

Textural Stability of Intermediate-Moisture Extrudates: Effects of Formulation

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ABSTRACT

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Effects of formulation on the textural stability of intermediate-moisture, flour-based, “jerky”-type extrudates were assessed. Potato-based extrudates containing various particulate-meat concentrations and different plasticizers (sucrose, fructose, glycerol, and glucose) were produced and subjected to accelerated storage for three weeks. The elastic modulus of the samples was measured before storage and then weekly. The relative fluidity and moisture mobility of the specimens were assessed by dynamic mechanical spectrometry (DMS), electron spin resonance (ESR), and nuclear magnetic resonance (NMR). Samples were also evaluated by fluorometry and X-ray diffraction to determine the extent of browning reaction and degree of molecular ordering, respectively. While elastic modulus increased appreciably during storage, firming was progressively reduced by entrained meat content and also by plasticizers, especially glycerol; plasticized and

meat-containing samples had correspondingly lower $\tan \delta$ peak temperatures as measured by DMS. Textural results were also in keeping with fluidity and local viscosity as assessed by ESR measurements. NMR T1 relaxation values, reflecting moisture mobility, increased during storage. Diffraction spectra were consistent with published observations of hydrated starch, suggesting that water may have been released due to increased association of proteinaceous constituents. Fluorescence measurements confirmed moderate Maillard browning in all samples and significant chlorogenic browning in glucose-containing samples, although these effects were unrelated to degree of firming. It was concluded that textural stability was optimized by interruption of the matrix by dispersed meat or by plasticization by low molecular weight constituents.

Intermediate-moisture extruded products are under development by the military for use as eat-out-of-hand operational ration components. These extrudates are formulated and processed to have meat “jerky”-type characteristics. Conventional beef jerky is a popular item because of its chewy texture and spicy flavor; it also is highly stable, is easily carried, and requires no preparation for consumption. However, there is a desire to have such eat-on-the-go ration components consist least partly of carbohydrates that are metabolized relatively quickly and thus could potentially increase physical performance. High-temperature, short-time extrusion is an effective means of producing a texturized, meat-flour composite.

Highly acceptable potato-based extruded prototypes containing 40% particulate beef were developed (Engel et al 1994). However, the texture of these products deteriorated throughout storage because the samples firmed appreciably. Increases in modulus over two to three weeks of accelerated storage were in the range of 600% (Wang and Karydas 1995). While other investigators have studied the effect of meat or fish muscle on extrudate properties (McKee et al 1994, Jean et al 1996, Gogoi et al 1996), there has been little research conducted concerning the physical stability of these products. Several studies have noted toughening in extrudates or other flour-based snack foods due to postprocessing moisture sorption (Barrett and Peleg 1992, Rhode et al 1993, Barrett et al 1995, Harris and Peleg 1996, Barrett and Kaletunc 1998). One hypothesis offered in these instances (Rhode et al 1993, Harris and Peleg 1996) was that failure due to brittle fracture requires less energy than does failure due to plastic deformation. However, the fact that initially moist extruded products also toughen suggests that another mechanism involving some physicochemical transformation is responsible for textural instability.

In this study, effects of formulation on the firming rates and thermal and spectroscopic properties of intermediate moisture extrudates

were assessed. Dispersed meat-to-flour matrix ratio and plasticizer type were varied. Extruded samples were subjected to accelerated storage. Modulus, $\tan \delta$ peak temperature (from mechanical spectroscopy), NMR T1 proton spin-lattice relaxation time, and the relative prominence of fast motion components in the electron spin resonance spectra of a probe molecule were determined. Additionally, structural transformations were investigated by X-ray diffractometry, and the relative extents of Maillard and chlorogenic browning were determined by solid sample fluorescence measurement. Extent of storage-induced firming was related to measured structural and moisture mobility changes.

MATERIALS AND METHODS

Sample Preparation

Meat-containing extrudate formulations. Potato-based extrudates (potato granules from Basic American) were formulated to contain 0, 10, 20, 30, and 40% meat mix content (wb). Meat was purchased as top-round roasts, cut into cubes (≈ 5 cm), trimmed of visible fat, and preground through a 10-mm hole die plate using a Hobart grinder. The coarsely ground beef was mixed with the binders and humectants (Table I), steam-kettle cooked to 82°C, and reground through a 3-mm hole die plate. The moisture content of the meat mix was determined by vacuum drying for 16 hr at 70°C. The native moisture content of the potato granules was also determined.

Meat mix, potato granules, and water were combined to yield 40, 30, 20, 10, and 0% meat formulations at equivalent moisture contents (i.e., sufficient water was added to the 0–30% meat formulations to obtain the same moisture content [30%] as the

TABLE I
Meat Mix Formulation

| Ingredient | % | Supplier |
|-------------------------|------|----------------------------------|
| Top round beef | 89 | Stop & Shop (Natick, MA) |
| Oat fiber | 6.0 | Canadian Harvest (Cambridge, MN) |
| Salt | 4.1 | Morton (Philadelphia, PA) |
| Vegetable oil | 0.5 | Sysco (Houston, TX) |
| Sodium tripolyphosphate | 0.25 | FMC (Lawrence, KS) |
| Ascorbyl palmitate | 0.05 | Pfizer (New York, NY) |
| Sodium nitrate | 0.04 | J.T. Baker (Phillipsburg, NJ) |
| Vitamin E | 0.03 | ADM Arkady (Olathe, KS) |
| Tenox 4A | 0.03 | Eastman (Kingsport, TN) |

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40% meat formulation). Potato and particulate meat mix were first blended using a Hobart mixer (low setting, 1 min), then water was gradually added. The formulations were mixed at medium speed for 5 min. Batches of 5 kg were prepared and allowed to equilibrate overnight at 5°C before extrusion.

