JJBS

Jordan Journal of Biological Sciences

Volume 2, Number 4, December 2009
ISSN 1995-6673
Pages 159 - 166

Polymorphism of Protein Fractions as Biochemical Markers for Identification of Wheat Varieties

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Abstract

The electrophoretic banding patterns of gliadin, albumin and globulin in wheat varieties grown in Sulaimanyah were determined by acid-polyacrylamide gel electrophoresis (Acid-PAGE) and Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). The varieties of bread wheat used in the study showed 21 different patterns: six of them were identified to be corresponding to Ω-gliadin, three to the γ-gliadin, six to the β-gliadin, and other six to the α-gliadin. The gliadin patterns of wheat bread varieties greatly differed from the patterns of Italian wheat variety (Costantino), and the variation detected in wheat varieties from Sulaimanyah was limited to forty seven patterns. Only Tammuz showed the presence of gamma 47. The gliadin gamma 47 is only genetic markers for technological properties in wheat. The electrophoretic banding patterns showed variations between the bread wheat varieties. The highest value of similarity was observed between the varieties Cham 4 and Cham 6 for gliadin analysis. The gliadin pattern revealed more polymorphisms than the albumin and globulin pattern. On the other hand, in durum wheat varieties, two types of omega gliadin: omega gliadin 33 and 35 and two types of gamma gliadin were found: gamma gliadin 42 and gamma gliadin 45. A total of 84 band patterns were identified (durum wheat), of which 2 different mobility bands were in the region of omega gliadins, 1 in the region of gamma gliadins, 2 in the region of beta gliadin and 2 in the region of alpha gliadins. Genetic diversity index was highest in Creso Kurde and followed by Ovanto.

الملخص

تتضمن الدراسة تحديد التركيب النموذجى لبروتينات كليادين والبومين وكلوبيولين لستة اصناف من الحنطة الناعمة المزروعة فى السليمانية بوساطة الفصل الكهربائى لهلام الحامض متعدد الاكريلامايد. وتم اكتشاف 21 نموذجاً او تراكيبياً مختلفة منها 6 نماذج لكليادين اوميكا و3 نماذج لكليادين كاما و6 نماذج لكليادين بيتا 6 نماذج لكليادين الفا. ان التركيب النموذجى لكليادين الموجود فى الاصناف الحنطة الناعمة المزروعة فى السليمانية تختلف عن تركيب النموذجى لكليادين الموجودة فى صنف الايطالى. ان بذور الصنف تموز هو الوحيد الذى يحتوى على كليادين كاما 47 التى تعتبر كدليل ايجابى لنوعية الحنطة. ان التركيب النموذجى لبروتين كليادين والبومين وكلوبيولين اظهرت الاختلاف الوراثى بين الاصناف المدروسة.كما لوحظت اوجة التشابة بين بذور الصنفين شام 4 وشام 6. واكدت النتائج ان التحليل النموذجى لكليادين اظهرت اختلافاً واضحاً بين الاصناف المدروسة مقارنة بالتحليل النموذجى لالبومين وكلوبيولين. وفى الحنطة الخشنة, تم اكتشاف نوعين من كليادين اوميكا 33 و 35 ونوعين من كليادين كاما 42 و 45. وجدت 2 نماذج مختلفة لكليادين فى المنطقة اوميكا ونموذج واحد فى منطقة كاما و تركيبين مختلفين فى المنطقة بيتا و تركيبين مختلفين فى المنطقة الفا. ان الاصناف كريزو كردى و اوفانتو من الاصناف الاكثر اختلافا عن باقى الاصناف المدروسة.كما ان تركيب النموذجى لبروتين البومين و كلوبيولين برزت الاختلاف الوراثى بين الاصناف المدروسة.

Keywords: Bread wheat, Durum wheat, Gliadins, Albumin, Globulin, Acid-PAGE, SDS-PAGE.

1. Introduction

The gluten proteins of wheat provide unique attributes to flour. The gluten proteins are classified based on their electrophoretic mobilities into glutenins and gliadins. Gliadins are monomeric proteins of molecular weight ranges between 30, 000 – 75, 000 Daltons. Gliadins are known to have extensive genotypic polymorphism (Branlard *et al.,* 1993). Based on the electrophoretic mobilities, the gliadins are classified into four different group's alpha (α), beta (β), gamma (γ), and omega (Ω) or (ω) gliadins. The genes coding for these proteins are present on the short arm of chromosome 1 and chromosome 6 of wheat. They are tightly linked genes present on the three homologous loci of chromosome 1 as Gli-A1, Gli-B1 and Gli- D1. In chromosome 6, they are present as Gli-A2, Gli-B2 and Gli-D2 (Wrigley and Shepard, 1973; Brown and Flavell, 1981). The Gli-1 genes code for the ω and γ-gliadins and the Gli-2 genes code for α and β-gliadins. The Gli-1 locus is tightly linked to the LMW glutenin loci Glu-3 in chromosome1. The gliadin

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loci are known to be inherited in simple Mendelian inheritance and multiple-allelism has been reported for both the Gli-1 and Gli-2 loci (Metakovsky *et al*., 1984; Metakovsky, 1991).

