A Starch Granule Protein Associated with Endosperm Softness in Wheat

To The Editor:

Endosperm texture is an important criterion of quality in wheat grain, for it affects both the milling performance of the grain and the quality of the resultant flour. In general, hard wheats are preferred for breadmaking and soft for cake and biscuit (cookie) making.

Despite the importance of wheat endosperm texture, and the volume of literature describing both methods for its measurement and the effects of various factors upon it, an exact description of the molecular basis for regulation of this grain property has proved elusive. Barlow et al (1973) performed micropenetrometer tests on isolated starch and storage protein preparations, and because they observed no differences in hardness for individual components from soft and hard wheats, they concluded that differences in hardness resulted from differences in the nature of the interface between the starch and storage protein. The material at this interface was shown to be rich in water-soluble proteins, which formed an electrophoretically complex group (Barlow et al 1973).

However, the biochemical component(s) responsible for differences in hardness were not identified.

Simmonds et al (1973) were also unable to identify any qualitative difference in the gliadin proteins extracted from hard and soft near-isogenic lines of wheat or in proteins extracted from the glutens from such wheats using 4*M* urea. Furthermore, analysis of 0.01*M* Na pyrophosphate-extractable material did not implicate any specific compound as acting as an adhesive between starch granules and the storage protein matrix (Simmonds et al 1973).

As a result of studies undertaken recently on the nature of the proteins associated with starch granules (Lowy et al 1981, Gough et al 1985), water-washed granules were shown to possess a family of proteins distinct from the bulk of the proteins of flour. Because of their ease of extraction, it was concluded that the smaller members of this protein family are associated with the surfaces of the granules (Lowy et al 1981; P. Greenwell, *unpublished*). The starch granule proteins of the whole family range in relative molecular mass (M_r) from about 5K to about 97K (Fig. 1), and some of these have been implicated in the cake-improving reaction of chlorine treatment of flour (Greenwell et al 1985).

During our studies of these proteins, we examined in greater detail the proteins of lowest M_r. Proteins extracted by swelling the

