

## AN ABSTRACT OF THE THESIS OF

Andres Encinas Mungarro for the degree of Master of Science  
in Crop Science presented on November 8, 1988  
Title: Durum Wheat Quality and its Relation to Electrophoretic Banding  
Patterns

Redacted for Privacy

abstract approved: \_\_\_\_\_  
Warren E. Kronstad

Physical properties of gluten and pigment content are of primary importance in determining quality in durum wheat (Triticum turgidum L. var. durum). The objective of this investigation was to evaluate the contribution of different  $\gamma$ -gliadin protein subunits in durum wheat pasta quality as measured by the sodium dodecyl sulphate microsedimentation (MST) test, protein, and pigment content. Twelve crosses involving parents,  $F_1$ ,  $F_2$ ,  $BC_1$ , and  $BC_2$  individual plants were evaluated for pigment content, protein content, and SDS sedimentation. Fourteen cultivars and segregating populations of two crosses were classified according to their electrophoretic banding patterns.

Cultivars with band 45 had stronger gluten properties than cultivars having band 42 as evidenced by the low SDS microsedimentation values. No clear association was observed between protein, pigment content and electrophoretic banding patterns. The close association between band 45 and gluten strength was again confirmed in segregating  $F_2$  individual plants of two crosses. Plants homozygous for band 45 were higher in SDS microsedimentation values, compared with plants heterozygous for band 45/42 and homozygous for band 42.

Transgressive segregation was observed for both gluten strength and pigment content in the  $F_2$  populations of all crosses, indicating selection will be effective to enhance these traits.

✓

DURUM WHEAT QUALITY AND ITS  
RELATION TO ELECTROPHORETIC  
BANDING PATTERNS

by

Andres Encinas Mungarro

A THESIS

submitted to

Oregon State University

in partial fulfillment of  
the requirements for the  
degree of

Master of Science

Completed November 1988

Commencement June 1989

APPROVED:

Redacted for Privacy

\_\_\_\_\_  
Professor of Plant Breeding and Genetics, in charge of major

Redacted for Privacy

\_\_\_\_\_  
Head of Crop Science Department

Redacted for Privacy

\_\_\_\_\_  
Dean of Graduate School

Date thesis is presented \_\_\_\_\_ November 8, 1988

Typed by \_\_\_\_\_ Andres Encinas M.

—

## ACKNOWLEDGEMENTS

I wish to express my deep appreciation to Dr. Warren E. Kronstad, my major professor, for his interest, encouragement and guidance during my course work and the preparation of this thesis.

I want to extend my thanks to Dr. S. Broich, Dr. R. G. Petersen, and Dr. N. W. Christensen for serving on my graduate committee and reviewing the manuscript.

I sincerely thank Dr. Pedro Brajcich, Dr. Arnoldo Amaya, Dr. Javier Peña, and the durum breeding program team at CIMMYT, México for their valuable help and interest in the development of this research.

Special thanks are extended to all the staff and members of the cereal project, and to fellow graduate students of the Department of Crop Science for their friendship and understanding.

Appreciation is extended to Crop Science Department of Oregon State University and the International Maize and Wheat Improvement Center (CIMMYT) for allowing me the opportunity to further my education.

I am deeply grateful to my wife Dolores, and my daughter Patricia, whose endless patience and love supported me throughout my graduate work.

## CONTRIBUTION OF AUTHORS

The contribution of each author to this thesis is described in detail;

Encinas Mungarro A.

Planned the experiment. Recorded agronomic data during crop development. Determined the electrophoretic banding patterns and protein content of experimental materials. Data management, statistical analysis and thesis writing.

Kronstad W. E.

Assisted in project planning. Advised on experimental materials and data analysis. Thesis editing.

Brajcich P.

Selected and developed the experimental material. Supervised experiment maintenance.

Amaya A.

Developed methodology for laboratory evaluations of gluten strength (microsedimentation test) and pigment content.

## TABLE OF CONTENTS

### CHAPTER

I	ABSTRACT	1
	INTRODUCTION	2
	MATERIAL AND METHODS	5
	RESULTS	9
	DISCUSSION	12
	REFERENCES	23
II	LITERATURE REVIEW	25
	Durum wheat	25
	Classification of wheat endosperm proteins	26
	Storage proteins	27
	Gliadin proteins	27
	Glutenin proteins	28
	Genetic control of endospermic proteins	29
	Inheritance of gliadins	30
	Inheritance of glutenins	32
	Relationship between gliadin components and pasta-making quality	33
	Pigment content	35
	BIBLIOGRAPHY	37

## LIST OF FIGURES

- |   |   |    |
|---|---|----|
| 1 | Frequency distribution for SDS microsedimentation values of $F_2$ , $BC_1$ , and $BC_2$ populations from the cross Quilafen/Shwa. Arrows indicate $F_1$ and parental means. | 19 |
| 2 | Frequency distribution for SDS microsedimentation values of $F_2$ , $BC_1$ , and $BC_2$ populations from the cross Altar/Alk. Arrows indicate $F_1$ and parental means.     | 20 |
| 3 | Frequency distribution for pigment content of $F_2$ , $BC_1$ , and $BC_2$ populations from the cross Quilafen/Shwa. Arrows indicate $F_1$ and parental means.               | 21 |
| 4 | Frequency distribution for pigment content of $F_2$ , $BC_1$ , $BC_2$ , populations from the cross Altar/Alk. Arrows indicate $F_1$ and parental means.                     | 22 |



## LIST OF TABLES

<u>Table</u>		<u>Page</u>
1	Characterization of fourteen durum wheat cultivars for protein content, pigment content, SDS micro-sedimentation values, and electrophoretic-banding patterns.	14
2	Classification of F <sub>2</sub> plants from cross Quilafen/Shwa into quality groups based on the presence or absence of specific gliadin-banding patterns and their relation to gluten strength as measured by the SDS micro sedimentation test.	15
3	Classification of F <sub>2</sub> plants from cross Altar/Alk into quality groups based on the presence or absence of specific gliadin-banding patterns and their relation to gluten strength as measured by the SDS microsedimentation test.	16
4	Descriptive statistics for gluten strength as measured by the SDS microsedimentation test for the F <sub>2</sub> populations of twelve durum wheat crosses.	17
5	Descriptive statistics for pigment content for the F <sub>2</sub> populations of twelve durum wheat crosses.	18

# DURUM WHEAT QUALITY AND ITS RELATION TO ELECTROPHORETIC BANDING PATTERNS

A. Encinas\*, W. Kronstad, P. Brajcich, and A. Amaya

## I ABSTRACT

Physical properties of gluten and pigment content are of primary importance in determining quality in durum wheat (Triticum turgidum L. var. durum). The objective of this investigation was to evaluate the contribution of different  $\gamma$ -gliadin protein subunits in durum wheat pasta quality as measured by the sodium dodecyl sulphate microsedimentation (MST) test, protein, and pigment content. Twelve crosses involving parents,  $F_1$ ,  $F_2$ ,  $BC_1$ , and  $BC_2$  individual plants were evaluated for pigment content, protein content, and SDS sedimentation. Fourteen cultivars and segregating populations of two crosses were classified according to their electrophoretic banding patterns. Gluten strength was closely related to a gliadin protein designated as band 45, and gluten weakness to a group of gliadin proteins including band 42. Electrophoresis banding patterns of gliadin proteins showed no association with either pigment content or protein content. Polyacrylamide gel electrophoresis and pigment extraction are valuable techniques for parental selection and early generation screening for pasta quality in durum wheat.

A. Encinas, and W.E. Kronstad, Dep. of Crop Science, Oregon State Univ., Corvallis, OR 97331; P. Brajcich, and A. Amaya, International Maize and Wheat Improvement Center (CIMMYT), Lisboa 27, PO Box 6-641, 06600 México D. F., México. Contribution from Oregon Agric. Exp. Stn. and CIMMYT, México. \*Corresponding author.

## INTRODUCTION

A major objective of durum wheat (Triticum turgidum L. var. durum) improvement programs is to develop cultivars with strong and elastic gluten. Strong gluten maintains the integrity of the pasta during cooking so a firm, resilient end product is obtained.

For durum and bread wheats (Triticum aestivum L. em Thell), breeders have emphasized increased protein quantity as a way of improving the pasta and bread making properties, respectively. However, this trait is polygenic and is greatly influenced by the environment. With the development of new techniques it appears that the quality may be as important as the quantity of the protein in determining end use acceptability.

