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GROUPING OF LOCAL AND INTRODUCED RYE SAMPLES AND THEIR OUTPUT LINES ON THE BASIS OF CLUSTER ANALYSIS

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Abstract

In the period 2009–2012 we conducted a rye variety trial on the experimental field of the Institute of Plant Genetic Resources (IPGR) situated in the town of Sadovo. 54 rye samples were examined, 16 of which were collected by expeditions within the country, and 37 received from abroad. The Bulgarian *Millennium* rye variety (current standard in IASAS) was used as a standard. A further biochemical assessment of the grain quality was performed.

The aim of the current study was to provide clarification with respect to the genetic similarity between the examined 54 native and introduced rye samples on the basis of some main biochemical parameters – *crude protein, %; lysine, %, and lysine in protein, %*, as well as their grouping using a hierarchical cluster analysis. As a result, 5 clusters were formed allowing further evaluation of the samples with regard to the studied biochemical parameters.

The present findings confirm that our classification would provide greater levels of objectivity in the evaluation of rye samples and the possibility to use them effectively in the different rye selection areas.

Keywords: rye, crude protein, lysine, cluster analysis.

INTRODUCTION

At present, many studies around the world have been focused on the determination of protein content in various plant species (Sogi et al., 2002, Tomotake et al., 2002; Rangel et al., 2003) aiming at increasing the nutritional value of the products.

The content of irrigated extract substances in rye is 62 to 71%, proteins – 10.8-15.9%, fats – 1.9-2.19%, ash – 2.15% and fibers – 1.8-2.7%, starch up to 69.8%. The amount of starch allows the production of spongy bread from the rye flour, despite the low levels of protein (clays, gluten). The total dietary fibre content in rye is between 147 and 209 g. kg – 1 in c.f. (Hansen et al., 2003). The high content of soluble arabinoxysilane, β-glucan and fructan in rye has a good physiological and healthy effect on humans (Mihalkova, 2007).

An analytical approach is also used in assessing the economic viability and sustainability of Bulgarian agriculture (Atanasov, 2015).

It is known that cereal grains are one of the main sources of plant protein, with rye yielding to wheat (Popov et al., 1966). Baeva (1981) found that the variation of protein and lysine under the influence of climatic conditions was greater in rye and triticale and weak in wheat. Therefore, we assume that the likely cause of the increased protein content is the changed agro-climatic conditions, incl. high temperatures during ripening.

The interests of Bulgarian producers today are focused on searching varieties with high productive potential, quickly adapting to the conditions of Bulgaria. At the same time, a special interest is manifested in the grain quality, which corresponds to the taste preferences of the Bulgarian consumer (Tosheva et al., 2018).

The advantage of rye is that the lysine content in its grain is slightly higher than that in wheat grain.

The aim of the study is to evaluate and group the local and introduced rye samples using a cluster analysis of the grain-tested chemical properties of the grain.

MATERIALS AND METHODS

During the period 2009-2012, a field experiment by the block method was conducted in the experimental field of ICGR – Sadovo, after a precursor of peas in 3 repetitions, with a yield area of 10 m², on cinnamon-forest soils. 54 samples were tested, 16 of which were collected during domestic expeditions and 38 were received from abroad. The Bulgarian *Millennium* variety (current standard in IASAS) was used as a standard. The *Danae* variety was used as a former 30-year standard for the country. During the vegetation, the standard agro machinery and equipment, generally used for rye cultivation in Bulgaria, were applied.

