Characteristics of Imitation Cheese Containing Native Starches

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ABSTRACT: Imitation cheeses containing 3% native maize, waxy-maize, wheat, potato or rice starch were manufactured and the microstructure, meltability, texture and dynamic rheology of these products were compared to a control (0% starch). Fat globules in starch-containing products (except potato) were smaller than in the control as evidenced by electron microscopy. All starches reduced meltability and cohesiveness of the imitation cheeses. Hardness was increased by wheat, potato or maize starch but reduced by waxy-maize or rice starch. Starches significantly reduced tan δ peaks compared to the control with potato starch having the greatest effect. Rice starch appears to have the most potential as a partial casein substitute in imitation cheese.

Key words: imitation cheese, starch, meltability, dynamic rheology, microstructure

Introduction

CHEESE IS WIDELY USED AS AN INGREDIENT IN PREPARED foods to add taste, texture, and nutritional quality. The high costs associated with natural cheese production and storage, however, has prompted industry to search for alternatives (Kiely and others 1991). Attempts to reduce cheese costs have led to the development of imitation cheeses based on casein and its derivatives and the use of vegetable fat to replace the more costly milk fat (Zwiercan and others 1987). Due to the high cost and certain functionality limitations of casein, a number of researchers have investigated lower cost vegetable proteins as casein substitutes, such as peanut (Chen and others 1979), cottonseed and soy protein isolates (Taranto and Yang 1981), but these have had limited success.

A number of patents relating to the use of starches as casein substitutes in imitation cheese have been issued. Burkwall (1973) used pre-gelatinized starches in the manufacture of shelf-stable, zero-melt cheese products and a high amylose maize starch was used to manufacture a zero-fat imitation cheese (Freck and Kondrot 1974). Pre-gelatinized and enzymatically debranched starches have been used to mimic the characteristics of caseinates in imitation cheese (Zwiercan and others 1987; Zallie and Chiu 1989). To the best of our knowledge, however, there has been no detailed study of the influence of native starches on the textural properties of imitation cheese.

The physical properties of melted imitation cheese determine its usefulness in a number of processed foods, for example, pizza, lasagne, cordon bleu products. The properties of the unmelted product are also critical to its use as an ingredient, however. To facilitate even distribution in processed or convenience foods, imitation cheese is usually sliced, diced or shredded. The physical structure of the imitation cheese must be amenable to these processes and should not stick or clump following processing. Texture Profile Analysis (TPA) (Yang and Taranto 1982; Abou El Nour 1998), dynamic rheology (Zhou and Mulvaney 1998; Mounsey and O'Riordan 1999) and scanning electron microscopy (SEM) (Taranto and Yang 1981; Song and Park 1986) have been reported as useful means of assessing these critical physical properties of imitation cheeses. The objective of this study was to investigate the effects of native maize, potato, waxy-maize, wheat or rice starches on the rheology, meltability and microstructure of imitation cheese.

Materials and Methods

Manufacture of Imitation Cheese

A control imitation cheese was manufactured with the following: 48.8% water, 24.5% rennet casein (82% protein) (Kerry Ingredients, Listowel, Ireland), 26% vegetable fat, (Trilby Trading Ltd., Liverpool, England), 2.18% emulsifying salts, [trisodium citrate 1.08%, citric acid, 0.62% (Jungbunzlauer, Pernhofen, Austria), disodium phosphate, 0.48% (Ellis and Everard, Ireland)], 1.67% sodium chloride, (Salt Union, Cheshire, England) and 0.1% sorbic acid (Hoechst Ireland Ltd., Dublin, Ireland). All ingredients (except citric acid) were blended in twin-screw cooker (Model CC-010, Blentech Corporation, Calif., U.S.A.) at 35 °C and heated to 78 °C using direct steam. Citric acid was added. After 5 min of mixing, the product was packaged, cooled to 4 °C and vacuum packed (Model C 10 H, Webomatic®, Bochum, Germany) 24 h later. Using a similar manufacturing process, a series of imitation cheeses were prepared by replacing 15% of the casein protein (3% w/w of the total product) in the control formulation with native maize, waxy-maize, wheat, potato or rice starch and reducing the concentration of emulsifying salts used to solubilize the casein by 15%. The maize and waxy-maize starch were supplied by National Starch and Chemical, Cambridge, England; wheat starch was supplied by Roquette, Lille Cedex, France; potato starch was supplied by AVEBE, Veendam, The Netherlands, and rice starch was supplied by REMY Industries, Leuven, Belgium. Three 4 kg batches of each cheese were manufactured.

