A New Method for the Estimation of Common Wheat (Triticum aestivum L.) in Pasta Products

F. García-Olmedo and R. García-Faure

Instituto Nacional de Investigaciones Agronómicas, Department of Cereal Chemistry and Technology Madrid-3 (Spain)

An electrophoretic component of the chloroform-methanol (2:1) extracted proteins from common wheat endosperm is barely detected in durum wheat. This sharp interspecific difference is expressed in percent units of another electrophoretic component which is present in both wheat species and designed protein ratio (PR). Macaroni production processing and/or variations of milling yield do not significantly affect the PR. A linear relationship exists between PR and the % of common wheat in a known mixture. Tentative interspecific limits for PR are established from a survey of 79 common wheat and 30 durum wheat varieties. Based on these limits, maximum and minimum possible common wheat content in an unknown mixture is calculated as a function of PR.

Introduction

Several methods have been proposed for detection and estimation of common wheat products in macaroni. These are based on the analysis of certain biochemical differences between the endosperms of common wheat (Triticum aestivum L.) and durum wheat (Triticum durum Desf.). Differences in certain lipid fractions do present exceptions (1-4). This means that some adulterations can go undetected if one of these methods is used alone. A method based on the electrophoresis patterns of water extracted proteins (5) does not show any exceptions, but its sensitivity leaves much to be desired in connection with the estimation of the percentage of T. aestivum products in an unknown mixture. This latter problem is especially difficult because any biochemical character is bound to show some intraspecific variability which will introduce a degree of uncertainty into the estimation. The concurrent use of more than one difference will evidently narrow the interval of uncertainty. but with present methods even this option does not lead to satisfactory results (6). For these reasons, sharper interspecific differences should be sought.

In this paper, the possible use of a new interspecific difference in the estimation of common wheat in macaroni is investigated.

Material and Methods

Samples. Seventy nine T. aestivum varieties and 30 T. durum varieties were used in this study (Tab. 1).

Milling fractions. In the fractionation experiment, samples of 2 kg of each of the wheat varieties were milled in a Buhler experimental mill, to give three break flours and three reduction flours plus bran and shorts. Bran and shorts were pooled and run through the mill again to give three re-milled fractions, flour, bran and shorts.

Extraction and electrophoresis of proteins. 10 gm samples of flour or ground macaroni were defatted with petroleum ether (40 ml) and the remaining solvent allowed to evaporate by spreading over filter paper. Protein was extracted with 40 ml chloroform: methanol (2:1) in a column 2,5 cm in diameter. The solvent was evaporated in vacuo at 45 °C. The resulting protein was dissolved in the electrophoresis buffer at a concentration of 40 mg/ml. The protein solutions were subjected to urea starch-gel electrophoresis in aluminium lactate buffer, pH 3,2, according to WOYCHIK et al. (7). The sample solutions were applied by soaking a piece of filter paper ($10 \text{ mm} \times 3 \text{ mm}$, Whatman No. 3) and inserting in the gel slot.

After electrophoresis, gels were stained in nigrosine solutions (0,05% in acetic acid: water 1:1 v/v) for 21 hours, washed with tap water and destained with 80% ethanol for 5 hours. The gels were transferred to tap water prior to densitometry. Sharp patterns were obtained by using 3 mm thick gels and staining with the underside up. Reflectance densitometry was performed in a Chromoscan densitometer (JOYCE and LOEBL).

Results and Discussion

The chloroform: methanol (2:1) extracted protein from the endosperm of common wheat yields an electrophoretic component (Fig. 1, band A) which is barely detected in the endosperm of durum wheat. The possible use of this sharp interspecific difference for the estimation of common wheat in macaroni has been investigated in this paper.

The amount of component A in the protein extract is expressed relative to a second component (Fig. 1, Band B), which is present in both wheat species. The height of densitogram peak A is measured in percent units of the height of peak B. Measurements are taken using as baseline the densitogram curve immediately to the right of peak A. Determination of the

 Table 1
 PR of flour and corresponding pasta

Variety	. PR	
	Flour	Pasta
Cabezorro	86	90
Dr. Mazet	78	77
Aragon-03	85	85



Fig. 1 Electrophoretic patterns of chloroform:methanol (2:1) proteins from T. aestivum and T. durum flours. Band A is quantitated by expressing peak height of A as percent units of peak height of B.



