Assessment genetic diversity of einkorn genotypes (*Triticum monococcum* L.) by gliadin electrophoresis

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Abstract: The genetic diversity of gliadins in twenty two einkorn accessions preserved under long-term seed storage condition in the National gene bank of Bulgaria were evaluated, using acid polyacrylamide-gel electrophoresis (Acid-PAGE). In total, 64 polymorphic bands and 22 gliadin patterns were identified. Thirty four different mobility bands and 21 gliadin patterns were identified in the ω-gliadin zone, 12 bands and 16 patterns were noted in the γ-gliadins, 17 patterns and 12 mobility bands were found for β-gliadins and six bands with five different α -gliadin patterns were determined. The genetic diversity index (H) was the highest for ω-gliadins (0.950), followed by β-gliadins (0.924) and γ- (0.914), respectively and the lowest value was detected in α-gliadin patterns (0.120). Cluster analysis based on the UPGMA method and Nei and Li similarity coefficients classified all the genotypes into 3 main groups. No relationships between genetic diversity, geographic origin and the genotypes were observed. The results of cluster analysis justify the high level of genetic variation among investigated einkorn accessions.

Key words: A-PAGE electrophoresis; einkorn; genetic diversity; gliadins

Ocenjevanje genetske raznolikosti genotipov enozrne pšenice (*Triticum monococcum* L.) z elektroforezo gliadinov

Izvleček: Genetska raznolikost gliadinov 22 akcesij enozrne pšenice, ki je bila shranjena v razmerah dolgotrajnega shranjevanja v nacionalni genski banki Bolgarije je bila ovrednotena s poliakrilamidno elektroforezo v kislem pH območju (Acid-PAGE). Celokupno je bilo ugotovljenih 64 polimorfnih prog in 22 vzorcev gliadinov. Določeno je bilo 32 različnih spremenljivih prog in 21 vzorcev gliadinov je bilo določenih znotraj ω-gliadinov, 12 prog in 16 vzorcev je bilo znotraj γ-gliadinov, 17 vzorcev in 12 spremenljivih prog je bilo v območju β-gliadinov in 6 prog s petimi različnimi vzorci v območju α -gliadinov. Indeks genetske raznolikosti (H) je bil največji za ω-gliadine (0,950), ki su mu sledile vrednosti za β- (0,924) in γ-gliadine (0,914). Najmanjše vrednosti indeksa so bile ugotovljene za α-gliadine (0,120). Klasterska analiza, ki je temeljila na UPGMA metodi in koeficientu podobnosti po Nei in Li je razvrstila vse genotipe v tri glavne skupine. Ugotovljeno ni bilo nobene povezave med genetsko raznolikostjo, geografskim poreklom in genotipi. Rezultati klasterske analize so potrdili veliko genetsko variabilnost med analiziranimi akcesijami enozrne pšenice.

Ključne besede: elektroforeza A-PAGE; enozrna pšenica; genetska raznolikost; gliadini

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1 INTRODUCTION

Genetic diversity has played a vital role in the success of crop improvement. Knowledge of genetic diversity has been successfully used for efficient germplasm management and utilization, genetic fingerprinting and genotype selection (Engles et al., 2002; Aliyeva et al., 2012).

The electrophoresis of seed storage proteins used for variety fingerprinting is reliable, simple, repeatable and economic procedure. It can be utilized by wheat breeders to detect variability among wheat genotypes to identify new sources of variation that could be used in crop improvement programs (Aliyeva et al., 2012).