Compositional Analysis

Compositional analyses of imitation cheese were done as described by Mounsey and O'Riordan (1999). An amylose/amylopectin assay kit (Megazyme International Ireland Ltd., Bray, Ireland) was used to determine the amylose content of the starches.

Melt Test

A modification of the Olson and Price (1958) method was used as described by Mounsey and O'Riordan (1999) to assess imitation cheese meltability.

Dynamic Rheology Test

Rheological characterization of the imitation cheese was undertaken as described by Mounsey and O'Riordan (1999). Parameters measured included the storage modulus (G'), loss modulus (G'') and loss angle ($\tan \delta$ (G''/ G')).

Texture Profile Analysis (TPA)

Textural properties were measured with an Instron Universal Testing Machine (Instron Model 4301, Instron Corp., Canton, Mass., U.S.A.). Cylinders of cheese 20 mm high and 18 mm dia were cut with a cheese borer, wrapped in celophane to prevent dehydration, and allowed to thermally equilibrate to 22 °C. Samples were compressed by 80% of their initial height using a 35 mm dia plate at a crosshead speed of 50 mm/min. The uniaxial compression test was performed in two successive cycles, and the textural parameters, hardness, and cohesiveness were calculated as described by Szczesniak (1963).

Microstructural Analysis Using Scanning Electron Microscopy (SEM)

Cryo scanning electron microscopy was used to examine the microstructure of the imitation cheese. (JEOL JSM-5410LV Scanning Microscope, JEOL Instruments, Tokyo, Japan). The cryo unit used was an Oxford Instruments Cryo Preparation System CT 1500 (Oxford Instruments, Oxford, England). Prisms approximately 1 × 3 × 8 mm were cut from blocks of imitation cheese using a scalpel, mounted on a specimen holder and cryofixed by rapidly plunging into nitrogen slush (–210 °C). Specimens were transferred (under vacuum at –180 °C) to the cryo chamber and the interior of the sample was exposed using a scalpel. The fracture face was etched at –88 °C by heating on the microscope stage in the microscope chamber. When sufficient water was sublimed (after approximately 5 min), the specimen was cooled to –180 °C. The specimen was then spattered with gold (3mA, 2 min) in the cryo chamber prior to analysis in the microscope chamber, using an accelerating voltage of 10 Kv. Two samples were analyzed from each replicate.

Statistical Analysis

The imitation cheese was manufactured in triplicate. All tests were replicated 4 times. PROC GLM of SAS (SAS Institute, Cary, N.C., U.S.A.) was used to determine differences between treatment means. Treatment means were considered significantly different at $P \leq 0.05$ unless stated differently.

Results and Discussion

ALL IMITATION CHEESE PRODUCTS HAD MEAN FAT, MOISTure and pH values of 26 ± 0.15%, 48.8 ± 0.2% and 5.83 ± 0.08%, respectively. The starch-containing products had lower protein values (17.2 ± 0.2%) than the control sample (20.3 ± 0.3%), due to the replacement of the casein with starches. The amylose contents of maize, waxy-maize, wheat, potato or rice starch were 25, 3.8, 28, 26, or 13%, respectively, which is in line with suppliers' specifications and published values (Swinkels 1985).

Electron micrographs were prepared in an attempt to relate the microstructure of the imitation cheeses to their meltability and rheology. SEM images of the control imitation cheese (no starch) (Figure 1a and b) show a uniform distribution of spherical fat globules (F) (dia 3 to 20 mm) in a smooth protein matrix (P). Thin protein films (TPF) can be seen coating the fat globules in Figure 1b, but the bulk of the protein in the imitation cheese appears to be involved in the formation of a matrix. The presence of undissolved emulsifying salts (ES) is also evident. These images are typical of those previously reported by other researchers (Song and Park 1986; Guinee and others 1998). The use of starch to partially replace casein resulted in marked differences in imitation cheese microstructure (Figure 2 to 6). The fat globule size of the products containing potato starch was similar to the control (Figure 2); however, products containing wheat (Figure 3), maize (Figure 4a and b), waxy-maize (Figure 5) or rice (Figure 6) starches appeared to have smaller fat globules (dia 2 to 10 mm), indicating more extensive fat emulsification (Rayan and others 1980; Savello and others 1989). It was also evident during the manufacture of the imitation cheeses that the oil of starch-containing products was emulsified more rapidly than the control. It may be that the native starches functioned as inert fillers in the early, low-temperature stag-