Several rapid and small-scale tests have been developed to assess gluten strength in wheat. According to du Cros et al. (1982), semolina dough strength may be measured on farinographs, mixographs or by using the Sodium dodecyl sulphate sedimentation (SDSS) test. Elastic properties of gluten can be tested by a Chopin alveograph or by a viscoelastograph. The results obtained using these physical tests are often influenced by environmental factors (Damidaux et al. 1980a, du Cros et al. 1982). They also have the disadvantages of requiring large samples of flour which prohibit their use for early generation evaluation.

Studies using polyacrylamide gel electrophoresis (PAGE) of durum wheat gliadin proteins have shown a close relationship between gluten strength and two  $\gamma$ -gliadins proteins of different relative mobility

designated as band 45 and band 42: band 45 is associated with strong gluten and good cooking performance, while band 42 is associated with weak gluten and poor cooking performance (Damidaux et al. 1980a, Kosmolak et al. 1980, du Cros et al. 1982). Genetic studies using disomic substitution lines revealed that the gene controlling bands 45 and 42 is located in the short arm of chromosome 1B (Damidaux et al. 1980b, Joppa et al. 1983b, du Cros et al. 1983). Further work by du Cros and Hare (1985) revealed that gliadin bands 45 and 42 are controlled by alleles at a single locus and that band 42 appears to be partially dominant to band 45 when in a heterozygous state.

Furthermore Josephides et al (1987), using 1B chromosome substitution lines concluded that chromosome 1B in durum wheat has a more important effect on gluten strength than any other chromosome or group of chromosomes. Gliadins forming the gluten complex are inherited as linkage blocks on chromosome 1B. Band 45 is linked to a low molecular weight subunit glutenin designated LMW-2. The band 42 conferring weak gluten is also linked to another low molecular glutenin subunit LMW-1. Therefore, the viscoelastic properties of gluten strength as measured by the SDSS test are more likely to be associated with the biochemical differences between the glutenin subunits LMW-1 and LMW-2 and not with the gliadin proteins (Payne et al. 1984a).

Pigment content is controlled by more than one gene with the gene action being primarily additive (Braaten et al. 1962; Johnston et al. 1983). Early generation evaluation and selection of progenies in segregating populations are possible using a modification of the

American Association of Cereal Chemists (1976) method (A. Amaya and J. Peña personal communication).

This paper describes the use of electrophoresis of gliadin components to predict durum wheat pasta quality.

## MATERIALS AND METHODS

Experimental populations selected represented diverse sources of durum wheat germplasm for assessment of quality and quantity of protein. Fourteen cultivars developed by the Durum Breeding Program at CIMMYT, Mexico, were selected based on their grain-yield potential and different quality parameters.

The crosses were made in winter 1985 at the Northwest Agricultural Research Center (CIANO), near Ciudad Obregon, in Northwest México. Subsequent backcrosses and the  $F_2$  populations were obtained in summer 1985 at El Batan Experimental site, near Mexico City, México. The parents and subsequent generations were planted in the fall of 1985 at CIANO for evaluation.

The six generations representing all crosses were planted in a randomized block design with three replications. The crosses consisted of one good quality parent crossed to parental lines representing good, medium, and poor quality types. Each replication included parents,  $F_1$ , and backcross populations represented by one row each and six rows for  $F_2$  populations for each cross. The rows consisted of 20 plants spaced 20 cm apart; the distance between rows was 30 cm. Planting was on November 20, 1985.

A total of 200 kg/ha of ammonium sulfate and 60 kg/ha of phosphorus was applied in two applications, one prior to planting and the second in January with the second irrigation.

After harvest, the grain was ground in a udy cyclone mill (U. D. Corp., Fort Collins, CO) using a 0.5-mm mesh sieve. The whole-meal

produced was used for determining the grain protein content by near-infrared reflectance spectroscopy with a Technicon Infralyser 400 (Technicon Corp., Tarrytown, N. Y.).

Gluten strength was evaluated by the "Sodium Dodecyl Sulfate Sedimentation Test" (SDSS Test), described by Axford et al. (1978), and modified by Dick and Quick (1983) as a "Sodium dodecyl sulfate microsedimentation test (MST)". This modification requires a smaller amount of material (1 g of ground sample) and less solution. It is fast, simple to perform, selective and reproducible when used to screen for gluten strength in durum wheat. The method was further modified by Amaya and Peña (Personal communication) for evaluating bread, durum wheat, and triticale. This further modification uses a continuous mechanical agitator for one minute after the addition of lactic acid and sodium dodecyl sulphate. After fourteen minutes, the volume of the sediment was measured in milliliters.

Pigment content was evaluated by the procedure of the American Association of Cereal Chemistry (AACC Method 14-50) and modified by Amaya and Peña (Personal communication) to be used in screening early segregating durum populations for pigment content at CIMMYT, Mexico. This modification consists of using 3 g of flour (whole flour or semolina), 15 ml of water-saturated butyl alcohol, and 1-hour extraction time, in contrast to an 8.0-g sample, 40 ml of water-saturated butyl alcohol, and 16 to 18 hours minimum for extraction, which is the method of the AACC. The same ratio 1:5 sample/alcohol is maintained, but the modification is faster and requires less sample. Amount of material to sample is a limiting factor in screening early segregating generations.

Fourteen parental lines, and seed of  $F_1$ ,  $BC_1$ ,  $BC_2$ , and  $F_2$  plant populations from two crosses, were classified according to the electrophoretic banding patterns of their gliadin proteins. To determine the banding patterns of gliadin polypeptides separated by polyacrylamide gel electrophoresis (PAGE), the procedure used was essentially that of Bushuk and Zillman (1978), with modifications proposed by Tkachuk and Metlish (1980), Lookhart et al. (1985), and Khan et al. (1985). Proteins were extracted with 1.0 ml of 70% aqueous ethanol to 0.5 g of whole meal and vortexing the suspension every 10 minutes for 1 hour at room temperature, and then centrifuged at 20,000 g, also at room temperature. The supernatant was decanted and diluted with 0.5 ml of aluminum lactate, pH 3.1 buffer (Lookhart et al., 1982), containing 60% (w/v) sucrose to increase the density and facilitate applying the sample to slot in the gel. Finally, 3% (w/v) methyl green tracking dye was added to serve as a marker during electrophoresis. The Bio-Rad dual-slab vertical-gel apparatus (Bio-Rad Laboratories, Richmond, CA) was used for PAGE. Electrophoresis was carried out in a vertical slab gel (160 x 140 x 3.0 mm) with 15 wells. The gel contained 6% (w/v) acrylamide with 0.3% (w/v) bisacrylamide, 0.024% (w/v) ascorbic acid,  $2 \times 10^{-4}$ % (w/v) ferrous sulfate-heptahydrate, and 0.1% (v/v) of 3% hydrogen peroxide. Each column was loaded with 15  $\mu$ l of sample. A constant current of 500 V was applied for 4 hours. Circulating coolant was maintained at 10°C for all runs. After removal from the glass plate, each gel was stained in a solution of 12% trichloroacetic acid and 5 ml of 0.5% Coomassie Brilliant Blue R-250 at room temperature and



destained with 12% Trichloro acetic acid. Bands were designated by using the nomenclature of Zillman and Bushuk (1979b).

## RESULTS

Differences among parental lines and crosses for protein, pigment content and SDS microsedimentation values were found. Results from the SDS microsedimentation test and electrophoretic banding patterns suggested a close association (Table 1). Cultivars with band 45 had stronger gluten properties than the cultivars having band 42 as evidenced by the low SDS microsedimentation values. The one exception was cultivar Sh1. No clear association was observed between protein content and electrophoretic banding patterns.

Cultivars with band 45 had also higher values for pigment content than cultivars with band 42, except for cultivars Miscv4.85 and Sh1.

The  $\gamma$ -gliadin band 45 was associated with gliadin band 35. Weak-gluten types with gliadin band 42 also contained gliadins 33, 35, and 38. According to Payne et al. (1984a), gliadins are inherited as linkage blocks on chromosome 1B. The same associations between these bands were observed in this study, regarding gliadin proteins.

The results for specific banding patterns of the  $F_2$  individual plants from cross Quilafen/Shwa are presented in Table 2. Plants homozygous for band 45 were higher in SDS microsedimentation values. Plants heterozygous for band 45/42 were intermediate, and plants homozygous for band 42 showed the lowest SDS microsedimentation values. In this cross differences were noted for gluten strength among the three groups; pigment content showed no difference; protein content differed between and among the high and low SDS microsedimentation groups.

