INFLUENCE OF THE STARCH GRANULE SURFACE ON THE RHEOLOGICAL BEHAVIOUR OF WHEAT FLOUR DOUGH

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ABSTRACT

The influence of the properties of the starch granule surface on the rheological behaviour of wheat flour doughs was studied in dynamic oscillation measurements (frequency sweep and strain sweep) and in stress relaxation measurements. A flour with a high protein content (15%) was diluted with wheat starch to obtain a protein content of 10%. The granule surface of the substituted starch was modified in three different ways: by heat treatment, by adsorption of a wheat protein fraction and by adsorption of lecithin to the granule surface. The effects of these modified starches were compared with the results obtained for nonmodified starch and protein or lecithin (in liposomes) added to the flour. Owing to the low concentration of the added protein and lecithin, no effect was observed when they were added to the bulk of flour. However, as a starch-surface modification the same components influenced the rheological parameters studied. Also the heat-treated starch had an effect on the rheological behaviour. The study established the importance of the properties of the starch-granule surface in wheat flour dough.

INTRODUCTION

From a rheological point of view the wheat flour dough can be regarded as a composite material where the starch granules act as fillers in the continuous gluten matrix (Hibberd 1970; Smith *et al.* 1970). In such a material, the modulus of the composite depends on the ratio of the modulus of the two materials; the larger the ratio, the higher is the reinforcing effect of the filler. Other properties such as volume fraction and shape of fillers, adhesion between filler and matrix

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also determine the rheological properties of the composite material (Nielsen 1974). The starch granules have been shown to have a more complex role in dough compared with an inert filler (Hibberd 1970).

The adhesion between the granules and the dough matrix has been given surprisingly little attention, even though the protein-starch interface in wheat flour dough is large. The surface area of A-granules in wheat starch has been estimated to be 0.25 m^2/g, and for B-granules 0.7 m^2/g (Morrison and Scott 1986). The starch-granule surface has been described as hydrophilic, whereas heat treatment and chlorination made it hydrophobic (measured as oil-binding capacity) (Seguchi 1984a,b). In another approach, the starch-granule surface was considered as a solid surface onto which the protein in solution was allowed to adsorb (Eliasson and Tjerneld 1990a). Differences in degree of adsorption were shown between starch variety, pH, salt concentration, heat treatment and protein. When wheat starch was added to a mixture of wheat proteins, the greatest adsorption was found for the protein fraction of the higher molecular weight.

The starch-granule surface has been discussed in relation to milling, where the wheat endosperm hardness is an important quality factor (Barlow *et al.* 1973; Grenwell and Schofield 1986). Protein-starch interactions, which are stronger in hard wheats, have been related to endosperm hardness. The soft endosperm splits along the cell walls, while in a hard endosperm the breakage also occurs through the granules. The hardness of the endosperm has also been related to the strength of the continuous protein matrix, which was considered to physically entrap the starch granules (Stenvert and Kingswood 1977).

The importance of the starch-granule surface during baking was emphasised by Sandsted (1961). Freeze-fracture studies of the ultrastructure of dough showed that starch-protein interactions are important in fermented dough and become stronger during baking (Fretzdorff *et al.* 1982).

The objective of the present study was to investigate the influence of the starch-granule surface on dough rheological properties at small deformation. The granule surface of native wheat starch was modified in three different ways: by heat treatment, by adsorption of lecithin, and by adsorption of a wheat protein fraction. The effects of these modified starches were compared with the results obtained for nonmodified starch and lecithin or protein added to the flour. A high protein containing flour was diluted by the starches to obtain a protein content of 10%.

MATERIALS AND METHODS

Materials

Spring wheat with a protein content of 14.9% supplied by Svalöf Weibull AB, Landskrona, Sweden, was a cross between W 31169 and Nemares (Johansson

and Svensson 1995). Native wheat starch was obtained from Lyckeby Stärkelsen AB, Kristianstad, Sweden (protein content 0.3% (N × 6.25), fat content 0.2%). The lecithin used was Epicuron 200 (Lucas Meyer, Hamburg, Germany). The protein fraction (M1, protein content 87% (as is)) was obtained from the variety Monopol by successive extraction with diluted hydrochloric acid (HCl) according to MacRitchie (1987). The preparation and characterization of this protein are described elsewhere (Eliasson and Lundh 1989).