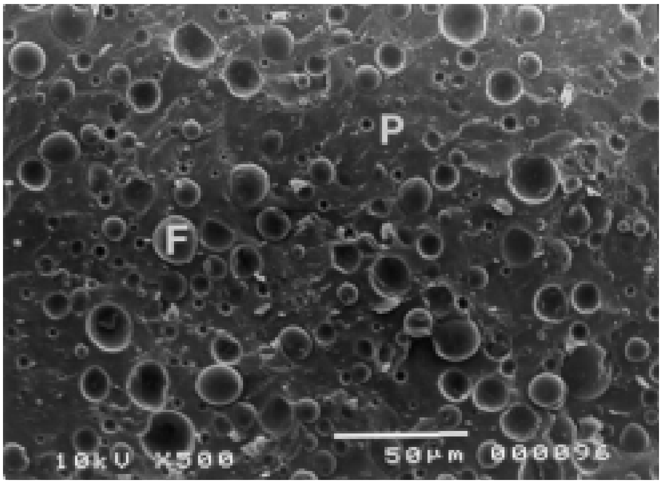

Figure 1a—Electron micrograph (x 500) of the control imitation cheese (0% starch). F = fat globule, P = protein matrix

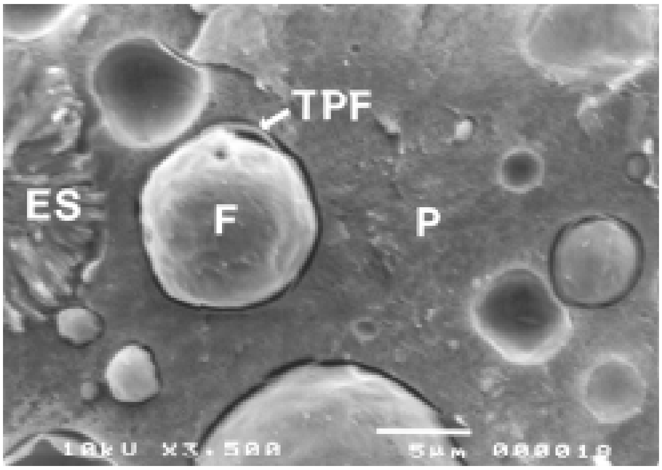

Figure 1b—Electron micrograph (x 3,500) of the control imitation cheese (0% starch). ES = emulsifying salt crystal, F = fat globule, P = protein matrix, TPF = thin protein film (indicated with a white arrow)

es of manufacture, leading to increased availability of water to hydrate the casein and increase its fat emulsifying properties. Additionally, the higher NaCl:casein ratio in the starch products (0.1:1) compared to the control of 0.08:1, may have promoted the solubilization of casein during heating (Aoki and others 1999) and ultimately increased its emulsifying properties. In the SEMs of imitation cheeses containing starch, irregularly shaped particles (S) are evident disrupting the protein matrix, which were not present in the control (Figure 1a and b). These particles are particularly evident at high magnification (× 3,500) as shown in Figure 4b for the maize- containing product and are possibly swollen starch granules and/or partially gelatinized starch fragments. Although the temperature used to manufacture the imitation cheeses (78 °C) was higher than the gelatinization temperature range of all the starches as determined by differential scanning calorimetry (data not shown), the relatively short cooking time (5 min) and low water levels possibly resulted in limited starch gelatinization. The changes in the fat globule size distribution and the disruption of the protein matrix by the native starches were manifested in the meltability and rheology of the imitation cheeses.

Table 1—Melt, hardness, and cohesiveness values of imitation cheese products with and without various native starches (3% w/w).

