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MICROSTRUCTURAL STUDIES OF TEXTURIZED VEGETABLE PROTEIN PRODUCTS: EFFECTS OF OIL ADDITION AND TRANSFORMATION OF RAW MATERIALS IN VARIOUS SECTIONS OF A TWIN SCREW EXTRUDER

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Abstract

In high-temperature short-time extrusion cooking with a twin screw extruder, effects of oil addition to defatted soybean flour and microstructural transformation of full-fat soybean flour during cooking, were investigated by use of a light microscope. At levels up to 15%, soybean oil was distributed in the protein and carbohydrate matrix as small, spherical drops under the experimental conditions used in this study. However, oil contents above 15% significantly prevented formation of well-aligned fibrous structures in the extrudates. During extrusion cooking, the starting materials began to break down by shearing and kneading forces in the feed section but gross cellular structures remained up to the cooking zone. After being introduced into the cooking zone, protein and carbohydrate were plasticized and appeared to be stream-like. Passage through the breaker plate and long cooling die induced formation of a fiber-like extrudate.

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Key words: Microstructure, soybean, texturized vegetable protein, extruder, extrudate, oil, protein, twin screw, barrel

Introduction

Extrusion cooking technology has been used to texturize many foodstuffs to produce expanded, texturized or fibrous structures. In the United States, such processes using a single screw extruder have found extensive application to prepare meat analogs mainly from defatted vegetable proteins, while in Europe, twin screw extruders have been primarily used to texturize cereals to produce bread analogs. These technologies were also introduced into the Japanese food industries and have been rapidly developed and applied in the Japanese market.

Over the past few years, the twin screw extruder has been especially appealing to the Japanese food industries because of its potential to operate for diverse purposes including compression, mixing, kneading, reaction, texturization, sterilization and inactivation of enzymes.

Because microstructural technologies have proven to be useful tools for studying the qualities and changes of texturized products, many researchers have examined effects of protein contents, protein solubility, pH, ionic strength and additives on texturized products prepared with single screw extruders (Faubion and Hoseney, 1982a; 1982b; Neumann et al., 1984; Noguchi et al., 1981; Rhee et al., 1981; Simonsky and Stanley, 1982). However, few papers have reported effects of lipid content (Faubion and Hoseney, 1982b), because a single screw extruder does not permit use of high lipid contents as a result of its design where materials are transported by frictional force between the barrel and screw. We have therefore examined the effects of oil addition on microstructure of soybean extrudates and describe our results here.

Secondly, this paper deals with microstructural transformations of the raw materials as they progress through the barrel of an extruder. Aguilera et al. (1976) set up 7 sampling points in a Wenger X-5 single screw extruder and investigated the progressive changes in soybean grits as they passed through the extruder. Our experiments were similar but used a Creusot-Loire BC-45 twin screw extruder with a long cooling die and our starting materials were full-fat soy meal plus a small amount of potato starch in order to better understand the transformation or deformation of cellular structures and added starches.

Materials and Methods

Materials

Effects of oil addition on microstructure of extrudates were studied with defatted soybean meal (Nisshin FT) containing 55% crude protein and 10.5% moisture and having a Nitrogen Solubility Index (NSI) of 31.1. It was premixed with 5, 10 or 15% soybean oil in a kneader and chopper.

A full-fat soyflour-starch mixture used to determine the transformation of materials in the extruder was made by grinding whole soybeans, sieving to 0.5 mm, and then mixing with 5% potato starch.

Extrusion-cooking conditions

A Creusot-Loire BC-45 co-rotating twin screw extruder with a long cooling die attached was used. Screw speed was 60 rpm except for the defatted flour containing 15% oil which was extruded at 120 rpm. Feed rate was 15 kg/hr. The barrel temperature in normal operation was 180°C. Screw geometry was Reverse-Forward-Reverse-Forward-Forward from the die side as shown in Fig. 1. Moisture contents of the sample flours were 60%.