Gliadins are alcohol-soluble storage proteins that accounts for about 40 %, by mass, of all proteins of wheat flour. They are one of the most polymorphic proteins in nature and one of the major protein components of the human diet (Dziuba et al., 2014; Metakovsky et al., 2018). They are monomeric proteins with a molecular mass in the range of 35,000 to 70,000 daltons. (Shewry et al., 2002; Kuktaite, 2004; Meintjes, 2004; D'Ovidio & Masci, 2004; Shuaib et al., 2007). When fractionated in acidic medium, they are divided into 4 major groups- α, β, γ and ω (Konarev, 1983; Bushuk, 1991). The genes controlling α- and β-gliadins were found to be located in the short arms of the chromosomes of the sixth (Gli-2) homologous group - 6A, 6B and 6D, and of the γ- and ω-gliadins of the first (Gli-1) homeology group - 1A, 1B and 1D (Payne et al., 1987; Dachkevitch et al., 1993; Nieto-Taladriz et al., 1996; Branlard, 2004). A small group of gliadin fractions are encoded by genes located at Gli-3, Gli-4, Gli-5 and Gli-6 loci (Pogna et al., 1993; Rodriguez & Carrillo, 1996; Metakovsky et al., 1997 a, 1997 b). An original methodology for their electrophoretic separation (A-PAGE) and a nomenclature for their designation (Bushuk & Zilman, 1978; Bushuk & Sapirstein, 1991) have been developed to detect the complex polymorphism of gliadins and to identify them. Intensive work with gliadins has been carried out in Ukraine and Russia (Metakovsky et al., 1984, 1986; Metakovsky & Sozinov, 1987; Metakovsky & Novoselskaya, 1991; Novoselskaya-Dragovich et al., 2003). Genetic polymorphism has been used to evaluate genetic diversity in wheat samples in Austria, Yugoslavia, Canada, Italy, France, Spain (Metakovsky et al., 1991, 1993, 1994, 1997 a, 1997 b, 2000; Metakovsky & Branlard, 1998; Ruiz et al. , 2002 a, 2002 b; Branlard et al., 2001), Japan (Tanaka et al., 2003), China (Wu et al., 2007), India (Sewa et al., 2005), Pakistan (Anjum et al., 2000), Bulgaria (Stoyanova, 2002), Africa (Mohd et al., 2007). Based on the gliadin spectra, catalogs have been drawn up in some countries to identify varieties (Sapirstein & Bushuk, 1986; Lookhart et al., 1983; Metakovsky et al.,

1991, 1994, 2018; Velkov, 1991; Stoyanova & Kolev, 1996; Ruiz et al., 2002 a, 2002 b).

The allelic compositions at Gli-A1 and Gli-A2 loci in *Triticum monococcum* L. ssp. *monococcum*, *Triticum monococcum* ssp. *boeoticum* (Boiss.) C. Yen and *Triticum urartu* Thumanjan ex Gandilyan had investigated (Metakovsky & Baboev, 1992, Saponaro et al., 1995; Zhao-cai et al., 2006). The high-quality gliadin alleles as Gli-A2b were found in einkorn wheat (Zhao-cai et al., 2006). In 40 accessions of *Triticum boeoticum* Boiss. and *Triticum urartu* Thumanjan ex Gandilyan collected from different regions of Iran, Ahmadi and Pour-Aboughadareh (2015) found that 92 % of the accessions carried Gli-A2 allele detected by Long et al. (2005) and Kawaura et al. (2005). The variation in gliadin seed storage proteins in Spanish einkorn was high, with seven allelic variants for the Gli-Am1 locus and fourteen for the Gli-Am2 locus found among the evaluated accessions (Alvarez et al., 2006; Alvarez & Guzmán, 2013).

The aim of this study was to compare the genetic diversity between einkorn genotypes with different geographic origin by electrophoretic patterns of seeds proteins.

2 MATERIAL AND METHODS

Twenty two einkorn accessions with different geographic origin preserved under long-term seed storage condition in the National gene bank of Bulgaria more than 20 years were evaluated by gliadin electrophoresis (Table 1).

Acid-PAGE (acid polyacrilamyde gel electrophoresis) was carried out according to the standard reference method of ISTA (Draper 1987; Anonymous 2003). Proteins were extracted from a bulk sample of 50 mg finely ground powdered seeds with 300 µl extracting solution (0.05 g Pyronin G; 25 ml 2-chloroethanol), stained overnight at room temperature, and centrifuged for 30 min at 17 000 g and 14 ºC. Then, 10 µl of the extracts were loaded into wells. Gliadin electrophoresis was performed on a vertical polyacrylamide gel with a thickness of 1.5 mm and an electrode buffer with a pH of 3.2 using a Consort E835 vertical unit (with gel cassette 200 × 200 mm). Electrophoresis was carried out at 20 mA for 5 hours and 15 minutes. Staining of gels was performed in a solution of Coomasie Brilliant Blue G-250: Coomasie Brilliant Blue R-250 (1:3), dissolved in trichloroacetic acid/methanol for 48 hours.

