Seed Proteins in Genome Analysis, Cultivar Identification, and Documentation of Cereal Genetic Resources: A Review[1]

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ABSTRACT

The specificity and polymorphism of some endosperm proteins from species of *Triticum, Aegilops, Secale, Hordeum, Agropyron,* and *Avena* were studied by immunochemical methods, polyacrylamide gel electrophoresis (pH 3.1), and isoelectric focusing. The ethanol-soluble proteins of seeds of these species showed a distinct genome specificity. These proteins were used in genome analysis of amphidiploids and in the study of the nature and origin of the genomes of polyploid wheats. Comparative electrophoretic studies of the prolamines led to creation of a standard electrophoretic spectrum and to the proposal of a nomenclature for the prolamine components of a given species or cultivar. This nomenclature provides the possibility for describing any seed sample by a specific "formula" for its proteins. A uniform system for documentation of cereal germ plasm is proposed, which would include registration of genome and protein formulas and would reflect the genus, species, cultivar, and biotype of any given sample.

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A protein is a primary product of a structural gene and serves as a marker for that gene. Because genes are coupled into genetic systems, proteins also may serve as markers for such systems, including chromosomes and the genome as a whole. Thus, the totality of protein markers gives considerable insight into genome or genotype structure (Johnson 1973; Khakimova and Gavrilyuk 1973; Konarev 1973, 1974; Konarev et al 1970, 1971, 1974, 1976b; Konarev and Gubareva 1977; Peneva and Migushova 1973). Consequently, proteins have frequently been used in recent times to resolve actual botanic, genetic, and breeding problems.

Use of proteins as markers is based on two kinds of specificity—antigenic specificity for identification of the relationship of the proteins to plant species and specificity of the electrophoretic spectrum of proteins with multiple forms (polymorphic proteins) for definition of intraspecific relationships. Markers of the first type (serologic markers) provide a means to identify mainly species and genomes, whereas markers of the second type, which are identified largely by electrophoretic methods, may be used for genotype and cultivar identification. Both types of protein markers, serologic and electrophoretically determined, may be used for registration of genetic plant resources. This is particularly important if the genomes and genotypes have characters of commercial value, such as high protein content or good baking quality, that are under complex genetic control.

Protein markers in phylogenetic and genetic investigations must be readily isolated and identified and also must have well-defined antigenic or polymorphic specificity. According to their specificities, protein markers may be divided into three groups as shown in Table I. Among cereal seed proteins, we found those of the alcohol-soluble fraction to be most suitable for identification of genomes and genotypes. This fraction consists of prolamine and nonprolamine proteins; the latter are apparently albumins and globulins.

For genome identification and analysis of wheat and its relatives, we used the nongliadin proteins of the alcohol-soluble fraction. These proteins possess high antigenic activity and distinct genome

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specificity. They are represented by many protein components that are controlled by a number of different chromosomes (Aragoncillo et al 1975, Waines 1973). Accordingly, these nongliadin proteins, as a group, serve as effective markers of species and genomes.

By means of serologic markers based on nongliadin proteins, we were able to define two types of the first wheat genome—genome A^b from *Triticum boeoticum* or *T. monococcum* and genome A^u from *T. urartu* or its close relatives (Konarev et al 1971, 1974, 1976b). Genome A^b is typical of the timopheevi group, which includes *T. timopheevi, T. araraticum, T. dicoccoides* from Iraq, *T. zhukovskyii, T. timonovum,* and *T. fungicidum.* Genome A^u is characteristic of the turgidum group, which includes *T. turgidum, T. dicoccoides* from Syria, *T. dicoccum,* and *T. durum.*

The second genome of timopheevi wheat was identified as genome B^{sp} from *Aegilops speltoides*, whereas the second genome of turgidum group wheats was genome B^l from *Ae. longissima* (Konarev et al 1976b, Peneva and Migushova 1973). Genome D of hexaploid bread wheats (*T. aestivum*) was related closely to the genome of *Ae. squarrosa* ssp. *strangulata* and was designated D^{st} (Khakimova and Gavrilyuk 1973, Konarev et al 1976b).

