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EFFECT OF EXTRUDER DIE TEMPERATURE ON TEXTURE AND MICROSTRUCTURE OF RESTRUCTURED MECHANICALLY DEBONED CHICKEN AND CORN STARCH

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Abstract
Proximate composition, textural properties and microstructural changes of mechanically deboned chicken/15% corn starch extrudates were evaluated when restructured at die temperatures of 25, 71, 82, 93, 104 and 115°C in a twin-screw extruder. Total solids and fat content decreased, whereas protein content increased as die temperatures were increased. When die temperature was increased from 71°C to 104°C, apparent stress at failure of extrudates increased by 44 kPa, but decreased at a die temperature of 115°C. Changes in the protein matrix, fat globules and starch granules due to changes in extruder die temperature were observed by scanning electron microscopy. Extrudates prepared at die temperatures of 71°C and 82°C exhibited microstructures similar to those observed for salt-soluble muscle protein gels. Extrudates prepared at die temperatures above 93°C exhibited microstructures more typical of gelatinized starch.

Introduction
Twin-screw extrusion (TSE) is one of the most popular new processes used by the food industry to produce expanded snack foods, ready to eat cereals and pet foods (Chinnaswamy and Hanna, 1988; Chinnaswamy et al., 1989). Twin-screw technology has been used extensively to prepare meat analogs from defatted vegetable proteins in the United States and to texturize cereals to produce bread analogs in Europe. This technology has also been extensively utilized in Japan (Gwiazda et al., 1987).

Mechanically deboned chicken (MDC) is used in a variety of comminuted products such as bologna and frankfurters (Froning et al., 1971; Gruden et al., 1972). Studies on extrusion and texturization of mechanically deboned poultry indicated the need for further improvement in the binding of restructured products (Lampila et al., 1985). The use of TSE was suggested to improve the texture of MDC (Lingle, 1987). Restructuring of MDC using TSE has been accomplished with the addition of various binding agents such as soy protein isolate, vital wheat gluten, wheat flour, corn starch and carrageenan (Megard et al., 1985; Alvarez et al., 1990).

Microstructural studies have been shown to be a useful tool for observing textural changes in processed meat products. Scanning electron microscopy (SEM) has been used to study microstructures of meat gels, emulsions and batters and their changes due to different ingredients and processing variables (Katsaras and Stenzel, 1984; Schmidt, 1984; Lee, 1985; Barbut, 1988). Studies demonstrating the effect of extrusion-cooking on the textural and microstructural properties of starch have been reported (Colonna and Mercier, 1983; Gomez and Aguilera, 1984; Diosady, 1985). Scanning electron microscopy has been used to investigate changes due to the
addition of wheat gluten, soy protein isolate and corn starch to different processed meat products (Siegel et al., 1979a; 1979b; Comer et al., 1986). Information on the microstructure and textural properties of mixtures of food ingredients such as MDC and corn starch during and after extrusion is limited (Chinnaswamy et al., 1989). Little information is available concerning the effect of specific extrusion conditions, such as barrel and die temperatures, on extrudate microstructure and texture. The objective of this study was to investigate changes in the texture and microstructure of restructured MDC containing 15% corn starch as a function of extruder die temperature.

Materials and Methods

Materials

Mechanically deboned chicken meat (without skin) was obtained from Nottawa Gardens Co. (Athens, MI) and held at -25°C until used (within two months). Corn starch (Argo, CPC International Inc., Englewood Cliffs, NJ) was used as a binder during extrusion. Thawed MDC was mixed with 15% corn starch, 1.5% salt and 0.5% sodium tripolyphosphate (Stauffer Chemical Co., Washington, PA) for 15 min in a Butcher Boy mixer (Model 250F, Lasar Mfg. Co., Inc., Los Angeles, CA) and held at 4°C for 24 hr prior to use.