Preparation of microscopic specimens

The initial soybean flours were mixed with lukewarm agar sol and coagulated. A small piece of the agar was then fixed with 1% osmium tetroxide in phosphate buffer (pH 7), dehydrated with an ethanol series, and then embedded in Epon resin. The resin was sliced, placed on glass slides and stained with Coomassie Brilliant Blue (CBB) for protein or periodic acid Schiff reagent (PAS) for carbohydrates; the samples were examined under a light microscope. The extrudates were treated the same as the initial flours except for the embedding in agar gel. The extrudates were examined lengthwise (along the direction of extrusion) and crosswise (at right angles to the direction of extrusion). The extruder contents were removed at locations A-G shown in Fig. 1 at 5 to 7 min. after stopping the extruder. The extruder contents and extrudates were treated as described above.

Rheological measurement of extrudates

The extrudates were punched out in the form shown in Fig. 2, and used to measure the maximum breaking strength with a Fudo Rheometer (model NRM-300 2D). The deformation rate was 6 cm/min. The values obtained were averages of a minimum of 8 samples.

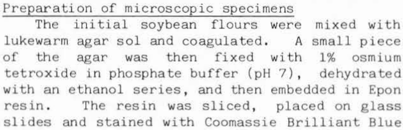


Fig. 1. Configuration of the extruder and sampling positions (A-G).

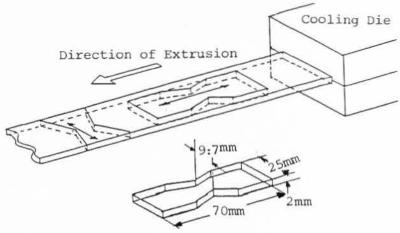


Fig. 2. Sample preparation for texture measurements.

Results and Discussion

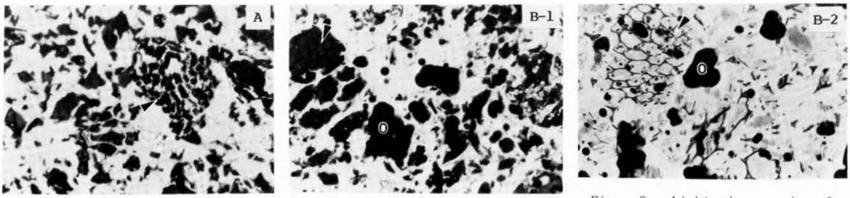
Figure 3 shows micrographs of the initial defatted flours with varying levels of added soybean oil. Gross cell disruption including agglomerated protein bodies and cell walls as observed. Added oil was not associated with protein but was scattered in big and small amorphous drops.

After extrusion cooking, the extrudates observed in lengthwise sections show well aligned fiber-like structures as shown in Fig. 4 (A - D). However, the lengthwise orientation of the fibers decreased, thickness of the fibrils increased and size of the air cells increased as the oil content increased. The fat globules observed in the fibrous protein matrix were smaller and more spherical than oil droplets in the initial flours. Under these extrusion cooking conditions, all of the added oil existed in this state.

The microstructures in crosswise sections also clearly show fibrous-like networks (Fig. 4 E - H). As in the lengthwise sections, thickness of the fibrils increased and size of the air cells increased as oil content of the extrudates increased.

The maximum breaking strengths of the extrudates is shown in Fig. 5. The values of breaking strength of the extrudates in lengthwise samples, decreased rapidly as oil content increased. In contrast, the crosswise samples decreased slightly at 5 and 10% but increased significantly at 15% added oil. These results, obtained by rheological measurement, agree with the microstructural changes observed on varying oil content; namely, the orientation of fibrils changed from lengthwise to crosswise at more than 10% oil. Under the extrusion cooking conditions used in this experiment, oil was expressed during cooking when more than 20% oil was present and light micrographs showed large oil drops outside of the protein matrix (data not shown).

Figure 6 shows the full-fat flour before (A) and after it had progressed through the forward (long and short pitch) and reverse screws (B-1 and B-2). In the raw flour, the added potato starch granules were clearly observed, whereas after mixing and kneading by the screws, no



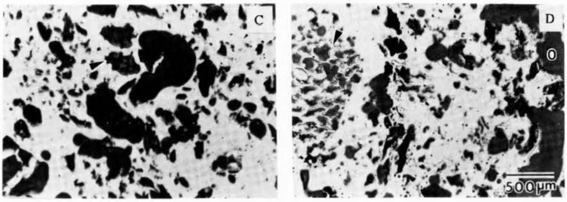


Fig. 3. Light micrographs of defatted soybean flours with 0 - 15% added soybean oil: (A) initial defatted soybean flour stained with CBB; (B-1) 5% added oil and stained with CBB; (B-2) 5% added oil and stained with PAS; (C) 10% added oil and stained with CBB; and (D) 15% added oil and stained with CBB. Magnification for all samples as shown in D. Arrows show gross cell disruption. O = oil droplets.