Starch Type	Melt (mm)	Hardness (N)	Cohesiveness
No starch (control)	185.6^{a}	110.2^{b}	0.252^{a}
Maize	99.3^{e}	116.4^{a}	0.19^{d}
Waxy-maize	117.5^{d}	75.2^{d}	0.189^{d}
Wheat	139.6^{c}	120.8^{a}	0.224^{b}
Potato	105.8^{e}	117.3^{a}	$0.201^{c,d}$
Rice	149.1^{b}	88.8^{c}	0.259^{a}

[1] For each column, means with the same letter do not differ significantly at $P \leq 0.05$.

The meltability of the imitation cheeses is shown in Table 1. All starches reduced the meltability and their relative effects followed the order maize > potato > waxy-maize > wheat > rice. Other researchers have associated poor meltability of processed cheese with small fat globule size (Rayan and others 1980; Savello and others 1989). However, a direct relationship between meltability and fat globule size is not evident from this study. For example, the fat globule size dis-

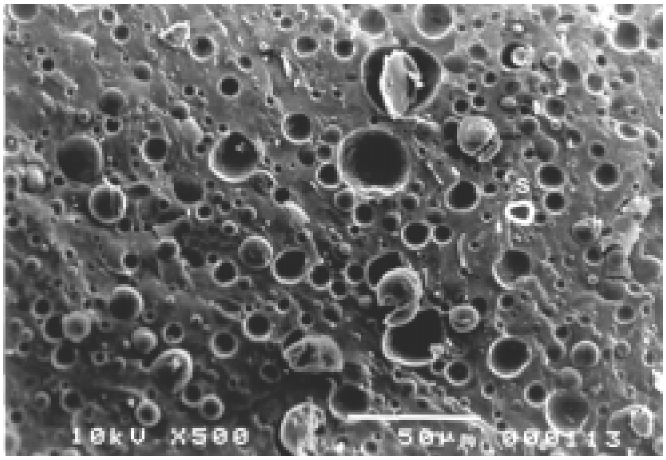

Figure 2—Electron micrograph (x 500) of imitation cheese containing 3% w/w native potato starch. S = starch particle (highlighted with a white border)

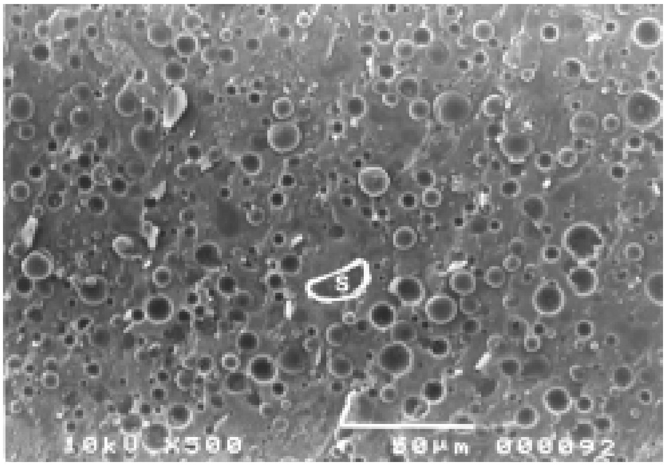

Figure 4a—Electron micrograph (x 500) of imitation cheese containing 3% w/w native maize starch. S = starch particle (highlighted with a white border)

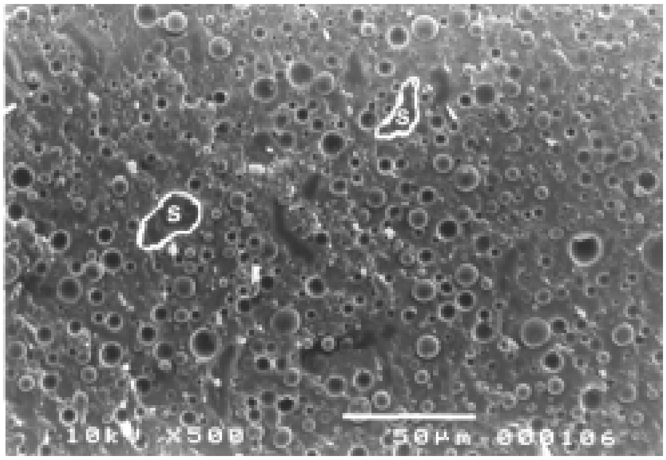

Figure 3—Electron micrograph (x 500) of imitation cheese containing 3% w/w native wheat starch. S = starch particle (highlighted with a white border)