Extrusion Conditions

A pilot scale Baker-Perkins MFP 50 D/25 twin-screw extruder (APV Baker Inc., Grand Rapids, MI) with a die composed of 3 horizontal parallel slits, each measuring 1.8 cm (width) x 0.3 cm (height), was used to restructure MDC at 350 rpm and a product feed rate of 1.0 Kg/min. The effective length:diameter ratio of the extruder was 25. Mixtures of MDC and corn starch were fed into the first feeding section port using a Moyno pump (Model IFF J4, Robbins and Meyer, Inc., Springfield, OH). Die temperatures of 71, 82, 93, 104, and 115°C were achieved by adjusting temperatures along the 7 zones of the extruder barrel as described by Alvarez et al. (1990). Average residence time in the extruder was 2.6 min. Extrudates from each of three extrusion runs were collected in duplicate in 2.8 cm (diameter) x 15 cm (length) plastic tubes and held at 4°C until analyzed within 5 days.

Product Analysis

Moisture, fat and protein were determined on raw MDC and extrudates after each extrusion run following AOAC (1984) procedures 24.003, 24.005 and 24.038-24.040, respectively. Apparent stress and strain at failure (Hamann, 1983) of duplicate 1.5 cm diameter x 1.5 cm length extrudate cores were measured using an Instron Universal Testing Machine (Model 4202, Instron Engineering Corporation, Canton, MA) at a crosshead speed of 10 mm/min with a 50 N compression cell by compressing between two lubricated parallel plates (Nuckles et al., 1990). Cores were removed from the extrudates with a 1.5 cm diameter cork borer and cut to 1.5 cm lengths using a template and razor blade.

Scanning Electron Microscopy

Specimen preparation. Extrudates were prepared for SEM as described by Klomparsens et al. (1986). Extrudates were equilibrated to 4°C and cubes of approximately 1.5 mm x 1.5 mm were cut from the extrudates with a chilled razor blade. Samples were fixed with 5% (v/v) glutaraldehyde for 2 hr, rinsed twice in 0.1 M Na phosphate buffer, pH 7.0 and post-fixed in 1% (w/v) OsO₄ in 0.1 M Na phosphate buffer, pH 7.0 for 2 hr followed by rinsing in the same buffer. The samples were dehydrated in a series of 25%, 50%, 75%, 95% and 100% (v/v) ethanol for 15 min each. The 100% ethanol dehydration step was performed three times. Samples were critical point dried using carbon dioxide (Balzers Union Model O10 Critical Point Dryer, Balzers, Liechtenstein) and mounted on 10 mm (diameter) x 5 mm (height) aluminum stubs (Electron Microscopy Sci., Ft. Washington, PA) using adhesive mounting tabs (M.E. Taylor Engineering, Brockville, MD). An Emscope Sputter Coater (Model SC500, Kent, England) was used to coat the samples with gold under a vacuum of 0.06 Torr. Observations were made using a JEOL scanning electron microscope (Model JMS-35CF, Osaka, Japan) equipped with a tungsten electron gun at an accelerating voltage of 15 kV.

Statistical Analysis

Triplicate extrusion runs were used to evaluate each treatment. Duplicate samples from each treatment were collected during each extrusion run. Proximate, textural and microstructural analyses were performed in duplicate on each sample collected. Statistical significance was determined using two-way analysis of variance (replicate x treatment). Means were separated using Tukey's Test and the mean square error term at the 5% level of probability (MSTAT, 1989).

Results and Discussion

Proximate Composition

On a total solids basis, MDC extruded at 25°C was composed of 38.3% protein and 24.7% fat (Table 1). Starch probably comprised the majority of the
the extrudates. The decrease in solids may be related to an increase in the water binding capacity of the MDC/corn starch extrudates due to gelation of proteins, gelatinization of starch and formation of a protein-starch matrix as die temperature was increased. Bhattacharya and Hanna (1987) reported that the water-holding capacity of non-waxy corn starch increased as barrel temperature was increased from 115°C to 164°C.

In general, fat contents of extrudates decreased (P < 0.05) while protein contents did not change (P > 0.05) as extrusion temperatures were increased. Extrusion of MDC/corn starch at a die temperature of 115°C exhibited a 1.9% increase in protein content and a 7.0% decrease in fat content compared to the 25°C treatment. Temperature and shear during extrusion may have caused melting of fat which was lost from the protein-starch matrix as drip. Alvarez et al. (1990) also reported a decrease in solids and an increase in fat of extrudates when die temperature was increased from 71°C to 82°C. At higher extrusion temperatures, Van Zuilichem and Jager (1990) found that moisture content of extrudates containing 80% lean pork and 20% corn starch decreased when extruded at temperatures of 160°C and above.

