 0,0001	0,8916	-0,9765	< 0,0001	0,9536

Values in bold are different from 0 with a significance level alpha=0,05; ❶- arranged in descending order; ❷ - arranged in ascending order

Table 3. Spearman correlationships between the new score indexes of class 2 group and their original values

Indexes	Score MP	Score GMP	Score STI
MP	0,989	0,978	-0,190
p-value	0,000	< 0,0001	0,101
R^2	0,978	0,957	0,036
GMP	0,980	0,985	-0,054
p-value	0,000	< 0,0001	0,640
R^2	0,960	0,971	0,003
STI	-0,145	-0,028	0,973
p-value	0,212	0,808	< 0,0001
R^2	0,021	0,001	0,947

*Values in **bold** are different from 0 with a significance level alpha=0,05*

Table 4. Spearman Correlations between scores on indices of the first group with parameter [RCI] and a second group with parameter [PCI]

Indexes	Group	*p-value*	*R2*
Resilience Capacity Index			
Score SSI	0,9800	0,0000	0,9604
Score TOL,	0,9773	< 0,0001	0,9551
Potential Capacity Index			
Score MP	0,8832	0,0000	0,7800
Score GMP	0,9305	< 0,0001	0,8658
Score STI	0,2382	0,0385	0,0567

*Values in **bold** are different from 0 with a significance level alpha=0,05*

Table 5. Spearman correlations between key values of environments (Yp и Ys) and both groups of 'new' parameters

Variable	Ys	*p-value*	R^2	Yp	*p-value*	R^2
RCI	0,3190	0,3616	0,1018	0,1139	-0,3374	0,3426
PCI	0,9692	< 0,0001	0,9394	0,6710	0,8192	0,0061
YSSI	0,9323	< 0,0001	0,8691	0,2885	0,5371	0,1104
YPSI	0,6422	0,0491	0,4125	0,9818	< 0,0001	0,9909

*Values in **bold** are different from 0 with a significance level alpha=0,05*

Table 6. Spearman correlations between the key values of the environments and combinations of formulas between both groups of indices

Combination №	Y_S	*p-value*	R^2	Y_P	*p-value*	R^2
1	0,9773	< 0,0001	0,9551	0,1533	0,1857	0,0235
2	0,9750	< 0,0001	0,9505	0,2061	0,0741	0,0425
3	0,1825	0,1145	0,0333	-0,3111	0,0064	0,0968
4	0,9908	< 0,0001	0,9817	0,2876	0,0120	0,0827
5	0,9857	< 0,0001	0,9716	0,3382	0,0029	0,1144
6	0,4503	< 0,0001	0,2027	0,4699	< 0,0001	0,2208
7	0,3037	0,0079	0,0922	0,9711	< 0,0001	0,9431
8	0,2338	0,0423	0,0547	0,9774	< 0,0001	0,9553
9	-0,5840	< 0,0001	0,3410	+0,5554	< 0,0001	0,3085
10	0,2870	0,0122	0,0824	0,9817	< 0,0001	0,9637
11	0,2097	0,0692	0,0440	0,9800	< 0,0001	0,9604
12	-0,6780	< 0,0001	0,4596	0,4492	< 0,0001	0,2018
13	0,9945	< 0,0001	0,9890	0,2542	0,0270	0,0646
14	0,9862	< 0,0001	0,9726	0,2133	0,0645	0,0455
15	0,9854	< 0,0001	0,9710	0,2711	0,0181	0,0735
16	0,2612	0,0229	0,0682	0,9865	< 0,0001	0,9733
17	-0,2572	0,0251	0,0662	0,8151	< 0,0001	0,6643
18	-0,2732	0,0172	0,0746	0,8095	< 0,0001	0,6552

Table 7. The difference between means of pre-separate groups according to scores of the rated parameters

Variable	Observations	Mean score	Std. deviation	Significance
YSSI-A	19	5,7	1,018	c
YSSI-B	19	4,6	1,012	bc
YSSI-C	19	4,5	1,364	b
YSSI-D	19	3,2	1,003	a
YPSI-A	19	2,2	1,123	c
YPSI-B	19	0,9	0,932	bc
YPSI-C	19	-0,2	1,181	ab
YPSI-D	19	-1,3	0,903	a