The data for cross Altar/Alk is presented in Table 3. Differences were noted only for SDS microsedimentation values among the three groups.

Descriptive statistics for gluten strength of the  $F_2$  populations from the twelve crosses are presented in Table 4. Differences were detected among the means and ranges of the  $F_2$ 's populations. Among the crosses the combinations of Amal/Altar and Altar//Boy/Yav showed the lowest SDS microsedimentation means, while Quilafen//Memo/Mexi showed the highest mean value. Higher SDS microsedimentation ranges were observed among crosses when both parents had band 45, while crosses involving one parent with band 42 always gave lower range values. The range of the  $F_2$  also indicates that transgressive segregation in both directions was present in all crosses for gluten strength.

Data of the  $F_2$ 's for pigment content is presented in Table 5. The lowest mean value was observed for the cross Amal/Altar, while Altar//Rok/Kmli showed the highest mean value. The range of the  $F_2$ 's also indicates that transgressive segregation in both directions was present in all crosses except for Quilafen/Shwa. In this cross, however, higher values favored the higher parent.

Frequency distribution for the SDS microsedimentation test of  $F_2$ ,  $BC_1$ , and  $BC_2$  populations, and means of  $F_1$  and parental lines of Quilafen/Shwa and Altar/Alk crosses are shown in Figure 1 and 2.  $F_1$  SDS microsedimentation values approached the lowest parent. In the other crosses however, the  $F_1$  was similar to the mid parents value. Mean values of the backcross generation tended to approach the mean of their respective recurrent parent. The most striking feature of the  $F_2$  distributions was the transgressive segregation that occurred in both

directions in all crosses. The SDS microsedimentation values for some  $F_2$  plants were higher than previously reported in durum wheat in routine screening of segregating lines, using the microsedimentation test.

Frequency distribution for pigment (carotene) content of  $F_2$ ,  $BC_1$ , and  $BC_2$  populations, and means of  $F_1$  and parental lines from crosses Quilafen/Shwa, and Altar/Alk are shown in Figures 3 and 4.  $F_1$  and  $F_2$  means were similar to the midparent values, backcrosses tended to approach their recurrent parent. Extreme  $F_2$  segregates surpassed parental values in the both crosses.

## DISCUSSION

Transgressive segregation was observed for both gluten strength (as measured by the SDS microsedimentation test), and pigment content in the  $F_2$  populations, indicating selection would be possible to enhance these traits. Based on the segregating patterns for SDS microsedimentation it would appear that further genetic progress could be made for gluten strength in the crosses where both parental lines with band 45 are involved. In those cases early generation screening for gluten strength could be avoided. In certain crosses, where any progenitor has band 42 extreme high segregation occurred. In these situations early generation screening will be required.

Electrophoresis of gliadin proteins showed no association with pigment content. Thus, the SDS microsedimentation values and pigment content are two genetically independent characteristics, and pigment content could be selected without relying on the electrophoretic banding patterns.

The gliadins in group 45 showed high correlation with gluten strength as measured by the SDS microsedimentation test, while group 42 showed a high correlation with gluten weakness. These results agreed with the findings of Damidaux et al. (1980b), Kosmolak et al. (1980), and du Cros et al. (1982), in which specific gliadin proteins are associated or linked with genes coding for gluten quality in durum wheat. Although these proteins do not play a direct role in SDS sedimentation (Payne et al. 1984), they are excellent genetic markers and can be used to select for durum wheat for good pasta making

qualities. Breeders can use protein molecular subunits in designing crossing programs as an indication of genetic potential for gluten strength, independent of environmental effects. Electrophoresis permits examination of parental lines and early generation material for protein quality without destroying the embryo of the kernel. Those individuals found to have the desired bands can be planted with the resulting progeny assumed to have the desired gluten strength. Parental material can be chosen for the presence of bands 45 only. Durum wheat breeders focusing on quality should avoid parental lines with band 42. Segregating populations with heterozygous band 42 should be analyzed, and only plants that are homozygous for band 45 should be selected. Plants heterozygous for bands 45 and 42 can be planted for further screening and selection.

Polyacrylamide gel electrophoresis is a preferred technique to separate and classify gliadin subunits. This technique is a reliable procedure for assessing gluten strength in durum wheat and offers several advantages; 1) it is simple to perform and interpretation of results is independent of the environment, 2) the genotype is readily obtained, and homozygous plants for band 45 can be easily distinguished, 3) the amount of sample required for electrophoresis is less than for any other method for testing gluten strength, and 4) finally it is a non-destructive method.

Table 1. Characterization of fourteen durum wheat cultivars for protein, pigment content, gluten strength as measured by the SDS microsedimentation test, and electrophoretic-banding patterns.

CULTIVAR	PROTEIN CONTENT	PIGMENT CONTENT	MST† VALUES	BANDING PATTERNS
	g kg <sup>-1</sup>	mg kg <sup>-1</sup>	mL	
Quilafen	154	6.5	12.1	45,35
Kill	142	5.0	10.8	45,35
Memo/Mexi	139	5.6	10.7	45,35
CBSV274.85	134	5.8	8.8	45,35
Aos	137	4.7	8.6	45,35
Altar	129	5.6	8.4	45,35
MiscV4.85	141	3.9	8.1	45,35
Chen	136	5.5	8.1	45,35
Rok/Kmli	138	7.6	7.1	45,35
Shl	139	3.9	6.5	45,35
Shwa	144	3.7	6.7	42,33,35,38
Alk	144	4.1	5.5	42,33,35,38
Amal	131	3.3	4.9	42,33,35,38
Boy/Yav	130	4.1	4.4	42,33,35,38
LSD	4.30	0.54	1.48	

† MST = Microsedimentation test

Table 2. Classification of  $F_2$  plants from cross Quilafen/Shwa into quality groups based on the presence or absence of specific gliadin-banding patterns and their relation to gluten strength as measured by the SDS microsedimentation test.

MST† GROUPS	BANDING PATTERNS	MST† VALUES	PIGMENT CONTENT	PROTEIN CONTENT
		mL	mg kg <sup>-1</sup>	g kg <sup>-1</sup>
HIGH	45	11.08 a‡	5.10 a	150 a
INTERMEDIATE	42/45	8.54 b	4.99 a	146 ab
LOW	42	6.64 c	4.97 a	141 b

† MST = Microsedimentation test.

‡ Means with the same letter within a column are not significantly different based on Fisher's Protected LSD test at the 0.05 probability level.



Table 3. Classification of  $F_2$  plants from cross Altar/Alk into quality groups based on the presence or absence of specific gliadin-banding patterns and their relation to gluten strength as measured by the SDS microsedimentation test.

MST† GROUPS	BANDING PATTERNS	MST† VALUES	PIGMENT CONTENT	PROTEIN CONTENT
		mL	mg kg <sup>-1</sup>	g kg <sup>-1</sup>
HIGH	45	9.34 a†	5.03 a	133 a
INTERMEDIATE	42/45	7.48 b	4.98 a	132 a
LOW	42	5.63 c	4.94 a	133 a

† MST = Microsedimentation test.

‡ Means with the same letter within a column are not significantly different based on Fisher's Protected LSD test at the 0.05 probability level.

Table 4. Descriptive statistics for gluten strength as measured by the SDS microsedimentation test for the  $F_2$  populations of twelve durum wheat crosses.

CROSS	N	MEAN mL	RANGE mL	STANDARD DEVIATION	PARENTAL MEANS	
					P1	P2
QUILAFEN//MEMO/MEXI	190	10.2 a‡	7.5-15.0	1.47	12.3	10.7
QUILAFEN/AOS	190	9.3 c	6.5-15.0	1.08	12.4	8.6
QUILAFEN/SHWA	190	8.3 e	6.0-15.0	1.01	12.1	6.7
KILL/ALTAR	190	9.8 b	6.5-15.0	1.67	10.8	8.3
MEMO/MEXI//ALTAR	190	9.5 c	6.5-14.0	1.80	10.7	8.5
AMAL/ALTAR	190	6.1 h	4.0-10.0	1.30	4.9	8.4
ALTAR/ROK//KMLI	190	7.7 f	5.0-10.5	1.06	8.4	7.1
ALTAR/ALK	190	7.0 g	4.0-13.0	1.80	8.2	5.5
ALTAR//BOY/YAV	190	6.1 h	3.0- 9.5	1.45	8.3	4.4
CHEN/CBS274.85	190	8.8 d	6.5-12.0	1.25	8.3	8.8
CHEN/MISCV4.85	190	8.5 de	6.5-12.5	1.07	8.3	8.1
CHEN//ROK/KMLI	190	7.4 f	5.4-12.0	1.02	8.1	7.1

‡ Means with the same letter within a column are not significantly different based on Fisher's Protected LSD Test at the 0.05 probability level.