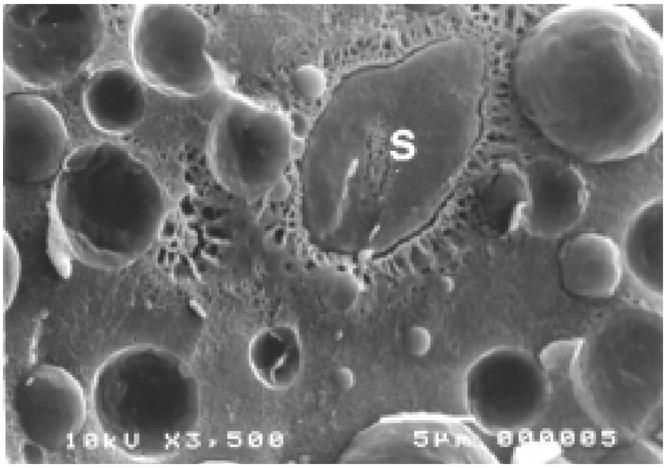

Figure 4b—Electron micrograph (x 3,500) of imitation cheese containing 3% w/w native maize starch. S = starch particle

tribution of imitation cheese containing potato starch was similar to that of the control, but its meltability was significantly lower and although rice- and wheat-containing products had the smallest fat globule sizes, their meltability was significantly higher than the other starch- containing products. These results suggest that other factors also influence the imitation cheese meltability such as the disruption of the protein matrix with swollen starch granules. It is likely that the small granule sizes of rice or the B-type granules of wheat starch and their limited swelling capacities up to about 75 °C (Lii and others 1995; Tester and Morrison, 1990) posed less resistance to melt compared to starches with large granule sizes and high swelling capacity such as potato starch (Kokini and others 1992; Swinkels 1985). The lack of thermoplasticity of cross-linked leached amylose, which was most likely to occur in products containing high amylose starches that are prone to retrogradation such as maize or wheat starch, was possibly another factor influencing meltability.

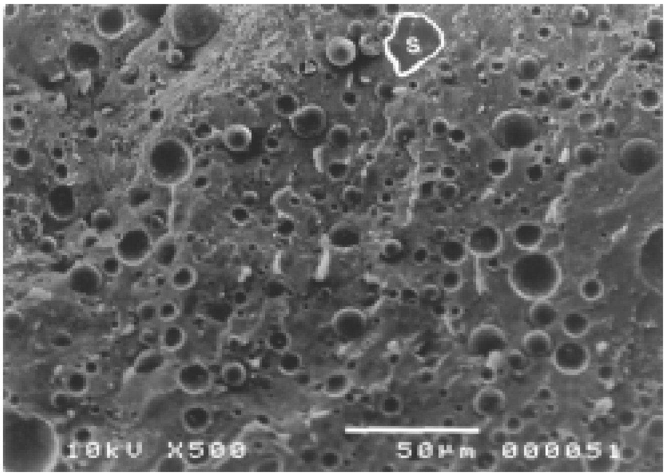

Figure 5—Electron micrograph (x 500) of imitation cheese containing 3% w/w native waxy-maize starch. S = starch particle (highlighted with a white border)

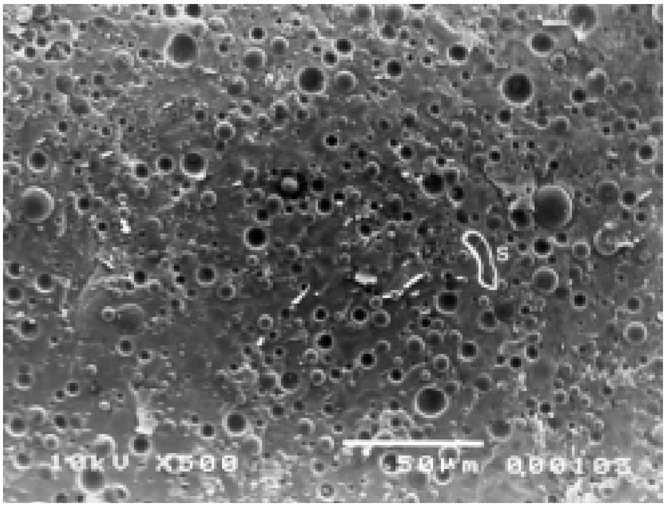

Figure 6—Electron micrograph (x 500) of imitation cheese containing 3% w/w native rice starch. S = starch particle (highlighted with a white border)