Plasticizer-containing extrudate formulations. Four plasticizers were individually tested at the 5% (of flour mix) level: sucrose (Domino), fructose (Staley), glycerol (KIC Chemical), and glucose (Mallinckrodt). For each batch, potato and plasticizer were pre-stirred and the mix was adjusted to 25% moisture content. A no-plasticizer control at 25% moisture content was also prepared.

Methods

Extrusion. Extrudates were processed on a Brabender extruder (controlled by a Haake Rheocord) operating at 20 RPM. Four temperature zones, from feed to die, were set at 35, 115, 115, and 135°C. Feed material was continually added to maintain a level of ≈2 cm in the intake funnel. The product was extruded through a 3-mm round

die opening. Current draw varied between 5 and 6 A. Cut extrudate sections (≈10 cm long) were allowed to cool to room temperature and then packaged six each in 10- × 15-cm trilaminate pouches. One multisorb oxygen scavenger sachet was added to each pouch. A moderate vacuum (7 psi) was pulled to reduce headspace during packaging. The samples were placed in a blast freezer and maintained at -20°C until analysis or commencement of storage studies.

Storage. The extrudates were placed in a 49°C environmental chamber at the start of all storage studies. Samples for textural, DMS, and NMR analyses were evaluated weekly for three weeks. Samples for ESR, fluorescence, and X-ray diffraction studies were evaluated after three weeks, one and three weeks, and two weeks of storage, respectively. Zero-time specimens were freshly thawed before evaluation. All analyses were conducted at room temperature.

Extrudate bulk density was determined through caliper measurement (average of three) of the diameter of weighed cylindrical specimens that were 5 cm long. Bulk density was determined using six replicates. The moisture content of the extrudates was determined by vacuum-drying.

Mechanical analysis. Samples were evaluated by uniaxial compression using a texture analyzer (TA.XT2, Texture Technologies) interfaced with a Zenith 286 computer. Specimens were sliced into cylinders 8 mm high. The extrudates were generally solid appearing rods with a few interspersed bubbles; regions with visible pores were excluded from analysis. All cut specimens were kept wrapped in plastic until compression to prevent moisture loss. Specimens were analyzed at room temperature (≈22°C). Specimens were compressed to 50% strain at a deformation rate of 0.2 mm/sec. The diameter of each specimen was measured before testing. Force-deformation data were converted to stress-Hencky strain data using a Minitab statistical program, and modulus determined through linear

TABLE II
Fitted Asymptotic Firmness^a

| Sample | E_f (kPa × 10 ⁻³) | r^2 |
|--------------------------|---------------------------------|-------|
| Control (0% meat) | 1.77 | 0.96 |
| 10% meat | 1.56 | 0.51 |
| 20% meat | 1.15 | 0.78 |
| 30% meat | 1.11 | 0.75 |
| 40% meat | 0.92 | 0.75 |
| Control (no plasticizer) | 2.14 | 0.94 |
| 5% glucose | 2.09 | 0.94 |
| 5% fructose | 1.96 | 0.93 |
| 5% sucrose | 1.91 | 0.93 |
| 5% glycerol | 1.84 | 0.94 |

^a From Eq. 1.

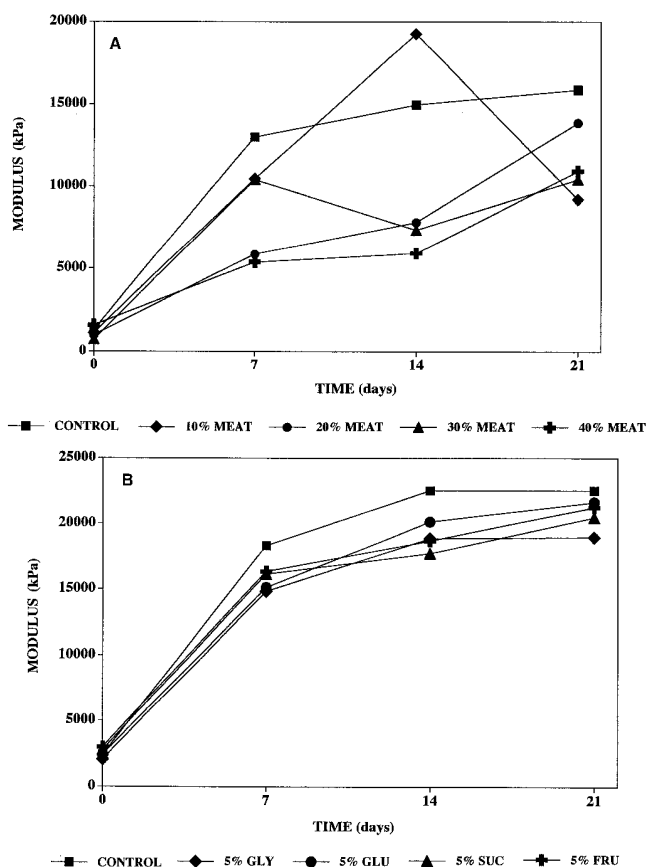


Fig. 1. Elastic modulus vs. storage time at 49°C showing extreme toughening of samples but beneficial effect of meat content and plasticizers for meat-containing extrudates (A) and plasticizer-containing extrudates (B).