Table 5. Descriptive statistics for pigment content for the  $F_2$  populations of twelve durum wheat crosses.

CROSS	N	MEAN mg kg <sup>-1</sup>	RANGE mg kg <sup>-1</sup>	STANDARD DEVIATION	PARENTAL MEANS	
					P1	P2
QUILAFEN//MEMO/MEXI	190	6.4 b‡	4.6-9.2	0.77	6.5	5.6
QUILAFEN/AOS	190	5.9 c	4.0-8.6	0.79	6.7	4.7
QUILAFEN/SHWA	190	5.0 e	4.0-7.0	0.63	6.5	3.7
KILL/ALTAR	190	5.0 e	4.7-6.8	0.64	5.0	5.6
MEMO/MEXI//ALTAR	190	5.6 d	4.6-8.1	0.79	5.7	5.4
AMAL/ALTAR	190	4.0 g	2.8-5.7	0.63	4.1	5.4
ALTAR//ROK/KMLI	190	7.2 a	4.4-10.8	1.10	5.6	7.6
ALTAR/ALK	190	5.0 e	3.4-6.5	0.60	5.7	4.1
ALTAR//BOY/YAV	190	4.7 f	3.4-6.2	0.50	5.5	4.1
CHEN/CBSV272.85	190	5.5 d	4.2-7.2	0.62	5.6	5.8
CHEN/MISCV4.85	190	4.7 f	3.3-6.6	0.62	5.5	3.9
CHEN//ROK/KMLI	190	4.7 f	3.0-7.2	0.76	5.6	7.6

‡ Means with the same letter within a column are not significantly different based on Fisher's Protected LSD Test at the 0.05 probability level.

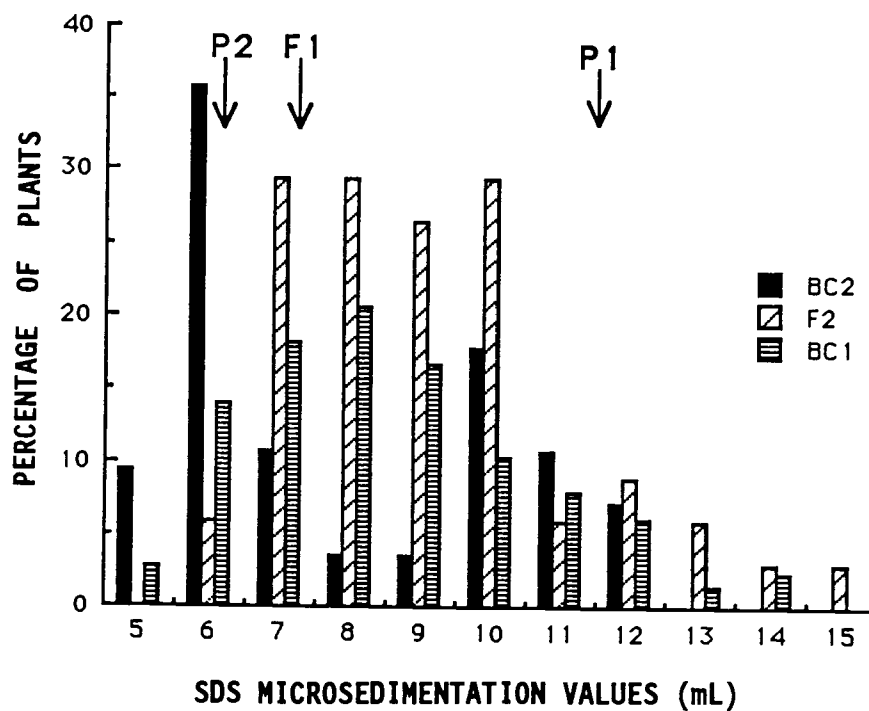


Figure 1. Frequency distribution for gluten strength as measured by the SDS microsedimentation test of F<sub>2</sub>, BC<sub>1</sub>, and BC<sub>2</sub> populations from the cross Quilafen/Shwa. Arrows indicate F<sub>1</sub> and parental means.

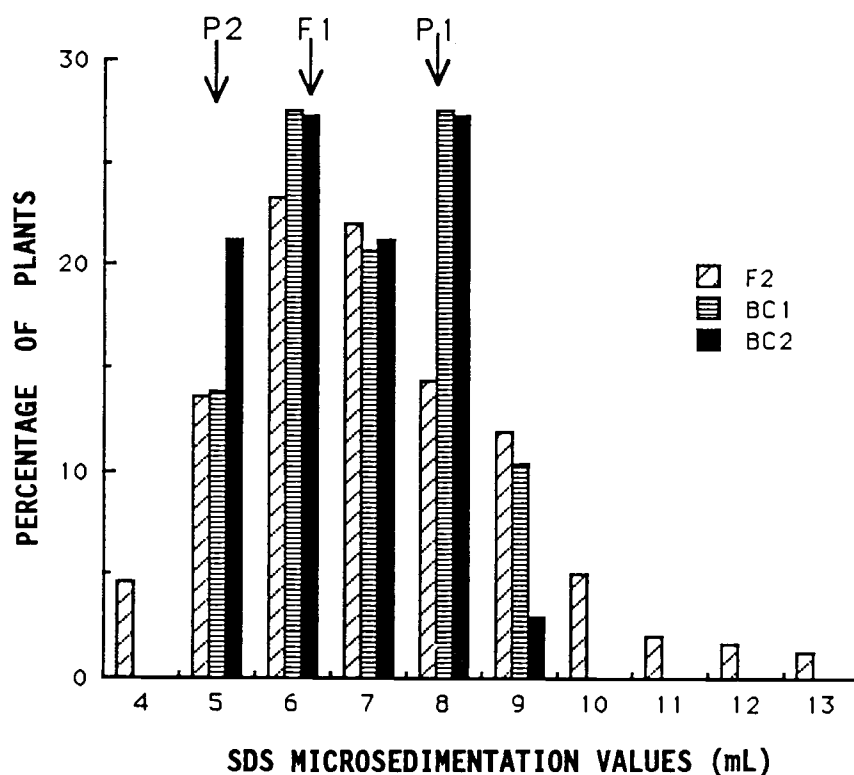


Figure 2. Frequency distribution for gluten strength as measured by the SDS microsedimentation test of F<sub>2</sub>, BC<sub>1</sub>, and BC<sub>2</sub> populations from the cross Altar/Alk. Arrows indicate F<sub>1</sub> and parental means.

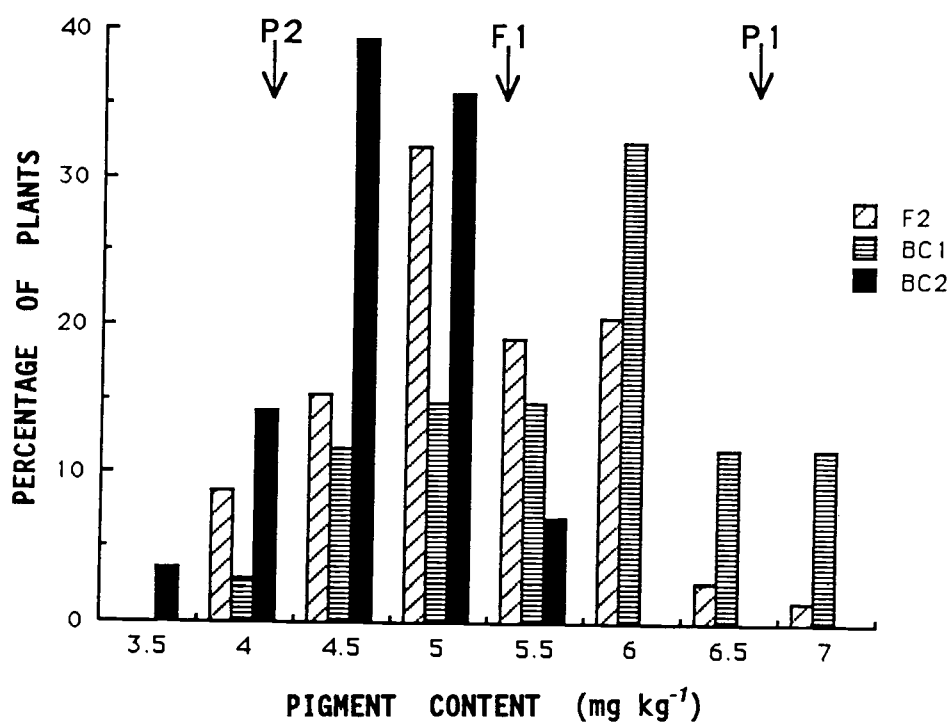


Figure 3. Frequency distribution for pigment content of F<sub>2</sub>, BC<sub>1</sub>, and BC<sub>2</sub> populations from the cross Quilafen/Shwa. Arrows indicate F<sub>1</sub> and parental means.