Specialized software BIO-1D++, version 11.07 was used to create databases for the gliadin spectra of the studied genotypes. Based on the results of electrophoretic band spectra, genetic similarity coefficient of Nei &

Table 1: List of accessions included in the study

Number of the accessions	Genus	Species	Spaut.	Subtaxa	Origin
BGR 19069	*Triticum*	*monococcum*	L.	var. *hornemannii*	Germany
BGR 19065	*Triticum*	*monococcum*	L.	var. *hornemannii*	Georgia
BGR 30035	*Triticum*	*monococcum*	L.	var. *hornemannii*	Russia
BGR 19078	*Triticum*	*monococcum*	L.	var. *laetissimum*	Germany
BGR 30022	*Triticum*	*monococcum*	L.	var. *laetissimum*	Germany
BGR 19061	*Triticum*	*monococcum*	L.	var. *laetissimum*	Spain
BGR 19079	*Triticum*	*monococcum*	L.	var. *nigricultum*	Germany
BGR 11001	*Triticum*	*monococcum*	L.	var. *atriaristatum*	Switzerland
BGR 12386	*Triticum*	*monococcum*	L.	var. *laetissimum*	Germany
BGR 19063	*Triticum*	*monococcum*	L.	var. *flavescens*	Spain
BGR 30030	*Triticum*	*monococcum*	L.	var. *eincorn*	Russia
BGR 30031	*Triticum*	*monococcum*	L.	var. *vulgare*	Russia
BGR 30028	*Triticum*	*monococcum*	L.	var. *hornemannii*	Switzerland
BGR 30026	*Triticum*	*monococcum*	L.	var. *hornemannii*	Switzerland
BGR 28720	*Triticum*	*monococcum*	L.	var. *macedonicum*	Germany
BGR 30036	*Triticum*	*monococcum*	L.	var. *macedonicum*	Russia
BGR 19055	*Triticum*	*monococcum*	L.	var. *flavescens*	Germany
B2E0417	*Triticum*	*monococcum*	L.	*var. vulgare*	Bulgaria
B3E0025	*Triticum*	*monococcum*	L.	*var. vulgare*	Bulgaria
BGR 28717	*Triticum*	*monococcum*	L.	var.*eincorn*	Russia
BGR 30027	*Triticum*	*monococcum*	L.	var.*albohornemannii*	Germany
BGR 26774	*Triticum*	*monococcum*	L.	var.*eredvianum*	Germany

Li (1979) was calculated for all possible pair of electrophoregrams. The similarity matrix was used to construct the dendrogram by the unweighted pair group average method (UPGMA).

The genetic diversity for each gliadin pattern was calculated as per Nei (1973), as $H = 1 - \Sigma pi^2$, where H is the genetic variation index and Pi is the proportion of a particular pattern in each group of α, β, γ and ω gliadins separately. The mean value of H was calculated for all the four groups of gliadins.

3 RESULTS AND DISCUSSION

In the Table 2 were presented number of gliadin bands, patterns, and the genetic variation index in gliadins for 22 investigated accessions of *Triticum monococcum* L. Among the 22 einkorn accessions analysed, 64 different bands were detected assuming that the bands with the same relative mobility represent the same protein. Each zone (α, β, γ and ω) was considered as a single locus and the different patterns as allelic variants.

The patterns within each gliadin group of α, β, γ and ω were identified by comparing banding patterns of each einkorn accession with all the other einkorn accessions (Ojaghi & Akhundova, 2010; Aliyeva et al., 2012).

A total of 34 different mobility bands and 21 gliadin patterns were identified in the ω-gliadin zone. Bands varied between two and seven in each ω-gliadin pattern, as patterns with five bands being the most frequent (40.9 %). Twenty one accessions presented its unique ω-gliadin pattern, while two genotypes had the same patterns, respectively 14-BGR 19055 and 15-B2E0417 with pattern 14 (Figure 1).

In the γ-gliadin zone, 12 bands and 16 different patterns were noted. The γ -gliadin pattern 5, were observed in accessions 5-BGR 30022 and 19-BGR 30027. The pattern 6 was detected in 6-BGR 19061 and 7-BGR 19079. The γ -gliadin patterns 10 and 12 were marked respectively in two groups of accessions – 11-BGR 19063, 14-BGR 19055 and 15-B2E0417; 13-BGR 30026, 18-BGR 26774 and 20-BGR 28717 (Figure 1).