Table 1. Total solids, protein and fat content of mechanically deboned chicken containing 15% corn starch extruded at different die temperatures.

<table>
<thead>
<tr>
<th>Die Temperature (°C)</th>
<th>Solids (%)</th>
<th>Protein (%)</th>
<th>Fat (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>25</td>
<td>39.3b</td>
<td>38.5b</td>
<td>24.7b</td>
</tr>
<tr>
<td>71</td>
<td>39.6b</td>
<td>37.4b</td>
<td>20.6b,c</td>
</tr>
<tr>
<td>82</td>
<td>36.8c</td>
<td>39.8b</td>
<td>20.4c</td>
</tr>
<tr>
<td>93</td>
<td>36.4c</td>
<td>40.0b</td>
<td>18.2c</td>
</tr>
<tr>
<td>104</td>
<td>36.3c</td>
<td>40.7b</td>
<td>18.2c</td>
</tr>
<tr>
<td>115</td>
<td>35.7c</td>
<td>40.3b</td>
<td>17.7c</td>
</tr>
</tbody>
</table>

The extrudates. The decrease in solids may be related to an increase in the water binding capacity of the MDC/corn starch extrudates due to gelation of proteins, gelatinization of starch and formation of a protein-starch matrix as die temperature was increased. Bhattacharya and Hanna (1987) reported that the water-holding capacity of non-waxy corn starch increased as barrel temperature was increased from 115°C to 164°C.

Textural Properties

Apparent stress at failure of extrudates increased (P < 0.05) from 8.3 kPa to 52.5 kPa as die temperature was increased from 71°C to 104°C (Table 2). A die temperature of 115°C resulted in a decrease (P < 0.05) in the apparent stress of extrudates to 31.6 kPa. In general, apparent strain at failure of extrudates increased as die temperature was increased, except in the 104°C treatment. Differences in extrudate composition might cause variations in apparent stress and strain (Foegeding and Ramsey, 1987). Also, the extent of starch gelatinization and protein denaturation as a function of die temperature may have influenced the textural properties of the extrudates.

Nuckles et al. (1990) reported values of 42.8 kPa and 0.76 for apparent stress and strain at failure, respectively, in frankfurter model systems containing MDC. Extrudates containing 15% corn starch exhibited similar apparent stress values when processed at die temperatures of 93°C. However, apparent strain at failure of MDC extrudates was lower than the model system frankfurters.

Effect of Temperature on Microstructure

Extrusion at a die temperature of 71°C produced microstructures exhibiting large areas of a fibrous, highly cross-linked honeycomb-like protein matrix, starch granules, fat particles and some voids of different sizes distributed throughout the matrix. Extrudates containing 15% corn starch exhibited similar apparent stress values when processed at die temperatures of 93°C. However, apparent strain at failure of MDC extrudates was lower than the model system frankfurters.

Table 2. Apparent stress and strain at failure of mechanically deboned chicken containing 15% corn starch extruded at different die temperatures.

<table>
<thead>
<tr>
<th>Die Temperature (°C)</th>
<th>Apparent Stress at Failure (kPa)</th>
<th>Apparent Strain at Failure (unitless)</th>
</tr>
</thead>
<tbody>
<tr>
<td>71</td>
<td>8.3d</td>
<td>0.40b</td>
</tr>
<tr>
<td>82</td>
<td>31.2c</td>
<td>0.38b</td>
</tr>
<tr>
<td>93</td>
<td>45.4b</td>
<td>0.44a</td>
</tr>
<tr>
<td>104</td>
<td>52.5a</td>
<td>0.32c</td>
</tr>
<tr>
<td>115</td>
<td>31.6c</td>
<td>0.47a</td>
</tr>
</tbody>
</table>

Means within columns followed by the same letter do not differ significantly (P < 0.05).
(Fig. 1). Similar protein matrices and fat globules have been identified in SEM studies of restructured and comminuted meat products (Comer et al., 1986; Barbut, 1988; Comer and Allan-Wotjas, 1988).