Starch Surface Modification

Heat Treatment. Starch and distilled water were equilibrated for one week at 8C in a sealed glass jar. The sample was thoroughly mixed twice during the period of time to maintain a reproducible water content of the starch. The water content of starch given by method 44-19 (AACC 1983a), was 24.6 ± 0.2% before heat treatment. Heat treatment was performed in petri dishes at 120C during 60 min, according to Seguchi (1984a). The starch was then left on a tray at room temperature to recover from the moisture loss in the oven.

Lecithin-treated Starch. Lecithin was mixed with distilled water on a magnetic stirrer and ultrasonicated to form a homogeneous lamellar liquid-crystalline dispersion (0.35 g lecithin in 350 mL distilled water). Starch was added to the lecithin dispersion and left on the magnetic stirrer for 1 h. The concentration was 1% lecithin on dry starch. The slurry was decanted on a tray and left to dry during the night. No shimmering surface of lecithin on top of the starch layer (on the tray) could be observed after drying, which was the case at higher concentrations of lecithin (2.5% and 5%). A layer of visible lecithin on top of the starch layer, on the tray, indicated that excess lecithin was added with respect to the granule surface adsorption. That lecithin was still present at a level of 1% was evident from the appearance of the dry starch powder after the lecithin treatment. The dry modified starch was finely divided using a mortar and pestle.

Adsorbed Protein. The wheat protein fraction was adsorbed to starch according to Eliasson (1990a). The solution containing the protein fraction, soluble in 0.01 mM HCl, was mixed with hydrated starch (18 g dry substance) on a magnetic stirrer for 30 min. The mixture was centrifuged to remove the starch from the protein solution. The starch was washed with 250 mL distilled water (on a magnetic stirrer for 30 min) and centrifuged as described above. To calculate the adsorbed amount of protein (5.8 mg/g dry starch), the protein concentration was determined by the biuret method before the adsorption (0.86 mg protein/mL), after the adsorption (0.30 mg protein/mL), and on the washing solution (0.14 mg protein/mL). The dry starch was crushed in a mortar and pestle and sieved through 30 DIN (Axel Kistner, Stockholm, Gothenburg, Sweden).

All the modified starches were investigated for water content according to the method 44-19 (AACC 1983a). Reference doughs for the lecithin- and protein-treated starches were prepared so that the same amount of lecithin or protein which was adsorbed on the granules also was added to the dough. A lamellar dispersion of lecithin (4.6 mg lecithin/g water) was prepared to result in 1% lecithin on dry added starch when included in the dough (0.33% on dry dough substance). The protein fraction soluble in 0.01 mM HCl (same as when it was adsorbed to the granule surface) was freeze dried. The same amount of protein as was adsorbed to the starch (1.9 mg protein/g dry material) was added to the dry flour in the mixer.

Dough Mixing

The farinograph water absorption of the flour was determined as described in (Larsson and Eliasson 199ba), where the method 54-21 (AACC 1983b) was adjusted to mixing in the 10 g mixing bowl. Flour (10 g) was mixed at 30C with the amount of distilled water according to the farinograph water absorpiton, i.e. 44.9% (total basis). The doughs were mixed for 6 min. When the starch-granule surface was studied, the flour was diluted by starch to obtain a protein content of 10% of the total mixture of flour and starch. A larger amount of water had to be added to develop appropriate consistency of the starch-diluted doughs. The amount of water added was based on the pronounced influence on the stress-relaxation modulus of the starch-water ratio (Larsson and Eliasson 1996a). The water addition to the flour diluted by starch was based on the ratio of starch to water (1.02) in the dough prepared with the original flour. The starch content of the original flour was estimated roughly as the dry flour subtracted by the protein (14.9%) and lipid (approximately 2%) contents. This approximation resulted in a water content of 46.5% for the dough diluted by starch. Water was added to this level in all doughs. All the doughs were mixed with the same amount of flour (5.81 g dry) and added starch (2.84 g dry). This resulted in a total estimated starch content of the final flour of 88.6%, where the added starch constituted 37.0% of the total starch in the dough. The protein fraction and lecithin were added (as a starch modification or directly to the dough) at a level of 0.2% and 0.33%, respectively (on dry starch + flour). The flour and the starch, or starch and freeze dried protein (cut into very fine pieces) were mixed for 3–5 min before the water was added to the mixing bowl. At least three doughs were mixed under each set of conditions and used for the rheological tests.

