Effect of Water Content on Changes in Semolina Proteins During Dough-Mixing

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ABSTRACT

Semolina from a 1 CW amber durum wheat composite was mixed in the farinograph at 30 and 50°C over a range of absorptions that corresponded approximately to a range of water contents from pasta doughs to bread doughs. At both 30 and 50°C, farinograph maximum consistency decreased rapidly as absorption was increased, while mixing time reached a minimum value around 45% absorption and increased when absorption was increased further. Results from determinations of dough protein extractability in dilute acetic acid and Osborne protein solubility fractionations showed that gluten breakdown, as measured by increased gluten protein solubility, did not occur at absorptions below 45%. Gel filtration elution profiles of acetic acid extracts showed that at absorptions of 45% and above, the proportion of protein in the high molecular weight fraction increased significantly. These data suggest that gluten development during dough mixing at absorptions below 45% is limited. For a series of wheats representing a variety of gluten strengths, strong gluten wheats were found to achieve maximum gluten breakdown during mixing at higher dough water contents than were weak gluten wheats.

Previous studies have shown that the amount of protein extractable from bread doughs by dilute acetic acid increases with mixing (Mecham et al 1962, 1963, 1965; Tanaka and Bushuk 1973; Tsen 1967). The rate of increase depends on mixing characteristics, which in turn are related to gluten protein properties. Although gluten protein properties also influence pasta dough mixing characteristics (Dexter and Matsuo 1977b, 1978a; Irvine et al 1961; Matsuo and Irvine 1975; Matsuo et al 1972), a recent investigation showed that differences in solubility changes of semolina proteins were not detectable during spaghetti processing for wheats possessing a range of gluten quality (Dexter and Matsuo 1977a). Furthermore, rather than an increase in protein extractability, a slight decrease was observed, possibly due to protein denaturation at the temperature of spaghetti processing (50°C). From these results, the water content of pasta doughs (ranging from about 27 to 35% absorption) was hypothesized to be insufficient to allow formation of a continuous gluten matrix, rendering them less susceptible to gluten breakdown than bread doughs. This was confirmed subsequently by a scanning electron microscopy study of pasta dough at various stages of spaghetti processing that showed that formation of a continuous network of protein sheets and fibrils did not occur in pasta doughs to the same extent as in bread doughs (Matsuo et al 1978).

The aim of this investigation was to ascertain the relationship between the water content of semolina-water doughs and gluten development, as reflected by changes in gluten protein solubility during dough mixing in the farinograph. In addition, the effects of variety and wheat class were determined for a series of samples representing a range in gluten characteristics.

MATERIALS AND METHODS

Wheat Samples

A grade No. 1 Canada Western (1 CW) amber durum wheat *(Triticum durum* Desf.) composite sample from the 1976 crop year was used for preliminary work. The other durum wheat samples used were grown in 1975 at the Agriculture Canada experimental plots at Glenlea, Man. They comprised five Canadian cultivars (Coulter, Macoun, Wakooma, Wascana, and Stewart 63) and one Argentine cultivar (Candeal Seleccion Le Pravision). Two Canadian hard red spring (HRS) wheat (*T. aestivum* L. em Thell) cultivars, Manitou and Glenlea, were included for comparative purposes. These samples were both composites of a number of samples grown across Western Canada during 1975.

Milling

One-kilogram samples of each wheat were tempered overnight and milled at 16.5% moisture in an Allis-Chalmers laboratory mill in conjunction with a laboratory purifier (Black 1966). The mill room was controlled for temperature (22°C) and humidity (60%). The long-milling flow of Black (1966) was modified as described previously (Dexter and Matsuo 1978b) to yield a durum wheat semolina extraction rate of about 70% and an HRS wheat farina extraction rate of about 63%.