Table 8. Scores of key values and indices of well-known wheat varieties

№	Variety	Class 1			Class 2			
		Score YP	Score SSI	Score TOL	Score YS	Score MP	Score GMP	Score STI
1	Sadovo 1	3	8	6	3	3	3	4
2	Pryspa	3	7	5	4	3	3	4
3	Pobeda	2	8	7	1	1	1	3
4	Karat	3	7	6	3	3	3	4
5	Neda	5	6	6	6	6	6	5
6	Lazarka	7	7	7	6	7	7	4
7	Enola	1	6	5	2	1	1	5
8	Iveta	4	8	7	3	3	3	3
9	Antonovka	7	7	6	7	7	8	4
10	Aglika	5	10	8	3	4	4	2

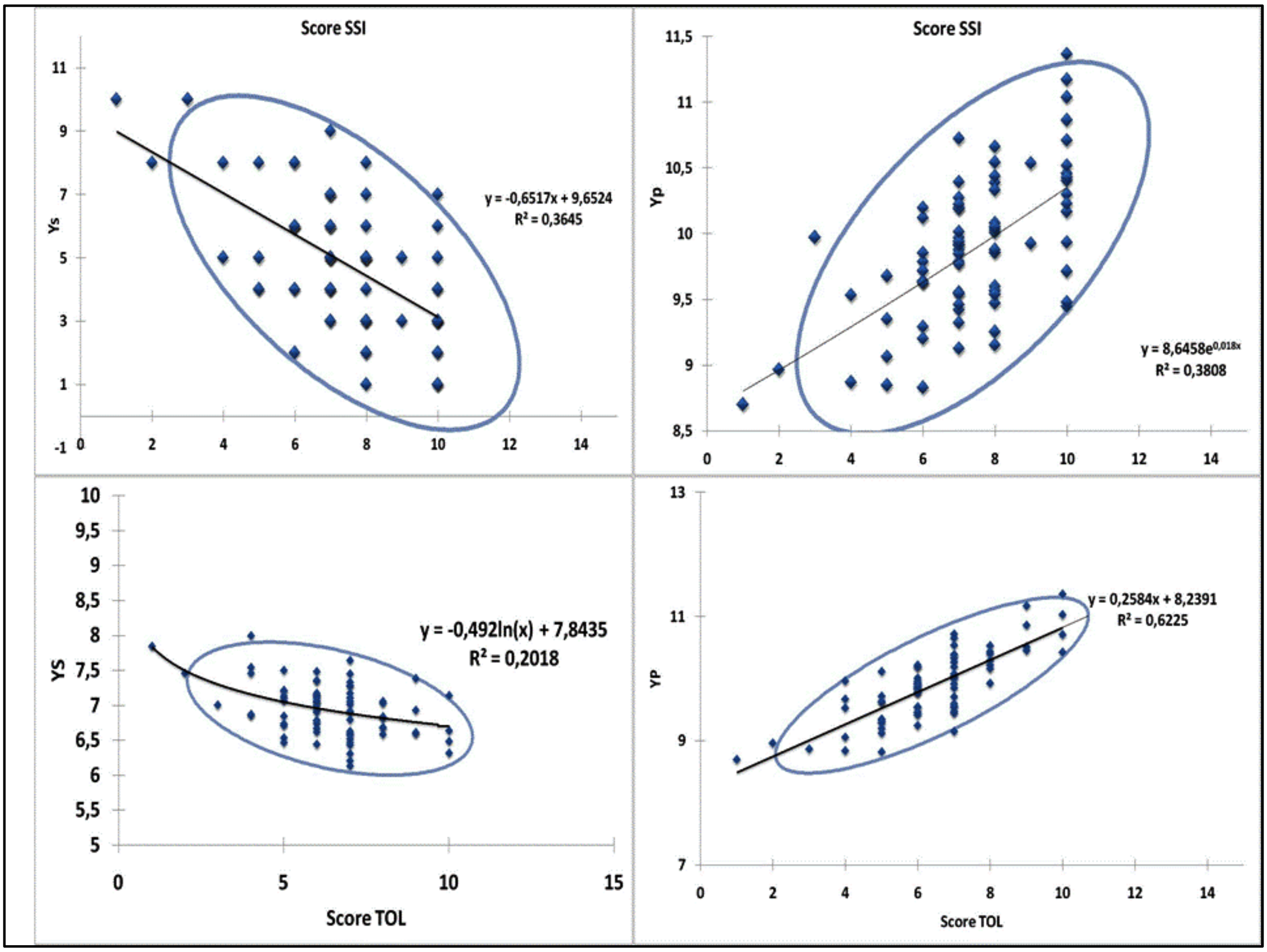

Fig.2. Regression relations between key values of environmental conditions and scores of the indices of the first group