Dough Rheological Measurements

The rheological behaviour of the doughs were studied using a Bohlin VOR rheometer (Metric Analys, Stockholm, Sweden) in three different modes. First

a frequency sweep in the linear region (strain = 0.00044) with the plate-plate geometry (diameter 15 mm, gap = 2 mm) was performed. After this the same sample was subjected to increasing strain at a frequency of 0.5 Hz (in the middle of the mechanical spectrum). The third measurement was a stress-relaxation test at a strain of 0.006, for which the cone-plate geometry was used. The higher strain value in the stress-relaxation measurement was chosen in order to separate the two cooperative flow processes (Wikström and Eliasson 1997). The dough subjected to stress relaxation was given a resting time of 45 min (at 30C, sealed in plastic), while the sample tested in the oscillatory mode only rested for 15 min before it was fixed in the rheometer geometry. Residual stresses were allowed to relax during at least 15 min before the strain was applied to the dough. The rheological measurements were repeated at least twice, using a new dough for each repetition. In the dynamic measurements error bars at 0.5 Hz give the maximum and minimum values of the storage and loss modulus, respectively. The reproducibility of the strain sweeps is shown by the error bars, indicating the maximum and minimum values at the highest strain investigated. For the stress-relaxation measurements, the mean values of the relaxation times (taken as the time where 50% and 10% of the initial stress remained, $t_{0.5}$ and $t_{0.1}$, respectively) and the standard deviations are shown in Table 1.

Differential Scanning Calorimetry (DSC)

The thermal properties of the native and modified starches were investigated on a Perkin Elmer DSC-2C calorimeter (Perkin Elmer Inc., USA) over the temperature range of 20C to 130C, with a scanning rate of 10C/min. The measurements were performed at the water-to-starch ratio 3:1, and under conditions described by Eliasson (1986).

RESULTS

In Fig. 1a–c the results from the dynamic measurements are shown as the frequency sweeps of the storage and loss modulus (G′ and G″, respectively). When the nonmodified starch was exchanged for the heat-treated one, the increase in both G′ and G″ was pronounced, and constant over the investigated frequency range (Fig. 1a). Similar results, an increase in both moduli, were obtained when starch with adsorbed protein was added to the flour (Fig. 1b). When the same fraction of protein, as was adsorbed to the starch, was added directly to the dough, the mechanical spectra of the nonmodified starch remained unchanged (Fig. 1b). The small amount of liposomal lecithin added to the dough (0.33%) could not be detected in G′ or G″. The slight increase, indicated for the storage modulus of the dough with lecithin-treated starch, was hardly significant (Fig. 1c). The