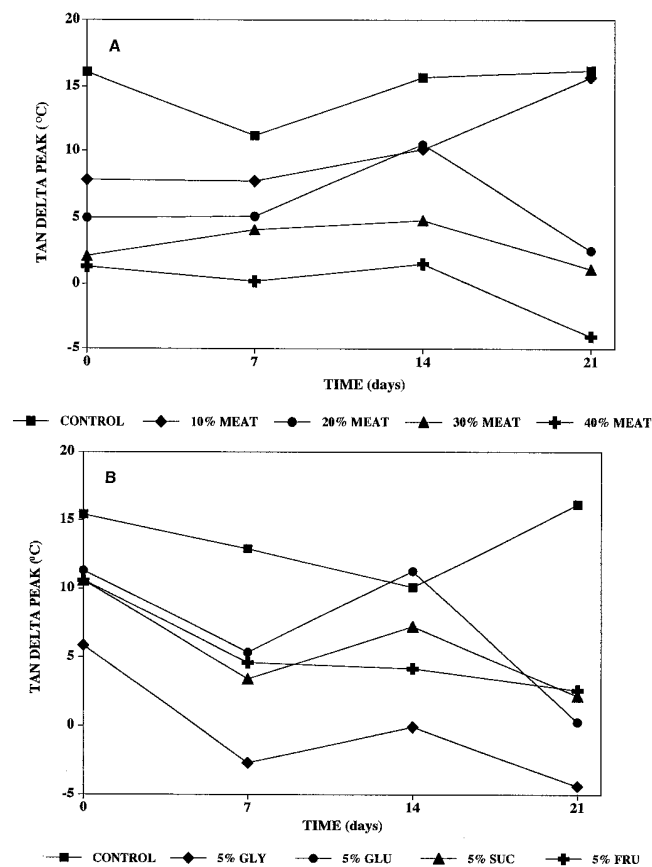


Fig. 2. Tan δ peak temperature vs. storage time at 49°C showing progressive reduction of tan δ peak temperature with meat content as well as beneficial effect of sugars for meat-containing extrudates (A) and plasticizer-containing extrudates (B).

regression of this relationship between 6% strain (to ensure full contact between the crosshead and sample) and 26% strain (because specimens buckled at high strain levels). Six replicates were run.

Mechanical spectrometry. A Seiko DMS-110 was used to determine thermal transitions in the extrudates, in accordance with the procedure of Barrett et al (1995). Samples were compressed flat (1–2 mm thick) on a Carver press operated at 500 kg, then carved into rectangles $\approx 3 \text{ cm} \times 1 \text{ cm}$. Exact dimensions were measured before analysis. The samples were subjected to oscillating flexing over a temperature range of -100 to 100°C , with a heating rate of $2^\circ\text{C}/\text{min}$. In each case, the peak temperature of the $\tan \delta$ curve for the 1 Hz frequency oscillation was recorded. Two to three measurements of $\tan \delta$ peak temperature were averaged.

Nuclear magnetic resonance. A 20/20 proton NMR analyzer (Process Control Technology) was used to assess moisture mobility in the samples. The operating frequency was 20 MHz. Samples were coarsely chopped and packed into the 10 mm diameter measuring tube (effective sample length was 50 mm). Saturation recovery methodology was used to measure the longitudinal proton relaxation time (T_1) of the samples. Samples were evaluated 35°C .

Electron spin resonance. The spin active nitroxide probe molecule, TEMPO (2,2,6,6-tetramethyl-1-piperidinyloxy), was used to assess microviscosity changes in selected extrudates. The spin active probe was dissolved in the water added to the control and to the sucrose- and glycerol-containing samples at a concentration of 1.25:1000 (w/w). This concentration gave a good signal-to-noise ratio and did not result in spin-spin interactions. The spectra were recorded on a spectrometer (Bruker EMX) at 9.8 GHz microwave frequency and at a power low enough to avoid saturation. At least two spectra were obtained for each sample. Line shape and the prominence of

lines corresponding to isotropic motion were evaluated to discern relative probe rotational mobility.

Fluorescence. Protein-bound, water and organic solvent-insoluble fluorescence of selected extrudates (control and plasticizer-containing) was measured by solid sample spectrofluorometry (Porter et al 1983, Hasegawa et al 1992, Porter et al 1993) using a spectrofluorometer (model 4800, SLM Aminco) operated in the ratiometric mode and using a tinted quartz rod in the reference compartment. Slit width was 4 nm and scan increment was 1 nm. Samples were scanned directly on the longitudinal surface and reported values are means of measurements taken at three widely spaced locations. An intraspectral ratiometric method, in which the ratios of excitation intensities 390/330 and 440/330 from an emission setting of 482 nm were observed, was used to compensate for intensity variability due to positioning changes (Porter et al 1993, Aubourg et al 1998). Because the 440 nm-stimulated emission arises after relatively more prolonged heating than does the 390-nm stimulated emission, the sum of the two excitation ratios was used as an index of Maillard polymerization progression from shorter to longer fluorophores (Porter et al 1993). Raw potato flour was additionally evaluated to determine the effect of the extrusion process on browning.