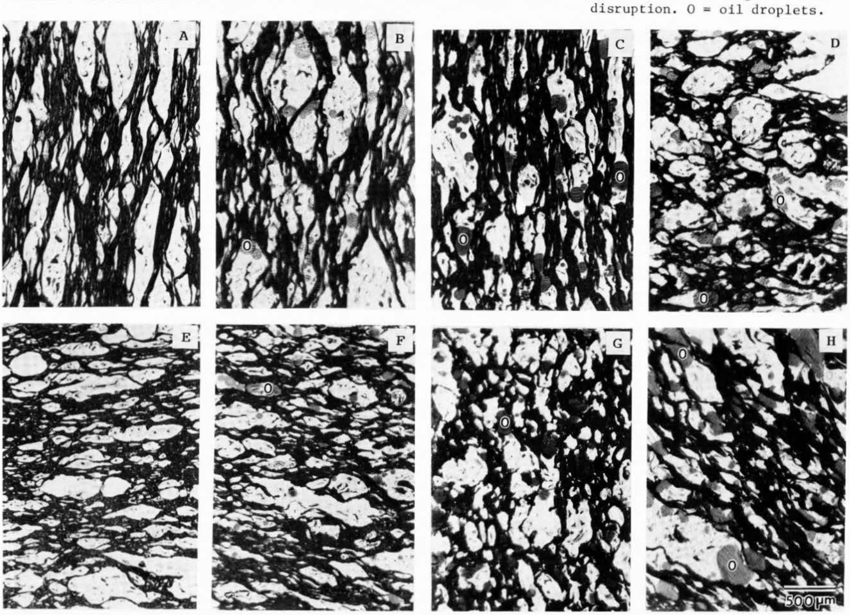


Fig. 4. Light micrographs of extrudates prepared from defatted soybean flours with 0 - 15% added soybean oil and stained with CBB.
A, B, C and D; extrudates in lengthwise sections with added soybean oil of 0, 5, 10 and 15%, respectively, stained with CBB. E, F, G and H; extrudates in crosswise sections with added soybean oil of 0, 5, 10 and 15%, respectively, stained with CBB.
O; oil droplets scattered throughout fibrous network. Magnification for all figures as shown in H.

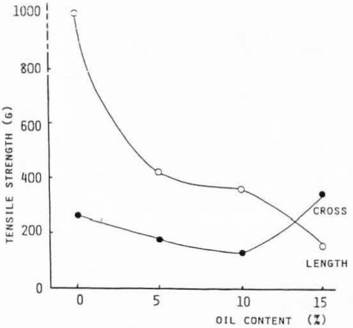


Fig. 5. Effect of oil addition on breaking strength of extrudates prepared from defatted soybean flour. Length-tensioned along the direction of extrusion and cross-tensioned at right angle to extrusion.

granules were found. However, the cellular materials retained their gross structures as shown by staining with CBB (B-1) and/or PAS reagent (B-2) and small oil droplets have begun to appear in the matrix. Both carbohydrate and protein were intertwined in "stream" formation. This agrees with Rhee et al. (1981) who reported that insoluble carbohydrate formed a matrix with plasticized protein in stream formation.

When the materials were introduced into the cooking zone (note location of induction heater in Fig. 1), protein and carbohydrate began to plasticize and the microstructure appeared to be more stream-like (Fig. 6, C - E). As the flour passed through the perforated breaker plate and the long die, fiber formation progressed to completion (Fig. 6 F, G). The results agree with the conclusion of Aguilera et al. (1976) that in a single screw extruder most of the structural changes, such as strand formation, occurred above 145°C and that plasticization took place just before the die.