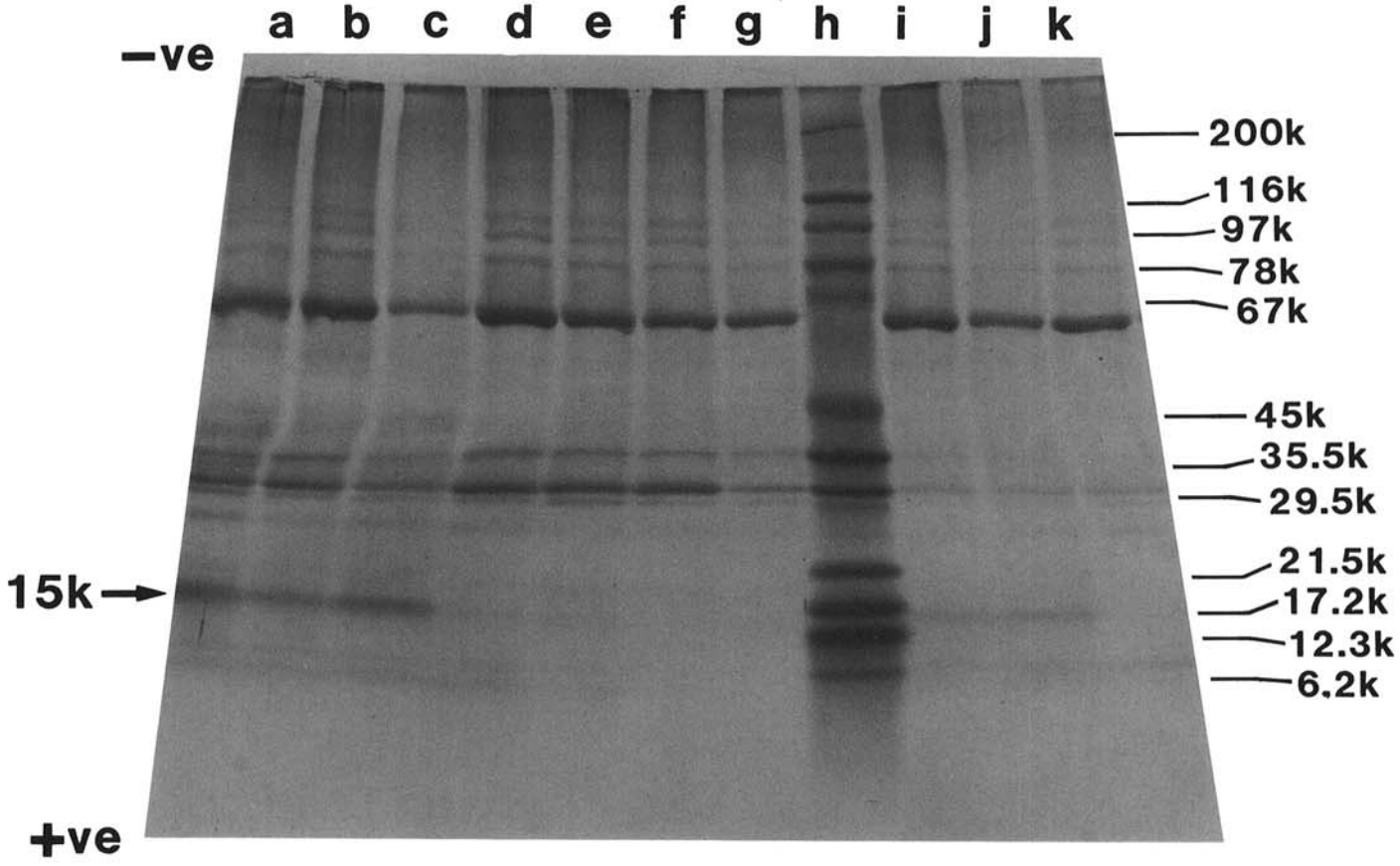

Fig. 1. Fractionation of proteins extracted at 50° C with 1.0 (w/v) sodium dodecyl sulfate (SDS) from starches from soft (tracks **a–c**) and hard (tracks **d–g**) UK-grown wheats, from a soft French wheat (track **i**), and from mixed grain samples of Canada Eastern Winter White (track **j**) and Canada Western Red Spring wheat (track **k**) by SDS-polyacrylamide gel electrophoresis in a gel containing a linear gradient of acrylamide from 7.5 to 25% (w/v). The individual wheat varieties were: track **a**, Norman; track **b**, Fenman; track **c**, Longbow; track **d**, Mission; track **e**, Brimstone; track **f**, Sicco; track **g**, Musket; track **i**, Beauchamp. The proteins in track **h** are M_r standards; the M_r values are indicated on the right hand side of the gel. The 15K protein present in soft wheat starches is arrowed on the left hand side of the gel.

granules at 50° C in 1% (w/v) sodium dodecyl sulfate (SDS) were fractionated by high-resolution SDS-polyacrylamide gel electrophoresis. The buffer system of Laemmli and Favre (1973) was used with separating gels containing a linear gradient of total acrylamides from 7.5 to 25% (w/v). This resulted in the proteins of lowest M_r being resolved into several discrete bands with apparent M_r values between 5 and 19K (Fig. 1).

It was noted, however, that whereas starches prepared from several soft bread wheat varieties gave a prominent band with an apparent M_r of 15K, those from several hard bread wheats gave only a very faint band of the same mobility (Fig. 1). To explore the generality of this result, we isolated starches from 100 UK-grown wheats, including seven durum wheats, by careful washing with distilled water. Analysis of the starch granule proteins showed that all the soft wheats possessed the prominent 15K band, the hard bread wheats had a faint or very faint 15K band, and the very hard durum wheats lacked that band completely.

Subsequently, we analyzed starches from hard and soft wheats with different genetic backgrounds (from Australia, Canada, Mexico, and the United States), and those wheats conformed to the pattern for UK wheats. Furthermore, whereas starch from the soft variety Cappelle Desprez had an intense 15K band, that from the derived single-chromosome-substitution line Cappelle Desprez (Bezostaya-5D), which is hard, had only a faint band. It appears, on the simplest interpretation, that the gene coding for the 15K band is on the same chromosome as the major gene controlling endosperm texture, which is known to be on the short arm of chromosome 5D (Law et al 1978). On this basis, the complete absence of the 15K band in the durum wheats is explained, because they have no D-genome and, therefore, neither the intense nor the faint form of the gene for the 15K band.

We have, therefore, demonstrated an unbroken positive association between the presence of a M_r 15K starch protein and endosperm softness, the dominantly inherited type of endosperm texture, for some 150 wheats of widely different genetic backgrounds. Thus, it appears that this protein plays an important role in conferring endosperm softness on wheats. The mechanism by which the protein causes this effect is not yet known. However, because the protein is associated with the surface of the starch granule, it may have some sort of "nonstick" property that reduces the adhesion between the granule and the protein matrix of the endosperm. Research now in progress to isolate and characterize this protein, to study its location in grain sections by immunohistochemical techniques, and to study its genetic control is intended to shed light on its mechanism of action.

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