Comparative immunochemical analysis of numerous representatives of species of wheat (*Triticum*) and of *Aegilops* enabled us to describe the evolutionary groups of polyploid wheats mentioned according to the following genome formulas: timopheevi group—$A^bA^bB^{sp}B^{sp}$; turgidum group—$A^uA^uB^lB^l$; and bread wheats—$A^uA^uB^lB^lD^{st}D^{st}$. These conclusions also are supported by other works (Aniol 1976, Chapman et al 1976, Chen et al 1976, Kimber 1974, Vitozzi and Silano 1976). Figure 1 summarizes the genome formulas and relationships of the various evolutionary groups and species. These formulas are characteristic generally of the groups, but within an evolutionary group a genome may sometimes be different for accessions that are apparently of the same species (Konarev et al 1970, 1971; Konarev and Gubareva 1977).

In connection with the use of nongliadin proteins to identify wheat genomes, we wish to emphasize that the electrophoretic pattern of these proteins is specific for a given species, but it does not always serve as a marker for the genome composition of the species. In some cases, electrophoretic patterns or particular bands of the patterns do not correspond to the serologic determinants characteristic of the genome or genomes. For example, presence of a particular albumin in the electrophoretic pattern of *T. urartu* and its absence in *T. boeoticum* and *T. monococcum* were taken by Johnson (1973) as an argument for the conclusion that the B genome of emmer wheat (turgidum group) was derived from *T. urartu*. However, the antigens of this albumin were found immunochemically in timopheevi wheats and in the *Sitopsis* group of *Aegilops* (Konarev et al 1976b), the latter group probably contributed genome B to polyploid wheats of the turgidum group. Also, the main albumin (Alb. 0.19) of bread wheats is absent from the electrophoretic pattern of durum wheats, but immunochemical examination of durum wheats shows the antigens characteristic of Alb. 0.19 (Konarev 1978). The specificity of electrophoretic patterns for species, but not for genomes, may be due to differences in electrophoretic mobilities resulting from differences in structural state (oligomers) or other polypeptide modifications that do not necessarily change the antigenic character of the proteins.

A discrepancy between the immunochemical and electrophoretic identifications of proteins may take the reverse form from the example above. The electrophoretic mobility of the slowest main band of the nongliadin proteins is common to all forms of einkorns (diploid species of *Triticum: T. urartu, T. boeoticum, T. monococcum*), but the proteins corresponding to this band are not identical when examined immunochemically. The antigens of this component are genome-specific corresponding to either the A^u or the A^b genomes (Fig. 2a,b,c).

Genome specific antigens are located in several zones of the

TABLE I
Protein Specificity and Its Application in Phylogenetic and Genetic Plant Analysis

Genus, Tribe, Family	Genus, Species, ssp., Genomes	Cultivar, Biotypes, Mutants
Evolutionarily conservative proteins with ancient functions—histones, cytochromes, etc.	Evolutionarily young proteins—storage proteins, some enzymes, etc.	Genetic polymorphic proteins—isoenzymes, prolamines, polypeptides of 7S and 11S globulins, etc.
Markers of higher taxons	Markers of species and genomes	Markers of alleles and allelic structure of genes
Phylogenetic and genome analysis		Genetic analysis

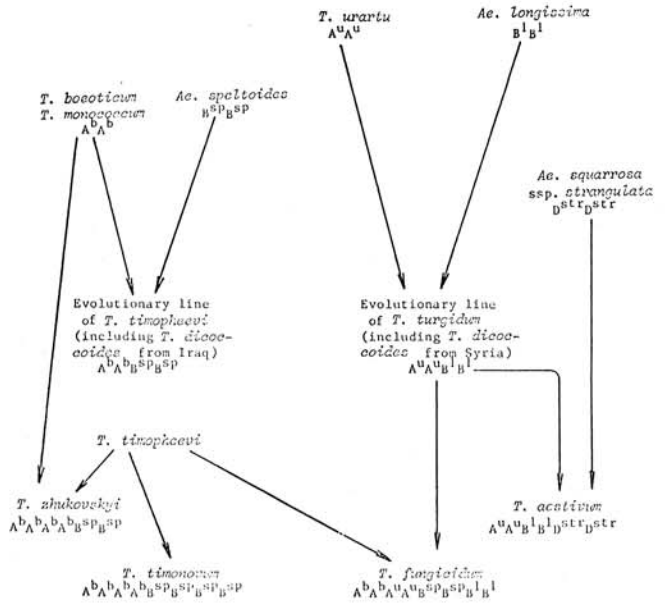

Fig. 1. Origin of polyploid wheat genomes based on the data of genome analysis by protein markers.