The hydration status of the protein matrix is also important in controlling the melting of imitation cheese (Zhou and Mulvaney 1998). The immobilization of water by the swollen starch granules or gelatinized starch may have resulted in dehydration of the protein matrix, leading to increased hydrophobic protein-protein interactions and ultimately poorer meltability. The honeycomb structure of the protein matrix in the area immediately adjacent to the starch structure (S) (as shown in figure 4B) may have arisen due to protein dehydration.

The effects of starches on the hardness of imitation cheese are shown in Table 1. Imitation cheeses containing high amylose (25 to 28% amylose) wheat, potato or maize starches had similar hardness values and were significantly harder than the control. However, rice starch with 13% amylose resulted in a significantly softer product than the control, and the imitation cheese with waxy-maize (3.8% amylose) had the lowest hardness value. The high hardness values of imitation cheeses containing high amylose starches may be due to hydrogen bonding of amylose leached during the imitation cheese cooking. It is also plausible that the shape of starch granules influenced the hardness of the imitation cheeses. It has been suggested that the reinforcing effect of a filler is enhanced when spheres are exchanged for more elongated or flat particles (Nielson and Landel 1994). Therefore the oval or lenticular granules of potato or wheat starch, respectively, (Swinkels 1985) possibly contributed more to the imitation cheese hardness than the more spherical granules of the other starch type.

Cohesiveness reflects the strength of the internal bonds of the cheese body (Szczesniak 1963). In order to obtain a cheese sample that shreds or slices easily, with minimum matting, a moderate cohesion is desirable (Yang and Taranto 1982). All starch-containing products, except that containing rice, had significantly lower cohesiveness than the control, possibly due to their lower protein content (Table 1). Bhaskaracharya and Shah (1999) have reported a positive correlation between protein content and cohesiveness of Cheddar and Mozzarella cheeses. Unlike the paracasein network in which inter- and intra-molecular bonds break and reform during large repeated straining, swollen starch granules would not be expected to contribute to the cohesion of the imitation cheese. The large starch granules (20 to 50 mm) coupled with the high swelling capacity of maize, waxy-maize and especially potato starches possibly disrupted the imitation cheese matrix resulting in fracturing of the protein network under large strain. In contrast, rice starch with its small diameter of about 7 mm and roughly spherical granules as well as limited swelling power appears to have caused little disruption of the imitation cheese protein matrix, resulting in a product with cohesiveness comparable to the control. The combined effect of the large lenticular A-type (up to 40 mm) granules and small spherical B-type (2 to 8 mm) granules of wheat starch (Vansteelandt and Delcour 1999) possibly accounts for the moderate reduction in cohesiveness of the products containing wheat starch.