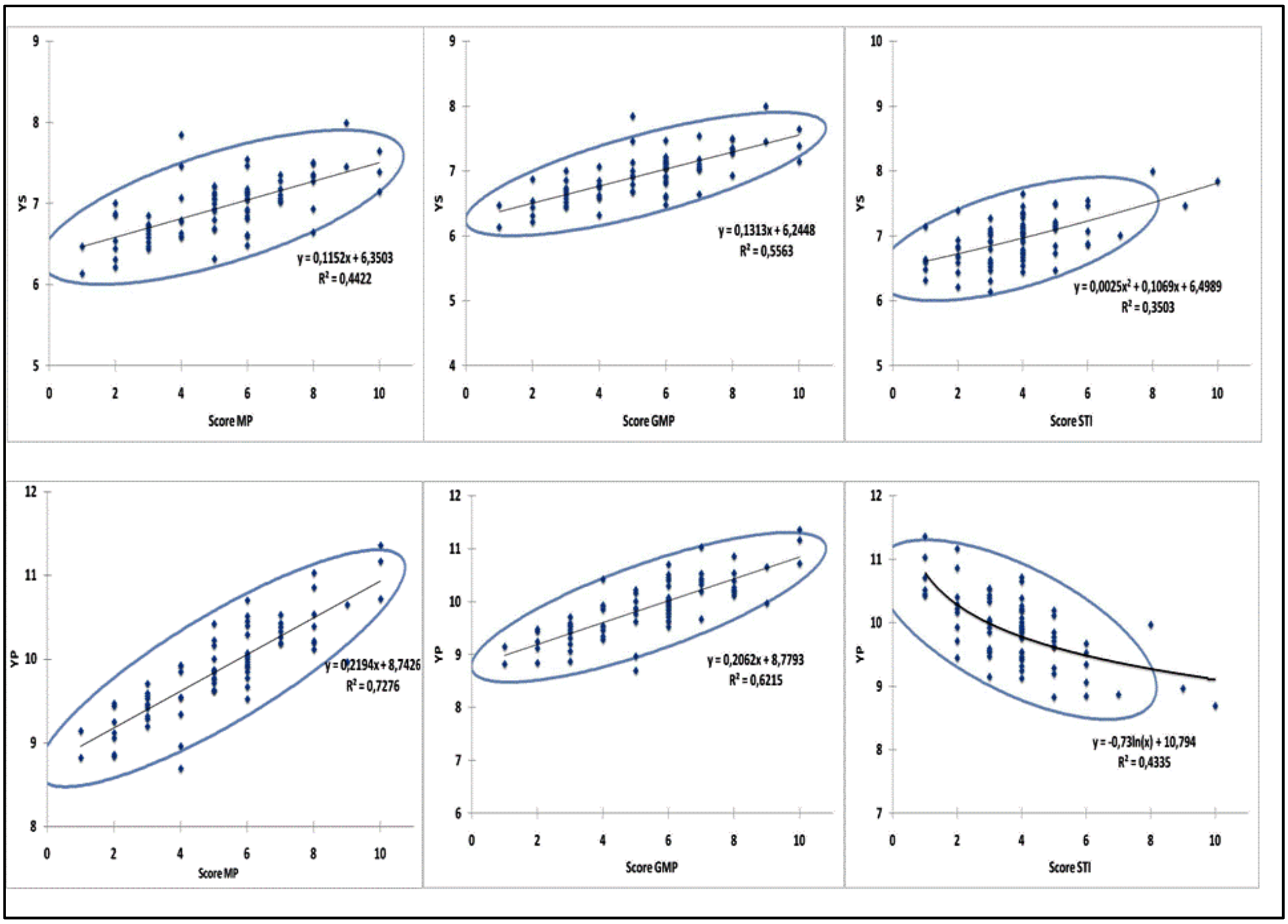

Fig.3. Regression dependencies between key values of environmental conditions and scores of indices of the second group