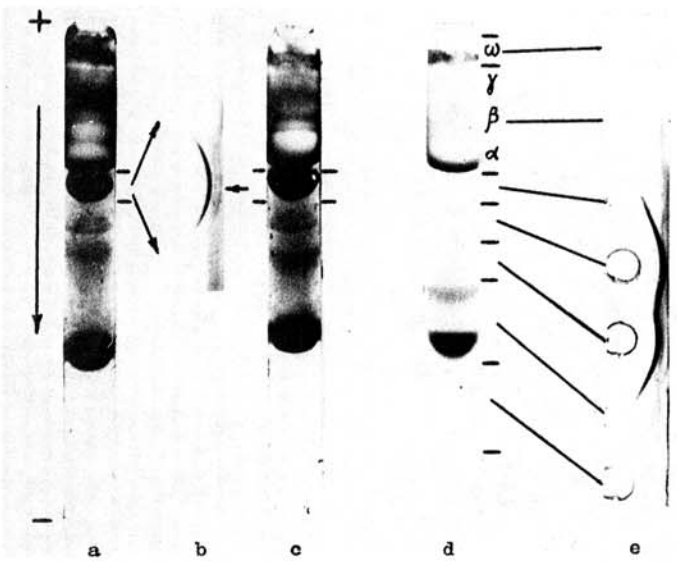

Fig. 2. Immunochemical analysis of separate electrophoretic zones of ethanol-soluble proteins of (**a**) *Triticum urartu*, (**c**) *T. boeoticum*, and (**d**) *T. durum* with antisera against (**b**) *T. boeoticum* and (**e**) *Aegilops longissima*.

electrophoretic patterns and are associated with different proteins of the nongliadin fraction (Fig. 2d,e). This contributes to the high degree of genome specificity of the serologic method, which makes it a more reliable marker of genomes and species. Further investigations of nongliadin proteins probably will lead to the discovery of intraspecies differences as well.

To understand intraspecies differences in terms of the proteins, it is necessary to understand the genetic variability of these proteins as manifested in a variety of molecular forms. The best methods for elucidating the nature of these forms are electrophoresis and isoelectric focusing. The differences in protein components that have the same function may result from gene variability associated with multiple loci. Alternatively, proteins coded for by genes at the same locus may have different forms (multiple alleles). Molecular forms of proteins resulting from different loci usually are representative of species and these multiple forms may serve as markers of species, genomes, genetic groups of organisms, and separate chromosomes. Variability that results from multiplicity of allelic components, on the other hand, tends to be more specific for cultivars, biotypes, and lines—the molecular forms of this type may be used for cultivar identification. Polymorphism of protein components may result from gene mutations or from quaternary structures composed of associated subunits, or secondary modification of proteins by amidation, deamidation, acetylation, phosphorylation, etc., of amino acid side chains.

Woychik et al (1961) were among the first to adapt gel electrophoresis in aluminum lactate buffer (pH 3.1) to native gliadin proteins. Their pattern for gliadins from Ponca wheat was grouped into four major fractions with subfractions designated as follows: $\alpha_1, \alpha_2, \beta_1, \beta_2, \beta_3, \beta_4, \gamma$, and ω. Subsequently, the γ-fraction was shown to have three subfractions and the ω-gliadin fraction was shown to have eight components (Bietz et al 1970, Charbonnier 1974, Huebner et al 1967). This has been thoroughly substantiated by more recent works. At the same time, the cultivar specificity of gliadins was demonstrated (Cluskey et al 1961, Graham 1963). Eventually, a number of laboratories proposed schemes for nomenclature that could be used with gel electrophoresis to designate a gliadin "formula" for any cultivar (Autran and Bourdet 1975; Günzel 1976; Konarev 1973; Konarev et al 1970, 1975, 1976a; Wrigley and Shepherd 1974). In this article, we will consider the nomenclature proposed by our laboratory (Konarev 1973; Konarev et al 1970, 1975, 1976a).