Quality Characteristics

Some quality data for the samples used in this study are presented in Table I. Standard AACC (1962) procedures were used for semolina ash, semolina yellow pigment, and spaghetti yellow pigment. Protein contents were determined by the Kjeldahl procedure (N × 5.7) as modified by Williams (1973). To assess gluten strength, a sensitive strain gauge was used to measure the force required to break a strand of wet gluten, as described by Matsuo (1978). Spaghetti was processed as described previously (Matsuo et al 1972), and spaghetti cooking quality was evaluated on the Grain Research Laboratory Spaghetti Tenderness Testing Apparatus (Dexter and Matsuo 1977b; Matsuo and Irvine 1969, 1971).

Sample Preparation

Fifty grams of semolina or farina (14% mb) was mixed with distilled water in a small stainless steel farinograph bowl (59 rpm) using the rear sensitivity setting (Irvine et al 1961). Mixing time, reported in minutes, was the time required to reach the peak of the curve. Samples were mixed over an absorption range that corresponded approximately to the range of water contents from pasta doughs to bread doughs. The absorptions used and their equivalent dough water contents are listed in Table II. In addition to mixing at 30°C, the normal bowl temperature, a series of samples was also mixed at 50°C, the temperature used in the previous study for spaghetti processing (Dexter and Matsuo 1977a). After mixing, the doughs were frozen rapidly and freeze-dried. The freeze-dried samples, each semolina and each farina, were ground in a coffee grinder and passed through a U.S. No. 100 sieve to insure uniformity of particle size from sample to sample.

Protein Extractions

One-gram samples were extracted in 17 ml of 0.05*M* acetic acid for 15 min in a Potter and Elvejhem homogenizer (Tanaka and Bushuk 1973) and centrifuged. The pellets were resuspended in

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TABLE I
Quality Data for Wheats Used in This Study

Property[a]	1976 Composite	Wheat Description							
		Coulter	Macoun	Wakooma	Candeal	Wascana	Manitou	Stewart 63	Glenlea
Wheat									
Class	Durum	Durum	Durum	Durum	Durum	Durum	HRS[b]	Durum	HRS
Grade	1 CW[c]	2 CW	3 CW	2 CW	2 CW	3 CW	1 CW	2 CW	1 Utility
Semolina									
Ash (%)	0.74	0.69	0.69	0.66	0.73	0.62	0.44	0.63	0.54
Protein (%)	14.2	13.5	13.2	13.4	12.3	13.7	13.6	13.1	11.8
Gluten strength (dynes × 10^4)	11.3	12.2	11.9	11.9	13.9	11.6	11.7	6.3	13.5
Yellow pigment (ppm)	7.00	5.58	6.80	5.41	5.47	6.49	2.21	3.83	1.91
Spaghetti									
Yellow pigment (ppm)	5.45	4.27	5.24	4.10	3.34	4.80	1.47	3.08	1.30
Pigment loss (%)	22.1	23.5	23.0	24.2	39.0	26.1	33.5	19.7	32.1
Cooking quality[d]									
Normal time	43.0	26.7	29.7	24.0	24.6	23.6	27.8	16.5	10.2
Overcooked 5 min[e]	29.5	25.6	22.1	14.6	10.0	4.9	...	...	...

[a]All analytic values are expressed on 14% mb.
[b]HRS = hard red spring.
[c]CW = Canada Western.
[d]Cooking quality expressed as recovery/compressibility × tenderness index. Normal cooking time was about 13 min.
[e]No value indicates strands were completely compressed and exhibited no recovery.

acetic acid and the samples recentrifuged. The two supernatants from each sample were combined, freeze-dried, weighed, and their protein contents (N × 5.7) were determined by the micro-Kjeldahl procedure of Mitcheson and Stowell (1970), with slight modifications. Reproducibility was about 5% for replicate extractions of each sample.