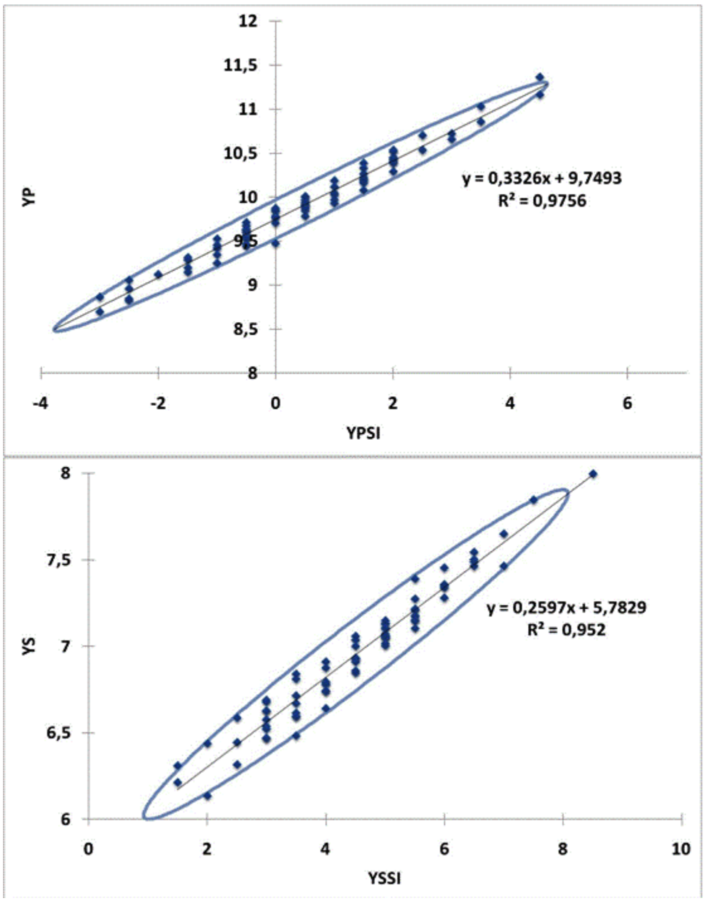

Fig.4. Interrelationships between the two derived parameters and key values of the environments

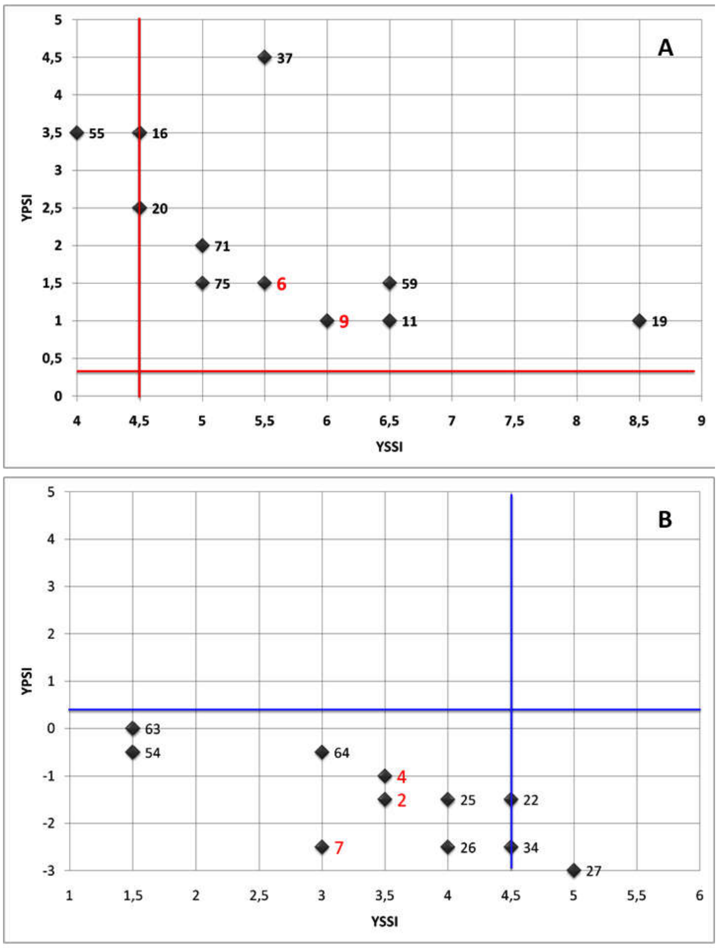

Fig.5. Location of points of varieties after arranging a high score (A) and lower scores (B) in respect to the index MP