Gliadin components have high content of glutamine amino acid. For instance ω-gliadins have more than 50% glutamine (Lasztity, 1984). The proline content is also high in gliadins exceeding what found in HMW glutenins. The high proline content plays an important role in the secondary structure of gliadins (Makarenko *et al*., 2002).

However, gliadins are poor in other basic amino acids such as lysine, arginine and histidine (Kasarda *et al*., 1974). The determination of the N-terminal sequence of the gliadins has supported the theory that a large number of gliadins is due to a genetic mutation of a common precursor during natural evolution of bread wheat. Most of α, β-gliadins and some of the γ gliadins have similar N-terminal sequence. The α, β, γ-gliadins have six to eight cysteine residues, as a result, three to four intramolecular disulphide bonds occur (Kasarda *et al.,* 1984). Although the HMW and LMW glutenins form the disulphide cross-linked gluten matrix a small proportion (5-10%) of α and γ-gliadins occupy the matrix cross-link function. The ω gliadins also may take part in the polymer formation (Kukataite *et al.,* 2004). The gliadins, like the LMW glutenin, can also acts as the chain terminators. The gliadins are generally contributed to the viscosity and extensibility of the dough. The glutenin: gliadin ratio is thought to be an important determinant of quality. The higher glutenin to gliadin ratio is shown to contribute to greater dough strength (MacRitchie, 1985). Another study suggests that an increase in the relative gliadin content is associated with increase extensibility and loss of dough strength (Edwards *et al.,* 2001). Glutenin enrichment can circumvent this and increase dough strength. Although gliadins have been associated to certain parameters of bread making quality, they are considered unimportant to dough strength (Gianibelli *et al.,* 2001). Changes in the glutenin to gliadin ratios modify the bread making quality. In the previous studies deletions in the Gli-1 loci were found to exhibit greater dough strength (MacRitchie & Lafiandra 2001). This study aimed to examine the variations of the gliadin, albumin and globulin patterns in wheat varieties.

2. Materials and Methods

2.1. Plant Samples

One hundred gram of grains of each wheat varieties (bread wheat: Costantino, Tammuz, Aras, Rabia, Cham 4, Cham 6 and durum wheat: Creso kurde, Creso Italy, Ovanto, Bakrajo 1, Acsad 65, Cemmitto) were collected from the department of Agriculture of Sulaimanyah. All varieties used in this study are genetically pure.

2.2. Gliadin Extraction and Electrophoresis

Gliadin patterns were determined by A-PAGE. Twenty gram of wheat grains was hammer-milled to a fine powder. Forty mg of flour was mixed with 200 µl of 60% (v/v) aqueous ethanol. The mixture was incubated in a closed microtube for 2h at 60°C and centrifuged for 10 min, at 14000 rpm. The supernatant was transferred to a microtube and diluted with 1 ml of acetone. The mixture was incubated for 10 min at RT. After centrifugation for 10 min at 14000 rpm, the pellet was resuspended in buffer A (30% Glycerol, 6 M Urea, 25 mM acetic acid, 0.01% w/v pyronin) (Sewa *et al.,* 2005).

To polymerize the gel, one drop (about 200 µl) of fresh 30% H_2O_2 was added to 75 ml of cold gel solution (8.3% acrylamide, 0.415% N,N'-methylenebis-acrylamide, 0.1% ascorbic acid, 0.00067% $Fe_2(SO_4)_3$ in 5.1 mM Allactic buffer, pH 3.1). Each sample (30 µl) was loaded onto a gel sized 180 (length) X 140 (width) X 2 (thickness) mm. Electrophoresis from the anode (the upper buffer) to the cathode (the lower buffer) was performed at 500 V for about 4 h until the tracking dye, pyronin, passed to the gel bottom. The gel was immersed into the staining solution containing 0.05% Coomassie Brilliant Blue R-250 (w/v) and 8% trichloroacetic acid for 5–16 h, and distained with distilled water (Metakovsky, 1991).

2.3. Albumin Extraction and Electrophoresis

Twenty gram of wheat grains was hammer-milled to a fine powder. Forty mg of flour was mixed with 400 µl of 0.5 M NaCl. The mixture was incubated in a closed microtube for 2h at 4°C, and centrifuged at 14000 rpm for 10 min; the supernatant was precipitated in 1 ml acetone. The pellet was suspended in solution [28.5% sample buffer (7% SDS, Tris-HCl 0.01 M, pH 6.8, 30% glycerol, 0.001% comassie bleu) and 5% 2-mercaptoethanol]. The samples were incubated at 80°C for 20 min before loading. Samples of 30 µl were loaded each the slots of SDS-PAGE. The gel consisted of a 15% separating gel, pH 8.4, beneath a 3% stacking gel, pH 6.8. Electrophoresis was carried out at room 27°C using a locally-made vertical electrophoresis apparatus. The running was performed at 15 mA/gel for 18 hours. Subsequently, the gels were stained by immersion in a solution consisting of 12.5% (w/v) trichloroacetic acid, 0.01% (w/v) Coomasie Brilliant Blue R250 and distained with distilled water (Tanaka *et al.,* 2003).