On the basis of comparative evaluations of electrophoretic spectra of gliadins (7.5% polyacrylamide gels, acetate buffer, pH 3.1) from samples of all the different species of wheat and their diploid relatives, we composed a standard spectrum for all band positions corresponding to the main protein components of gliadin fractions. This standard spectrum is divided into α, β, γ, and ω fractions and subfractions in accord with the nomenclature of Woychik et al (1961) and makes it possible to write a gliadin formula for the electrophoretic pattern of any sample—biotype, cultivar, or line (Fig. 3). For example, the gliadin formula for the cultivar Ponca would be written as follows:

$$\alpha_{567}\,\beta_{23_1 45}\,\gamma_{234}\,\omega_{\bar{2}6\ddot{8}\bar{9}10}.$$

To provide additional flexibility to the standard formula, additional conventions are used to indicate deviations from the standard such as higher intensity (band number underlined), lower intensity (band number overlined), slightly greater mobility (subscript 1 to band number), slightly lesser mobility (subscript 2 to band number), split band or doublet (double mark over band number). The standard spectrum also can be used to write formulas for cultivars and biotypes of *Hordeum* (barley), *Secale* (rye), *Avena* (oats), *Agropyron, Aegilops,* and *Triticale*. Examples of cultivar registration formulas are given in Table II.

The high degree of polymorphism and specificity of gliadin proteins suggests that they must contain a rich supply of genetic and phylogenetic information that has yet to be discovered. The genetic control of gliadins was studied by Shepherd (1968) and

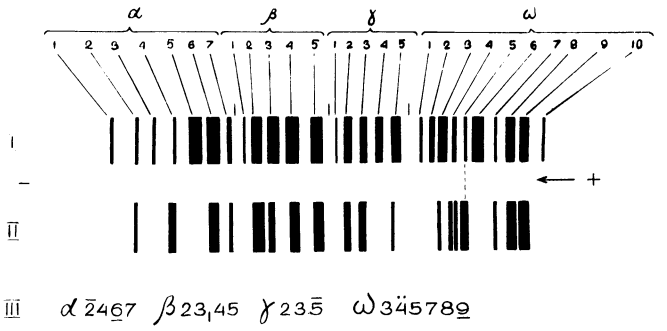

Fig. 3. Standard spectrum of (**I**) gliadin: (**II**) gliadin spectrum of Chinese Spring; and (**III**) gliadin formula of this cultivar.

TABLE II
Protein Formulas of Some Wheat, Barley, and Oat Cultivars

Cultivar	Protein Formula			
	α	β	γ	ω
		Wheat		
Chinese Spring	$\bar{2}\ 4\ \underline{6}\bar{7}$	$23_1 45$	$23\ \ \bar{5}$	$\ddot{3}\underline{4}\ 78\ \underline{9}$
Odesskaya 16	$\bar{2}\ 456\bar{7}$	$\underline{2}\underline{3}_1 45$	$23\ 4$	$2\ 4\ \underline{6}78_1\ddot{9}10$
Bezostaya 1	$\underline{56}7$	$23\ 45$	$23\ 4$	$2\ 4\ \underline{6}\ 8\ \underline{9}$
Charkovskaya 46	$5\underline{6}7$	$23\ 4\underline{5}$	$2\ \ \ 5$	$\underline{3}\bar{4}\bar{5}\underline{6}\bar{7}\underline{8}$
Kubanka	$5\underline{6}$	$23\ 45$	$23_2 4_1 5$	$\underline{3}\ddot{4}\ \ddot{6}7$
		Barley		
Vinner	456	$23\ \underline{5}$	$1\ \ 4\ 5$	$5\underline{6}7$
Tchernomorec	$4\underline{5}\ 7$	$1\ 3\ 45$	$2\ 4$	5678
Iugnii	45	$23\ 4\underline{5}$	$3\ 4$	5678
		Oats		
Zolotoj dojd	$\bar{0}12\bar{3}\ \ 67$	$1\ 3\ \bar{4}$		
Pshebui II	$12\ \ 4\bar{5}$	$\bar{1}\ 3\ 4\bar{5}$		

Wrigley and Shepherd (1973), who used aneuploid lines of the hexapolid wheat cultivar 'Chinese Spring.' They showed that gliadin genes are located on chromosomes of homoeological groups 1 and 6. This work was reproduced and adapted to the gliadin nomenclature described in this paper by Mitrofanova (1976), who studied ditelosomic and compensated nullisomic-tetrasomic lines of Chinese Spring and other genetic variations (lines with replaced or added chromosomes, AABB tetraploids derived from hexaploid cultivars, etc.).