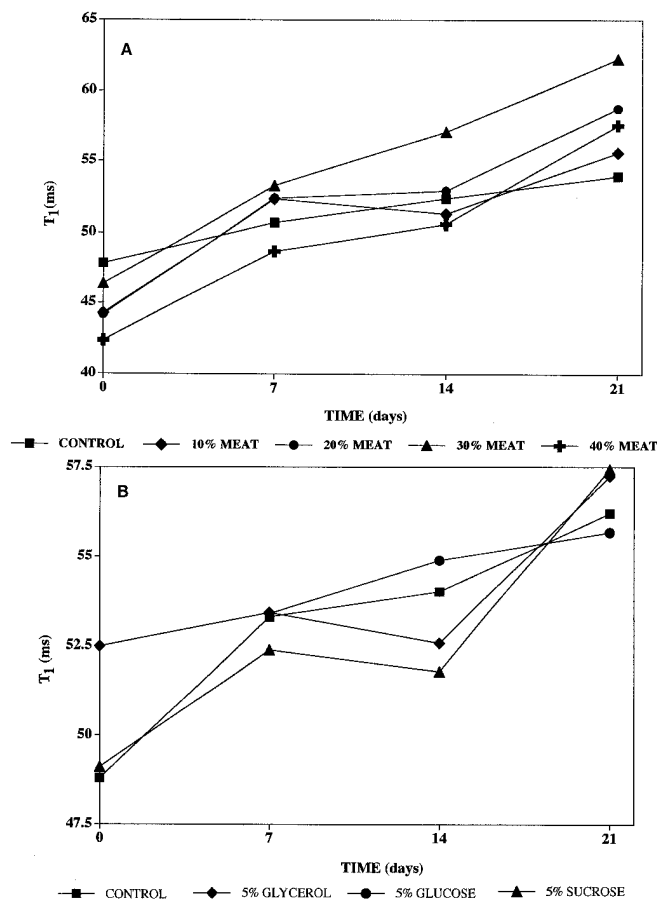


Fig. 3. T_1 relaxation time vs. storage time at 49°C showing increased T_1 values with storage time, most likely due to water released as a result of macromolecular association for meat-containing extrudates (A) and plasticizer-containing extrudates (B).

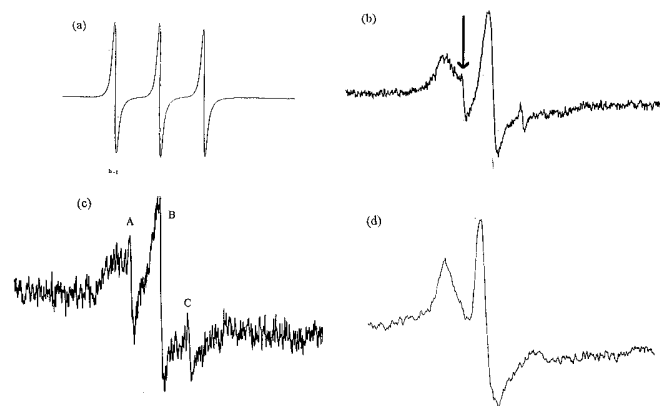


Fig. 4. Representative three-line spectra of TEMPO: (a) in water, h_{-1} low field line; (b) control sample before storage (arrow indicates fast motion component); (c) glycerol-containing sample before storage (A–C indicate contribution from isotropic motion); (d) control sample after storage at 49°C . Spectra show decreased prominence of fast motion component after storage and relatively increased prominence of fast motion component in glycerol-containing specimen before storage.

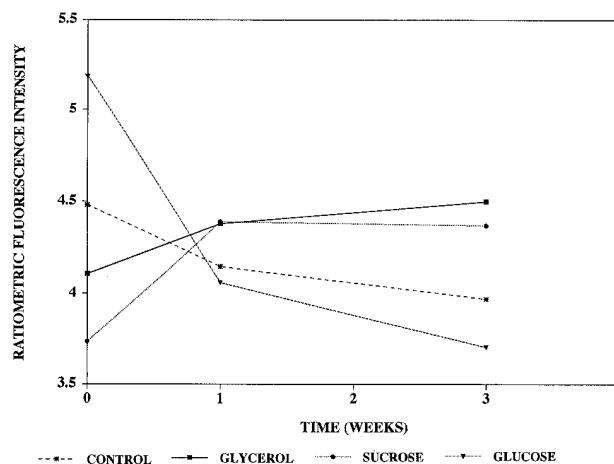


Fig. 5. Fluorescence ratio sums for pure potato, glycerol-containing, sucrose-containing, and glucose-containing samples at 0, 1, and 3 weeks storage at 49°C showing reduction in fluorescence ratio for glucose-containing extrudate and control, most likely as a result of quenching due to chlorogenic acid quinones and amine addition products arising from Maillard-induced phenolic browning.

X-ray diffraction. X-ray measurements of control and plasticizer-containing samples were conducted on a θ - θ powder diffractometer (Bruker D5005) with Gobel mirror incident optics, 0.2 mm incident beam exit slit, thin film detector side collimation, and a one-dimensional scintillation counter detector. Specimens were sliced into disks ≈ 3 mm high and ≈ 20 sections were placed adjacently in an aluminum sample holder. Cut surfaces of the extrudates were presented to the X-ray beam. Sample diffraction was measured over an angular range from $2\theta = 5$ – 120° . Data were collected at 0.05° intervals that were counted for 5 sec at each interval. Total scan time was 3 hr, 11 min. Plots of diffracted X-ray intensity versus diffraction angle (2θ) were obtained for each specimen.