3. Data Analysis

Electrophoregrams for each variety of wheat were scored and the presence (1) or absence (0) of each bands noted. Presence and absence of bands were entered in a binary data matrix. Analysis was carried out using a statistical package SPSS-PC, version 15, by using the Dice similarity.

4. Results and Discussion

4.1. Variation in Gliadin pattern

4.1.1. Bread wheat

Electrophoretic separation of gliadin components exists in the bread wheat material (Figure 1). The Acid-PAGE analysis showed alpha, beta, gamma and omega namely three classes of gliadins. According to the nomenclature of Bushuk and Zillman (1978), the reference Costantino (reference variety) possessed omega gliadin Ω-35, 36, 38 (Table 1). Cham 4, Cham 6 and Rabia possessed only omega gliadin Ω-36. Aras possessed omega gliadin Ω-35

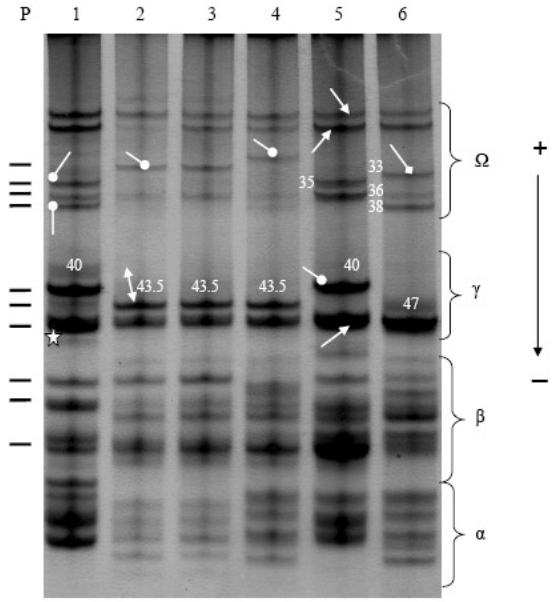

Figure 1. Electrophorogram separation of gliadin components in bread wheat varieties. 1: Costantino; 2: Cham 6; 3: Cham 4; 4: Rabia; 5: Aras; 6: Tammuz. The subunits of gliadins are designed according to Metakovsky, 1991, Bushuk and Zillman, 1978. Arrows correspond to the protein encoded by Gli-D1, oval arrow refers to the protein encoded by Gli-B1, diamond arrow refers to the protein encoded by Gli-A3, star refers to the protein encoded by Gli-A1, double arrows refer to the protein encoded by Gli-B1. P: polymorphic bands.

Table 1. Gliadin subunits composition of six varieties of bread wheat.

Varieties	Omega gliadin (Ω)	Gamma gliadin (γ)
Costantino	35, 36, 38	40
Cham 6	36	43.5
Cham 4	36	43.5
Rabia	36,38	43.5
Aras	35, 36	40
Tammuz	33, 36, 38	47

and 36. On the other hand, Tammuz showed three omega gliadin Ω-33, 36, and 38. According to the nomenclature of Pogna *et al.,* (1995) and Metakovsky (1991), Costantino carries alleles Gli-A1a, Gli-A3a, Gli-B1m, Gli-B3a and Gli-D1k (Table 2). Cham 4 and Cham 6 contain the same alleles in all loci (Table 2). The alleles Gli-B3a are represented in all varieties. The allele composition at the Gli-1 loci of the genotypes analyzed here is reported in Table 1. According to Pagne *et al.,* (1987) the omega gliadins and the D-subunits of glutenin, coded by the Gli-B3 locus, are expressed in low amount and allelic variation is rather limited. Bread wheat Aras and Costantino possess the gamma gliadin component designated 40, whereas Cham 4, Cham 6, and Rabia possess the gamma gliadin 43.5. In addition, Tammuz possesses the gamma gliadin 47 (Table 1). This gamma locus is controlled by the Gli-B1 gene. The studies (Damidaux *et al.,* 1987; Payne, 1984, Joppa *et al.,* 1983) demonstrated the usefulness of gliadin gamma 45 and gamma 42, encoded at Gli-B1 locus as biochemical marker of good and poor pasta quality, respectively. Alpha gliadin patterns varied among the varieties.

Table 2. Allele's composition at the Gli-1 and 3 loci of bread wheat varieties. The alleles are designed According to Pogna *et al*., 1995 and Metakovsky *et al*., 1984.

Varieries	Gli-A1	Gli-A3	Gli-B1	Gli-B3	Gli-D1
Costantino	a	a	m	a	k
Cham 6	m	b	b	a	b
Cham 4	m	b	b	a	b
Rabia	b	b	c	a	b
Aras	b	b	d	a	k
Tammuz	m	a	e	a	k

The viscoelastic properties of gluten in both durum wheat and common wheat are influenced each of gliadin and glutenin patterns. Significant associations have been also detected between wheat quality and specific gliadin components (Damidaux *et al.,* 1987; Pogna *et al.,* 1982; Sozinov and Poperelya, 1980). Pagne *et al.,* (1987) and Pogna et al., (1990) suggested that allelic variation at the Glu-3 loci coding for LMW subunits of glutenin is probably responsible for differences in gluten quality, previously thought to be associated with the closely linked Gli-1 loci coding for gliadins. However, alleles at Gli-1 maintain their interest as genetic markers of quality. Dachevitch T. *et al.,* (1993) showed the genetic background of gliadins coded by the group 1 chromosomes in bread wheat Costantino. The authors identified some alleles coded to gliadins subunits in Costantino.