Table 1. Collection Rye

№	Sample	Country	Catalogue number	BGR
Local forms rye				
1	St Milenium	Bulgaria	A2BM0326	39862
8	Rye from Karnobat town	South Bulgaria	A2000295	3736
10	Rye from Lukovit town	North Bulgaria	A9E0037	40545
16	Rye from Hisar town	South Bulgaria	A9E0050	40800
12	Rye from Pokrovan village	South Bulgaria	A9E0053	40801
3	Rye from Begunci village	South Bulgaria	A9E1386	41045
4	Rye from Dimcha village	North Bulgaria	A9E1387	41046
5	Rye from Veliko Tarnovo	North Bulgaria	A9E1388	41047
6	Rye from Gorna Orqhovica	North Bulgaria	A9E1389	41166
7	Rye from Krasnovo village	South Bulgaria	A9E1390	41167
9	Rye from Karnobat	South Bulgaria	A9E1391	41048
11	Rye from Manole village	South Bulgaria	A9E1392	41049
13	Rye from Ruse	North Bulgaria	A9E1393	41076
14	Rye from Strelcha	South Bulgaria	A9E1394	41168
15	Rye from Stroevo village	South Bulgaria	A9E1395	41169
17	Low rye	South Bulgaria	B1BM0176	41998
Introduced rye samples				
2	Danae	Germany	569	892
18	Igusinskaja	Belarus	A7000261	41040
19	Sjabrouka	Belarus	A7000264	41042
20	Spadchina	Belarus	A7000270	41044
21	Verasen	Belarus	A7000263	41041
22	Zaveja-2	Belarus	A7000265	41043
23	Picasso	Germany	A6000412	39865
24	Recrut	Germany	A6000411	39393
25	Pollino	Germany	A6000413	39394
26	Eskudino	Germany	A6000414	39866
27	Ascari	Czech Republic	B0000065	41178
28	Albedo	Czech Republic	B0000066	41179

29	Aventino	Czech Republic	B0000067	41180
30	Conduct	Czech Republic	B0000068	41181
31	D. diament	Czech Republic	B0000069	41182
32	Evolo	Czech Republic	B0000070	41183
33	Fugato	Czech Republic	B0000071	41184
34	Matador	Czech Republic	B0000072	41185
35	Recrut	Czech Republic	B0000073	41186
36	Selgo	Czech Republic	B0000074	41187
37	Visello	Czech Republic	B0000075	41188
38	Elvi	Estonia	A9000471	41170
39	Sangaste	Estonia	A9000468	41171
40	Tulvi	Estonia	A9000470	41172
41	Vambo	Estonia	A9000469	41173
42	Intensivne 95	Ukraine	A9000466	41174
43	Veronika	Ukraine	A9000467	41175
44	Hasto	Ukraine	B0000060	41189
45	Hamara	Ukraine	B0000061	41190
46	Harakovskaq 88	Ukraine	B0000062	41191
47	Harakovskaq 95	Ukraine	B0000063	41192
48	Harakovskaq 98	Ukraine	B0000064	41193
49	Botanical Garden №23	China	A9000039	41176
50	Botanical Garden №24	China	A9000040	41177
51	AC Rifle	Canada	A9000461	41996
52	Musketar	Canada	A9000459	41994
53	Prima	Canada	A9000460	41995
54	AC Remington	Canada	A9000462	41997

Crude protein content was determined by Keldal method (Mashev et al., 1989) and lysine – colorimetric (Ernakov et al., 1972).

Experimental data were evaluated by variance analysis, for each of the samples the minimum (Min), the maximum (Max) and the mean (Mean) values, the mean error (CE), the SD and the coefficient of variation (CV). The assessment of genetic proximity was performed by comparing the following indicators: *Crude protein, %; Lysine, %* and *Lysine in Protein, %.*

A similar approach to the classification of the gene pool is also used in the evaluation of oats (Antonova, N., G. Rachovska, 2013) and tomatoes (Ivanova et al., 2010). The clustering of the 54 specimens was done through hierarchical cluster analysis. The method of intergroup binding was used (Ward, 1963; Dyuran, Odelly, 1977).