TABLE II
Absorptions (14% mb) Used for Semolina-Water Dough Preparation and Their Equivalent Dough Water Content

Absorption (%)	Dough Water Content (%)
30	33.8
35	36.3
40	38.6
45	40.7
50	42.7
55	44.5
60	46.3

TABLE III
Acetic Acid Extractability of Proteins in Semolina-Water Doughs From 1 CW Durum Wheat Composite Semolina Following Mixing in Farinograph[a]

Absorption (%)	30°C		50°C
	Mixed 5 min	Mixed 30 min	Mixed 30 min
30	71.4	66.8	65.4
35	70.6	69.2	68.3
40	71.5	73.5	60.8
45	73.2	77.7	61.2
50	76.7	80.2	59.0
55	79.7	86.7	59.1
60	81.0	88.6	63.3

[a]Extractability of original semolina protein was 71.4%. Results expressed as percent total nitrogen.

Protein Solubility Fractionations

Some samples were fractionated into five protein solubility classifications by the modified Osborne procedure of Chen and Bushuk (1970). Each fraction was freeze-dried and weighed and the protein content (N × 5.7) determined by the modified micro-Kjeldahl method of Mitcheson and Stowell (1970). Replicated results agreed to about 5% for each solubility fraction.

Gel Filtration

Gel filtration was performed on a 4.5 × 50-cm bed of Sephadex G-150 with upward flow at 60 ml/hr. The column was calibrated for molecular weight by the method of Whitaker (1963) using previously described standards (Dexter and Matsuo 1977a). Samples for analysis (2.8 g) were extracted in 50 ml of 0.05*M* acetic acid for 15 min in a Potter and Elvejhem homogenizer (Tanaka and Bushuk 1973) and centrifuged in a Beckman L-2 ultracentrifuge for 20 min at 150,000 × *g*. Forty milliliters of extract (200–250 mg of protein) was injected onto the column for each trial. The effluent was monitored at 280 nm in an LKB 8300 Uvicord II ultraviolet analyzer. Fractions were collected, freeze-dried, and weighed and their nitrogen contents determined by the modified micro-Kjeldahl method of Mitcheson and Stowell (1970). Recovery of total nitrogen from the column was approximately 90%.

RESULTS AND DISCUSSION

Farinograph Characteristics

Figure 1 illustrates the effect of temperature and absorption on the farinograph mixing curves obtained for the semolina from the 1 CW amber durum composite. Previously it was demonstrated that either increasing absorption over a narrow range or increasing temperature results in a concomitant decrease in farinograph mixing time and maximum consistency (Bayfield and Stone 1960, Hlynka 1962, Irvine et al 1961, Moore and Herman 1942, Skovholt and Bailey 1932). In the current study, however, where the range in absorptions was much wider, at both 30 and 50°C, farinograph mixing time was found to be at its minimum value around 45% absorption (dough water content 40%), and increased when absorption was raised or lowered from that level. At equivalent absorptions, the farinograph maximum consistency and mixing time were greater at 30 than at 50°C. The shape of the curves appeared to change at absorptions above 45%, becoming more rounded at peak consistency. This suggested that a fundamental

change is occurring in dough structure at this moisture level, possibly reflecting the attainment of gluten development. Previous work supported this hypothesis, showing that although the amount of bound water in dough is about 25% (Bushuk and Mehrotra 1977, Davies and Webb 1969, Lee 1970, Toledo et al 1968, Vail and Bailey 1940), not until 12–13% free water is present (36–40% dough water content) does sufficient fluidity exist to allow the formation of a truly extensible dough system (Daniels 1975). Furthermore, Wood et al (1972) showed that the critical dough water content for maximum lipid binding in work-free doughs is also about 40%, which they postulate to be a result of the spontaneous rearrangement of wheat proteins caused by water during gluten structure development.

Protein Solubility and Molecular Weight Distribution

Changes in protein extractability in acetic acid following mixing for various times in the farinograph were determined for the 1 CW composite semolina at 30 and 50°C (Table III). When mixed at 30°C, significant gluten breakdown (increasing solubility) was not evident until the absorption was raised to 45%. Protein extractability at 45% absorption and above increased significantly

TABLE IV
Osborne Solubility Distribution of Proteins From 1 CW Durum Wheat Composite Semolina and Some Selected Semolina-Water Farinograph Doughs[a]