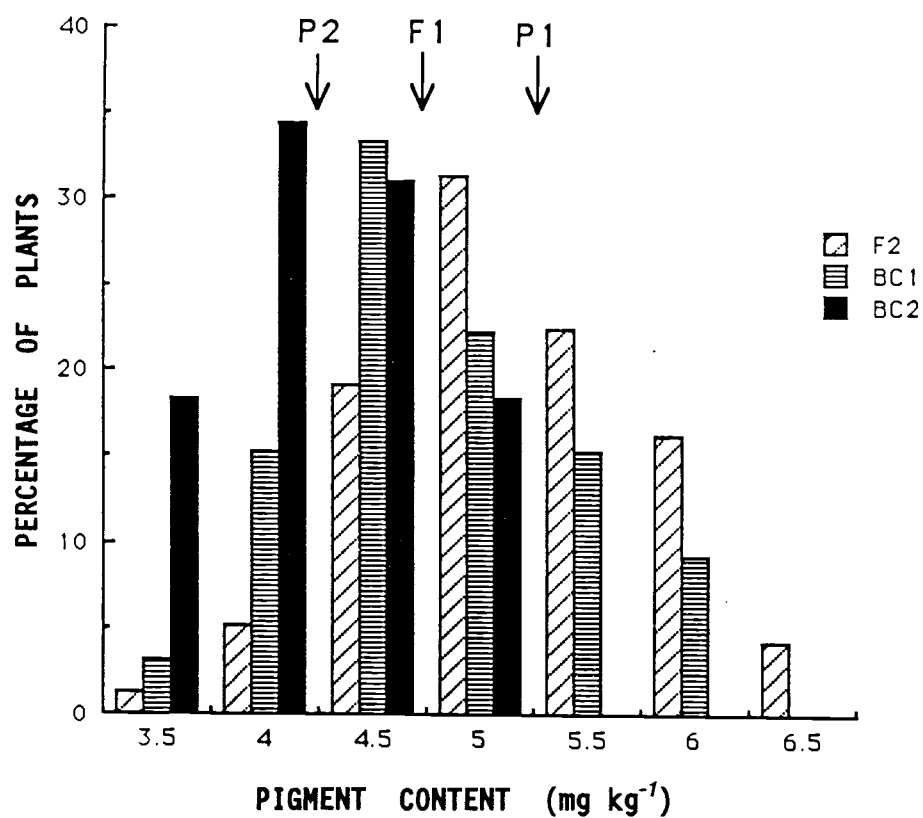


Figure 4. Frequency distribution for pigment content of F<sub>2</sub>, BC<sub>1</sub>, and BC<sub>2</sub> populations from the cross Altar/Alk. Arrows indicate F<sub>1</sub> and parental means.

## REFERENCES

- American Association of Cereal Chemistry. 1976. Approved methods. Pigments, method 14-50.
- Axford, D.W., E.E. McDermott, and D.G. Redman. 1978. Small scale tests of bread-making quality. *Milling Feed and Fertilizer*. 66:18-20.
- Braaten, M.O., K.L. Lebsack, and L.D. Sibbitt. 1962. Inter-generation relations of physical properties of dough and carotenoid pigment content in durum wheat. *Crop Sci.* 2:277-280.
- Bushuk, W. and R.R. Zillman. 1978. Wheat cultivar identification by gliadin electrophoregrams. I. Apparatus, method and nomenclature. *Can. J. Plant Sci.* 58:505-515.
- Damidaux, R., J.C. Autran, and P. Feillet. 1980a. Gliadin electrophoregrams and measurements of gluten viscoelasticity in durum wheat. *Cereal Foods World*. 25:754-756.
- Damidaux, R., J.C. Autran, P. Grignac, and P. Feillet. 1980b. Determinisme genetique des constituants gliadins de Triticum durum Desf. associes a la qualite culinaire intrinseque des varietes. *C. R. Acad. Sci. Paris, Serie D* 291:585-588.
- Dick, J.W. and J.S. Quick. 1983. A modified screening test for rapid estimation of gluten strength in early-generation durum breeding lines. *Cereal Chem.* 60:315-318.
- du Cros, D.L., and R.A. Hare. 1985. Inheritance of gliadin proteins associated with quality in durum wheat. *Crop Sci.* 25:674-677.
- du Cros, D.L., C.W. Wrigley, and R.A. Hare. 1982. Prediction of durum-wheat quality from gliadin-protein composition. *Aust. J. Agric. Res.* 33:429-442.
- du Cros, D.L., L.R. Joppa, and C.W. Wrigley. 1983. Two-dimensional analysis of gliadin proteins associated with quality in durum wheat: chromosomal location of genes for their synthesis. *Theor. Appl. Genet.* 66:297-302.
- Johnston, R.A., J.S. Quick, and J.J. Hammond. 1983. Inheritance of semolina color in six durum wheat crosses. *Crop Sci.* 13:743-746.
- Joppa, L.R., K. Khan, and N.D. Williams. 1983b. Chromosomal location of genes for gliadin polypeptides in durum wheat Triticum turgidum. *L. Theor. Appl. Genet.* 64:289-293.
- Josephides, C.M., L.R. Joppa, and V.L. Youngs. 1987. Effect of chromosome 1B on gluten strength and other characteristics of durum wheat. *Crop Sci.* 27:212-216.



- Khan, K., A.S. Hamada, and J. Patek. 1985. Polyacrylamide gel electrophoresis for wheat variety identification: Effect of variable on gel properties. *Cereal Chem.* 62:310-313.
- Kosmolak, F.G., J.E. Dexter. R.R. Matsuo, D. Leisle, and B.A. Marchylo. 1980. A relationship between durum wheat quality and gliadin electrophoregrams. *Can. J. Plant Sci.* 60:427-432.
- Lookhart, G.L., B.L. Jones, S.B. Hall, and K.F. Finney. 1982. An improved method for standardizing polyacrylamide gel electrophoresis of wheat gliadin proteins. *Cereal Chem.* 59:178-181.
- Lookhart, G.L., D.B. Cooper, and B.L. Jones. 1985. Effect of temperature and alternate buffer systems on resolution of wheat gliadin proteins by polyacrylamide gel electrophoresis. *Cereal Chem.* 62:19-22.
- Payne, P.I., E.A. Jackson, and L.M. Holt. 1984. The association between  $\gamma$ -gliadin 45 and gluten strength in durum wheat varieties: A causal effect or the result of genetic linkage?. *J. Cereal Sci.* 2:73-81.
- Tkachuk, R., and V.J. Metlish. 1980. Wheat cultivar identification by high voltage electrophoresis. *Ann. Technol. Agric.* 29:207-212.
- Zillman, R.R., and W. Bushuk. 1979b. Wheat cultivar identification by gliadin electrophoregrams. III. Catalogue of electrophoregrams formulas of canadian wheat cultivars. *Can. J. plant Sci.* 59:287-298.



## II LITERATURE REVIEW

Wheat followed by rice and maize represent the major sources of food in the world. The many end uses for semolina or flour are what makes wheat such an important crop. Therefore, in most wheat breeding programs a major objective is to improve the milling, pasta making and baking properties as they improve the quality of the various end products. For durum and bread wheats breeders have emphasized increase protein quantity as a way of improving the milling and baking properties. However, with the development of new techniques it now appears that the quality of the protein may be as important as the quantity.

### Durum wheat

Durum wheat is an allotetraploid ( $2n = 28$ ) with a genomic designation of AABB. The cultivated Triticum turgidum L. var durum originated from T. dicoccoides as the free threshing character was accumulated sometime around 300 B.C. (Morris and Sears, 1967). The wild tetraploid ancestor of durum wheat is a product of the hybridization of two diploid species, followed by a spontaneous doubling of the chromosome. One of the diploid wheats, T. monococcum, contributed the A genome to T. turgidum. The species that contributed to the B genome is not known yet.