As a measure of similarity, the Euclidean intergroup spacing was used:

$$D(x,y)=\sqrt{\sum_{i=1}^{n}(x_i-y_i)^2}$$

A dendrogram was built in order to represent graphically the formed clusters. The dashed horizontal dendrogram line shows the escalated distance at which the clusters were formed. The statistical processing of empirical data was performed using the Descriptive statistics and Cluster analyzes methods using the SPSS 13 statistical program. The data is standardized.

RESULTS AND DISCUSSION

Table 2 presents the analysis of the grain chemical composition, as well as data of the minimum, maximum and average values, the error of the average ones, the standard deviation and the coefficient of variation of the studied rye samples.

It was found that the crude protein content of the tested samples ranged from 10.0% to 17.21%, at an average value of the study indicator 13.67 ± 0.2%. It was higher than the standards for 6 samples (2 from Bulgaria, 2 from the Czech Republic, 2 from Canada). Results for the content and yield of lysine and lysine in the protein were encouraging. Lysine in grain ranged from 0.47% to 0.90%. The lysine content in protein was from 3.07% to 7.04%. 25% of the samples had a higher content of lysine than the standards. The variability of the measured variables expressed by the coefficient of variation (CV) is average.

A hierarchical cluster analysis was applied to identify the similarity and proximity of the tested genotypes. Evaluation of the genetic proximity between the samples was performed based on the examined chemical parameters. The clustering results were presented using a dendrogram (Figures 1 and 2) showing the sequence of the grouping of objects and clustering. With the conduction of the cluster analysis, five major clusters are formed.

The first cluster consists of two sub-clusters. The first sub-cluster covers 9 samples, six of which are of local origin. The samples from this cluster are similar in terms of lysine content and lysine content in the protein (Fig. 1). Comparing the Euclidean distances between them, it can be seen that there is practically no difference between A7000264, A7000263, A9E1389 and A9000468; A2000295, A9E1395 and A9E1387; A9E1386 and A9E1391.

The second sub-cluster comprises 12 samples – A6000414, B0000060, A7000265, B1MM0176, A6000411, A7000270, A9E0050, B0000068, A9E1388, A9E1393, A9000470, A9000466, eight of which are introduced, and four are native rye varieties. The samples included in this sub-cluster are characterized by similar indicators of protein content and lysine content in protein. The values of the examined indicators of samples in the first sub-cluster are higher than the values of the indicators of samples in the second sub-cluster.

In the second cluster, the local rye forms are A9E0053, A9E1394, A9E1390, A9000469, A9E0037, *Millennium* and *Danae* standards, as well as A900040 of Chinese origin. The samples from this cluster are distinguished with higher values of the crude protein, lysine and lysine content in protein standards.

The third core cluster includes nine samples, one from Bulgaria - A9E1392, and the other B0000075, A9000469, A9E1392, A7000261, A9000471, B0000073, B0000074, B0000075, B0000063, B0000072 – introduced. The samples within this cluster are identical by their lysine content in protein.

Samples included in the fourth and fifth clusters (Fig. 2) are introduced rye samples, mainly from the Czech Republic and Canada.

The fourth cluster includes 8 samples – B0000061, B0000064, B0000069, B0000065, B0000071, A6000412, B0000070 and B0000062 are identical in terms of lysine content and lysine content in the protein. The values of the examined indicators of samples included in this cluster are lower than all tested samples.

The fifth cluster includes 7 samples numbered A9000461, A9000459, B0000066, B0000067, A9000462, A9000039 and A9000460. They are characterized by a low protein content of lysine.