Seventeen β- gliadin patterns and totally 12 different mobility bands were found. The bands in the gliadin

Table 2: Number of gliadin bands, patterns, and the genetic diversity in gliadins for 22 accessions of *Triticum monococcum* L.

№	Accession number	Number of gliadin bands				
		α	β	γ	ω	Total bands
1	BGR 19069	3	4	2	5	14
2	BGR 19065	3	6	1	5	15
3	BGR 30035	4	4	4	4	16
4	BGR 19078	3	4	3	2	12
5	BGR 30022	4	3	4	5	16
6	BGR 19061	3	7	4	2	16
7	BGR 19079	3	4	4	3	14
8	BGR 11001	3	4	5	4	16
9	BGR 12386	4	3	4	6	17
10	BGR 19063	3	4	4	4	15
11	BGR 30031	3	5	3	3	14
12	BGR 30028	3	6	4	5	18
13	BGR 30026	4	5	4	4	17
14	BGR 19055	3	5	3	6	17
15	B2E0417	3	4	3	6	16
16	B3E0025	3	5	2	5	15
17	BGR 28720	3	5	4	3	15
18	BGR 26774	3	5	4	7	19
19	BGR 30027	3	5	4	5	17
20	BGR 28717	3	5	4	5	17
21	BGR 30036	3	5	4	5	17
22	BGR 30030	3	6	1	5	15
Range of gliadin bands		3-4	3-7	1-5	2-7	12-19
Number of gliadin patterns		5	17	16	21	22
Genetic variation index (H, %)		0.120	0.924	0.914	0.950	0.727

patterns varied from 3 to 7 bands, as patterns with five bands being the most frequent (45 %). Thirteen accessions had specific patterns in the β- gliadin zone, while 1-BGR 19069 and 4-BGR 19078 with pattern 1, 2-BGR 19065, 20-BGR 28717 and 22-BGR 30030 with pattern 2, 3-BGR 30035 and 10-BGR 19063 with pattern 3, 5-BGR 30022 and 9-BGR 12386 with pattern 4 had the similar patterns (Table 2, Fig. 1).

Six bands were recorded in α -gliadin region and only five different α -gliadin patterns were determined. Two numbers of accessions (3-BGR 30035 and 13-BGR 30026) had specific patterns, respectively patterns with numbers 2 and 5. α -gliadin pattern with number 1 included ten accessions, pattern number three -2 accessions (5-BGR 30022 and 9-BGR 12386) and pattern number four - 8 accessions (6-BGR 19061, 7-BGR 19079, 8-BGR 11001, 10-BGR 19063, 11-BGR 30031, 12-BGR 30028,

14-BGR 19055, 18-BGR 26774). The accessions with three bands in the α-gliadin zone predominate (81.82 %).

Considering the four zones together, 22 gliadin patterns were identified, as in BGR 26774- 19 bands and in BGR 30028- 18 bands were detected. The lowest numbers of bands were found in BGR 19078 genotype (Table 2).

According to the work of Metakovsky and Baboev (1992), two independent gliadin blocks were determined for each einkorn accession. The upper block was composed by ω - and γ -gliadins, and in some cases also by one slow moving β-gliadin, whereas the lower block was formed by β - and α -gliadins. These two blocks were encoded by the *Gli-A1m* and the *Gli-A2m* loci on the short arm of chromosomes 1 and 6, respectively. Ciaffi et al. (1997), in study of 74 accessions of *T. monococcum* from Italy, Greece, Turkey and Russia found more allelic variation at *Gli-A1m* than at *Gli-A2m*. In contras Alvarez et
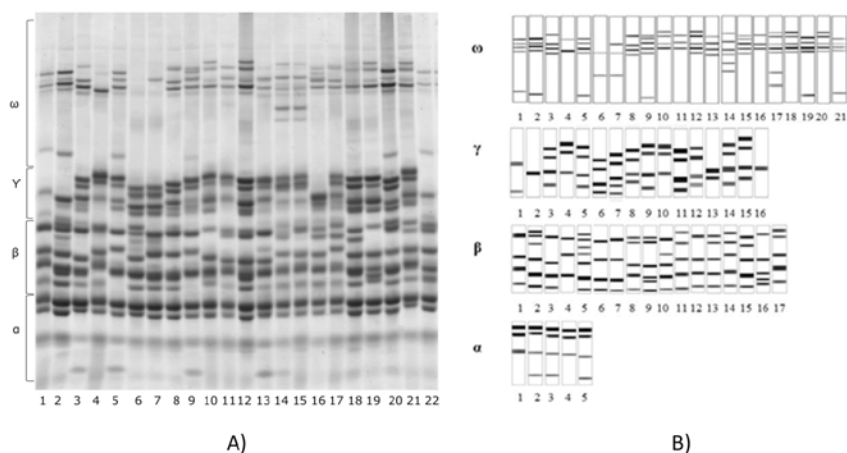
Figure 1: A) Gliadin patterns of accessions of *Triticum monococcum* L. after acid polyacrylamide gel electrophoresis (A-PAGE) (1- BGR 19069, 2- BGR 19065, 3- BGR 30035, 4- BGR 19078, 5-BGR 30022, 6- BGR 19061, 7- BGR 19079, 8- BGR 11001, 9- BGR 12386, 10- BGR 19063, 11- BGR 30031, 12- BGR 30028, 13- BGR 30026, 14- BGR 19055, 15- B2E0417, 16- B3E0025, 17-BGR 28720, 18- BGR 26774, 19- BGR 30027, 20- BGR 28717, 21- BGR 30036, 22- BGR 30030); B). Ideograms of different gliadins patterns in the α, β, γ and ω regions