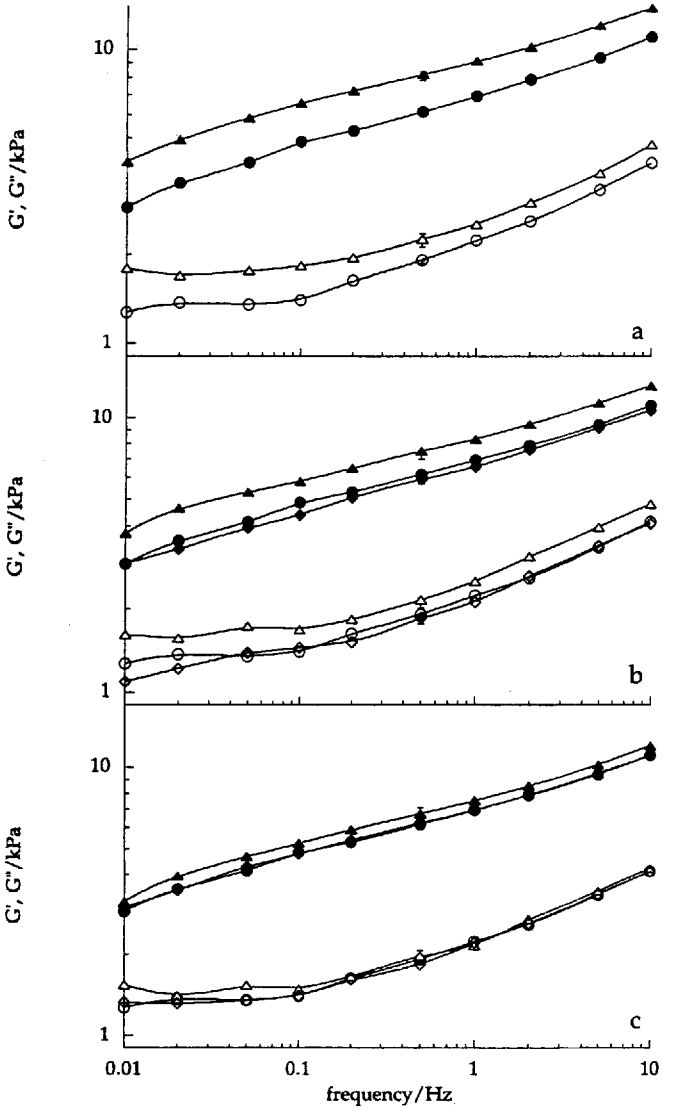

FIG. 1a–c. THE MECHANICAL SPECTRA FOR DOUGHS INCLUDING (a) NONMODIFIED STARCH (●) AND HEAT-TREATED STARCH (▲), (b) NONMODIFIED STARCH (●), PROTEIN-TREATED STARCH (▲) AND PROTEIN + NONMODIFIED STARCH (♦), (c) NONMODIFIED STARCH (●), LECITHIN-TREATED STARCH (▲) AND LECITHIN + NONMODIFIED STARCH (♦)
Filled symbols G′ and open symbols G″.

frequency dependencies of G′ and G″ were not influenced by protein or lecithin added, neither as a granule-surface modification nor added directly to the dough.

The stress-strain behaviour is presented in terms of the reduced storage modulus, G′/G″ (strain = 0.00044), as a function of strain (Fig. 2a–c). The nondiluted flour dough showed a linear region at strains lower than 0.001 (results not shown). When the flour was diluted by starch, the linear region decreased, or the doughs seemed to flow somewhat even at very low strain levels. Heat treatment of the added starch decreased the strain tolerance, observed as a stronger decrease in the reduced modulus beyond a strain of 0.02 (Fig. 2a). A tendency towards an increase in tolerance to strain seemed to be just detected when the protein fraction was adsorbed at the granule surface (Fig. 2b). For the lecithin-treated starch no difference was observed (Fig. 2c). There was no influence on the strain-sweep tests when the protein fraction (Fig. 2b) or lamellar lecithin (Fig. 2c) was added to dough diluted by nonmodified starch.

The stress-relaxation modulus was affected in the same way as was the storage modulus in Fig. 1a–c, although it differed less, and is not shown here. Examples of the stress-relaxation spectra are shown in Fig. 3a–c as the modulus divided by the initial modulus (G/G_0) plotted as a function of time. In the present study the reproducibility of the stress-relaxation measurements is shown by $t_{0.5}$ and $t_{0.1}$ (Table 1). The heat-treated starch slowed down the stress relaxation considerably, compared with the reference dough with native starch (Fig. 3a). When the protein- and lecithin-modified starches were included, the effect was less, with the slower decay of stress more clearly illustrated by the two relaxation times, $t_{0.5}$ and $t_{0.1}$ (Table 1). The two relaxation times were influenced in a similar way as was the storage modulus in Fig. 1a–c. The protein fraction and the lamellar phase of lecithin added to the dough including nonmodified starch did not have any effect on the stress-relaxation spectrum (Fig. 3b–c and Table 1).