Dynamic rheology has the potential to provide information relating to the heat-induced changes in the viscoelasticity of imitation cheese that can be useful both at the manufacturing stage as well as at the final end use, for example, in pizza-like products. The effects of starches on the rheological parameters G', G'', and tan δ (G''/G') of imitation cheese measured as a function of temperature are shown in Figures 7, 8 and 9, respectively. The storage modulus (G') is an indication of a material's ability to store energy while maintaining structural integrity (that is, its elastic properties), while the loss modulus (G″) is an indication of the material's ability to dissipate energy (that is, its viscous properties). Tan δ is a useful index of a material's viscoelasticity, as values <1 indicate gel-like behaviour. For all products at 22 °C, the value of G′ was significantly greater than G″, indicating the dominant elastic character of the imitation cheeses. G′ (Figure 7) and G″ (Figure 8) values of all starch-containing products decreased significantly with increasing measuring temperature from 22 to 85 °C, indicating a softening of the cheese matrix. This agrees with results of similar work in natural cheese (Hsieh and others 1993; Rosenberg and others 1994) and model imitation cheese (Zhou and Mulvaney 1998; Mounsey and O'Riordan 1999). When imitation cheese is heated, it is likely that the casein network dissociates and the fat globules liquify, which results in a plasticization of the protein matrix, allowing it to deform and flow. This would explain the inverse relationship between the modulii and temperature. The crossover temperature (G′ = G″, tan δ = 1) of 55.9 °C for the control imitation cheese in this study is the same as that reported by Zhou and Mulvaney (1998) for a model imitation cheese. While the G′ and G″ of all the imitation cheeses decreased with increasing temperature, the changes in both modulii relative to the control were influenced by the starch types. Imitation cheeses containing maize or rice starch had G′ and G″ values similar to the control in the temperature range of 22 to 45 °C, but significantly lower G′ values than the control at temperatures > 50 °C. In the case of wheat or potato starch-containing products, G′ values were significantly higher than the control up to 70 or 80 °C respectively, and G″ values were significantly higher up to 55 or 70 °C, respectively. Products with waxy-maize starch had significantly lower G′ and G″ values than the control at all measuring temperatures. In agreement with hardness values, dynamic results at 22 °C indicated that the products containing high amylose starches were more elastic (higher G′) than the control, while the zero amylose waxy-maize starch resulted in the lowest G′ values. In general, the differences in G′ and G″ between the starch containing products at 22 °C were maintained at higher temperatures.

The tan δ values for all products were similar to that of

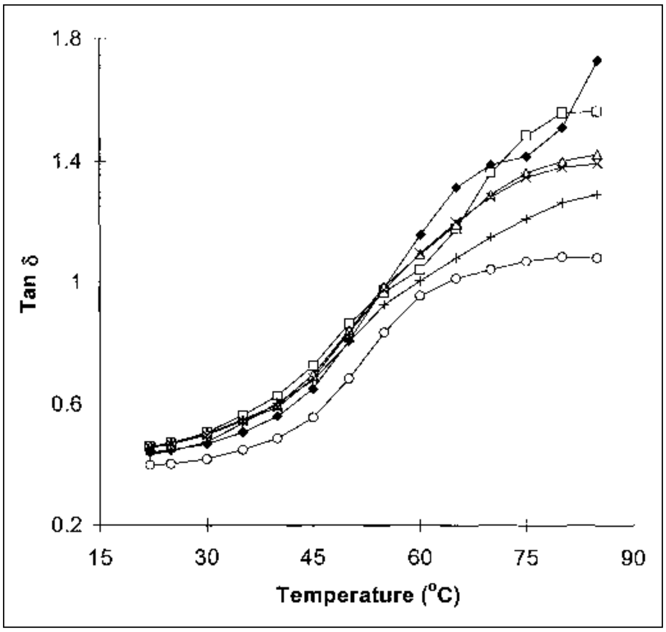

Figure 8—Effect of heating temperature on the loss modulus (G") of imitation cheese containing 0% starch (control) (u), or 3% w/w native waxy-maize, rice (D), wheat ('), maize (+) or potato (o) starch

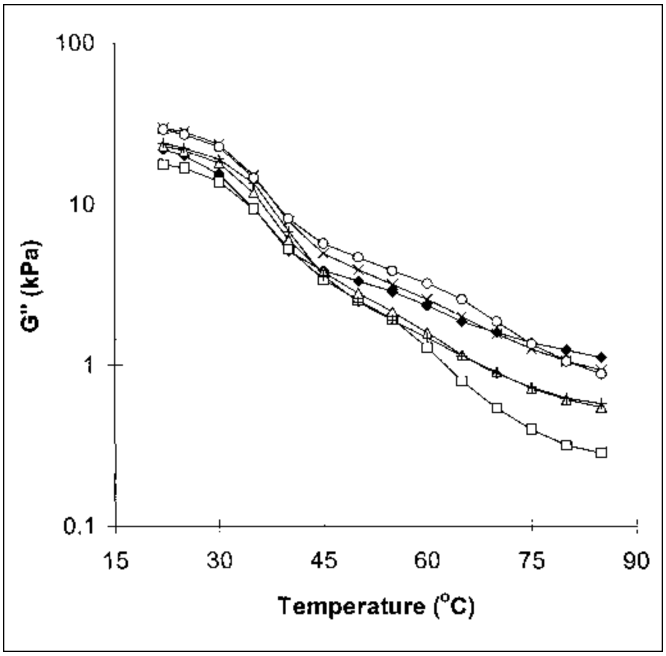

Figure 7—Effect of heating temperature on the storage modulus (G') of imitation cheese containing 0% starch (control) (u), or 3% w/w native waxy-maize, rice (D), wheat ('), maize (+) or potato (o) starch