Result of Acid-PAGE also revealed the variations among the varieties of wheat according to the beta gliadin patterns.

4.1.2. Durum wheat

The Acid-PAGE analysis showed four classes of gliadins namely alpha, beta, gamma and omega (Figure 2). The varieties contain two types of omega: 33 and 35. Only Creso Kurde contained omega 33 and 35 and the rest of varieties possessed omega 35 (Table 3). On the other hand, the acid-PAGE showed one type of gamma gliadin: 45 which is considered as a good marker for pasta making quality (Pogna, 1990). On the other hand, the polymorphism of omega and gamma gliadin is revealed by one and two dimensional gel (Nieto-Taladriz, 1993). Similar studies (Damidaux *et al.,* 1987; Payne, 1984; Joppa *et al*., 1983) demonstrated the usefulness of gliadin gamma 45 and gamma 42, encoded at Gli-B1 locus as biochemical marker of good and poor pasta quality, respectively. The viscoelastic properties of gluten in both durum and common wheat are influenced each of gliadin and glutenin patterns. Significant associations were also detected between wheat quality and specific gliadin components (Damidaux *et al.,* 1987; Pogna *et al.,* 1982; Sozinov and Poperelya, 1980). Pagne *et al*., (1987) and Pogna *et al*., (1990) suggested that allelic variation at the Glu-3 loci coding for LMW subunits of glutenin is probably responsible for differences in gluten quality, previously thought to be associated with the closely linked Gli-1 loci coding for gliadins.

In this study, Acid-PAGE of grain storage proteins gliadin was performed in order to analyze molecular weight of gliadin subunits and investigate genetic diversity

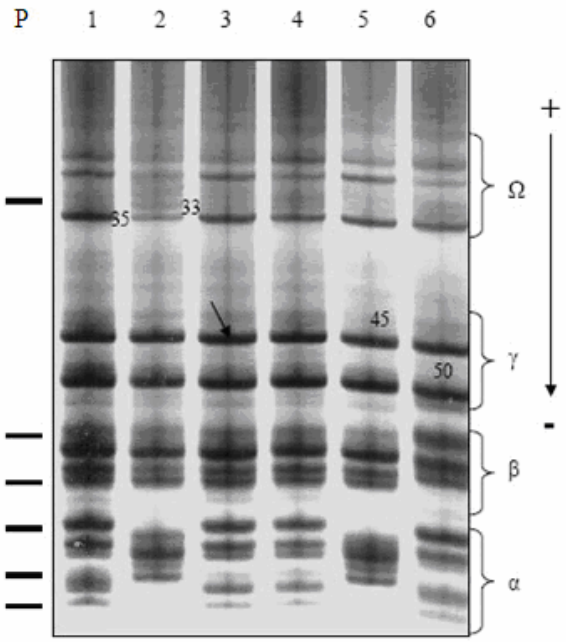

Figure 2. Electrophorogram showing banding pattern of durum wheat protein (Gliadin). 1: Creso Italy; 2: Creso Kurde; 3: Cemmitto; 4: Acsad 65; 5: Ovanto; 6: Bakrajo 1. The subunits of gliadins are designed according to Pogna *et al*. 1990. Arrow corresponds to the protein encoded by locus Gli-B1-2. P: Polymorphic bands.

Table 3. Omega and gamma gliadin subunits composition and locus of gliadin of durum wheat varieties.

Varieties	Omega gliadin	Gamma gliadin	Locus
Creso Italy	35	γ-45 and 50	Gli-B1-2
Creso Kurde	33, 35	γ-45 and 50	Gli-B1-2
Cemmitto	35	γ-45 and 50	Gli-B1-2
Acsad 65	35	γ-45 and 50	Gli-B1-2
Ovanto	35	γ-45 and 50	Gli-B1-2
Bakrajo 1	35	γ-45 and 50	Gli-B1-2

Among different wheat varieties.The electrophorograms showing proteins banding pattern of different wheat varieties are given in figure 2. Eighty four bands were obtained among which 6 bands showed variation, but the other bands were common in all varieties. The results revealed that Ovanto showed only one present band (Figure 2). At low molecular weight of gliadin (alpha gliadin), there are three bands polymorphic, reflecting more diversity.

4.2. Genetic Diversity and Cluster Analysis

4.2.1. Bread wheat

The electrophorogram proteins banding pattern, for seeds of all varieties of wheat detected by Acid-PAGE (Figure 1), showed segregation for gliadin.

