

## COMMUNICATION TO THE EDITOR

### A Specific Color Reaction of Albumin, Globulin, and Gliadin Preparations from Wheat<sup>1</sup>

DEAR SIRs:

Pence et al. (1) attempted fractionation of a salt extract of flour by salting out with ammonium sulfate. Fractions precipitating at 0.4M ammonium sulfate concentration were considered as gliadins; a pure albumin preparation was obtained

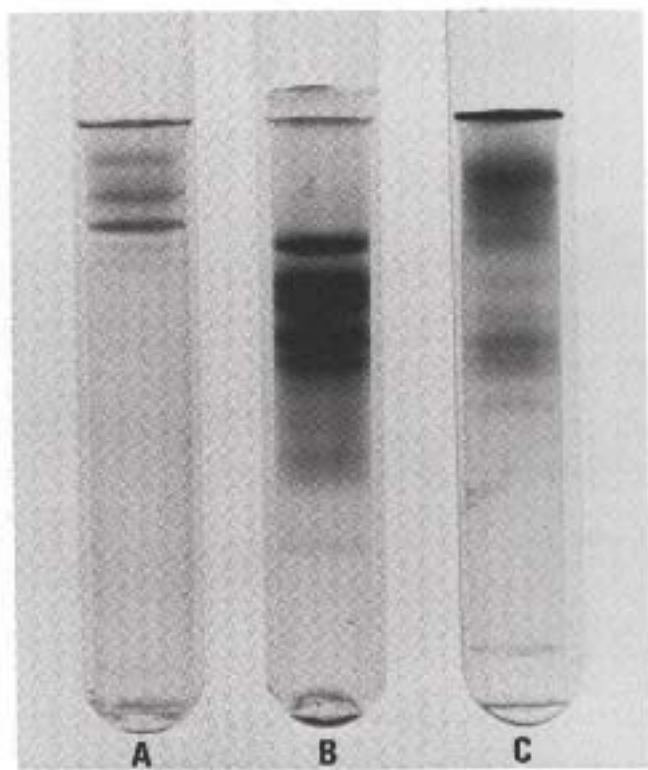


Fig. 1. Disc electrophoretic patterns of gliadin (A), albumin (B), and globulin (C) preparations from bread wheat, var. Turro. The protein fractions were obtained according to Pence et al. (2) with some slight modifications (4,5). Migration was from top to bottom in *Tris*-glycine-buffer, pH 9.5, according to Silano et al. (4,5). Acrylamide concentration was 7.5% (w/v). The time of electrophoretic runs was 135 min. for the gliadins and 95 min. for the albumins and globulins.

<sup>1</sup>This work was supported in part by the "Consiglio Nazionale delle Ricerche," Roma.

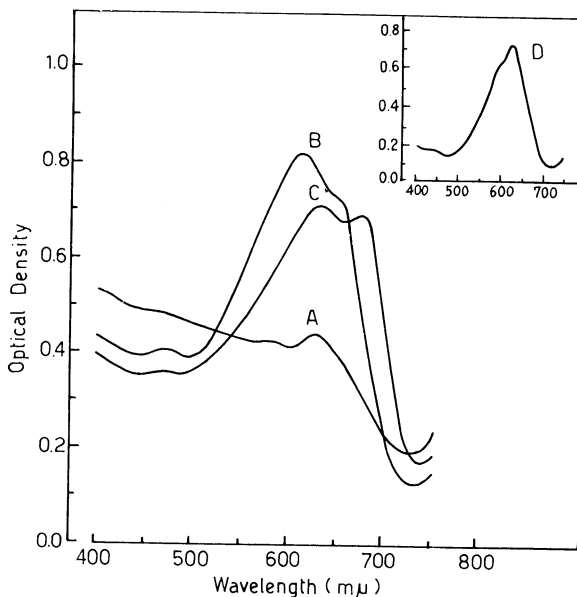


Fig. 2. Comparison of absorption spectra of aniline blue-black (D) and of polyacrylamide gel electrophoretic gliadin (A), albumin (B), and globulin (C) bands stained with aniline blue-black. The polyacrylamide gels reported in Fig. 1 were immersed in acetic acid (7.5% by volume) in the glass cuvet of a Beckman DB spectrophotometer and the absorption spectra were recorded using acetic acid (7.5% by value) as a blank. For the measurement of globulin band spectra, the first 1.5 cm. from the top of the gel was cut off.

between 0.4M and 1.74M, and the globulins were separated overnight by dialysis of the extract against water. The relative proportions of the three types of proteins present in an extract were calculated by Pence et al. (2) from the tryptophan and amide nitrogen values. These chemical criteria to evaluate the purity of protein preparations are based on the high content of tryptophan (1) in albumin fractions and of amide nitrogen (3) in gliadins.

There have also been several attempts at correlating some physicochemical determinations, such as electrophoretic mobility and molecular weight, with the above-mentioned solubility characteristics of wheat proteins. Considerable overlapping of mobility and molecular-weight characteristics of albumins, globulins, and gliadins was shown. Therefore, no method is yet available to rapidly evaluate the purity of wheat protein preparations.

By polyacrylamide gel electrophoresis in a *Tris*-glycine-buffer system (4,5) of the albumin, globulin, and gliadin fractions from a large number of defatted durum and bread wheat varieties, we obtained experimental confirmation of Pence's fractionation and classification as shown in Fig. 1. Treating the gels with aniline blue-black<sup>2</sup>, 0.5% (w./v.) in acetic acid (7.5% by volume), stains gliadin bands (A) reddish-brown, albumin (B) blue-black, and globulins (C) blue-green (Fig. 2). The

<sup>2</sup>Commercial product, available from Canal Industrial Corporation, Rockville, Md.

color differences are clear-cut and reproducible, and presumably depend on structural characteristics which differentiate the above-mentioned protein classes.

We believe that these singular staining properties, coupled with the disc electrophoresis technique (4,5), along with solubility characteristics and chemical differences, can contribute to establishing a rigorous classification of these proteins. Moreover, when bands of different color are evident in the disc electrophoretic patterns of protein preparations from wheat seeds, the presence of different protein classes can be inferred.

V. SILANO  
F. POCCHIARI

Laboratori di Chimica Biologica  
Istituto Superiore di Sanita  
Roma, Italy

#### Literature Cited

1. PENCE, J. W., and ELDER, ANGELINE H. The albumin and globulin proteins of wheat. *Cereal Chem.* 30: 275 (1953).
2. PENCE, J. W., WEINSTEIN, N. E., and MECHAM, D. K. A method for the quantitative determination of albumins and globulins in wheat flour. *Cereal Chem.* 31: 29 (1954).
3. BAILEY, C. H. The constituents of wheat and wheat products. Reinhold: New York (1944).
4. SILANO, V., DE CILLIS, U., and POCCHIARI, F. Varietal differences in albumin and globulin fractions of *Triticum aestivum* and *T. durum*. *J. Sci. Food Agr.* 20: 260 (1969).
5. MINETTI, M., PETRUCCI, T., POCCHIARI, F., SILANO, V., and AVELLA, R. Varietal differences in water-soluble gliadin fractions of *Triticum aestivum* and *T. durum* seeds. *J. Sci. Food Agr.* 22: 72 (1971).

[Received July 10, 1970. Accepted February 9, 1971]