Durum wheats are used for pasta products such as macaroni and spaghetti. Like bread wheat cultivars, durums require a high grain

protein content, usually above 13%, however, proteins such as the glutenins tend to be less elastic, and have harder kernel texture. durum glutenins are usually weaker than those found in common bread wheat. However, more recently developed durum cultivars have stronger glutenins and these produce a better loaf of bread, although it is still poor by common wheat standards. Interestingly, the durums with stronger glutenins also generally give pasta products with stronger consistency. Durum grain including both the seed coat and endosperm is physically very hard, being much harder than the hard common wheats. As a consequence durum wheat can be milled to give good yields of semolina, which is the purified middling from the grain. According to Honsey (1976), durum wheat is the preferred class of wheat for the manufacture of pasta products. The rheological properties of stiff doughs made from durum wheat semolina are well suited for the pasta manufacturing process. In addition, durum wheats generally yield a product that is bright yellow in color and when cooked it retains its shape and does not become soft and mushy.

### Classification of wheat endosperm proteins

The classification of wheat endosperm proteins based primarily on solubility was proposed by Osborne in 1907 and is still in use. He proposed four classes of proteins: a) albumins, which are soluble in water, b) globulins, soluble in salt solutions, c) prolamins (gliadin in wheat), soluble in aqueous alcohols, and d) glutenens (glutenin in wheat), insoluble in any of the previous solvents. In practice, wheat

endosperm proteins have often been classified according to their extractability by the different solvents in a sequential extraction. This type of classification can obviously lead to ambiguity because a given protein could be assigned to different classes depending on the order in which the solvents are used. Other extraction conditions, such as temperature, mechanical treatment during extraction, solvent/endosperm ratio, number of extractions with a given solvent, and particle size of the milled endosperm, could also affect the results (Wrigley et al. 1982).

### Storage proteins

Most of the protein in wheat and other cereal grains is storage protein. Gluten is the primary storage protein in the wheat caryopsis and it is composed of two proteins, gliadins and glutenins, which affect dough functionality and quality.

### Gliadin proteins

Gliadins, the prolamine fractions of wheat grains are a mixture of protein components with similar compositions. They are soluble in 70% ethanol. These gliadin proteins are also extracted by dilute acids, such as 0.01 M acetic acid. They have an average molecular weight of approximately 40,000, are single chained, and extremely sticky when hydrated. They also have little or no resistance to extension, and appear to be responsible for the dough's cohesiveness (Hosey, 1976).

Woychik et al. (1961a) classified gliadins into alpha, beta, delta, and omega, according to their relative mobility upon gel electrophoresis with aluminum lactate buffer with pH ranging from 3.1 to 3.3. Alpha, beta, and delta gliadins have about the same amino acid composition, molecular weights ranging from 30,000 to 45,000 and similar N-terminal sequences. Omega gliadins however, show differences in molecular weight, amino acid composition, and N-terminal amino acid sequences. Molecular weights ranging from 65,000 to 80,000 have been reported for omega gliadins (Charbonnier et al., 1980).

### Glutenin proteins

According to Bietz et al. (1973), glutenins form a complex of proteins having high molecular weights and are made up of numerous polypeptidic subunits. They generally represent about 30 to 40% of the total flour protein and are considered the most important contributors to the strength and viscoelastic properties of wheat dough. Glutenins have a broad spectrum of molecular weight, ranging from about 40,000 to over several million, with mean values from 150,000 to 3,000,000 (Jones et al., 1961, Nielsen et al., 1962). There is considerable evidence that these proteins are dispersed and that the molecular weight is an important wheat variety characteristic involved in the bread-making quality (Khan et al., 1977). Rheological properties (related to gluten viscoelasticity or strength) may in fact derive from the distribution of molecular weights. The high molecular weight (HMW) glutenins constitute a small fraction of the storage proteins contained in the seed of

Triticum species. Variation in HMW glutenin composition among common wheat cultivars is known to have a profound effect on their bread making properties (Burnouf and Bouriquet, 1980, Moonen et al., 1983).

Glutenins might be expected to have a similar influence on the bread making quality of durum wheat as well as on the quality of spaghetti (Payne et al., 1984). In both durum and bread wheat species variation in HMW glutenin subunit composition is being used as an aid in cultivar identification.

### Genetic control of endospermic proteins

Research on protein content in wheat indicates that this character is controlled by a great number of genes which can be accumulated in a single progeny through breeding. Research started by Simmonds (1962) and carried on by several other workers (Ewart, 1968, Bushuk and Wrigley, 1971) on amino acid content of the different protein fractions has made it possible to explain the lysine-protein relationship and to suggest new possible methods for selection. Electrophoretic techniques (Jones et al, 1959, Woychik et al., 1961b) have shown that the solubility fractions were heterogeneous and were made up of a considerable number of protein components. Each component resulted as the primary product of a structural gene and so served as a marker for one gene, one chromosome and or one entire genome. This evidence has promoted the use of protein components in studies aimed at sorting out the relationship between protein fractions and components and their pasta and bread-making quality; and solving genetics and breeding

problems. Therefore, it is obvious that the comprehension of mechanisms of regulation, inheritance and genetic control are extremely important from the practical as well as from the theoretical points of view.

### Inheritance of gliadins

Gliadins undoubtedly are proteins which have so far received the greatest attention from scientists. This is due to their technological significance since together with glutenins they are responsible for the viscoelastic properties of gluten and their impact on nutritional value. They are poor in essential amino acids and responsible for the low nutritional value of the total wheat proteins. A peculiar aspect of gliadins is their high level of heterogeneity, which is strictly determined by specific genotypes. A genotype may produce an electrophoretic pattern of gliadins which differ from that of any other genotype with regard to the number, mobility, and intensity of the components. This pattern is independent of environment (Lee and Ronalds, 1967; Wrigley, 1970; Zillman and Bushuk, 1979). Due to this fact, they can be used for: a) variety identification (Ellis, 1971; Wrigley and Shepherd, 1973); b) for detecting species and genera in food preparation (Wrigley, 1977); c) for checking off-types in pure seed production (Appleyard et al., 1979); d) for building up pedigrees (Wrigley and Shepherd, 1974); e) and more generally in a wide range of breeding studies. Different methods have been proposed for expressing the gliadin electrophoretic profile by means of formulae (Bushuk and Wrigley, 1974; Jones et al., 1980; Wrigley et al., 1982). An

interesting research subject is the chromosomal location of genes coding for gliadin components. Boyd and Lee (1967), by using ditelocentric lines of Chinese Spring, were able to show that chromosome 1D was involved in the synthesis of some slow moving components even though they could not exclude the effects of other chromosomes. Wrigley and Shepherd (1974) were able to assign almost all the gliadin components to specific chromosomes and confirmed that the genes coding for these proteins are located in the short arm of the homologous chromosomes 1 and 6. More specifically, chromosomes 1A, 1B, and 1D essentially control delta and omega components whereas alpha and beta components are controlled by chromosomes 6A, 6B, and 6D. The control of chromosomes from groups 1 and 6 for the gliadin components was confirmed for cultivars other than Chinese Spring, thus supporting the idea that synthesis and control of these proteins are governed by a single common model in different materials. By using substitution lines of Cheyenne in Chinese Spring background, Kasarda et al. (1976) confirmed these findings and assigned to specific chromosomes in the groups 1 and 6 as many as 13 out of the 25 components that the proteins of Cheyenne exhibit on electrophoresis in aluminum lactate pH 3.2. In particular, Kasarda et al. (1976) showed that the analysis of the synthesis of A gliadins (a fraction of the alpha gliadins with unusual aggregating properties, which are present only in a few wheat varieties), is controlled by genes on the short arm of chromosome 6A.

Genetic studies using disomic substitutions lines revealed that the gene controlling bands 45 and 42 in durum wheat is located in the



short arm of chromosome 1B (Damidaux et al. 1980a, Joppa et al. 1983b, du Cros et al. 1983).

du Cros and Hare (1985) concluded that two gliadin proteins that are associated with gluten strength in durum wheat were controlled by two alleles at one locus. They also concluded that dosage effects were present with slight dominance of band 42 over band 45.