al. (2006) found 7 and 14 alleles for the *Gli-A1*m and *Gli-A2m* between the 48 evaluated einkorn accessions. Zhaocai et al. (2006) identified 14 and 19 allele variants, respectively at *Gli-A1m* and *Gli-A2m* of 41 *T. monococcum* accessions. Ruiz et al. (2007) noted that *Gli-A2m* was the most polymorphic and the most useful to distinguish between 17 einkorn varieties.

In this study, it was observed that ω, γ, and β areas had high allelic variants but the least allelic variants were established in area α. The highest allelic variants had ω gliadins (Fig.1).

In investigated 22 einkorn genotypes, no identical gliadin patterns were found in the gliadin spectra. The genetic diversity based on the patterns was calculated for each of the four zones. The genetic variation indexes varied between 0.120 and 0.950. Omega (ω) zone was found to have the biggest diversity (H = 0.950), followed by β (H = 0.924) and γ (H = 0.914) and the least diversity being that of α (H = 0.120). The mean value of H calculated for all the four groups of gliadins was also high (H = 0.727) (Table 2). It indicated that high genetic variation existed in the einkorn wheat (Zhao-Cai et al., 2006).

The genetic similarities (GS) among 22 einkorn accessions estimated by Nei and Li coefficient (1979) are presented in Table 3. It ranged from 0.36 to 0.97. The highest GS (0.97) was found between accessions: BGR 30022 (from Germany) and BGR 12386 (from Germany), BGR 19055 (from Germany) and B2E0417 (from Bulgaria), respectively, which indicates that the genetic diversity within these pairs of accessions was very low. The lowest value of GS was 0.36, which was observed

between accession BGR 30028 (from Switzerland) and BGR 30030 (from Russia).

Cluster analysis of gliadin bands using the UPGMA method, as well as Nei and Li (also called Dice) similarity coefficients is presented in Fig. 2. The correlation between the cophenetic value matrix and the original similarity coefficient matrix was high (r = 0.739) indicating a good fit of the cluster analysis performed (Ma et al., 2012). The dendrogram allowed distinguishing three main clusters. Cluster 1 included 6 genotypes. It was divided into three subgroups, The first subgroup included only genotype1-BGR 19069 (var. *hornemannii* from Germany). Subcluster 2 combined 3 genotypes: 2-BGR 19065 (var. *hornemannii* from Georgia), 22-BGR 28717 (var. *eincorn* from Russia) and 20-BGR 30030 (var. *einkorn* from Russia), which had pattern 2 in the β zone and pattern 1 in α zone. Subgroup 3 included 4-BGR 19078 (var. *laetissimum* from Germany) and 21-BGR 30036 (var. *macedonicum* from Russia), which had pattern 1 in α zone. In cluster 2 were included 15 genotypes. The cluster was divided to two subgroups. The first subgroup grouped 3-BGR 30035 (var. *hornemannii* from Russia), 13-BGR 30026 (var. *hornemannii* from Switzerland), 5-BGR 30022 (var. *laetissimum* from Germany), 9-BGR 12386 (var. *laetissimum*, Germany), 10-BGR 19063 (var. *flavescens* from Spain), 12- BGR 30028 (var. *hornemannii* from Switzerland), 18-BGR 26774 (var. *eredvianum* from Germany), 19-BGR 30027 (var. *albohornemannii* from Germany), 6-BGR 19061 (var. *laetissimum* from Spain) and 7-BGR 19079 (var. *nigricultum* from Germany). The second subgroup included 5 genotypes, grouped also into three subgroups. In the first subgroup was separated