RESULTS

Physical Properties

The moisture contents (wb) of the extrudates were 22–24% for meat-containing samples and 16–18% for plasticizer-containing extrudates (and did not change significantly during storage). In each case, the control was the highest moisture content sample. Bulk densities were 1.2 ± 0.04 g/cm³ and 1.3 ± 0.09 g/cm³ for the meat-containing and plasticizer-containing batches, respectively. All samples appeared to be solid rods randomly interspersed with a few bubbles (i.e., they were generally nonexpanded materials except for the presence of occasional holes).

Mechanical Properties

The elastic moduli of the specimens are shown in Fig. 1A, for meat-containing samples, and Fig. 1B, for plasticizer-containing samples. As is indicated, modulus increased appreciably during storage for

each sample, with the maximum rate of firming occurring within the first week of storage. After the first week of storage, samples continued to firm at a much reduced rate, generally approaching a plateau by the third week. The data for plasticizer-containing specimens were well described by an asymptotic function:

$$E - E_0 = E_f[1 - \exp(-bt)] \quad (1)$$

where E is modulus in kPa, E_0 is initial modulus, t is time in weeks, and E_f and b are fitted constants (Table II). Meat-containing samples also showed maximal firming during the first week of storage, but data for these specimens had lower r^2 values for fits to Eq. 1 (Table II).

The efficacy of both plasticizers and entrained meat in reducing ultimate firmness is apparent from the figures. Increasing meat content progressively reduced modulus in the stored samples: average stored modulus values (combining data for one, two, and three week pulls) were reduced by 11, 37, 36, and 49% by incorporating 10, 20, 30, and 40% meat, respectively ($P = 0.67, 0.17, 0.03,$ and 0.06 , respectively). Plasticizers at the 5% level lowered average stored modulus moderately (10–17%), glycerol had the most pronounced effect ($P_{\text{glycerol}} = 0.17$). These results are reflected in the asymptotic modulus values (E_f) obtained from Eq. 1, which decrease progressively with meat content and which are also lowered by the plasticizers (Table II).

Clearly, meat had a pronounced softening effect on the extrudates, and results implicate flour as the texturally unstable phase. Possible explanations for this functional plasticization are interruption of the flour-based extrudate matrix by meat constituents or a relatively greater affinity of water for flour-based components (i.e., a nonuniform microscopic distribution of water), such that moisture migrates from the meat phase to the continuous flour-based phase until the water activities are equal.

Sugars and glycerol had a slightly beneficial effect on texture, most likely by increasing fluidity or moisture mobility. These results are analogous to those observed for sucrose in corn meal extrudates equilibrated at high humidity levels (Barrett et al 1995), in which compressive resistance was progressively reduced by sucrose content.

Dynamic Mechanical Properties

Tan δ peak temperature at each storage interval was progressively shifted to lower temperatures by increasing meat content (Fig. 2A) and also lowered by each plasticizer (Fig. 2B), in keeping with textural results. Comparing control and stored samples (averaging across all storage intervals), tan δ peak temperature was reduced

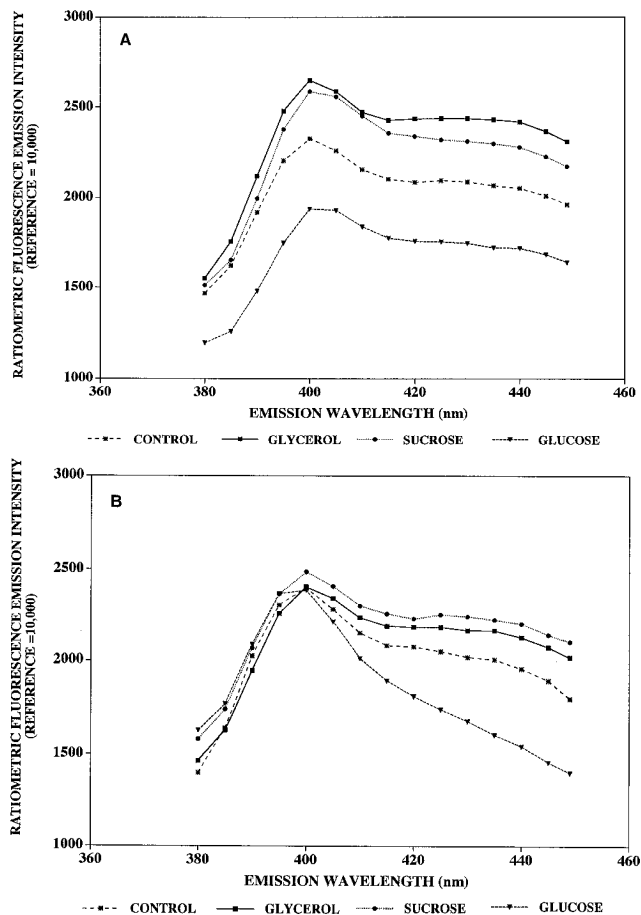


Fig. 6. Fluorescence emission intensity (excitation at 330 nm) showing loss of chlorogenic acid emission (440-nm plateau) in glucose-containing specimen due to Maillard-induced phenolic browning reaction before (A) and after (B) one week of storage at 49°C .