The protein matrix observed at 71°C is similar to the microstructure of gels prepared from bovine myosin (Hermansson et al., 1986) and chicken breast salt soluble protein (Wang et al., 1990). Incomplete protein gelation and minimal starch gelatinization in samples extruded at 71°C may have caused the low apparent stress values (Wang et al., 1990).

Fat globules in the 71°C extrudates varied from 1 to 10 μm. Fat particles found in meat emulsions ranged in size from 1-100 μm (Lee, 1985), whereas fat globules of 10-50 μm were identified in SEM micrographs of poultry meat batters (Barbut, 1988).

Extrudate microstructures also contained numerous voids. The voids may have originated from lipid globules, air or water droplets. Voids may have formed in the extrudates when air was incorporated during mixing in the extruder barrel or during product expansion due to pressure drop at the die. Air pockets have been reported to influence the texture of restructured beef products (Bernal and Stanley, 1986).

Extrudate microstructures also contained regions which appeared to be gelatinized starches (Owusu-Ansah et al., 1990). Muscle protein gels formed by heating above 65°C may exhibit decreased water holding capacity and protein extraction and gelation which forms a heat set matrix to enclose components such as fat, starch and water (Siegel and Schmidt, 1979; Samejima et al., 1981; Trout and Schmidt, 1984). The extruder die temperature and residence time were insufficient for complete starch gelatinization as indicated by the presence of intact starch granules. At a die temperature of 93°C, the extrudates contained regions which appeared to be predominantly a protein matrix and were similar to microstructures observed in extrudates processed at die temperatures of 71°C and 82°C (Fig. 3). Other distinct regions appeared to be gelatinized starch and were expected to contribute different textural properties to the extrudates.

Starch gelatinization occurs between 90°C and 150°C, depending on the moisture content and residence time in the extruder (Lawton et al., 1972). Protein gelation and starch gelatinization at 93°C resulted in an increase in apparent stress at failure of the extrudates. Apparent strain at failure increased in extrudates processed at a die temperature of 93°C in comparison to extrudates processed at lower temperatures, which may be related to degree of starch gelatinization. Gelatinized starch would be expected to contribute different textural properties to the extrudates than intact starch granules (Oakenfull, 1987; Bhuiyan and Blanshard, 1982).

Microstructures of extrudates obtained at a die temperature of 82°C (Fig. 2) did not differ markedly from samples extruded at 71°C, except the protein matrix was more compact and composed of thicker, more globular protein strands. The apparent stress at failure of the extrudates were greater than those processed at a die temperature of 71°C. Regions of a cross-linked protein matrix, intact starch granules and fat globules ranging in size from 0.3 to 6 μm were observed.

The characteristics of the protein matrix may indicate that extrusion die temperatures of 71°C and 82°C resulted in protein denaturation and gelation. Thermally induced unfolding and aggregation of salt soluble muscle proteins form a matrix that contributes to the texture, water holding and fat holding properties of meat products (Smith, 1988). Chicken salt soluble proteins exhibit thermal transition temperatures of 57°C to 60°C when measured by differential scanning calorimetry (Rijowski and Mast, 1988). A combination of mechanical action, ionic strength, and temperature causes tissue disruption,
Restructured Mechanically Deboned Chicken

decreased strength due to extensive protein aggregation (Hermansson, 1986; Wang, 1990). Thermal treatment of proteins may result in structural changes due to hydrolysis of peptide bonds, formation of new covalent cross-links and modification of amino side chains (Cheftel et al., 1985). Plasma protein gels (Hermansson, 1983) and chicken salt-soluble protein gels (Wang et al., 1990) exhibit shrinkage and partial disruption of the gel network leading to decreases in gel strength when heated above a critical temperature. Chinnaswamy and Hanna (1988) observed that increasing barrel temperatures above 110°C caused starch degradation and textural problems in corn starch extrudates.