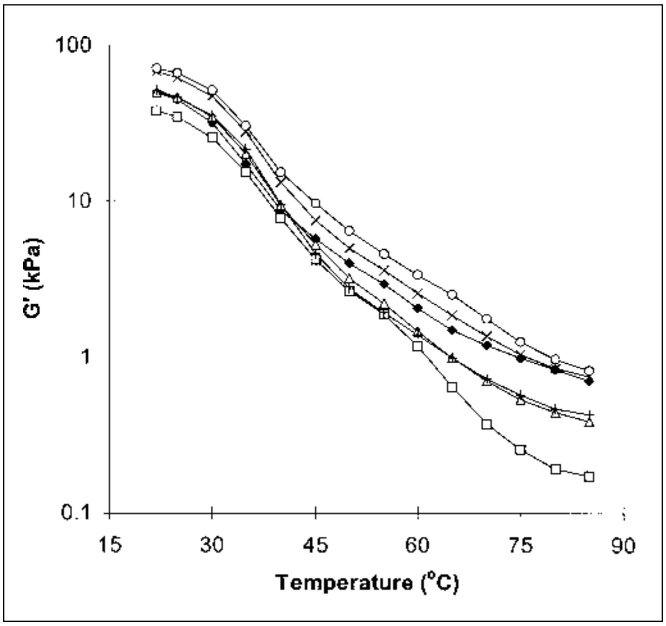

Figure 9—Effect of heating temperature on the tan δ of imitation cheese containing 0% starch (control) (u), or 3% w/w native waxy-maize, rice (D), wheat ('), maize (+) or potato (o) starch

the control imitation cheese (0.44) at 22 °C indicating solid structural behaviour (Figure 9). Tan δ values for all the imitation cheese products increased with increasing measuring temperature up to 85 °C as the elastic component of the sample decreased to a greater extent than the viscous component, indicating a weakening in network arrangement (Sanchez and others 1994). Tan δ values for the products containing waxy-maize, rice or wheat starch were similar to the control product at 22 to 55 °C but on further heating to 85 °C, the tan δ values products containing rice or wheat starch were lower than the control. Tan δ values for products with waxy-maize starch were higher than the control at 60 to 65 °C, similar at 70 to 80 °C but lower at 85 °C. Tan δ values for products containing maize starch were similar to the control at 22 to 50 °C but significantly lower at higher temperatures. Products with potato starch had significantly lower tan δ values compared to the control at all temperatures. Peak tan δ values of imitation cheeses have been shown to correlate well with meltability (Mounsey and O'Riordan 1999). The starches decreased the peak tan δ values of imitation cheeses compared to the control in the following order, potato > maize > wheat > rice > waxy-maize, which suggests an inverse relationship between the amylose content of the starches used and the peak tan δ values. The factors affecting meltability already discussed (such as increased fat emulsification, a poorly hydrated protein matrix, the presence of retrograded amylose and starch granules/fragments in imitation cheeses containing starch) probably account for the reduced tan δ values compared to the control, especially at temperatures greater than 55 °C.

Conclusions

THE EFFECTS OF PARTIALLY REPLACING CASEIN WITH STARCH on the meltability and rheological properties of imitation cheese were dependent on the origin of the starch used. Starches such as maize, potato and wheat reinforced imitation cheese structure but this strengthening effect was at the expense of imitation cheese melt where dehydration of the protein matrix, coupled with lack of thermoplasticity of the embedded starch, probably impaired softening and flow at high temperatures. Rice starch with its small granular size, relatively low amylose content and limited swelling capacity had the least effect on the imitation cheese melt and resulted in products with acceptable rheological properties. Thus rice starch appears to have potential as a low-cost partial casein replacer in imitation cheese.

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MS 19991138

This work was supported by funding from the Irish Department of Food, Forestry and Agriculture.

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