Sample Description	Albumins (%)	Globulins (%)	Gliadins (%)	Soluble Glutenins (%)	Insoluble Residue (%)
Semolina	10.9	7.6	39.0	11.5	28.2
Mixed at 30°C					
30% Absorption	9.0	3.3	42.3	10.2	31.9
40% Absorption	9.8	3.8	43.9	11.1	26.3
50% Absorption	8.6	4.8	47.3	11.7	25.1
60% Absorption	10.5	5.0	39.6	14.2	27.1
Mixed at 50°C					
30% Absorption	8.5	2.9	39.2	9.4	35.4
45% Absorption	9.1	4.1	32.6	6.7	44.8
60% Absorption	9.9	4.1	31.9	7.8	42.3

[a]Results expressed as percent total nitrogen. Doughs mixed 30 min.

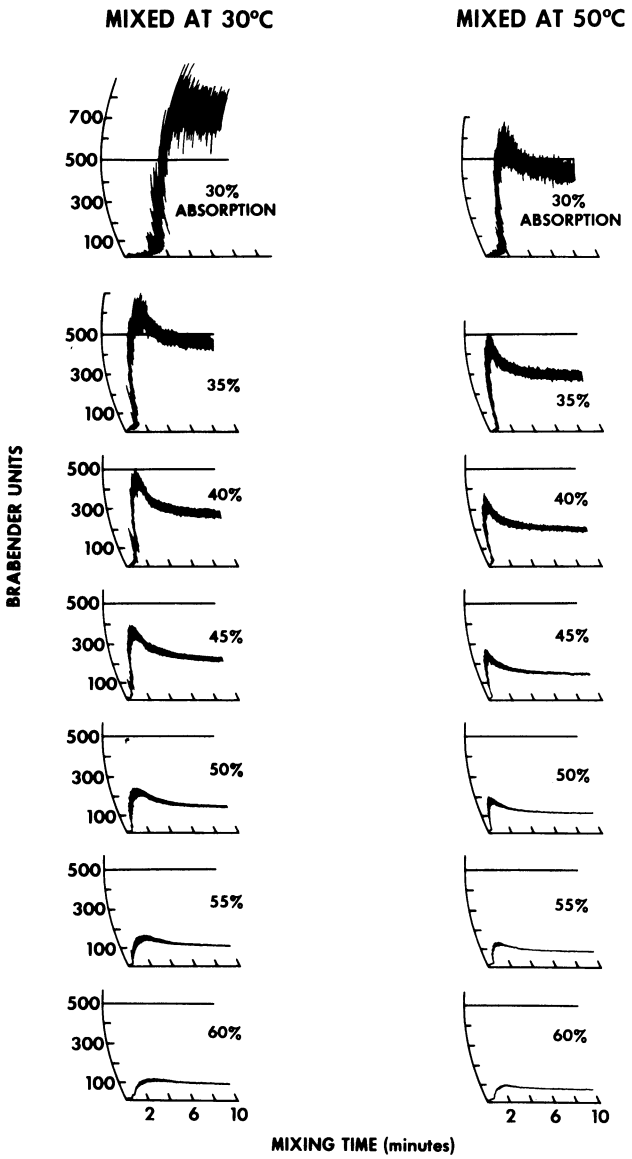

Fig. 1. Effect of absorption at two temperatures on farinograph mixing characteristics for semolina from 1 CW amber durum wheat composite from 1976 crop year. Farinograms obtained using rear sensitivity setting as described by Irvine et al (1961).