Gluten strength and the respective electrophoretic bands were found to be linked with the gene Rgl for glume color in durum wheat (Leisle et al. 1981, Hare et al. 1986, McClung and Cantrell 1986). They reported a close association between gluten strength and glume color. White glume color is linked with strong gluten and the presence of band 45, while brown color is linked with weak gluten and band 42. This result indicated that glume color can be a good predictor of gluten quality, and can be used directly in the field to screen early generations. However, this trait is restricted only to those environments where glume color differences can be easily distinguished (McClung and Cantrell, 1986).

### Inheritance of glutenins

Research on glutenins has centered mainly on bread wheat. In this species (genome AABBDD) the HMW subunits of glutenin were shown to be coded by three complex loci (Glu-A1, Glu-B1, and Glu-D1), located on the homeologous chromosomes of group 1 (Bietz et al., 1975, Lawrence and Shepherd, 1981). Payne and Lawrence (1983), following fractionation by sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) of

about 300 common wheat cultivars, identified three different alleles coded by Glu-A1, 11 by Glu-B1, and six by Glu-D1. Later studies permitted detection of further variants and establishment that each common wheat cultivar contains one or two subunits coded by Glu-D1 (Payne et al., 1984). To date, only durums (genome AABB) from Italy have been studied extensively with regard to HMW glutenin subunits composition. In a recent survey involving 52 Italian cultivars, Pogna et al. (1985) distinguished nine different Glu-A1 and Glu-B1 alleles, including three variants in durum wheat which had not been detected among common wheat.

#### Relationship between gliadin components and pasta-making quality

It has been known for a long time that the amount and quality of proteins have considerable influence on the technological properties of wheat. Several reports indicate that pasta-making properties of semolina and bread-making properties of flour are related to the amount of glutenin proteins and more directly to the gliadin - glutenin components (Bushuk and Zillman, 1978; Matsuo and Irvine, 1975; Wasik and Bushuk, 1975). Attempts have been made to correlate the presence or absence of specific gliadin or glutenin components to the pasta- and bread-making quality of durum and bread wheats. Relationships between the electrophoretic profiles of gliadins and the pasta-making quality of several durum wheat varieties have been investigated by Damidaux et al. (1978). The results provided a criterion for dividing durum wheat cultivars into two groups: one group characterized by the presence of

one component in the  $\gamma$ -gliadin region, known as 45, with good pasta-making quality, and the other group including cultivars that are characterized by a component called 42 with poor pasta-making qualities. The results were confirmed by Kosmolak et al. (1980) when analyzing durum cultivars from Canada. Wasik and Bushuk (1975) pointed out that the glutenins extracted from good pasta-making cultivars had a predominant component of molecular weight 53,000, compared to another components of molecular weight 60,000. In bread wheat Payne et al. (1981b) found a positive correlation between the presence of a high molecular weight glutenin subunit controlled by chromosome 1A and the bread-making quality. Relationships between blocks of gliadin components and quality were shown by Sozinov and Popereleya (1980). Wrigley et al. (1977) pointed out the relationship between certain gliadin bands and the characteristics of hardness and dough-mixing strength. In order to explain these results, two different hypotheses were suggested. The first assumes that the gliadin and glutenin coding genes are closed linked to the genes that control the bread and pasta-making quality, while according to the second hypothesis, which receives the most credit, specific gliadin or glutenin components should be directly involved in the bread-making qualities. A variety of rapid and small scale tests have been developed to assess gluten strength in wheat. However, such tests have several disadvantages for testing early generation breeding material, due to the sample size of flour required. Semolina dough strength can be measured by farinographs, mixographs, or by the SDS-sedimentation test (Dexter et al., 1980). The results obtained when using these physical tests are often influenced by

environmental factors (Damidaux et al. 1980a, du Cross et al, 1982). Therefore, breeders need reliable and quick techniques that can evaluate the genetic potential for gluten strength without a significant influence from the environment. These techniques should be able to "finger-print" the genes by identifying gene products, such as the protein involved in the determination of gluten strength. The identification of such gene products is now possible by using electrophoretic techniques as reported by Damidaux et al. (1978) and Kosmolak et al. (1980). They concluded that the presence of a specific gliadin is correlated with gluten strength in durum wheat, and that an allelic band may be correlated with weak gluten and poor cooking characteristics. Breeders can select for durum wheat quality on the basis of electrophoresis of gliadin subunits. This technique offers the advantages that the genotype of the seed is indicated, without environmental influence, and that very small samples are required for this analysis. So in the case of early generation screening half of the grain can be used for electrophoresis, leaving the germ for further propagation.

### **Pigment content**

The yellow color of semolina is determined by the level of xanthophyll pigments present in the endosperm of durum wheat. If the enzyme lipoxygenase is at low levels, the semolina is highly correlated with pasta color (Irvine and Anderson 1953).

Heritability values for semolina color are high (Braaten et al.; Johnston et al. 1983). The gene action is primarily additive and the intensity of the expression is only slightly influenced by the environment.

Walsh (1970), described three methods for evaluating semolina color: Visual evaluation, pigment extraction, and light reflectance. Pigment extraction is the most reliable method, however, requires 10 g of sample, is destructive and very slow.

A reflectance colorimeter method described by Johnston et al. (1981), requires only 1 to 2 g of sample and it is suitable for screening in early generations. According to Johnston et al. (1980) and Quick et al. (1980b), this method permitted the separation of high and low color in a segregating population where sample sizes were limited. They concluded that early generation selection for semolina color is possible by using this kind of rapid and efficient procedure.

## BIBLIOGRAPHY

- American Association of Cereal Chemistry. 1976. Approved Methods. Pigments, method 14-50.
- Appleyard, D.B., J. McCausland, and C.W. Wrigley. 1979. Checking the identity and origin of offtypes in the propagation of pedigree wheat seed. *Seed Sci. Technol.* 7:459-466.
- Bietz, J.A., M.W. Shepherd, and J.S. Wall. 1975. Single-kernel analysis of glutenin: Use in Wheat genetics and breeding. *Cereal Chem.* 52:513-517.
- Bietz, J.A., R.F. Huebner, and J.S. Wall. 1973. Glutenin the strength protein of wheat flour. *Baker's Dig.* 47:26-31.
- Boyd, W.J.R. and J.W. Lee. 1967. The control of wheat gluten strength at the genome and chromosome levels. *Experientia.* 23:332-333.
- Braaten, M.O., K.L. Lebsack, and L.D. Sibbit. 1962. Inter-generation relations of physical properties of dough and carotenoid pigment content in durum wheat. *Crop Sci.* 2:277-280.
- Burnouf, T. and R. Bourniquet. 1980. Glutenin subunits of genetically related european hexaploid wheat cultivars; they relation to bread-making quality. *Theor. Appl. Genet.* 58:107-111.
- Bushuk, W. and C.W. Wrigley. 1971. Glutenin in developing wheat endosperm. *Aust. J. Biol. Sci.* 16:305-317.
- Bushuk, W. and C.W. Wrigley. 1974. Proteins. Composition, structure and function. In: Inglett, G.E. (ed.): *Wheat Production and Utilization*: 119-145.
- Bushuk, W. and R.R. Zillman. 1978. Wheat cultivar identification by gliadin electrophoregrams. I. Apparatus, method and nomenclature. *Can. J. Plant Sci.* 58:505-515.
- Charbonnier, L., T. Terce-Laforgue, and J. Mosse. 1980. Same physicochemical properties of Triticum vulgare, and gliadins. *Ann. Technol. Agric.* 29:175-190.
- Damidaux, R., J.C. Autran and P. Fillet. 1980a. Gliadin electrophoregrams and measurments of gluten viscoelasticity in durum wheat. *Cereal Foods World.* 25:754-756.