The DSC endotherms for the nonmodified and the heat-treated starches were compared to check whether any gelatinization had taken place in the heat-treated starch. The enthalpies of the endothermic transitions, and the onset temperatures were 12.2 ± 0.6 J/g, 54.0 ± 0.5C and 11.8 ± 0.7 J/g, 54.5 ± 0.2C for the nonmodified and the heat-treated starches, respectively. This shows that the modified starch was not gelatinised during the heat treatment.

DISCUSSION

In a composite material, such as wheat flour dough, the properties of the continuous matrix, the volume fraction and shape of fillers and the adhesion between filler and matrix determine the rheological properties of the material (Nielsen 1974). The rheological properties of the gluten phase have been given most attention in research on wheat flour dough as gluten composition is important for

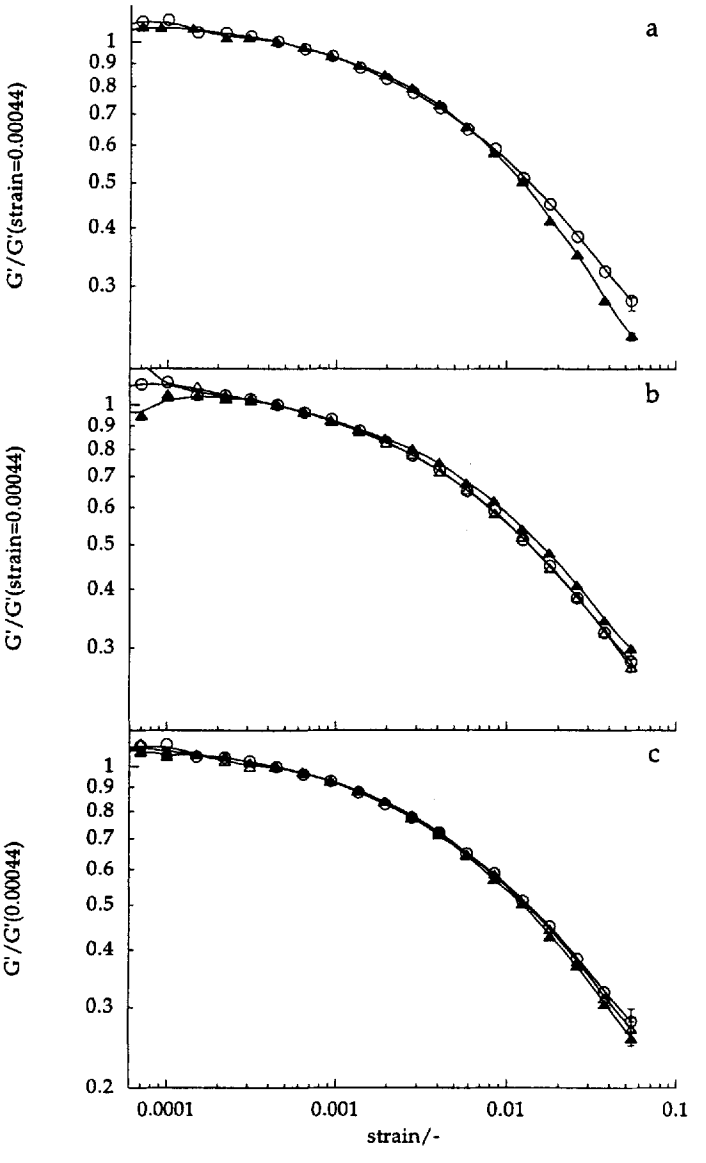

FIG. 2a–c. THE STRAIN SWEEPS FOR DOUGHS INCLUDING (a) NONMODIFIED STARCH (○) AND HEAT-TREATED STARCH (▲), (b) NONMODIFIED STARCH (○), PROTEIN-TREATED STARCH (▲) AND PROTEIN + NONMODIFIED STARCH (△), (c) NONMODIFIED STARCH (○), LECITHIN-TREATED STARCH (▲) AND LECITHIN + NONMODIFIED STARCH (△)