Thirty four bands were obtained, and 11 bands were showed variations, but the other bands were common in all varieties. Cluster analysis of wheat grain storage proteins was carried out, depending on the results of Acid-PAGE and using the UPGMA analysis to find the diversity among the given varieties of wheat as shown in the dendrogram (Figure 3). The diagram indicated five main groups: group 1= Cham 6 and Cham 4, group 2= Rabia, group 3= Tammuz, group 4= Costantino, group 5= Aras. At Dice dissimilarity of distance 1, Cham 6 and Cham 4 showed more similarity than others varieties.

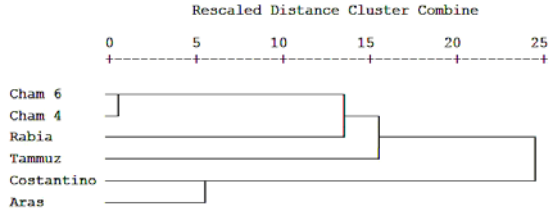

Figure 3. Dendrogram of bread wheat varieties showing the dissimilarity among the varieties based on A-PAGE- Gliadin.

At distance 13, there are 3 groups: group 1= Cham 6, Cham 4, Rabia, group 2= Tammuz, group 3= Costantino and Aras. At distance 17, Cham 6, Cham 4, Rabia and Tammuz are in the group 1 and Costantino and Aras are in the group 2. At dissimilarity distance 24 the diversity divides into two groups: group 1= Cham 6, Cham 4, Rabia and Tammuz and group 2= Costantino and Aras. Costantino and Aras showed more dissimilarity distance with the rest of the varieties.

4.2.2. Durum wheat

Dendrogram cluster analysis of wheat grain storage proteins (Gliadins) resulted from Acid-PAGE (Figure 2), using the UPGMA analysis, showed the diversity among the given varieties on the bases of dissimilarity distance of Dice (Figure 4). The diagram revealed four main groups: group 1= Creso Kurde and Ovanto, group 2= Cemmitto and Ascad 65, group 3= Bakarajo 1, group 4= Creso Italy. At Dice dissimilarity of distance 1, the varieties Creso Kurde and Ovanto and the varieties Cemmitto and Ascad 65 showed more similarity than others varieties.

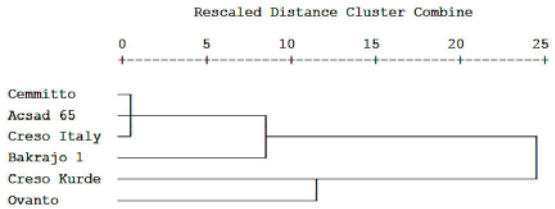

Figure 4. Dendrogram of durum wheat varieties showing the dissimilarity among the varieties based on A-PAGE- Gliadin.

At distance 5, there are 3 groups: group 1= Creso Kurde and Ovanto, group 2= Cemmitto, Ascad 65 and Bakarajo 1, group 3= Creso Italy. At distance 15, the varieties Creso Kurde and Ovanto are in the group 1 and the varieties Cemmitto, Ascad 65, Bakrajo 1 and Creso Italy are in the group 2. At dissimilarity distance 22, the diversity divides into two groups: group 1= Creso Kurde and Ovanto, group 2= Cemmitto, Ascad 65, Bakarajo 1, Creso Italy. The varieties Creso Kurde and Ovanto showed more dissimilarity distance with the rest of the varieties.

The presence of some patterns may correlate with a higher adaptive value of germplasm to the particular environment (Metakovsky *et al.,* 1991; Sewa *et al*., 2005). The association between genotypes of wheat and their

environment has also been reported by Nevo *et al.,* (1988, 1995). However, no significant relationship between genotypes of wheat and their environments was reported (Dreisigacker *et al.,* 2004). Different combinations of gliadin patterns were prevalent in different regions, suggesting the adaptive properties of individual alleles or the chromosome segments, in which these alleles reside (Nevo *et al.,* 1995; Metakovsky and Branlard, 1998). Different gliadins might have some advantage over other gliadins in adaptation to the conditions, prevailing in these zones or these are closely linked with genes having adaptive values to the specific environment, though that needs to be confirmed by genetic analysis (Sewa *et al.* 2005).

4.3. Variation at The Albumin and Globulin Subunit

4.3.1. Bread wheat

Albumins and globulins were characterized by rich protein pattern. The numbers of band varied from 13 to 15, and they were defined by molecular weight 106–2 kDa (Figure 5). The protein pattern of albumins and globulins was divided into two relatively wide areas 66–23 kDa and 16–2 kDa. The electrophoretic pattern of tested varieties showed high similarity. The differentiating areas were located at the four zones with molecular weights of 92–87 kDa, 68–58 kDa, 49–39 kDa and 10-4 kDa for the 1st, 2nd , 3rd and the 4th zone, respectively**.**

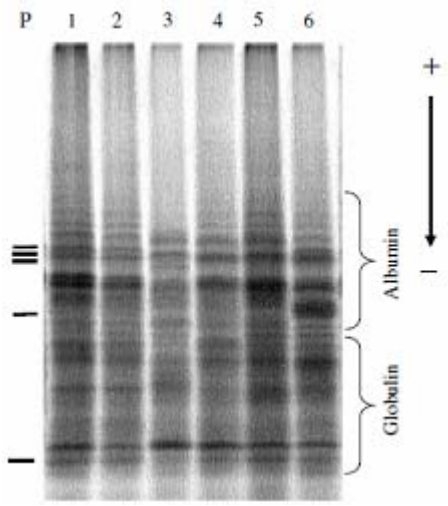

Figure 5. Electrophorogram showing banding patterns of bread wheat protein soluble (Albumin and globulin). 1: Costantino; 2: Cham 6; 3: Cham 4; 4: Rabia; 5: Aras; 6: Tammuz. P: polymorphic bands.