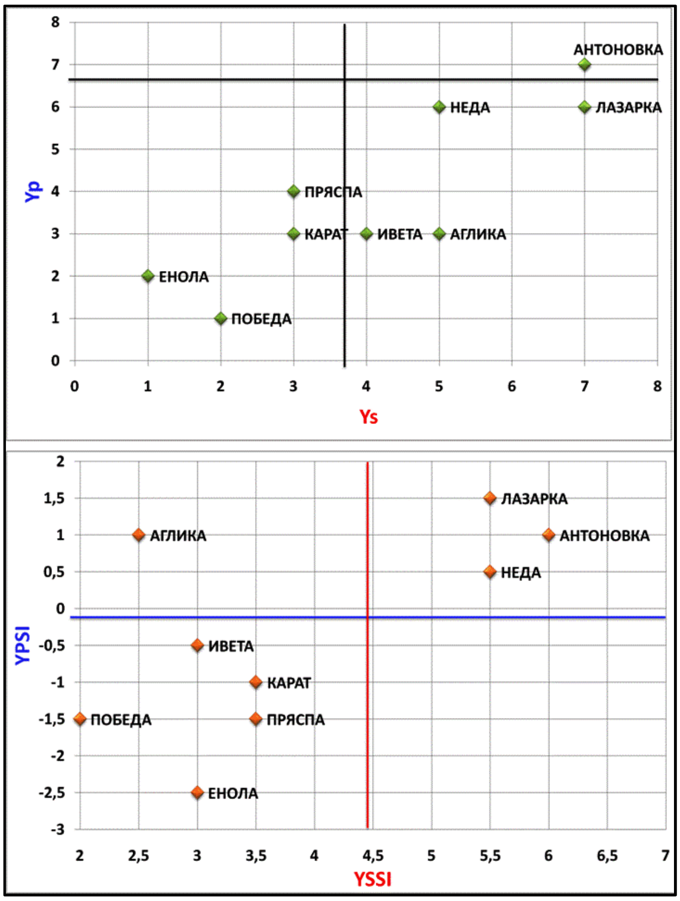

Fig.6. Arrange well known wheat varieties in two ways without and after transformation of the data

We should establish correlations between scores evaluation of each index and key values of the indication under favorable conditions (Yp) and under stress (Ys). It is extremely necessary to understand whether this approach is right one for our study. The relationship between these indexes is represented in Figures 3 and 4, for each group is shown separately. Dependencies exist between individual indexes with key values of the environment (Yp and Ys). Overall, these dependencies are almost linear with small exceptions. Generally stronger is the correlations between key values and indices that characterize the potential environmental conditions (Fig.4). This point is proven by the higher values of the coefficient of determination (R^2). After this type of analysis it is clear that scores evaluation of the indexes works properly and can be accepted as an approach for assessing and clustering the genotypes. How to make the grouping of the various classes of indices in a manner to have a correlation between the conditions of stress and potential good weather conditions of the season? On this question we seek answer through simple statistical averaging the values of both classes' indices.

The parameters, which came out of them, are [**RCI**] - **R**esilience **C**apacity **I**ndex and [**PCI**] - **P**otential **C**apacity **I**ndex. The first one is the result of the average of the index values of the first group, the second - from those of the second group. Once again we have been placing the grading assessment of the received parameters and comparing them with the indices of the respective group (Table4). The four main indices out of five showed a strong link between parameters that are produced with their participation. Only the score of an STI show low correlation with the group in which it participates. The next step is to check the suitability of the other two parameters directly related to the combination of different groups of indices. It's about following parameters: Score index stress [**YSSI**], (**Y**ield **S**tress Score Index - YSSI = *(STIs + SSIs)/2*) and score index of potential conditions **[YPSI],** (**Y**ield **P**otential **S**core **I**ndex - YPSI = *(MPs + STIs)/2- (SSIs+TOLs)/2).* Both parameters are proposed by Thiry *et al*. (2016) in order to find the point of intersection between both groups of surrounding conditions. The relationship, which has each of both parameters with the key value of the environment, is strongly positive (Fig.4). This colloborate the results which the authors of this approach received (Thiry *et al*., 2016), even in this case reliability of correlation *($R^{2)}$* is very high and the trust area in the figure (ellipse) is very narrow, which is irrefutably proof.