Table 2. Variation analysis of grain chemical composition of rye

	Mean	Min	Max	CE	SD	CV, %
Crude protein, %	13.67	10.0	17.21	0.2	1.76	12.87
Lysine, %	0.7	0.47	0.90	0.01	0.1	14.28
Lysine in protein, %	5.1	3.07	7.04	0.1	0.9	17.64

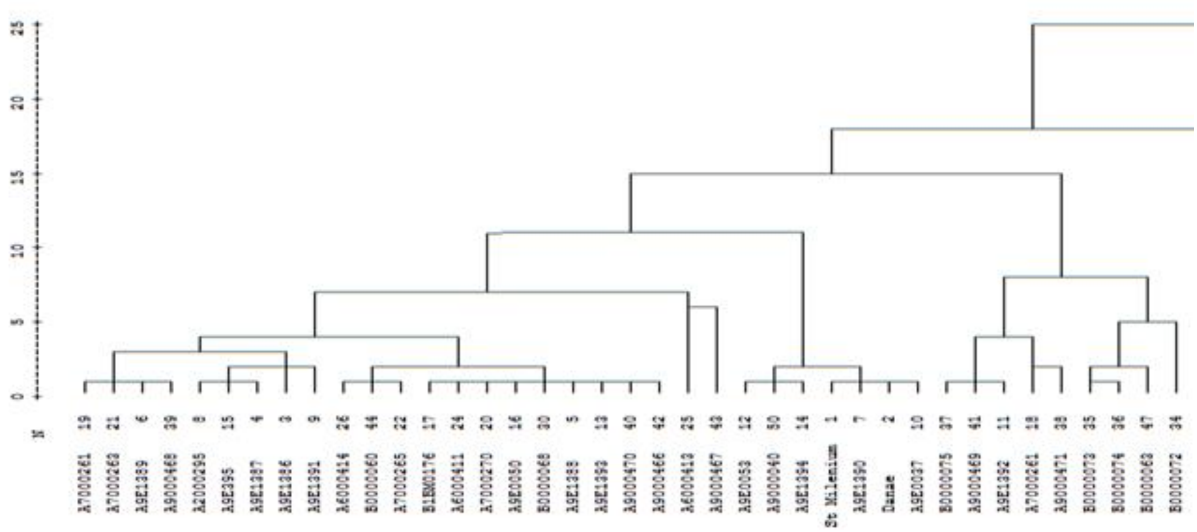

Fig. 1. *A dendrogram on the base of the average intergroup distances for I, II and III cluster*

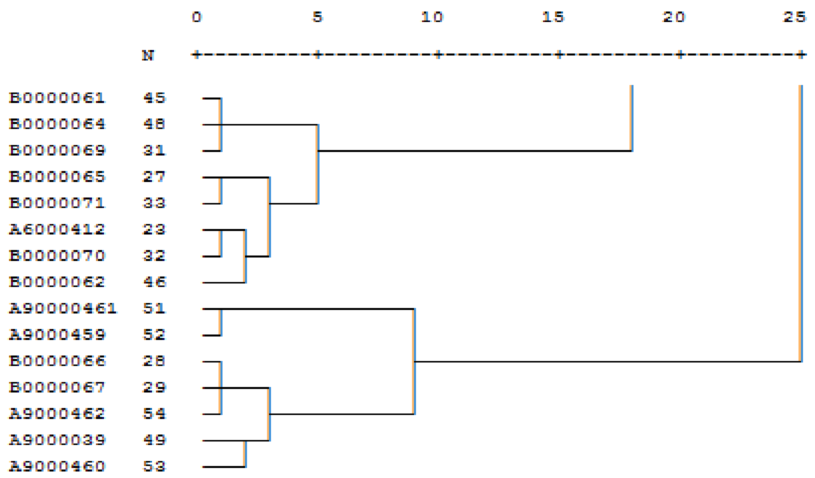

Fig. 2. *A dendrogram on the base of the average intergroup distances for IV and V cluster*

All tested samples of their indicators are of interest and can be used in the selection schemes.

CONCLUSIONS

1. The collection of local and introduced rye samples is grouped into five clusters of different genetic proximity. Clusters include samples of similar qualities that are a valuable genetic resource and can be used in the selection of varieties with high biological potential.

2. The classification makes it possible to increase the objectivity of the evaluation and to increase the possibilities for the collection of rye to be used in different directions: direct implementation in production, selective improvement activity, international exchange.

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