Table 3: Average genetic similarity matrix of Nei and Li (1979) coefficient based on A-PAGE gliadin patterns for the 22 einkorn accessions

	L1	L2	L3	L4	L5	L6	L7	L8	L9	L10	L11	L12	L13	L14	L15	L16	L17	L18	L19	L20	L21	L22
L1	1																					
L2	0.55	1																				
L3	0.67	0.58	1																			
L4	0.69	0.67	0.64	1																		
L5	0.67	0.65	0.81	0.64	1																	
L6	0.53	0.58	0.69	0.57	0.63	1																
L7	0.5	0.62	0.8	0.46	0.73	0.87	1															
L8	0.53	0.65	0.56	0.5	0.69	0.69	0.73	1														
L9	0.52	0.63	0.85	0.55	0.97	0.61	0.71	0.67	1													
L10	0.55	0.6	0.77	0.74	0.71	0.65	0.76	0.71	0.75	1												
L11	0.43	0.69	0.73	0.69	0.6	0.67	0.64	0.6	0.58	0.75	1											
L12	0.56	0.48	0.76	0.47	0.65	0.71	0.75	0.76	0.63	0.85	0.69	1										
L13	0.52	0.63	0.85	0.62	0.85	0.73	0.77	0.73	0.88	0.81	0.84	0.74	1									
L14	0.45	0.69	0.67	0.69	0.67	0.67	0.65	0.67	0.65	0.63	0.77	0.69	0.76	1								
L15	0.47	0.58	0.69	0.71	0.69	0.63	0.67	0.69	0.55	0.65	0.73	0.53	0.73	0.97	1							
L16	0.69	0.73	0.77	0.67	0.77	0.71	0.76	0.65	0.75	0.73	0.76	0.73	0.75	0.69	0.65	1						
L17	0.76	0.67	0.71	0.74	0.77	0.65	0.62	0.58	0.75	0.8	0.83	0.67	0.75	0.69	0.65	0.87	1					
L18	0.67	0.53	0.8	0.58	0.74	0.69	0.67	0.69	0.78	0.82	0.73	0.81	0.67	0.67	0.74	0.71	0.76	1				
L19	0.77	0.56	0.85	0.69	0.79	0.67	0.65	0.55	0.76	0.88	0.71	0.8	0.71	0.59	0.61	0.69	0.75	0.89	1			
L20	0.71	0.88	0.67	0.69	0.67	0.55	0.45	0.55	0.76	0.69	0.58	0.51	0.71	0.65	0.61	0.5	0.63	0.61	0.76	1		
L21	0.71	0.56	0.61	0.76	0.55	0.48	0.52	0.48	0.65	0.63	0.52	0.57	0.53	0.53	0.48	0.56	0.81	0.72	0.76	0.71	1	
L22	0.83	0.93	0.58	0.74	0.58	0.52	0.55	0.45	0.5	0.6	0.48	0.36	0.56	0.56	0.58	0.6	0.67	0.41	0.56	0.75	0.75	1