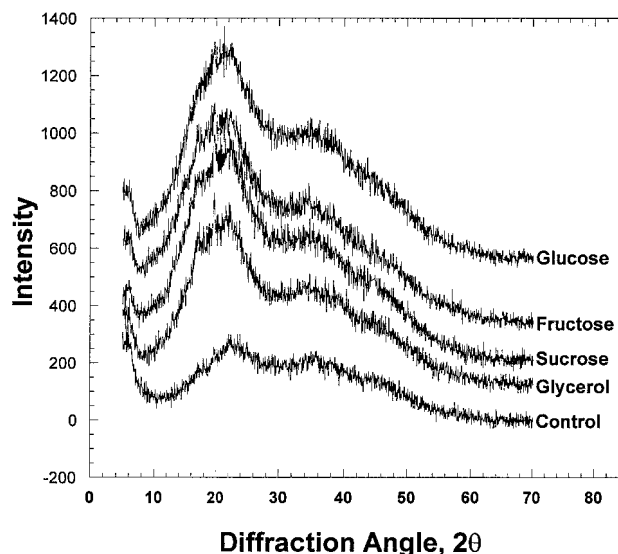


Fig. 7. X-ray diffraction spectra for extrudates (control and plasticizer-containing) before storage. Curves have been shifted in intensity for clarity.

4.6, 9.3, 11.9, and 15.5° in samples with 10, 20, 30, and 40% meat content, respectively ($P_{10\%} = 0.10$; $P_{20,30,40\%} < 0.01$). A similar analysis shows that average $\tan \delta$ peak temperature was shifted down by 6.5, 7.7, 8.1, and 14.1° in samples with added glucose, sucrose, fructose, and glycerol, respectively ($0.02 < P < 0.09$), a result in keeping with previous findings of Barrett et al (1995) concerning plasticization of cornmeal extrudates with sucrose.

Textural and DMS results are highly correlated. Regression coefficients for average stored modulus vs. average $\tan \delta$ peak temperature are 0.93 for meat-containing samples and 0.86 for plasticizer-containing samples.

The effect of storage time on $\tan \delta$ peak temperature was somewhat different for the two types of samples. For meat-containing samples, $\tan \delta$ peak temperature remained relatively constant among pulls, but for plasticizer-containing samples this parameter fell by an average of 9° between zero and three weeks of storage. Regression of $\tan \delta$ peak temperature against storage time yielded a distinctly negative correlation for all plasticizer-containing samples (average Student-*t* ratio was -1.9) but not for meat-containing samples (average Student-*t* ratio was 0.4). That the transition temperatures did not rise during storage may suggest that textural changes were due to constituent association or aggregation (with accompanying moisture redistribution) rather than to covalently bonded structures, which might function as antiplasticizers.

Spectroscopic Properties

Nuclear magnetic resonance. T1 proton relaxation time increased progressively during storage for all samples (Fig. 3A and B). Most likely, the redistribution of water and the release of water due to the aggregation of macromolecules (i.e., a squeezing out effect) contributed to the higher T1 measurement. Also, because the ring-down time for the PCT analyzer is ≈ 10 μ sec, it is possible that changes in molecular populations with lower mobility (and shorter T1 values) occurred but were beyond the measurement capability of the instrument. For example, Ruan et al (1996) reported different relaxation time distributions for baked flour-based products.

Initial T1 values varied slightly with formulation (i.e., in general were somewhat lower for meat-containing samples and higher for plasticized samples), but these differences were not maintained throughout storage.

Electron spin resonance. ESR spectra in general are indicative of the effect of the solvent or the molecular environment on the rotational mobility of small solutes such as nitroxide free radicals. Figure 4a shows a representative spectrum of TEMPO in water at a concentration of 1.25:1000 (TEMPO to water, w/w). This symmetrical three-line spectrum indicates that the majority of the probe population is in isotropic motion, reflecting no constraints or restrictions to rotation. The effect of the extrudate matrix on TEMPO rotation is shown in Fig. 4b for the potato-only control before storage. The spectrum deviates substantially from isotropic motion, indicating that the probe is primarily in microenvironments of restricted motion and partly in microenvironments of high mobility, as reflected by the presence of a fast motion component (arrow) in the low field line of the spectrum. Addition of plasticizers to the extrudates did not result in appreciable differences among

spectra before storage, with the possible exception of the glycerol-containing specimen, for which the contribution from the unrestricted probe population was more pronounced (Fig. 4c).

After storage at 49°C, powder type spectra were obtained, showing no fast motion lines as indicated by a comparison of the control samples before and after storage (Fig. 4b and d, respectively). The spectrum for the stored specimen is characteristic of anisotropic motion (i.e., the mobility of small solutes, such as TEMPO, is highly constrained by the macromolecular matrix). Powder spectra were also obtained after storage for samples with sucrose and glycerol. Increased storage time did not further affect the line shape of the spectra for any of the samples. The decrease in TEMPO mobility after storage reflects increased microscopic viscosity and is consistent with textural results. It is likely that the decreased probe mobility is due to entrapment in regions of the polymeric matrix from which water was released during storage. Such redistribution of moisture during storage is consistent with NMR results showing increased T1 relaxation time with storage time.