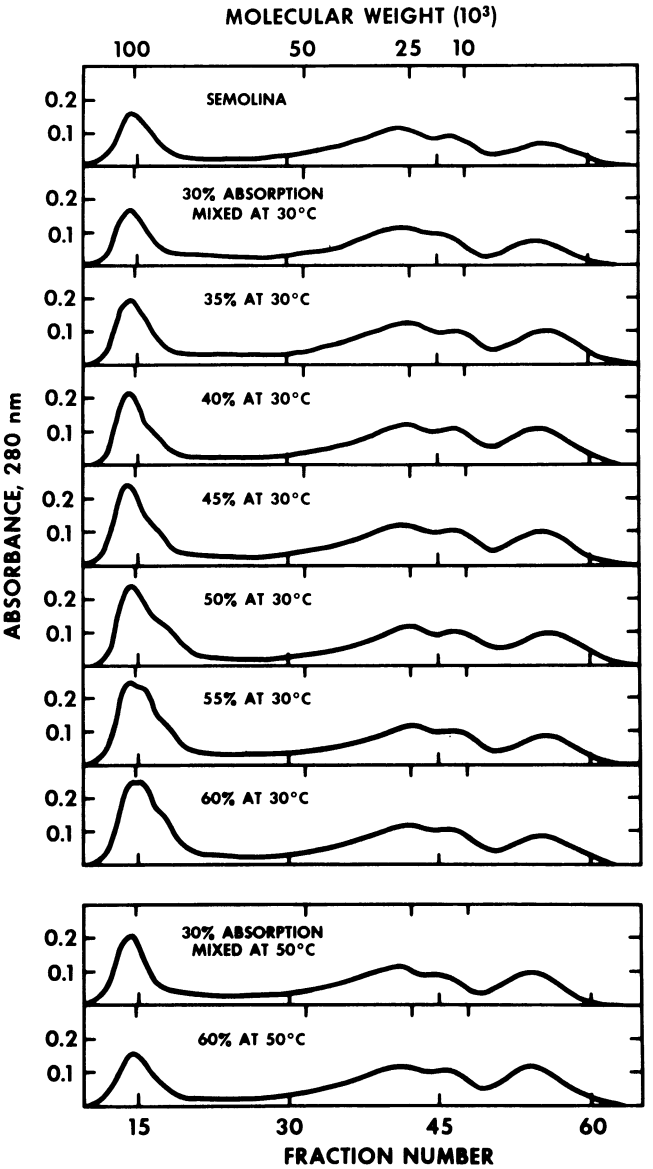

Fig. 2. Gel filtration elution profiles (Sephadex G-150) of some acetic acid protein extracts prepared from 1 CW amber durum wheat semolina and various farinograph doughs.

with increased mixing time. These trends suggest that at 30°C, gluten development is not initiated during dough mixing at absorptions below 45%. In contrast, when mixed at the 50°C laboratory pasta processing temperature (Dexter and Matsuo 1977a), protein extractability decreased at each absorption compared with that of the semolina, especially at 40% absorption and above, presumably due to protein denaturation. The decrease in solubility arising from protein denaturation would offset any increase in solubility due to gluten breakdown.

Further information was gained on the nature of the proteins involved in the solubility changes by subjecting some of the samples to Osborne solubility fractionations (Table IV). At both 30 and 50°C for doughs mixed at 30% absorption, we found a marked decrease in the proportion of nongluten proteins, especially the globulins, and an increase in insoluble residue compared with the semolina. This result was similar to that found for pasta doughs during spaghetti processing at 50°C (Dexter and Matsuo 1977a). At higher absorptions, however, temperature had a marked effect on the protein distributions of the doughs. At 30°C, gliadins and soluble glutenins increased somewhat, while residue protein decreased. This result was in agreement with the results of Tanaka and Bushuk (1973). At 50°C, a decrease in the proportion of gliadins and soluble glutenins appeared to be significant, concomitant with a large increase in residue protein. Since gluten denaturation is taking place at 50°C only at absorptions greater than pasta absorptions, one may infer that rearrangement of gluten proteins (gluten development) occurs only at absorptions greater than pasta absorptions.