Under this extrusion cooking condition, the solid fibrous structure obtained with defatted soybean (Fig. 4) could not be prepared from full-fat flour. However, when the extrudate was

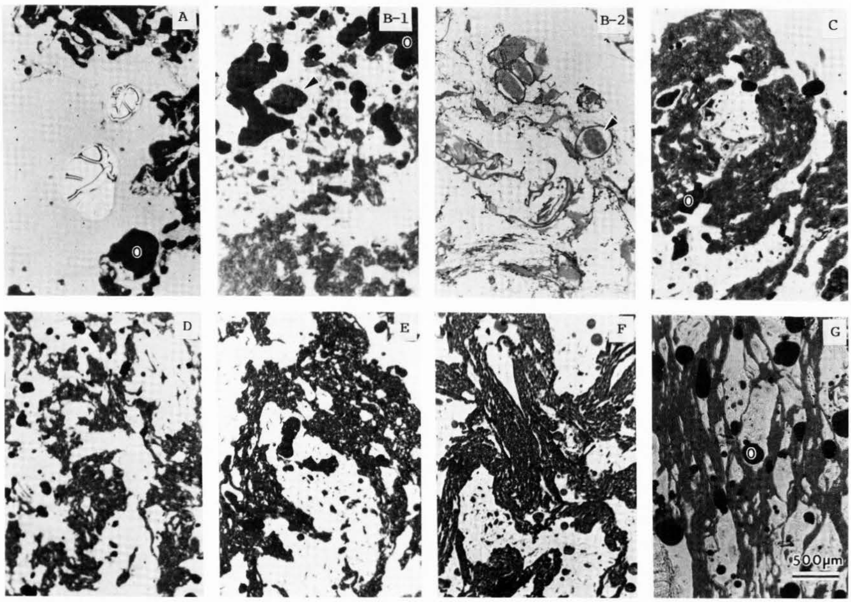


Fig. 6. Changes in microstructures of full-fat soybean flour and added starch during high-temperature short-time extrusion cooking; (A) initial full-fat soybean flour + potato starch; and (B - G) extrudates taken from positions B - G of the extruder (Fig. 1) Stained with CBB except for B - 2 which was stained with PAS. O = oil droplets.

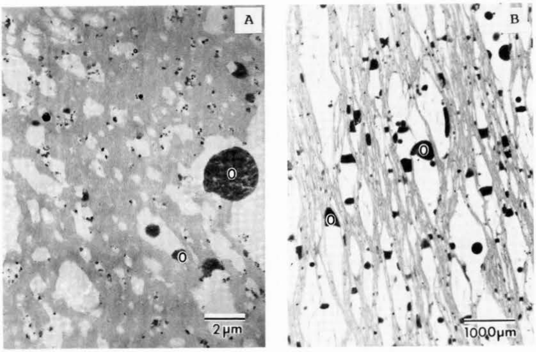


Fig. 7. Electron (A) and light (B) micrographs of extrudate prepared from the one-to-one mixture of full-fat soybean flour and defatted soybean flour. O: oil droplets. Small black spots scattered throughout fibrous matrix in Fig. 7-A are also oil globules.

prepared under the same condition but by addition of defatted soybean flour to the full-fat flour in a 1:1 ratio, a highly fibrous structure could be extruded (Fig. 7). This fact suggests that oil inhibits formation of fibrous structure. However, more than 20% oil may be used by selecting more reverse or kneading screws in the higher length/diameter barrel of a twin screw extruder.

Acknowledgement

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Discussion with Reviewers

A.-M. Hermansson: Can the authors speculate on the mechanisms behind the effect of the oil content on the alignment and formation of fibrous structures of the extruder?

Authors: Although the interactive reactions during extrusion cooking are still obscure, it has been speculated that hydrogen, ionic and hydrophobic bondings contribute as do disulfide bonds to intramolecular reactions between protein and carbohydrates. Intramolecular peptide bonds are also likely to form. Oil may interact with soybean protein by hydrophobic bonding and might prevent direct protein - protein reactions. But fundamental experiments are necessary to clarify this point.

W.J. Wolf: What is the evidence that all of the added oil was present in the form of globules or droplets? Can you rule out the possibility of protein - lipid interaction at the molecular level which would not be detected by light microscopy?

Authors: At present we cannot completely rule out protein-lipid interactions at the molecular level. However, Fig. 7-A, obtained with the electron microscope, likewise indicates that the lipid is dispersed as minute globules in the mesh-like structure.