The L1 to L22 correspond to the following number of accessions: L1- BGR 19069, L2- BGR 19065, L3- BGR 19063, L4- BGR 30035, L4- BGR 30035, L6- BGR 19078, L5- BGR 30022, L6- BGR 19078, L7- BGR 19061, L8- BGR 11001, L9- BGR 12386, L10- BGR 19063, L11- BGR 30031, L12- BGR 30028, L13- BGR 30026, L14- BGR 19055, L15- B2E0025, L16- B2E0417, L17- BGR 28720, L18- BGR 26774, L19- BGR 30027, L20- BGR 28717, L21- BGR 30036, L22- BGR 30030
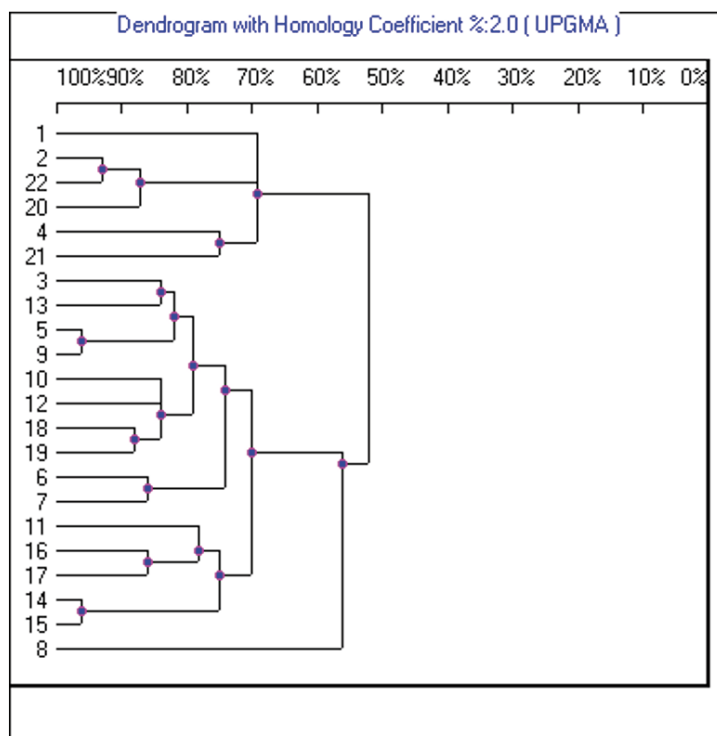
Figure 2: UPGMA dendrogram base on A-PAGE and Nei and Li similarity index (1- BGR 19069, 2- BGR 19065, 3- BGR 30035, 4- BGR 19078, 5-BGR 30022, 6- BGR 19061, 7- BGR 19079, 8- BGR 11001, 9- BGR 12386, 10- BGR 19063, 11- BGR 30031, 12-BGR 30028, 13- BGR 30026, 14- BGR 19055, 15- B2E0417, 16- B3E0025, 17-BGR 28720, 18- BGR 26774, 19- BGR 30027, 20- BGR 28717, 21- BGR 30036, 22- BGR 30030)

11-BGR 30031 (var. *vulgare* from Russia), into second respectively 16-B3E0025 (var. *vulgare* from Bulgaria) and 17-BGR 28720 (var. *macedonicum* from Germany), while in the third were 14-BGR 19055 (var. *flavescens* from Germany) and 15-B2E0417 (var. *vulgare* from Bulgaria), which had pattern 14 in the ω-gliadin zone and pattern 10 in the γ-gliadin. In the last cluster 3 was separated genotype 8-BGR 11001 (var. *atriaristatum* from Switzerland). The results of cluster analysis justify the high level of genetic variation among einkorn genotypes. No relationships between genetic diversity, geographic origin and the genotypes were observed. Zhao-Cai et al. (2007) also confirmed that the genetic relationships in diploid wheats based on the gliadins were associated with the species or subspecies rather than the geographical origin. Khabiri et al. (2013) and Medouri et al. (2015) evaluated the relationships among 17 populations of *Aegilops cylindrica* Host and *Aegilops geniculata* Roth by gliadin polymorphism and found that genetic diversity did not follow the geographical distribution. Zaefizadeh et al. (2010) also found no correlation between genetic diversity and the geographical distribution of durum wheat landraces studied. A considerable differentiation of common wheat germplasms from different countries and breeding centres was discovered earlier using gliadin alleles as wheat genotype markers (Metakovsky et al., 1991, 1994; Chernakov and Metakovsky, 1994; Metakovsky and Branlard, 1998).

Additional investigation must to do to analyze population structure of einkorn wheat and relationships between agro-morphological data and gliadins.

4   CONCLUSION

The investigated 22 einkorn genotypes were characterized with high genetic diversity on the basis of their alcohol soluble proteins-gliadins by the acid-PAGE method. The ω-zone was the most polymorphic, followed by β, γ and α, respectively. The highest genetic similarities (GS) was found between accessions: BGR 30022 and BGR 12386, BGR 19055 and B2E0417, respectively. The lowest GS was observed between accession BGR 30028 and BGR 30030. The results of cluster analysis justify the high level of genetic variation among einkorn accessions. No relationship between genetic diversity, geographic origin and the genotypes was observed.

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