Gel filtration profiles of acetic acid extracts for doughs mixed at 30°C provided corroborative evidence for incomplete gluten development below 45% absorption. At 30°C, increase in high molecular weight material as shown in Fig. 2 was not significant until an absorption of 45% was reached. Determination of the nitrogen present (Table V) in fractions 10–27, the high molecular weight fractions, confirmed that until an absorption of 45% was reached, there was no proportionate increase, but as the absorption was increased further to 60%, the value increased rapidly, both in proportion of total nitrogen and in actual amount. Although the lower molecular weight fraction decreased in proportion to the total extractable protein, it exhibited an increase in actual amount at absorptions above 45%. This may be at least partially due to the reported equilibrium between glutenin and gliadin fractions that Dalek-Zawistowska et al (1975) observed. The gel filtration profiles of acetic acid extracts from 50°C doughs mixed at 30 and 60% absorption (Fig. 2), and their nitrogen distributions (Table V), were consistent with their protein solubility data (Tables III and IV).

Varietal Differences

The effect of absorption on farinograph mixing time at 30°C (Fig. 3) and acetic acid protein extractability after 15 min of mixing (Fig. 4) were determined for a series of wheats with varying gluten characteristics (Table I). Generally the results for each wheat followed the same trends observed for the 1 CW amber durum wheat composite semolina (Fig. 1, Table III). The two strongest gluten varieties (Table I), Candeal and Glenlea, however, appeared to exhibit a minimum mixing time at 50% absorption rather than

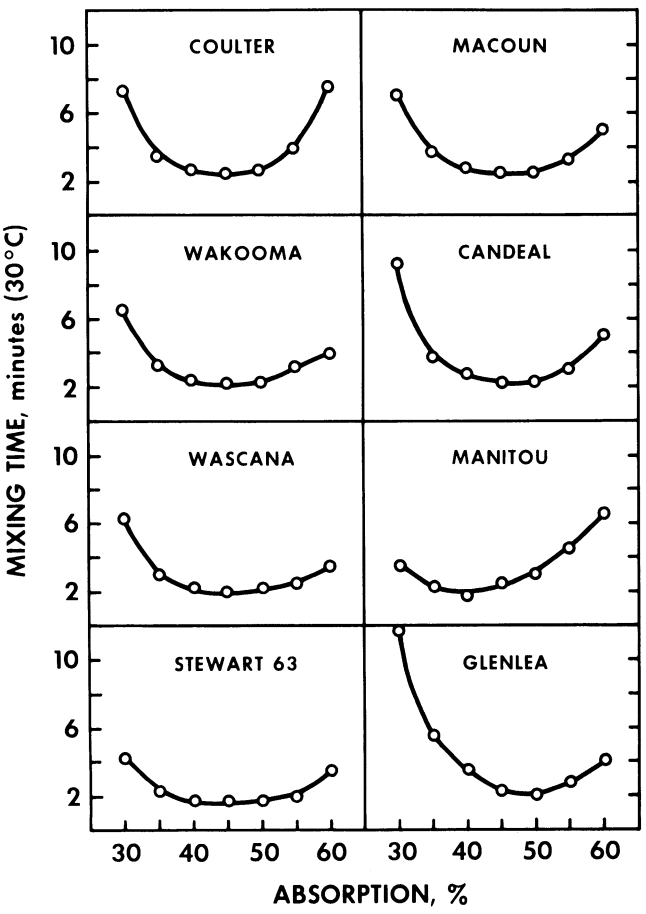

Fig. 3. Effect of absorption on farinograph mixing time at 30°C for semolina from six durum wheat cultivars (Coulter, Macoun, Wakooma, Candeal Seleccion Le Pravision, Wascana, and Stewart 63) and two hard red spring wheat cultivars (Manitou and Glenlea).