Fig. 1. Scanning electron micrograph of mechanically deboned chicken and 15% corn starch mixtures extruded by twin-screw extrusion at a die temperature of 71°C. (f) fat globule, (s) starch granule, (p) protein matrix. Bar length is 1 µm.

Fig. 2. Scanning electron micrograph of mechanically deboned chicken and 15% corn starch mixtures extruded by twin-screw extrusion at a die temperature of 82°C. (s) starch granules, (p) protein matrix, (f) fat globule. Bar length is 1 µm.

Fig. 3. Scanning electron micrograph of mechanically deboned chicken and 15% corn starch mixtures extruded by twin-screw extrusion at a die temperature of 93°C. (cp) protein-starch matrix, (p) protein matrix. Bar length is 1 µm.

Fig. 4. Scanning electron micrograph of mechanically deboned chicken and 15% corn starch mixtures extruded by twin-screw extrusion at a die temperature of 104°C. (cp) protein-starch matrix. Bar length is 1 µm.

Fig. 5. Scanning electron micrograph of mechanically deboned chicken and 15% corn starch mixtures extruded by twin-screw extrusion at a die temperature of 115°C. (b) bacteria or lipid globules, (c) cracks, (cp) protein-starch matrix. Bar length is 1 µm.
The results indicate that die temperature had significant effects on the textural properties of extruded MDC containing 15% corn starch. It was possible to obtain different textural attributes in the finished product by changing the die temperature. Scanning electron microscopy was useful for observing changes in proteins, lipids and starch granules in extruded products due to changes in die temperature.

Conclusions


Siegel DG, Church CE, Schmidt GR. (1979a). Gel structure of nonmeat proteins as related to their ability to bind meat pieces. J. Food Sci. 44:1276-1279.


Discussion with Reviewers

R. Chinnaswamy: How do you make 1.5 x 1.5 cm cores out of 1.8 x 0.3 cm dies? Authors: The MDC/corn starch leaving the extruder is in a molten or fluid-like condition, thus the strips of material exiting the die flow back together to form a cohesive mass which will take the shape of the collection container. Cores of 1.5 x 1.5 cm were cut for textural analyses.

R. Chinnaswamy: Your micrographs show differential distribution of fat, protein and starch granules. Such non-uniform distribution characteristics of protein, fat and starch bodies may also influence stress and strain values. Is this correct? Authors: Non-uniform distribution of ingredients would be expected to influence textural properties of extrudates, however in this case, we do not think this caused any problems with the textural analyses as the standard deviations for apparent stress and strain were small. Also, the components are...
quite small when compared to the sample size used. The micrographs show any area of 150 μm², whereas the surface area of the core used for textural analysis was approximately 1.8 x 10^6 μm².

R. Chinnaaswamy: Do you have any data or literature which would support your relationship between extrudate elasticity and starch gelatinization?

S. Barbut: Can you explain the significant decrease in apparent strain at 104°C and then the increase observed at 115°C?

Authors: We cannot definitely explain the relationship between starch gelatinization and elasticity in our extrudates, however a wide variety of temperature dependent reactions occur during extrusion which might influence the textural properties of the finished product. Fat droplet size is changing due to shear, fat is melting, starch is gelatinizing, proteins are gelling and starch and protein are competing for available water. The relative proportion of starch granules to gelatinized starch will also affect the textural properties of the extrudates. Thus, at any extrusion temperature a very complex set of chemical and physical interactions will determine product texture. Some of the possible types of ingredient interactions were reviewed recently by Lanier (1991).

S. Barbut: You indicated that fat content decreased significantly when extrusion temperature increased. Were there any visible signs (i.e. a more greasy surface) in the lower fat treatments?

Authors: None of the extrudates appeared greasy. At the higher extrusion temperatures, some free fat was observed dripping out at the die. Also, fat volitilization at high extrusion temperatures is another possible explanation for the decrease in fat content of the extrudates.

S. Barbut: In Fig. 5 you identified "b" as bacteria or lipid. Can the authors further elaborate on this almost perfectly round structure?

Authors: We cannot positively identify this structure. We did not observe this structure in the 104°C treatment and only a few (less than 5) were observed in the 115°C treatment.

Additional References