The tabulation of chromosomal control of the synthesis of gliadin protein components (Table III) indicates that chromosomes of homoeologous group 1 control ω-gliadins and slow-moving γ-gliadin components, whereas chromosomes of group 6 control mainly α, β, and fast-moving γ-gliadin components. A similar distribution of chromosomal control may occur in other cereal species (Shepherd 1968, 1973). As a consequence of this localization of genes for gliadin proteins, six groups of gliadin protein components correspond to the 1A, 1B, 1D, 6A, 6B, and 6D chromosomes. These groups must be displayed to serve as markers for those choromosomes; in some cases these chromosomes may be specific for certain cultivars or biotypes. This is easily seen in diploid species, which have a single genome. For example, the ω-gliadin pattern (controlled by the 1D chromosome) of *Ae. squarrosa* is represented by more than 100 variants in the biotypes of this species that have been studied, whereas about 30 genetic variants of the α-gliadin pattern (controlled by the 6D chromosome) were found for these biotypes (Khakimova and Gavrilyuk 1973). Some variations of the electrophoretic gliadin spectra for *Ae. squarrosa* ssp. *strangulata* and ssp. *eusquarrosa* are presented in Table IV. This large number of variations is typical for

TABLE III
Chromosomal Control of Gliadin Components (cv. Chinese Spring)

By Shepherd (1968)		By Mitrofanova (1976)	
Components	Chromosome and Its Arm[a]	Components	Chromosome and Its Arm[a]
1 2	1D(S)	ω 9	1D(S)
3	1D(S)	8	1D(S)
4		7	1D(S)+?
K 1	1A(S)	5	1B(S)
2	0	4	1B(S)+?
3	1B(S)		
4	1B(S)	3	1B(S)
6		γ 5	1A(S)
L 1	1A+1D	3	1A+1D
2	1A+B	2	1B+1D+6B(S)
3a	0	β 5	6B(S)+?
3b	1B	4	6B(S)+?
4	6B+?	3_1	6B(S)
6	0	2	0
7	1A+6A		
M 2	1B(S)	α 7	1B(S)
3	0	6	6D(α)+?
5	6D(α)		
6	6A(α)	4	6A
7	6A(α)	2	6A

[a]0 = Control by three pairs of homoeologous chromosomes is assumed. ? = Control by other chromosomes is assumed.

TABLE IV
Some Biotypes of *Aegilops squarrosa* Registered by Gliadin Spectrum

Catalog No. (VIR)	Gliadin Formulas			
	α	β	γ	ω
		ssp. *strangulata*		
76	$\overline{34}$ $\underline{6_1}7$	12_1 45	$\underline{1}$ $\underline{3\overline{4}}$	$\underline{3}$ $\underline{56\overline{789}}$
110	3 $\underline{6_1 7}$	$\underline{12}$ $\underline{3}$ $\underline{5}$	234	3 $\underline{5}$ $\underline{7}$8910
131	$\overline{3}$ $\underline{6_1\overline{7}}$	$\underline{2}$ $\underline{3}$ 5	$\underline{23}$ 5	$\underline{5678\overline{9}}$
163	$\overline{34}$ 6_1	$\underline{2}$ $\underline{3}$ $\underline{5}$	$\underline{34}$	$\underline{3}$ $\underline{56\overline{789}}$
236	$\overline{3}$ $\underline{6_1 7}$	$\underline{12}$ $\underline{3}$ $\underline{45}$	$\underline{\overline{234}}$	3 $\underline{5\overline{6}}\ddot{\overline{7}}89$
		ssp. *eusquarrosa*		
33	$\overline{3}$	$\underline{12}$ $3_2$45	$\overline{2}\underline{34}$	4 $\ddot{\overline{6}}789$
248	$\overline{23}$	$12_2\underline{3}$ $\underline{45}$	$\underline{1}$ $\underline{3}$	4 $\underline{6}\ddot{7}89$
362	$\overline{3}$	$\underline{12}$ $\underline{3}$ $\underline{45}$	$\underline{23}$	$\underline{6\overline{789}}$
475	$\overline{3}$	$\underline{2}$ $\underline{3}$ $\underline{45}$	$\underline{1}$ 34	$\underline{5678\overline{9}}$
965	$\overline{3}$	$\underline{12}$ 3	$\underline{1}$ $\underline{3}$	$\underline{456}789$

the gliadin patterns of other related species, including the cereals, and the variation provides a means to identify biotypes and cultivars and to discover heterogeneity within cultivars. We have found that some wheat cultivars are mixtures of two or more genotypes, which can be identified through their gliadin electrophoretic spectra. (Table V).