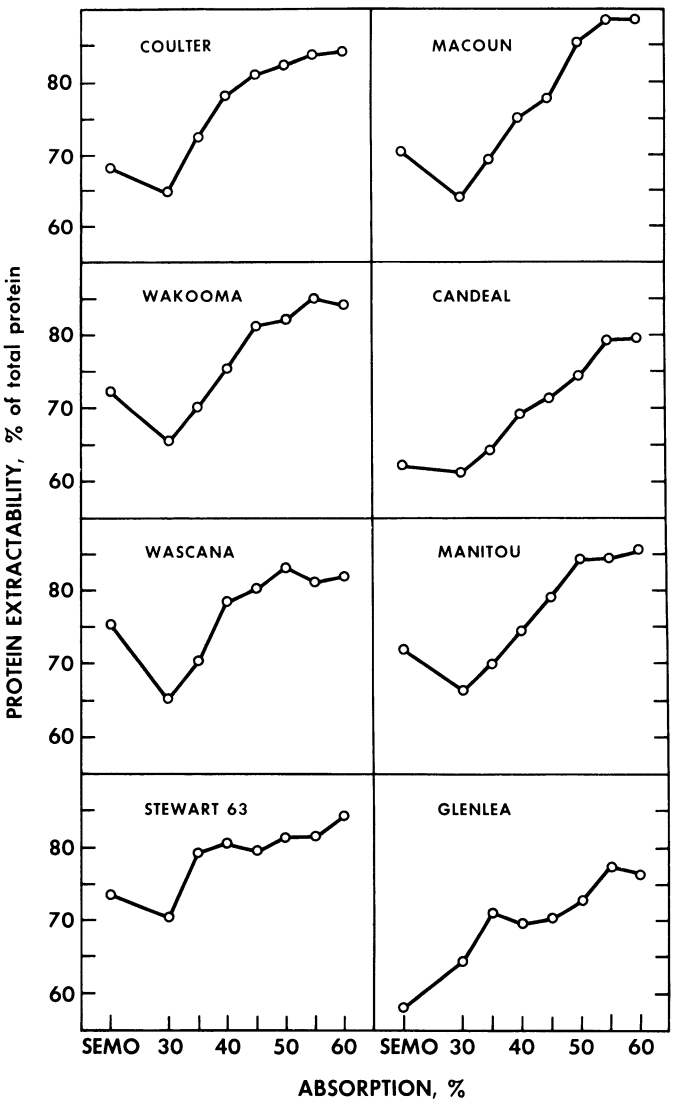

Fig. 4. Effect of absorption on acetic acid extractability of proteins following mixing in farinograph (15 min at 30°C) for semolina from six durum wheat cultivars (Coulter, Macoun, Wakooma, Candeal Seleccion Le Pravision, Wascana, and Stewart 63) and two hard red spring wheat cultivars (Manitou and Glenlea).

TABLE V
Distribution of Acetic Acid-Soluble Proteins Within Gel Filtration Fractions[a] for Semolina-Water Farinograph Doughs Prepared From 1 CW Durum Wheat Composite Semolina[b]

Sample Description	Total Protein in Extract (mg)	% Protein Recovered[c]	
		Fractions 10–27	Fractions 28–63
Semolina	214	24.2	75.8
Mixed at 30°C			
30% Absorption	201	26.2	73.8
35% Absorption	209	26.2	73.8
40% Absorption	222	25.7	74.3
45% Absorption	234	26.3	73.7
50% Absorption	242	28.2	71.8
55% Absorption	262	32.9	67.1
60% Absorption	267	31.1	68.9
Mixed at 50°C			
30% Absorption	197	22.2	77.8
60% Absorption	191	20.2	79.8

[a]See Fig. 2.
[b]Doughs mixed 30 min.
[c]Results normalized to 100% recovery.

45% (Fig. 3), and also did not exhibit maximum gluten breakdown as reflected by increased protein extractability until higher absorption (Fig. 4). In contrast, Stewart 63, a weak gluten variety (Table I), reached a minimum mixing time by 40% absorption and also underwent maximum gluten breakdown at much lower absorptions (35%) than did the other varieties. Therefore, differences in gluten strength may be partially responsible for differences in water requirements for full gluten development. This may be due to differences in their ability to compete for available water, since previous studies in our laboratory have suggested that weak gluten lines tend to have higher wet gluten per unit protein values.[2] The ability of gluten to compete for available water would be expected to influence pasta dough rheologic properties, and could be one of the factors responsible for the relationship between pasta dough farinograph properties and gluten quality (Dexter and Matsuo 1977b, 1978a; Matsuo and Irvine 1970, 1975).

[2]Unpublished data.

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