The similarity matrix and the resulting dendrogram (Figure 6) characterized similarity coefficients of the tested varieties. Although the results showed high similarity, the tested varieties were firmly identified. Tammuz possesses the five bands polymorphic, and Cham 4 possesses only one subunit. The highest similarity was found between Rabia and Aras. On the other hand, the lowest value of the similarity coefficient was found between Cham 4 and the rest of varieties**.** The diversity, in high molecular weight protein subunits, is the result of gene silencing in some varieties encoding these proteins (Lawrence and shephred, 1980).

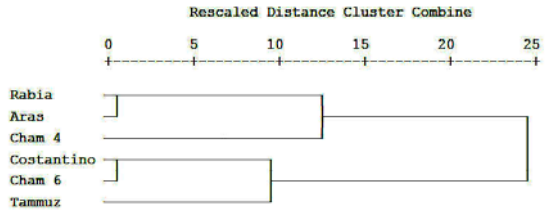

Figure 6. Dendrogram of bread wheat varieties showing the dissimilarity among the varieties based on SDS-PAGE- Albumin and globulin.

4.3.2. Durum wheat

Albumins and globulins were characterized in durum wheat by SDS-PAGE (Figure 7). The differentiating areas were located to zone: 40-60kDa. The SDS-PAGE gel did not show any difference among the varieties at the level globulin. The number of bands present is similar for all varieties.

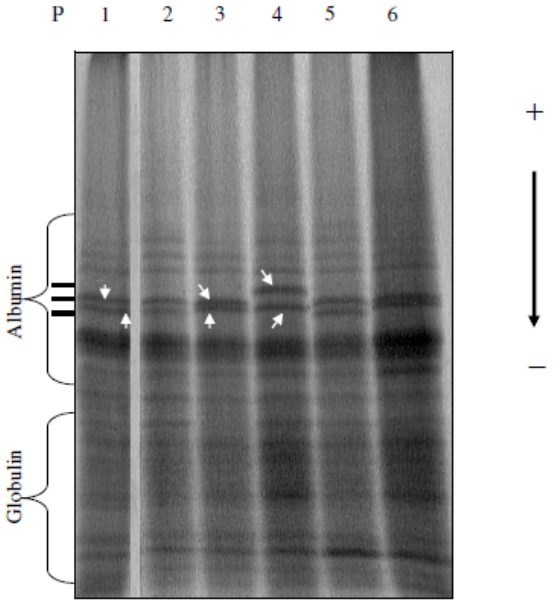

Figure 7. Electrophorogram showing banding pattern of durum wheat protein soluble (Albumin and globulin).1: Bakrajo 1; 2: Acsad 65; 3: Creso Kurde; 4: Creso Italy; 5: Cemmitto; 6: Ovanto, P: Polymorphic bands.

The similarity matrix and the resulting dendrogram (Figure 8) characterised similarity coefficients of the tested varieties. The highest similarity was found between Creso Kurde and Ovanto and also for Acsad 65, Cemmitto and Bakrajo 1 (Figure 8). On the other hand, the lowest value of the similarity coefficient was found between the group which contains Creso Kurde, Ovanto and Creso Italy and the group containing Acsad 65, Cemmitto and Bakrajo 1.

The seed proteins are very suitable and useful genetic markers, because they are not influenced by external conditions, and so they enable to identify tested plant genotypes (Černý and Šašek, 1996). The finding predicates higher level of the intra-varieties polymorphism in these cultivars. Patterns of albumins and globulins among varieties were very similar and their characters corresponded with the detection by American authors, who used water dissolved proteins for identification of two common wheats in non-reduction conditions (Kim and Bushuk, 1995). Apart from the gluten proteins, water-

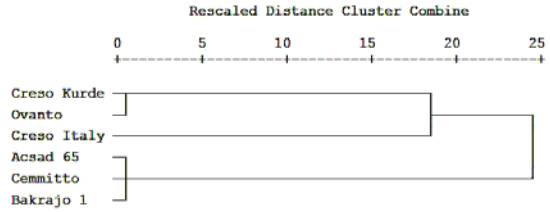

Figure 8. Dendrogram of durum wheat varieties showing the dissimilarity among the varieties based on SDS-PAGE- Albumin and globulin.

soluble albumins and salt-soluble globulins constitute from 10 to 22% of total flour protein (Singh and MacRitchie, 2001a). Albumins such as α-amylase/trypsin inhibitors (Buonocore *et al.,* 1985; Shewry *et al.,* 1984) and purotionins (Garcia- Olmedo *et al.,* 2002) may have dual roles as nutrient reserves for the germinating embryo and as inhibitors of insects and fungal pathogens prior to germination. Puroindolines influence grain hardness (Morris, 2002). Generally, albumins and globulins are not thought to play a critical role in flour quality, although, minor importance on bread-making quality has been reported (Schofield and Booth, 1983). Both protein fractions are important from nutritional point, because of rather high amounts of essential amino acids. This experience is confirmed with the comparison of polymorphism among water soluble protein (albumins and globulins) and seed storage proteins (gliadins) of Dvořáček *et al*. (2001a).