The gliadin spectrum is useful also for discovering the intracultivar heterogeneity (or heterozygosity) of cross-pollinated crops such as rye. The genetic makeup of four different rye cultivars as represented by their gliadin formulas is shown in Table VI. Rye cultivars consist of mixtures of genotypes many of which are commonly shared. The cultivars differ from one another by the predominance of one or more particular genotypes, as illustrated in Table VI where the proportion of seeds displaying a particular genotype is given for four different rye cultivars. The various rye genotypes reflected in the gliadin spectra may be linked to characters of biological and commercial importance. Accordingly, registration of cultivars can be helpful in selection and seed production of cross-pollinated crops.

The preceding discussion illustrates various ways in which the genetic constitution of biotypes and lines are expressed in the gliadin spectrum, which serves to mark genotypes. Some gliadin components are, however, monomorphic in that they appear in all representatives of a species, subspecies, genome, or genetic grouping of biotypes (cultivars). These monomorphic components serve as markers for the corresponding chromosome of the species or genome. For example, two coherent components represented as ω_{89} are monomorphic for the D genome. They appear in gliadins of all representatives of *T. aestivum, Ae. squarrosa*, and other species of *Aegilops* that have a genome similar to the D genome. These components are absent from gliadins of nullisomic 1D-tetrasomic 1A and nullisomic 1D-tetrasomic 1B lines of Chinese Spring and from gliadins of the ditelocentric 1DL variant of Chinese Spring, which is missing the short arm of the 1D chromosome. Accordingly, the ω_{89} components serve as a very effective marker of the 1D chromosome, its short arm, or the D genome itself.

An α-gliadin component designated α_6 is monomorphic for all representatives of *Ae. squarrosa* ssp. *strangulata*, but is absent from all representatives of the ssp. *eusquarrosa*. Accordingly, this component serves as a marker for the D^{st} genome. The presence of α_6 in gliadins of *T. aestivum* may be considered evidence for the origin of the D genome of bread wheat being *Ae. squarrosa* ssp. *strangulata*. Other α-gliadins, including the aggregative A-gliadin, are controlled by chromosome 6A (Kasarda et al 1976, Shepherd 1968, Wrigley and Shepherd 1973).

In the ω-gliadin pattern of representatives of *Secale* (rye), the ω_{234} triplet is monomorphic. This triplet serves as an effective marker for the first rye chromosome (1R) in the genetic analysis of triticale and wheat lines with added 1R chromosomes.

The components that serve as markers for genome or species are recognized solely on the basis of their positions (and to some extent, intensities) in the electrophoretic spectrum. In some cultivars the component may appear as a close variant in the electrophoretic pattern (Table II).

Comparative analysis of the electrophoretic patterns of gliadins from many representatives of different cereal genera has shown that all four fractions (α, β, γ, and ω components) are represented in the electrophoretic spectra of *Triticum, Agropyron*, some species of *Aegilops*, and some species of *Hordeum*. The gliadins of species from the sitopsis group of *Aegilops*, such as *Ae. speltoides* and *Ae. longissima*, which seem to possess a B-type genome, have almost no α-gliadins; α-gliadin bands are weak or nearly absent. This is true

TABLE V
Polymorphism of cv. Krasnodarskaya 39 Based on Gliadin Spectra

Type of Spectrum	Gliadin Fractions				To 100 Plants
	α	β	γ	ω	
I	567	23145	1 34	6 89	61
II	4 67	23145	1 3	3 6789	38
III	4567	123 45	23	3 6 89	1
			Total Cultivar Formula		
	567	23145	1234	3 6789	

TABLE VI
Intracultivar Polymorphism of Rye Based on Gliadin Spectra

Type of Spectra	Composition of Gliadin Fractions				Types of Spectra Within Cultivar per 100 Plants			
	α	β	γ	ω	Vjatka-2	Gitkins-kaya	Petcus	Belta
I		45	12 5	12345	10	21	26	15
II		5	1	2345	25	7	24	5
III		45	1	2345	4	10	2	40
IV		5	1	1234	17	8	4	10
V		45	1	12345 67	10	29	4	···
VI		45	12 5	2345	12	···	14	30
VII		45		12345	12	8	2	···
VIII		345		2345	···	3	16	···
IX		45	1 5	12345	6	2	···	···
X		5	123	1234	4	4	···	···
XI		345	5	2345 7	···	···	8	···
XII		45	1 5	1234	···	8	···	···