Fluorescence

Figure 5 shows the fluorescence ratio sum for samples after zero, one, and three weeks storage at 49°C. (Each extruded specimen, even before storage, exhibited a pronounced increase in fluorescence ratio indicative of strong Maillard browning relative to raw potato flour.) During storage, further moderate Maillard browning occurred in the glycerol- and sucrose-containing samples. However, the fluorescence ratio sum for the glucose-containing product was reduced by $\approx 30\%$ despite severe browning in this sample as assessed visually (no color changes were visually detectable in the other samples).

Such anomalous behavior may be explained by the occurrence of a phenolic browning reaction involving chlorogenic acid. Our research (W. L. Porter and W. G. Yoemans, *personal communication*) and that of others (Guyot et al 1997) has demonstrated that chlorogenic acid (and other antioxidants) can inhibit the oxidative pyrazine formation step of Maillard browning through a nonenzymatic reduction reaction that produces a quinone. This chlorogenic acid-derived quinone browns dramatically in the presence of amine groups native to potato. Furthermore, both the quinone and its amine adducts quench fluorescence.

This hypothesis was supported by the disappearance, in glucose-containing samples, of fluorescence emission intensity at 440 nm, an emission wavelength characteristic of chlorogenic acid (determined through separate experiments involving excitation of a chlorogenic acid solution deposited on filter paper). Emission spectra for the extrudates (Fig. 6A and B) indicate distinct loss of chlorogenic acid in the glucose-containing specimen during accelerated storage, and slight loss of this substrate in the control sample, as well. Spectra for the sucrose- and glycerol-containing specimens appear unchanged.

It is unlikely, however, that either browning reaction had a significant effect on firming. Regarding chlorogenic browning, the heavily browned glucose-containing sample exhibited less firming than did the control. Regarding Maillard browning, sucrose- and glycerol-containing extrudates, in which fluorescence increased, remained softer than did samples showing no evidence of Maillard polymerization during storage.

X-ray Diffraction

Unstored specimens had qualitatively similar diffraction spectra that were primarily amorphous in character (Fig. 7). This result is consistent with the work of Mercier et al (1979), in which the loss of strong crystalline features in potato starch due to extrusion (using similar conditions of hydration and temperature as those employed in this study) was observed. Unstored plasticizer-containing extrudates exhibited enhanced intensity and peak sharpening in the range of $2\theta = 15^\circ$ to $2\theta = 28^\circ$ (Table III), plus enhanced intensity in the region at $2\theta \approx 17^\circ$. These features are characteristic of barely crystalline structures (Zobel 1988). Maximum intensity of starch reflections in all unstored specimens occurred at $2\theta \approx 22.1^\circ$.

TABLE III
Maximum Reflection Intensity in Unstored Extrudates

| Sample | Angle (2θ) | Spacing, d(Å) ^a | Intensity ^b |
|----------|----------------------|----------------------------|------------------------|
| Control | $\approx 22.1^\circ$ | ≈ 4.0 | 82 |
| Fructose | $\approx 22.1^\circ$ | ≈ 4.0 | 257 |
| Glucose | $\approx 22.1^\circ$ | ≈ 4.0 | 254 |
| Glycerol | $\approx 22.1^\circ$ | ≈ 4.0 | 225 |
| Sucrose | $\approx 22.1^\circ$ | ≈ 4.0 | 252 |

^a $d = \lambda / (2\sin\theta)$.

^b Arbitrary units, estimated accuracy $\pm 15\%$.

Accelerated storage produced a clearly defined crystalline structure in all the extrudates (Fig. 8). The observed development of crystalline diffraction peaks in the stored samples is consistent with changes in potato starch diffraction patterns induced by increased moisture content observed by Nara et al (1978). In contrast to effects observed in unstored specimens, the intensity of the primary starch peaks was not increased in additive-containing samples relative to the control sample.

Four primary reflections were present in the stored plasticizer-containing extrudates: a weak reflection at $2\theta = 5.8\text{--}6.1^\circ$, a medium intensity reflection at $2\theta = 15.0\text{--}15.2^\circ$, and strong reflections at $2\theta = 17.0\text{--}17.5^\circ$ and $2\theta = 22.8\text{--}22.9^\circ$. X-ray peaks at these positions represent peaks typically assigned numbers 1, 3, 4, and 6 that are characteristic of a type C starch pattern (Zobel 1988). While potato starch generally gives a type B pattern, conversion to type C in the presence of heat and moisture has been reported (Zobel 1988, Kawabata 1994). Peak intensities for selected samples are shown in Table IV.

The stored flour-only extrudate exhibited five primary reflections: weak reflections at $2\theta = 5.9^\circ$ and $2\theta = 15.0^\circ$, and strong reflections at $2\theta = 17.1^\circ$, $2\theta = 22.6^\circ$ and $2\theta = 23.7^\circ$. Peaks in these positions are nominal features of a type B starch pattern (Zobel 1988), characteristic of potato and tuber starches. However, the peaks in the $2\theta = 22.6^\circ$ and $2\theta = 23.7^\circ$ doublet are closer than is typical for type B starch (and are shifted toward the 22.9° type C peak), most likely indicating the initial stage of transition to type C structure.

Patterns from both unstored and stored extrudates additionally show a peak at $2\theta \approx 19.7^\circ$ (Figs. 7 and 8). A diffraction peak in this position is generally associated with the formation of V structure in starches characteristic of amylose complex formation, which has been demonstrated to occur in potato extrudates containing small amounts of fatty acids (Mercier et al 1979).