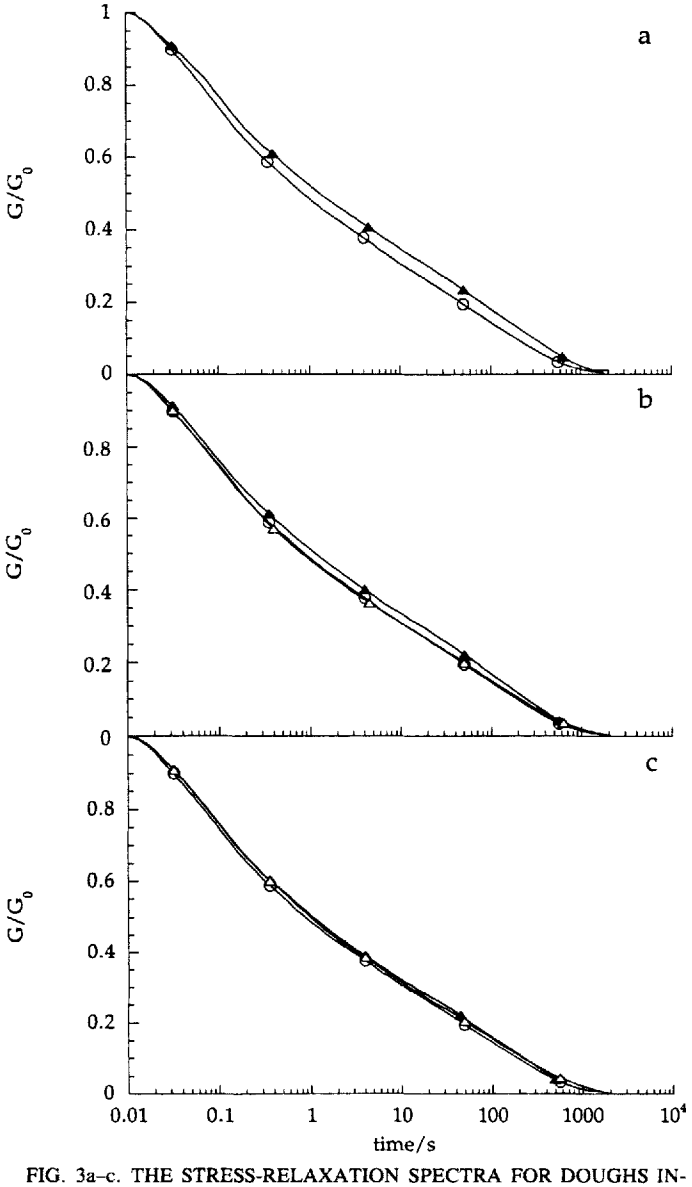

FIG. 3a–c. THE STRESS-RELAXATION SPECTRA FOR DOUGHS INCLUDING (a) NONMODIFIED STARCH (○) AND HEAT-TREATED STARCH (▲), (b) NONMODIFIED STARCH (○), PROTEIN-TREATED STARCH (▲) AND PROTEIN + NONMODIFIED STARCH (△), (c) NONMODIFIED STARCH (○), LECITHIN-TREATED STARCH (▲) AND LECITHIN + NONMODIFIED STARCH (△)

TABLE 1.
MEAN VALUES AND STANDARD DEVIATIONS FOR THE STRESS-RELAXATION TIMES OBTAINED FROM FIG. 3a–c

Dough containing	$t_{0.5}$/s	$t_{0.1}$/s
Non-modified starch	0.91±0.02	210±19
Heat-treated starch	1.41±0.03	308±5
Protein-treated starch	1.10±0.08	255±14
Protein+non-modified starch	0.86±0.11	200±10
Lecithin-treated starch	1.05±0.04	252±12
Lecithin+non-modified starch	0.91±0.11	210±21

the mixing and baking performance (MacRitchie 1987). The influence of starch concentration (Hibberd 1970; Rasper and deMan 1980), and particle size of granules, *i.e.* starch variety (Rasper and DeMan 1980) have been shown to influence the rheological behaviour of reconstituted wheat flour doughs.