If we make a simple comparison between the two new groups of parameters (Table 5) and the key values of the conditions we find that the second group of parameters is much more acceptable for our purposes. Upon them there is a correlation between the parameters which are from opposite groups' comparison. Until now, all comparisons were made for each separate group between key values of the environment. For the first time we discover corelations between them. Particularly indicative of the goal that we pursue are the strong links between the key value of the conditions under stress (Ys) and parameters [YSSI] (**0,9323)** and [YPSI] (**0,6422),** which means we are closer to identify a criterion for a separation of the studied four groups of variety according to their response. Except these two groups of parameters (Thiry, *et al*. 2016) offer another 18 variants of formulas, which make different combinations of the values of the 5 indeces they used. Whether any of these combinations is suitable to be used for grouping varieties, the data of Table6 gives the answer. As we expected reliable correlations do exist, but they depend on the different combinations and environmental conditions. Compared to (Ys), these are the combinations 1, 2, 4, 5, 13, 14, and 15; against (Yp) - these are the combinations: 7, 8, 10, 11 and 16. In fact, only one combination № 9 has a relatively high correlation with the two values and they are with opposite direction. To make grouping we must accept this combination for a valid one. By grading the varieties according to the score in relation to this combination has proven that it does not separate genotypes in groups, on the contrary they dissipate into space in the scatter plot, (data not shown).

DISCUSSION

It comes to the logical question: which should be the criterion for grouping data from indexes or parameters to differentiate properly variety of groups? Thiry, *et al*. (2016) does not offer a final solution for that. On the contrary, they opined that we could partly to distinguish the varieties by some of the studied parameters. According to them by using the parameter **[YSSI]** we can not divide group A from B and group C by D. When using parameter **[YPSI]** is not possible to separate A from C and B from D. This of course does not satisfy us as conclusion, which is why we had a reason to look for some way to separate the groups. We made a several attempts to separate them with new combinations of formulas but none of the tested practice succeeds. Finally we decided to classify data varieties according to the index **MP** in ascending order. It is directly connected with both sets of environmental conditions because of that we have choosen it. After that we rated values to the parameters [**YSSI**] and **[YPSI]** into four groups each including 19 varieties (out of 76). The groups were named A, B, C, D, where A was the group with the highest values, and the D with the lowest ones. We have assumed if this point of view is true the grouping should be like: 19 varieties of group A will be located in the upper right part of the figure (see Fig.1), while those in group D - at the bottom left. What Fig.5 showed, surpassed our wildest expectations. The arrangement of the varities turned out almost as we expected. Two of the varieties under № 27 and № 55 are out of our expectations. Four other varieties (16, 20, 22, 34) are placed on the borders between two different groups. This means that the applied approach works effectively, but not in all cases. Probably we faced again the problem with inability to separate all varieties just in four exact groups. Therefore we sought statistical difference between the values of pre-formed groups of four varieties (A, B, C, D), to check if this hypothesis is true. The results show that with this approach is difficult to make difference between the groups B and C both compared to the two parameters. On the contrary, group A and group D are divided efficiently. In conclusion, we can say that the complex and complicated in certain moments approach suits our study. With some reservations, the data associated with so many indices, parameters and combinations of indices due to persistent doubts about their effectiveness as a result. We doubted, even when we arranged varieties in Figure 5. We compared the scores of the 10 well-known cultivars involved in the study but we received confusing results that will be in no help to divide the varities into groups towards their response to the environmental conditions (Table 8).

Extremely difficult to navigate if there are trends in the arrangement of numbers (scores), as well as a contrast. We can put varieties 6 and 9 in group A. Variety № 7 (Enola) combines both high indexes of Class 1 and low of Class 2, what shall we do with it?

We put on the scatter plot a point for each variety representing the scores of key environmental values (Ys) and (Yp). Surprisingly we received similar results as those we had after "complex and lengthy" calculating indices and parameters (Fig.6). We got exactly the same information towards the data varities data which is shown in Table8. The points of the different varieties after grouping by parameters [**YSSI**] and **[YPSI],** however, fully meet our information about the response of the tested varieties in both types of environmental conditions. This proves that the dividing parameters are workable.

Conclusion

We had confirmed the effectiveness of a comprehensive new approach for successful separation of varieties according to their grain yield in different growing conditions. By grading assessment and relatively easy and quick transformation of well-known breeding indices parameters it is quite possible to evaluate each group of varieties, which are grown in seasons with different levels of productivity? We proposed a specific approach using the mean value of the two seasons the varieties in each group, therefore to be properly assessed and matched towards standard varieties.

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