DISCUSSION

The extreme firming of intermediate-moisture extrudates was significantly mitigated by addition of particulate meat and slightly lessened by addition of plasticizers, especially glycerol. Mechanical spectrometry and ESR results are consistent with textural measurements in indicating decreased glass transition temperatures and increased fluidity (lower viscosity) in less firm samples. That NMR proton T1 measurements show increased relaxation times after storage and that DMS $\tan \delta$ peak temperatures do not increase during storage suggest that molecular transformations responsible for

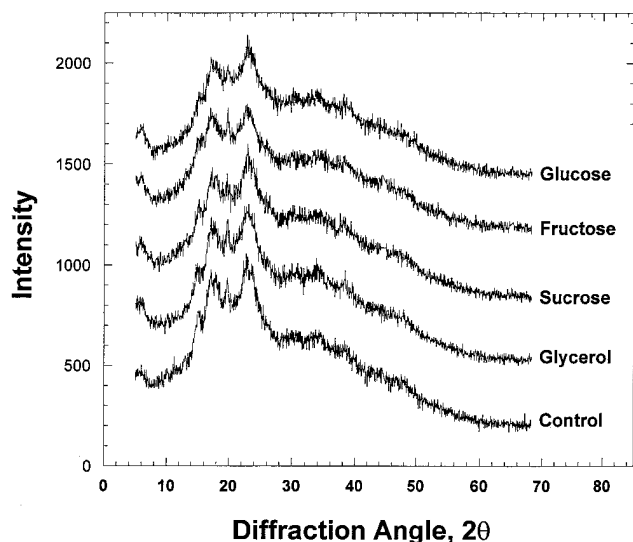


Fig. 8. X-ray diffraction spectra for extrudates (control and plasticizer-containing) after two weeks of storage at 49°C . Curves have been shifted in intensity for clarity.

textural changes are more likely aggregation of proteinaceous constituents (creating a population of relatively more mobile water) rather than chemical complexation.

Chemical analyses did not reveal any changes related to firming. While Maillard products were discerned in most samples, the extent of this browning mechanism during storage was generally moderate. While extreme chlorogenic browning was observed in the glucose-containing specimens, the degree of firming of this sample was no greater than that of the control, in which much less phenolic complexation was present.

Increased sharpness of peaks in X-ray diffraction spectra of flour-based products has been considered by some authors indicative of retrograded starch (Van Soest et al 1994, Kim et al 1997). However, interpretations of the physical significance of diffraction data or other measurements of crystallinity (e.g., calorimetry) have varied. Specifically, some studies (Zobel and Senti 1959, Rogers et al 1988) report poor correlation between textural changes and measured crystallinity in baked products.

Moreover, Nara et al (1978) reported the development of sharpness in diffraction patterns of potato starch after exposure of specimens to high relative humidity levels, during which peaks became progressively more prominent as sample moisture content increased. Thus, the results in our potato extrudates could possibly have been caused by interphase migration of water to starch regions, due to release of water from aggregated protein moieties. Different microregions in flour-based products can have different affinities for water and different tendencies for self-association. Guerrieri and Cerletti (1996) demonstrated a relatively greater affinity of water for starch than for gluten in heat-treated wheat flours and postulated that increased protein hydrophobicity due to thermal processing promoted slow conformational changes and aggregation during aging.

Furthermore, several authors implicate protein associations, or protein-starch associations, as causative of firming. For bread, Willhoft (1973) proposed that dehydration of gluten contributes to textural changes; Martin and Hosney (1991) and Martin et al (1991) hypothesized that the development of gluten-starch linkages promoted firming.

Extrusion processing has been used to create texturized plant-based proteins, for example from soy, due to its ability to form aligned macromolecular structures. In wheat flour extrudates, alignment of proteins was photomicrographically demonstrated by Kaletunc (1999) and Batterman-Azcona et al (1999). Additionally, extrusion-induced interactions between proteins and starch have been observed by Wang and Schaich (1999). It is possible that in relatively lower protein systems, such as in extruded potato flour, a continuous phase may consist of a network containing both protein and starch. An interpretation of increased association of proteinaceous regions during storage as a significant contributor to structural rigidity is most in keeping with the results of the diverse analyses of this study.

TABLE IV
Peak Intensities for Selected Specimens After Storage^a

| Sample | Peak | Angle (2θ) | Spacing, $d(\text{\AA})^b$ | Intensity ^c | |
|----------|---------|---------------------|----------------------------|------------------------|------|
| Control | 1 | 5.94° | 14.9 | 32.0 | |
| | 3 | 15.0° | 5.89 | 43.0 | |
| | 4 | 17.1° | 5.19 | 278 | |
| | 6a | 22.6° | 3.93 | 309 | |
| | 6b | 23.7° | 3.75 | 283 | |
| | Glucose | 1 | 6.10° | 14.5 | 46.0 |
| 3 | | 15.1° | 5.86 | 112 | |
| 4 | | 17.0° | 5.22 | 215 | |
| 6 | | 22.8° | 3.90 | 240 | |
| Glycerol | | 1 | 5.95° | 14.8 | 29.0 |
| | | 3 | 15.0° | 5.90 | 119 |
| | 4 | 17.1° | 5.18 | 251 | |
| | 6 | 22.8° | 3.90 | 261 | |

^a After correction of background and amorphous portions.

^b $d = \lambda / (2\sin\theta)$.

^c Arbitrary units, estimated accuracy $\pm 15\%$.

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