The effect on G′ and G″ for the heat-treated starch was pronounced. The frequency dependence was, however, not influenced, compared with the dough containing the non-modified starch (Fig. 1a). The native starch-granule surface is considered to be hydrophilic, so that the starch granules, surrounded by a thin liquid film, form a continuous aqueous phase, in the developed dough. Heat treatment of starch has been shown to render the granule surface hydrophobic, indicated as an increased oil-binding capacity (Seguchi 1984a). Such a modification of the granule surface may certainly influence the microscopic structure of dough. A larger affinity to the hydrophobic gluten may be one possibility. When the adhesion between filler and matrix is good, an effect of adhesion properties does not always appear in small amplitude measurements (Nielsen 1974). The distribution of the granules in the matrix may be influenced when the surface properties of a filler are changed. Thus, another opportunity for the granules with an increased oil-binding capacity (lower affinity towards water) can be to agglomerate. A greater increase in G′ than was predicted was observed for filled gels, where the higher G′ was attributed to aggregation of the filling particles (van Vliet 1988). The adhesion between the filler and matrix increases in importance when the external stresses exceed the frictional forces between the phases or within aggregates, such as outside the linear region in a strain-sweep test (Nielsen 1974). For example, the stability of a gel can be illustrated by the effect on the reduced modulus by increasing strain (Clark and Ross-Murphy 1987). The formation of particle aggregates in a continuous matrix is then expected to result in an increase in G′, but induces instability towards increasing strain. In the pre-

sent work both an increase in G′ (Fig. 1a) and a higher strain sensitivity (Fig. 2a) were observed for the dough containing the heat-treated starch.

It may be argued that not only the granule surface, but also the granule shape or size was affected by the heat treatment. In this case such an effect also has to be considered for the evaluation of the rheological behaviour of the doughs including heat-treated starch. The reinforcing effect of a filler is enhanced when spheres are exchanged for more elongated or flat particles (Nielsen 1974). A modification of the shape and size of the granules may also influence the effective packing volume of the granules in the composite dough. Such a deformation of the granule shape or size may arise from partial gelatinisation of the starch granule if the local water content is high enough. The heat-treated starch was not gelatinised, as shown by the DSC measurements. A relevant comparison may also be the heat-moisture treatment (100C, 16 h and < 30% water in sealed containers) of wheat starch, which has shown that the granule size and shape remained unaffected by the treatment (Kulp and Lorenz 1981; Hoover and Vasanthan 1994). The heat treatment in the present study was different from the cited heat-moisture treatment as it was performed in open petri dishes.

Heat-moisture treatment has also showed an increase in water-binding capacity for the starch (Kulp and Lorenz 1981). The possibility that the heat treatment in the present study influenced the water-binding capacity of the granules may be a reasonable explanation for a higher modulus. According to Smith and coworkers a moderate increase in strain dependence could be observed when the water content was increased in water-binding capacity for the heat-treated starch would have the same effect as a reduction in the dough-water content, which according to Smith and coworkers imply a higher strain tolerance. Earlier studies on a broad variety of wheats, also showed that a small change in water content for spring and winter wheats, did not influence the stress-relaxation times to the same extent as is shown in Table 1 (Larsson and Eliasson 1996a). The stress relaxation, which resulted when the flour was partially substituted by nonmodified starch, was slowed down when the starch was heat treated (Fig. 3a), indicating that an increased amount of junction zones was created by the heat treatment of starch. These junction zones were weak, which is evident from the fact that a reduced strain tolerance was observed when the heat-treated starch was included (Fig. 2a).

The effect on the mechanical spectra when the protein fraction was adsorbed at the granule surface was similar to the effect by the heat-treated starch, but smaller. Over the frequencies investigated G′ and G″ increased (Fig. 1b). Also a slower stress relaxation (longer relaxation times (Table 1)) for the starch with protein adsorbed at the granule surface was observed (Fig. 3b). The effect of the surface treatment by protein in the stress-relaxation test was, as in the dynamic measurement, less than the one of heat-treated starch. An improvement of the

structure, which was responsible for the network was evident from both the mechanical spectra and the stress-relaxation measurement. Differences between the doughs containing the starches of different surface treatment appeared in the strain-sweep tests. A tendency towards increased strain tolerance was just indicated for the protein-modified starch (Fig. 2b). The differences between the nonmodified and the protein-modified starches were hardly significant, why the important result is that the protein-modified starch did not increase the strain sensitivity of the dough, as the heat-treated starch did. This indicates that a stronger structure was created when the flour was partially substituted by the protein-treated starch compared with the heat-treated one. Such a behaviour suggests a stronger interaction with the gluten matrix for the starch with protein adsorbed to the granule surface.