In conclusion, the results showed that the gamma gliadin 47 which is linked to the strong quality of wheat for baking, is less frequent in the varieties tested. The two marker systems were analysed for a complex similarity evaluation of tested varieties. On the other hand, the gliadin pattern revealed more high level of polymorphisms than the albumin and globulin pattern. Identification of gliadins alleles in wheat varieties in Kurdistan may be useful for selection aims in breeding programs to determine the relationship between gluten viscoelastic properties and allelic variations at Gli-1, Glu-3 and Glu-3 in wheat varieties.

Acknowledgements

The author thanks the agriculture department of Sulaimanyh for providing the seeds and LNCV for supporting the work and Unita di ricerca per la maiscoltura, Via stezzano, 24, Bergamo, Italy especially Dr. Rita Redaelli for help and for supporting the work.

References

Branlard G, Dardevet M, Nieto-Taladriz MT, and Khelife D. 1993. Allelic diversity of omega gliadins as revealed by SDS-PAGE. Their possible implication in quality variation. Gluten 93:234–243.

Brown JWS and Flavell RB. 1981. Fractionation of wheat gliadin and glutenin subunits by two-dimensional electrophoresis and the role of group 6 and group 2 chromosomes in gliadin synthesis. Theor. Appl. Genet. 59: 349-359.

Buonocore V, De Biasi MG, Giardina P, Poerio E, Silano V. 1985. Purification and properties of an alfa-amylase tetrametric inhibitor from wheat kernel. Biochimica et Biophysica Acta. 831: 40-48.

Bushuk W and Zillman RR. 1978. Wheat cultivars identification by gliadin electrophoregrams. Apparatus, method and nomenclature. Can. J. Plant. Sci. 58: 505-515.

Černý J, Šašek A. 1996. Bílkovinné signální geny pšenice obecné. ÚZPI, Praha: 1–66.

Dachkevitch T, Redaelli R, Biancardi AM, Metakovsky EV and Pogna NE. 1993. Genetics of gliadins coded by the group 1 chromosomes in the high-quality bread wheat cultivar Neepawa. Theor. Apple Genet. 86(2-3): 389-399.

Damidaux R, Autran JC, Grignac P and Feillet P. 1987. Mise en evidence de realtions aplicables en selection entre l electrophoregramme des gliadines et les proprietes viscoelastiques du gluten de Triticum durum. Desf. C.R. Acad. Su. Ser. D. 287:701.

Dreisigacker S, Zhang PM, Warburton L, Van Grinkel M, Hoisington D, Bohn M and Melchinger AE. 2004. SSR and pedigree analyses of genetic diversity among CIMMYT wheat lines targeted to different megaenvironments. Crop Sci 44: 381-388.

Dvořáček V, Čurn V, Moudrý J. 2001a. Methodological approach to evaluation of biochemical markers in spelt wheat (*Triticum spelta* L.). Scientia Agric. Bohem. 32:171–182.

Edwards NM, Dexterl JE, and Scanlon MG. 2001. The use of rheological techniques to elucidate durum wheat dough strength properties. The sixth Italian Conference on Chemical and Process Engineering. 2:825-830.

Garcia-Olmedo F, Carmona MJ, Lopez-Fando JJ, Fernandez JA, Castagnaro A, Molina A, Hernandez-Lucas C, Carbonero P. 2002. Characterization and analysis of thionin genes. In Plant Gene Research. Genes Involved in Plant Defense. (New York, Springer) 283-302 pp.

Gianibelli MC, Larroque OR, MacRitchie F and Wrigley CW. 2001. Biochemical, genetic and molecular characterization of wheat glutenin and its component subunits. Cereal Chem. 78(6): 635-646.

Joppa LR, Khan K and Williams ND. 1983. Chromosomal location of genes for gliadin polypeptides in durum wheat *Triticum turgidum* L. Theor. Appl. Genet., 64: 289-293.

Kasarda DD, Da-Roza DA and Ohms JI. 1974. N-terminal sequence of alpha 2-gliadin. Biochem Biophys Acta. 209: 154-162.

Kasarda DD, Lafiandra D, Morris R and Shwery PR. 1984. Genetic relationships of wheat gliadin proteins. in: Proc 3rd Symp. Seed Proteins. Kulturpflanze. 32: 41-61.

Kim HR, Bushuk W. 1995. Changes in some physicochemical properties of flour proteins due to partial reduction with dithiotreitol. Cereal Chem. 72: 450–456.

Kukataite R, Larsson H and Johansson E. 2004. Variation in protein composition of wheat flour and its relationship to dough mixing behavior. J. Cereal Sci. 40:31-39.