also of *Ae. squarrosa* ssp. *eusquarrosa* (Table IV). In rye, the gliadins are represented in the spectrum mainly by slow-moving β-components and by fast-moving ω-components; the α-components and fast-moving β-components are absent. This is in accord with the data of Autran et al (1977) concerning the N-terminal amino acid sequences of whole gliadins from different cereal grains. They found that the N-terminal sequence of rye gliadin was similar to the sequence of the γ_3-component from common wheat but not to that of α-gliadins, which tends to be a major type of sequence in gliadins from *T. dicoccoides, T. boeoticum,* and other species of *Triticum* (Autran et al 1977).

Many representatives of *Hordeum* and *Agropyron* are very poor in γ-gliadin components, and the gliadins of *Avena* consist mainly of α- and β-gliadin components. The gliadins of the diploid einkorns, which probably contributed the A-genome to polyploid wheats, are especially rich in α-gliadins. The gliadins of species of *Aegilops*, which may have contributed the B and D genomes to bread wheats, are rich in ω-gliadin components. In the emmer and timopheevi wheats (AABB), the α-gliadins belong mainly to the A genome (6A chromosome). In *T. aestivum* (AABBDD), however, the α-gliadins are controlled by both the A and D genomes (chromosomes 6A and 6D). In the electrophoretic patterns of *Triticale*, the greater part of the pattern comes from the wheat parent, especially in the α- and γ-zones.

Accordingly, there are at least four ways to use alcohol-soluble seed proteins in marking genetic systems. The genome (and equivalent diploid species) can be determined best through use of genome-specific antigens that are associated mainly with the nongliadin proteins. Separate chromosomes of a genome are best marked by monomorphic electrophoretic components of the gliadins, such as the ω_{89}-components, which are specific for chromosome 1D, and the ω_{234}-components, which are characteristic of the 1R chromosome of rye. Specific cultivars or biotypes are identified best through polymorphic gliadin components, especially those of the α- and ω-gliadin zones. In the future, it may be possible to designate chromosomes by means of the polymorphic components. Finally, the sum of the monomorphic and polymorphic gliadin components defines the genotype as a whole.

The alcohol-soluble fraction contains protein markers of all main genetic systems. This fraction is easy to prepare and all analyses can be performed on material from a single extraction.

One last illustration of the use of protein markers is the analysis of the genome structure of triticale, which is an amphiploid of wheat and rye. The genome composition of triticale can be evaluated by serologic markers analyzed with antisera to the proteins of rye (R genome) and *Ae. squarrosa* (D genome of wheat). This is illustrated for the ethanol-soluble proteins of these species in Fig. 4a. The wheat genomes could be analyzed also by antisera to the proteins of *T. aestivum* and *T. durum*. The genotypes of the triticale samples can be analyzed through the electrophoretic spectra of the gliadin proteins (Figs. 4b and 5).

On the basis of this serologic and electrophoretic evaluation, the triticale samples are registered in the form of genome and gliadin formulas as follows:

Triticale Rosner (2n = 42)

$$A^uA^uB^lB^lRR \qquad \alpha_{567}\ \beta_{234}\ \gamma_{1345}\ \omega_{234}$$

Triticale AD 20 (2n = 56)

$$A^uA^uB^lB^lDDRR \qquad \alpha_{34567}\ \beta_{345}\ \gamma_{1345}\ \omega_{23456_2 89}.$$

In the gliadin spectrum of triticale, the wheat components predominated. This sometimes makes it possible to identify the wheat parent cultivar, as in the example of octoploid triticale AD COC 32-19 (Bulgar) obtained from the hexaploid wheat Bezostaya 1 and rye C-2 (Fig. 4b). The formulas of the three species are as follows:

Triticale AD COC 32-19 $\quad \omega_{\underline{234}\bar{6}89}$

Wheat Bezostaya 1 $\quad \alpha_{567}\,\beta_{2345}\,\gamma_{234}\,\omega_{\bar{2}\,\bar{4}689}$

Rye C-2 $\quad \beta_{\bar{3}\underline{45}}\,\gamma_{\bar{1}}\,\omega_{\underline{234}}.$

As indicated, the ω_{234} components mark the 1R chromosome and the ω_{89} components mark the 1D chromosome. Presence of the R and D genomes in this cultivar of triticale is confirmed by serologic tests.