When wheat flour dough was subjected to ultracentrifugation, it separated into five phases (liquid, 'gel', gluten, starch and unseparated) (Larsson and Eliasson 1996a,b). The water content of the gluten phase was independent of the total amount of added water under defined mixing conditions, so that excess water is recovered in the liquid phase. The recovered amount of unseparated phase was dependent on the dough-water content, but also on variety. The adhesion properties between the starch granules and the gluten phase may be one reason for the differences between varieties.

As a consequence of the low concentration of the added protein (0.2% on dry weight), no effect was observed in any of the rheological tests, when it was directly added to the dough. This can be compared with earlier studies on the addition of the protein fraction to dough. No effect on the storage modulus was observed when the protein fraction was added to dough at a level of 1% (Eliasson and Lundh 1989). It may be argued that the exposure of starch to acid during the protein adsorption caused an effect on the starch, which influenced the rheological parameters. That the observed effect could be attributed to the protein adsorption was controlled by subjecting starch to the procedure used for protein adsorption, but in absence of the protein. No effect of the acid-treatment was observed in the rheological parameters (results not shown).

The slight increase in G′ indicated when lecithin was adsorbed at the granule surface was not enough to establish an effect (Fig. 1c). The stress relaxation seemed to be moderately slowed down in a similar way to when the added starch was heat or protein treated (Fig. 3c). This effect was shown in $t_{0.1}$, but not in $t_{0.5}$ (Table 1). No effect was observed in the strain sweep test by the lecithin treatment of starch (Fig. 2c). As a consequence of the low concentration (0.33% on dry flour plus starch) of the added lecithin dispersion, no effect was observed when it was directly added to the dough. This can be compared with earlier studies on the addition of lecithin in the lamellar liquid-crystalline phase to dough. The addition of 2% lecithin in the lamellar phase has been shown to increase the stress-relaxation modulus of wheat flour dough (Eliasson and Tjerneld 1990b; Larsson

and Eliasson 1996b,c), but no effect was observed in the long relaxation time, $t_{0.1}$ (Larsson and Eliasson 1996b). It can be concluded that different effects are obtained depending on how the lecithin is added, *i.e.* as a starch modification or in its lamellar liquid-crystalline phase. The increase in $t_{0.1}$ observed with the lecithin-treated starch, indicated a small effect on the starch network. Even though lecithin forms the lamellar phase also at very low water contents (down to approximately 15% water) according to the phase diagram (Bergenstål and Fontell 1983), the phase behaviour at lower water contents is complex (Tardieu *et al.* 1973). It cannot be ruled out that other phases may have formed at the granule surface when the local water content decreased during the air drying. The water content of the lecithin-treated starch after water evaporation at room temperature was 15.2 $\pm$ 0.0% (on dry substance). Whether the lamellar phase or some other lecithin phase was present at the granule surface after the preparation, it seemed to remain at the granule surface in the dough after mixing when the measurements were performed, which may explain the increase in $t_{0.1}$.

CONCLUSIONS

The properties of the starch-granule surface are important for the rheological behaviour of wheat flour dough. This was shown for doughs made by starch-diluted flour, where the starch fraction was modified in different ways. (1) Heat treatment of the starch increased the storage and loss moduli of the dough, slowed down the stress relaxation, and reduced the strain tolerance of the dough. Weaker aggregates or an increased agglomeration of the heat-treated starch granules was suggested to explain this behaviour. (2) Protein adsorbed to the granule surface, also induced an increase in the dynamic modulus and slowed down the stress relaxation. The strain tolerance was unaffected. A stronger interaction with the gluten phase could explain such a behaviour. (3) Lecithin treatment of the starch did not influence the mechanical spectra or the strain-sweep test significantly, but slowed down the stress relaxation slightly.

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