Laszitity R. 1984. The chemistry of cereal proteins. CRC Press: Boca Raton, Fl, U.S.A.

Lawrence GJ, Shepherd KW. 1980. Variation in glutenin protein Subunits of wheat. Aust. J. Biol. Sci. 33: 221-233.

MacRitchie F. 1985. Studies of the methodology for fractionation and reconstitution of wheat flours. J. Cereal Sci. 3: 221-230.

MacRitchie F and Lafiandra D. 2001. The use of near-isogenic wheat lines to determine protein composition-functionality relationship. Cereal Chem. 78(5): 501-506.

Makarenko SP, Trufanov VA and Putilina TE. 2002. Infrared Spectroscopic Study of the Secondary Structure of Wheat, Rye, and Barley Prolamins. Russ. J. Plant Physiol. 49:1608-3407.

Metakovsky EV, Novoselskaya AY and Sozinov AA. 1984. Genetic analysis of gliadin components in winter wheat using two-dimensional polyacrylamide gel-electrophoresis. Theor. Appl. Genet. 69: 31-37.

Metakovsky EV.1991. Gliadin allele identification in common wheat II. Catalogue of gliadin alleles in common wheat. J. Genet. Breed. 45: 325-344.

Metakovsky EV and Branlard G. 1998. Genetic diversity of French common wheat germplasm based on gliadin alleles. Theor. Appl. Genet. 96:209–218.

Morris CF. 2002. Puroindolines: the molecular genetic basis of wheat grain hardness. Plant Molecular Biology 48, 633-647.

Nevo E, Beiles A and Krugman T. 1988. Natural selection of allozyme polymorphisms: A microgeographic differentiation by edaphic, topographical and temporal factors in wild emmer wheat (*Triticum dicoccoides*). Theor. Appl. Genet. 76:737–752.

Nevo E, Pagnotta MA, Beiles A and Porceddu E. 1995. Wheat storage proteins: Glutenin diversity in wild emmer, *Triticum dicoc*and *coides*, in Israel and Turkey. 3. Environmental correlates and allo- zymic associations. Theor. Appl. Genet. 91:415–420.

Nieto-Taladriz MT, Perretant MR and Rousset M. 1993. Effect of gliadins and HMW and LMW subunits of glutenin on dough properties in the F_6 recombinant inbred lines from a bread wheat cross. Theor Appl Genet. 88: 81-88.

Payne PI, Jackson EA, Holt LM and Law CN. 1984. Genetic linkage between endosperm storage protein genes on each of the short arms of chromosomes 1A and 1B in wheat. Theor. Appl.Genet. 67: 235.243.

Payne PI. 1987. Genetics of wheat storage protein and the effect of allelic variation on bread making quality. Ann. Rev. Plant Phys-iol. 38:141–153.

Pogna NE, Redaelli R, Vaccino P, Biancardi AM, Dal Belin Peruffo A, Curioni A, Metakovsky EV and Pagliaricci S. 1995. Production and genetic characterization of near-isogenic lines in the bread-wheat cultivar Alpe. Theor. Appl. Genet. 90:650-658.

Pogna NE, Boggini G, Corbellini M, Cattaneo M, Peruffo, ADB. 1982. Association between gliadin electrophoretic bands and quality in common wheat. Can. J. Plant Sci. 4:913–918.

Pogna NE, Autran JC, Mellini F, Lafiandra D, Feillet P. 1990. Chromosome 1B-encoded gliadins and glutenin subunits in durum wheat: genetics and relationship to gluten strength. J. Cereal Sci. 11: 15-34.

Schofield JD. 1983. Wheat proteins and their technological significance. *In* Developments in food proteins, Hudson, B. J. F., ed. (London, Applied Science Publishers), 1-65 pp.

Sewa R, Nisha J, Vinamrata D, Singh RP and Shoran Jag. 2005. Analyses of Acid-PAGE Gliadin Pattern of Indian Wheats (*Triticum aestivum* L.) Representing Different Environments and Periods. Crop Sci. 45:1256-1263.

Shewry PR, Lafiandra D, Salsedo G, Aragoncillo C, Garcia-Omedo F, Lew E.J.L., Dietler MD, Kasarda DD. 1984. N-terminal amino acid sequences of chloroform/methanol-soluble proteins and albumins from endosperms of wheat, barley and related species. Federation of European Biological Societies Letters 175.

Singh H and MacRitchie F. 2001a. Application of polymer science to properties of gluten. J. Cereal Sci. 33, 231-243.

Sozinov AA and Poperelya FA. 1980**.** Genetic classification of prolamins and its use for plant breeding. Ann. Technol. Agric. 29: 223-245

Tanaka H, Tomita M, Tsujimoto H and Yasumuro Y. 2003. Limited but specific variations of seed storage proteins in Japanese common wheat (*Triticum aestivum* L.). Euphytica. 132: 1573-5060.

Wrigley CW and Shepard KW. 1973. Electrofocusing of grain proteins from wheat genotypes. Ann. N.Y. Acad. Sci. 209: 154-162.