- Damidaux, R., J.C. Autran, P. Grignac, and P. fillet. 1978. Mise en evidence de relations applicables en selection entre l'electrophoregramme des gliadines et les proprietes viscoelastiques du gluten de T. durum Desf. C. R. Acad. Sci. Paris. 287:701-704.
- Dexter, J.E., R.R. Matsuo, F.G. Kosmolak, D. Leisle, and B.A. Marchylo. 1980. The suitability of the SDS-sedimentation test for assessing gluten strength in durum wheat. Can. J. Plant Sci. 60:25-29.
- du Cros, D.L., and R.A. Hare. 1985. Inheritance of gliadins proteins associated with quality in durum wheat. Crop. Sci. 25:674-677.
- du Cros, D.L., L.R. Joppa, and C.W. Wrigley. 1983. Two-dimentional analysis of gliadins proteins associated with quality in durum wheat: Chromosomal location of genes for their syntesis. Theor Appl. Genet. 66:297-302.
- Ellis, R.P. 1971. The identification of wheat varieties by the electrophoresis of grain proteins. J. Nat. Inst. Agric. Bot. 12:223-235.
- Ewart, J.A.D. 1968. Fractional extraction of cereal flour proteins. J. Sci. Food Agric. 19:241-245.
- Hare, R.A., D.L. du Cros, and W.C. Barnes. 1986. Genetic linkage between glume color and certain gliadin proteins in durum wheat. Crop. Sci. 26:831-833.
- Honsey, R.C. 1976. World proteins needs. In: Mc Graw-Hill Yearbook of Science and Technology. McGraw- Hill: New York. pp. 61-77.
- Irvine, G.N., and J.A. Anderson. 1953. Variation in principal quality factors of durum wheat with a quality prediction test for wheat or semolina. Cereal Chem. 30:334-342.
- Johnston, R.A., J.S. Quick, and B.J. Donnelly. 1980. Note on comparision of pigment extraction and reflectance colorimeter methods for evaluating semolina color. Cereal Chem. 57:447-448.
- Johnston, R.A., J.S. Quick, and B.J. Donnelly. 1981. Methods for evaluting semolina color in early generations. Crop. Sci. 21:472-473.
- Johnston, R.A., J.S. Quick, and J.J. Hammond. 1983. Inheritance of semolina color in six durum wheat crosses. Crop Sci. 23:607-610.
- Jones, R.W., G.E. Babcock, N.W. Taylor, and F.R. Senti. 1961. Molecular weights of wheat gluten fractions. Arch. Biochm. Biophys. 94:483-493.

- Jones, R.W., G.L. Lookhart, S.B. Hall, and K.F. Finney. 1980. Polyacrylamide gel electrophoretic pattern of gliadin proteins from the 80 most commonly grown U. S. wheat varieties. 65th. Ann. Meet. Amer. Assoc. Cereal Chem. 59:181-188.
- Jones, R.W., N.W. Taylor, and F.R. Senti. 1959. Electrophoresis and fractionation of wheat gluten. Arch. Biochem. Biophys. 84:363-376.
- Joppa, L.R., K. Khan, and N.D. Williams. 1983b. Chromosomal location of genes for gliadin polypeptides in durum wheat Triticum turgidum L. Theor. Appl. Genet. 64:289-293.
- Kasarda, D.D., J.E. Bernardin, and C.C. Nimmo. 1976. Wheat Proteins. In: Advances in Cereal Science and Technology. (Y. Pomeranz, ed.). Am. Assoc. Cereal Chem., St. Paul, Minnesota. pp.158-236.
- Kasarda, D.D., J.E. Bernardin, and C.O. Qualset. 1976. Relationship of gliadin components to chromosomes in hexaploid wheats (*T. aestivum* L.). Proc. Nat. Acad. Sci. (Wash.). 73:3646-3650.
- Khan, K., and W. Bushuk. 1977. Glutenin: Structure and fuctionality in bread-making. Proc. 10th. Conf. Wheat Utilization Research. Tucson. 101-115.
- Kosmolak, F.G., J.E. Dexter. R.R. Matsuo, D. Leisle, and B.A. Marchylo. 1980. A relationship between durum wheat quality and gliadin electrophoregrams. Can. J. Pl. Sci. 60:427-432.
- Lawrence, G.J., and K.W. Shepherd. 1981. Chromosomal location of genes controlling seed proteins in species related to wheat. Theor. Appl. Genet. 59:25-31.
- Lee. J.W., and J. A.Ronalds. 1967. Effect of environment on wheat gliadin. Nature. 231:844.
- Leisle, D., F.G. Kosmolak, and M. Kovac. 1981. Association of glume color with gluten strength and gliadin proteins in durum wheat. Can. J. Plant Sci. 61:149-151.
- Matsuo, R.R., and G.M. Irvine. 1975. Rheology of durum wheat products. Cereal Chem. 52:131-135.
- McClung, A.M., and R.G. Cantrell. 1986. Inheritance of glume color, color and gluten strength in durum wheat. Euphytica 35:885-890.
- Moonen, J.H.E., A. Scheepstra, and A. Graveland. 1983. The positive effects of the high molecular weight subunits 3 + 10 and 2\* of glutenin on the bread-making quality of wheat cultivars. Euphytica. 32:735-742.



- Moorris, R, and E.R. Sears. 1967. The cytogenetics of wheat and its relatives. In: K.S. Quisenberry and L.P. Reitz (eds), Wheat and Wheat improvement, pp. 19-87. American Society of Agronomy, Madison, WI.
- Nielsen. H.C., G.E. Babcock, and F.R. Senti. 1962. Molecular weight studies on glutenin before and after disulfide-bond splitting. Arch. Biochem. Biophys. 96:252-258.
- Osborne, R.C. 1907. The proteins of wheat kernel. Carnegie Institute. Washington Publ. 84:1-119.
- Payne, P.I., and C.J. Lawrence. 1983. Catalogue of alleles for the complex gene loci, GLU-A1, GLU-B1, and GLU-D1 which code for high-molecular subunits of glutenin in hexaploid wheat. Cereal Res. Comm. 11:29-35.
- Payne, P.I., E.A. Jackson, and L.M. Holt. 1984. The association between  $\gamma$ -gliadin 45 and gluten strength in durum wheat varieties: A causal effect or the result of genetic linkage?. J. Cereal Sci. 2:73-81.
- Payne, P.I., K.G. Corfield, L.M. Holt, and J.A. Blackman. 1981b. Correlation between the inheritance of certain high molecular weight subunits of glutenin and bread-making quality in progenies of six crosses of bread wheat. J. Sci. Food Agric. 32:51-60.
- Pogna., N., F. Mellini, and D.B. Peruffo. 1985. The role of PAGE in varietal identification and in developing varieties of durum wheat with good spaghetti-making quality. Inter. Symp. on Durum Wheat; Applied Research and Export of Industrial Products, Foggia, Italy. Genet. Agri. Monograph. VII:199-212.
- Quick, J.S., R. A. Johnston, V.S. Vasiljevic, and Y. Holm. 1980b. Recent advances in evaluation of durum wheat quality. Proc. 3rd. Wheat Conf. Madrid, pp. 729-739.
- Simmonds, D.H. 1962. Variation in the amino acid composition of Australian wheats and flour. Cereal Chem. 39:445-455.
- Sozinov, A.A., and F.A. Popereleya. 1980. Genetic classification of prolamines and its use for plant breeding. Ann. Technol. Agric. 29:229-245.
- Walsh, D.E. 1970. Measurement of spaghetti color. Macaroni J. 52:20-22.
- Wasik, R.J. and W. Bushuk. 1975. Relation between molecular weight distribution of endosperm proteins and spaghetti-making quality of wheats. Cereal Chem. 52:322-328.

- Woychik, J.A., J.A. Boundy, and R.J. Dimler. 1961a. Starch gel electrophoresis of wheat gluten proteins with concentrated urea. *Arch. Biochem. Biophys.* 94:477-482.
- Woychik, J.H., J.A. Boundy, and R.J. Dimler. 1961b. Amino acid composition of proteins in wheat flour. *J. Agr. Food Chem.* 9:307-310.
- Wrigley, C.W., and K.W. Shepherd. 1973. Electrofocusing of grain proteins from wheat genotypes. *Ann. N. Y. Acad. Sci.* 209:223-235.
- Wrigley, C.W., and K.W. Shepherd. 1974. Identification of Australian wheat cultivars by laboratory procedures: Examination of pure samples of grain. *Aust. J. Exp. Agr. Anim. Husb.* 14:796-804.
- Wrigley, C.W., J.L. Autran, and K.W. Bushuk. 1982. Identification of cereal varieties by gel electrophoresis of the grain proteins. *Advances in Cereal Science and Technology*. Vol. 5. Am. Assoc. Cereal Chem. St. Paul, Minnesota.
- Wrigley, C.W. 1970. Protein mapping by combined gel electrofocusing and electrophoresis. Application of the study of genotypic variations in wheat gliadins. *Biochem. Gen.* 4:509-516.
- Wrigley, C.W. 1977. Characterization and analysis of cereal products in foods by protein electrophoresis. *Food Technol. Aust.* 29:17-20.
- Zillman, R.R., and W. Bushuk. 1979. Wheat cultivar identification by gliadin electrophoregrams. III. Catalogue of electrophoregram formulas of Canadian wheat cultivars. *Canad. J. Plant. Sci.* 59:287-298.