The 1R chromosome (or part of it) occurs in genotypes of some wheat cultivars such as Kaukas, Aurora, Zorba, Orland, and Wentzel. This was confirmed by the presence of the ω_{234} components in these cultivars. The absence of the whole R genome

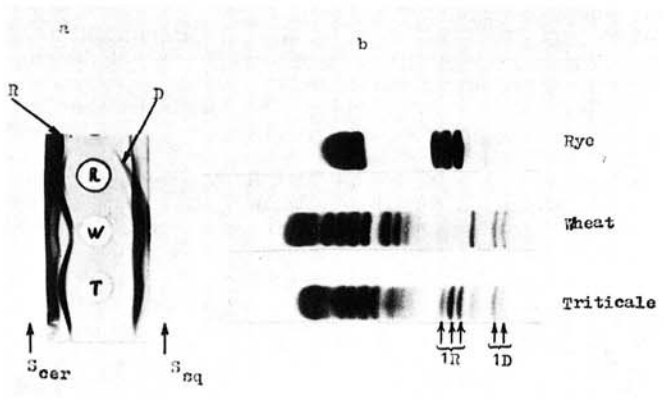

Fig. 4. Comparative (**a**) immunochemical and (**b**) electrophoretic analysis of ethanol-soluble proteins from grain of triticale (2n = 56) AD COC 32-19 and its parents rye C-2 and wheat Bezostaya 1. **Scer** and **Ssq** = Antisera against ethanol-soluble proteins of *Secale cereale* (R genome) and *Aegilops squarrosa* (D genome); **1D** and **1R** = first chromosomes of D and R genomes; **R, W,** and **T** = ethanol-soluble proteins of designated cultivars of rye, wheat, and triticale.

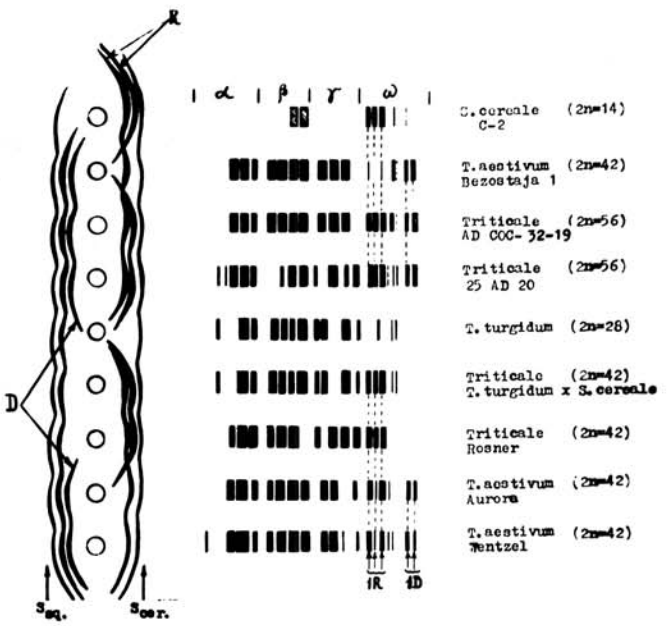

Fig. 5. Immunochemical and electrophoretic analysis of alcohol-soluble proteins of triticales and wheats with different genome compositions. **Ssq** and **Scer** = antisera against alcohol-soluble proteins of *Aegilops squarrosa* and *Secale cereale*; **D** = serologic marker of D-genome; **R** = serologic marker of R-genome; **1D** and **1R** = electrophoretic chromosomal markers.

was evident from immunochemical results (Fig. 5).

In summary, the use of protein markers provides the basis for a rational, straightforward, and practical system of registration of genetic cereal resources. The system described provides a means for designating genome and genotype formulas based on protein components. These formulas reflect the genus, species, cultivar, and line or biotype of a sample and are accordingly rich in genetic and phylogenetic information. The significance of this information will increase in the future. The protein formulas described can be used both in searching for initial materials in a breeding program and for testing experimental materials for the inclusion of desirable genetic structures in the lines at the early stages of a breeding program. Registration of cultivars and biotypes in the form of protein formulas lends itself to computer storage and retrieval of information. This can be especially important in programs of germ plasm conservation for wheat and other cereals where such information must be stored for use far in the future.

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