Fig. 2 Ash and PR versus milling yield in one T. aestivum and one T. durum variety.

described coefficient, designed PR (protein ratio) is reprodu-

cible (var. coeff. $\frac{S}{X}$ < 0,05).

The dependence of PR on milling yield has been studied in connection with the setting of tentative limits for intraspecific variability of PR in both wheats. In Fig. 2, nine milling fractions from each of two varieties, one *T. durum* and one *T. aestivum*, have been arranged in order of ash content from low to high, and the average values for ash and PR have been plotted against milling yield. Normal variations of milling yield do not significantly affect PR values. Furthermore, PR is unchanged by macaroni processing, as can be deduced from Tab. 1.

In order to establish tentative interspecific limits for *PR* values, a survey of flours from 79 *T. aestivum* and 30 *T. durum* varietis

Table 2 PR values of flours from T. aestivum varieties

Variety	PR	Variety	PR
Sample B (HRW)	104	Candeal	79
Cama	100	Yacktana	79
Orca	100	Opal	79
Stella	100	Mocho	78
Tavares	100	Rieti	78
Champlein	96	Selkirk	77
Manella	95	Opal	77
Cleo	93	Kloka	77
Dr. Mazet	93	Colorado	77
Rojo Eslava	92	Gredos	1
Flevina	92	Languedoc	76
Ouern	92	Troll	76
38230-24	91	P. Marsall	75
Jufy I	91	Thatcher	7
Atle	90	Hembrilla	75
Gaby	90	Pais	7
C. Desprez	89	Rojo	73
Marquis	88	Aragon-03	72
Eno	88	Blanguillo	71
Felix	88	Manitou	71
Apollo	87	Canaleja	71
Tadorna	85	Montnegre	70
Jufy	85	Magdalena	69
Tenero	84	Mara	68
Cabezorro	84	Aradi	68
Rojo Basto	84	Libero	67
Mentana	84	Ariana	67
Toseta	83	Dimas	67
Jufi	83	Barbilla	67
Pelle	83	Funo	67
Jeja	83	Caribo	65
Strampelli	83	Niki	65
Sample A (HRW)	83	Virgilio	64
Amyntas	82	Navarro-122	63
Ardica	80	Hybrid 4b	62
Chamorro	80	F. Aurora	61
Estrella	80	Cascon	60
Generoso	80	P. Gemelli	60
Rex	80	Ebro	60
Pane-247	79		



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 Table 3 PR values of flour from T. durum varieties

Variety	PR	Variety	PR	Variety	PR
Electra	18	Hibrido-D	14	Pelissier	4
Claro Fino	18	Farto	13	Golden Ball	3
Alaga	17	Ledesma	13	Capeiti	3
Jerez-36	17	63849-A	13	Mindum	0
Methoni	17	Limnos	13	Stewart-63	0
S. Capelli	17	R. Argelino	12	Lakota	0
Valenciano	16	Sample B-2	9,6	Leeds	0
Capeiti	15	Sample B-1	6,7	Wellis	0
Bidi-17	15	Patrizio-6	6,1	Raspinegro	0
Grifoni	14	DT 191	4	Andalucia	0

Acknowledgments

Fig. 3 PR versus % of T.aestivum in series of binary mixtures (continuous lines). Maximum and minimum possible content of T.aestivum in an unknown mixture as a function of PR (discontinuous lines).

has been carried out. Results are summarized in Tab. 2 and 3. Upper and lower limits for PR are 104 and 60 in *T. aestivum* and 18 and 0 in *T. durum* respectively.

The linear relationship between PR and percent of T.aestivum has been checked in 3 series of binary mixtures of known varieties (Fig. 3).

In unknown mixtures, the maximum and minimum possible *T.aestivum* content can be calculated as a function of the observed *PR* values. This can be done graphically as shown in Fig. 3. In the most unfavorable case, PR = 50, the interval of uncertainty is about 50%, which means that better results are obtained by this method alone than by the combined use of methods previously available in our laboratory (6).

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