Optimization of Initial Moisture, pH, and Extrusion Temperature of an Acetone-Extracted Maize Gluten Meal and Soy Extrudate for Use in Pet Foods[1]

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Cereal Chem. 65(3):267–270

ABSTRACT

This study was conducted to optimize textural properties of acetone-extracted maize gluten meal (AEMGM) and defatted soy flour (DSF) extrudates to achieve an extruded product for use as a canned pet food ingredient. Optimal blend moisture, pH, and extruder exit temperature were determined using a three-variable response-surface statistical design and analysis. Acidifying the blend, increasing the moisture level, and increasing the extrusion temperature improved structural integrities of the resultant extrudates. Upon retorting, all extrudates remained intact with no evidence of disintegration. As extruder exit temperature increased, the amount of trypsin inhibitor activity in the resultant AEMGM-DSF extrudates decreased. The blend ratio, moisture, and pH level did not affect trypsin inhibitor activity. Optimum textural properties of AEMGM-DSF blends occurred using a 50:50 AEMGM-DSF ratio with moisture level of 32%, a pH of 6.25, and an extruder exit temperature of 153°C. Optimum extruded products were characterized by a low water-holding capacity and inactivation of trypsin inhibitor activity.

During wet milling of maize, ground endosperm is separated into starch and a high-protein gluten fraction, maize gluten meal (MGM). MGM, with a protein content of approximately 60%, is composed of zein and glutelin protein that are rich in leucine and glutamic acid but limiting in lysine and tryptophan (Shroder and Heiman 1979). MGM has been used in poultry feed as a valuable xanthophyll source (Reiners et al 1973), for protein fortification in pet foods, and as a partial flour replacement for bakery products (Feldberg 1965).

Protein complementation in blended MGM extrudates can contribute to nutritional and functional improvements in food products (Mustakas et al 1970, Conway and Anderson 1973, Peri et al 1983, Neumann et al 1984, Koeppe et al 1987). Extrusion conditions can alter both nutritional and functional qualities (Cummings et al 1972, Smith 1975, Mercier and Feillet 1975, Bhattacharya et al 1986, Koeppe et al 1987). Extrusion variables include both ingredient factors (ingredient ratios, initial moisture content, and pH) and processing factors (extrusion temperature gradients, pressure and shear rate differences). Mechanical and thermodynamic variations of extrusion processing can affect product structure, texture, density, viscosity, solubility, rate of rehydration, and shape of the final texturized product. Comprehensive reviews of extrusion processing have been written by Kinsella (1978) and El-Dash (1981).

The objective of this study was to optimize extrusion conditions of acetone-extracted maize gluten meal (AEMGM)-defatted soy flour (DSF) extrudates to achieve a canned pet food ingredient.

MATERIALS AND METHODS

Sample Preparation

MGM, containing approximately 70% moisture, was obtained from Penford (Cedar Rapids, IA). DSF was obtained from Archer Daniels Midland Co. (Decatur, IL). Samples were stored at −20°C until needed. AEMGM was prepared using the extraction procedure of Harris et al (1987). AEMGM and DSF were blended (20:80, 30:70, and 50:50, respectively; w/w, db), and all treatments were taken from these initial blends.

Response-Surface Analysis

Optimal blend moisture, pH, and extruder exit temperature were determined using a three-variable response-surface statistical design and analysis (Myers 1976) (Table I). Fifteen treatments for each AEMGM-DSF blend were produced that differed in pH, moisture content, or extruder exit temperature (Tables II and III).

The extrudates were developed as follows: three levels of AEMGM-DSF blends were used as individual treatments in the ratios of 20:80, 30:70, and 50:50 MGM and DSF, respectively; test variables included moisture content, pH, and extruder exit temperature. Each treatment was subjected to the same experimental design (Tables I and II).

Extrusion and Preliminary Evaluation

Extrusion was performed as described by Koeppe et al (1987) without the die in the manner of Feldbrugge et al (1978) using a C. W. Brabender extruder (model 2802) equipped with a 19-mm diameter single screw having a 20:1 length-to-diameter ratio. Screw speed was 145 rpm. Extruder exit temperatures are reported in Table II.

Extrudate Evaluation

AEMGM-DSF extrudates were evaluated for response-surface data on the basis of color, bulk density (BD) (Harris et al 1988), water-holding capacity (WHC) (Harris et al 1988), structural integrity, retortability, and trypsin inhibitor (TI) activity. Color was evaluated using a HunterLab color difference meter and methods as described by Harris et al (1987). Structural integrity was determined by placing 5 g of extrudate into 50 ml of boiling distilled water for 5 min and observing disintegration. Retortability was determined by placing 5 g of each extrudate in a

TABLE I
Three-Variable Statistical Design for Determining Optimum Extrusion Conditions for Acetone-Extracted Maize Gluten Meal and Defatted Soy Flour Extrudates[a]

X-1	*X*-2	*X*-3
−1	−1	−1
1	−1	−1
−1	1	−1
1	1	−1
−1	−1	1
1	1	1
1	1	1
−1.682	0	0
1.682	0	0
0	−1.682	0
0	1.682	0
0	0	−1.682
0	0	1.682
0	0	0[b]

[a]From Myers 1976.
[b]Sequence repeated six times.

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standard glass microbiological petri dish and autoclaving at 121° C and 18 psi pressure for 15 min. Structural integrity and retortability were graded using a subjective scale, 1–5: 5 = no fragmentation; 1 = total disintegration of the extrudate. TI activity was determined using the method of Kakade et al (1974). One TI unit (TIU) was defined as the amount of inhibitor that caused 10% inhibition of trypsin in 10 min under the described assay conditions. Analyses were performed in triplicate. Bovin trypsin (lot 24F-8011) and *N*-α-benzoyl-DL-arginine-*p*-nitroanalide (BAPNA) were obtained from Sigma Chemical Company (St. Louis, MO).

Final Product Evaluation

AEMGM-DSF extrudates produced using the optimum conditions were evaluated as described. In addition, the proximate analysis (AOAC 1980, AACC 1983) of the extruded product was determined.

RESULTS AND DISCUSSION

Color

Higher *L* (+ lightness, − darkness) values were observed from AEMGM-DSF extrudates containing higher concentrations of DSF (70 and 80%) as opposed to extrudates processed from an equal blend of AEMGM and DSF (50%) (Table IV). Extrudates containing AEMGM (50%) exhibited greater *b* (+ yellow, − blue) values, probably because of the additional concentration of residual carotenoid pigments.

TABLE II
Response-Surface Values for pH, Moisture, and Extruder Exit Temperature of Acetone-Extracted Maize Gluten Meal and Defatted Soy Flour Extrudates[a]

Variable	pH	Moisture (%)	Extruder Exit Temperature (° C)
Range			
Low	3	20	140
Medium	6	30	160
High	9	40	180
−1.682	2.64	13.18	126.26
−1	4.00	20.00	140.00
0	6.00	30.00	160.00
1	8.00	40.00	180.00
1.682	9.36	46.82	193.82

[a]Values correspond to *X*-1, *X*-2, and *X*-3 in Table I. From Myers 1976.

TABLE III
Response-Surface Variables and Sample Codes for Acetone-Extracted Maize Gluten Meal and Defatted Soy Flour Blends

	Variables		
Sample	Slurry pH	Initial Moisture (%)	Extruder Exit Temperature (° C)
1	2.64	30	160
2	4.00	20	140
3	4.00	20	180
4	4.00	40	140
5	4.00	40	180
6	6.00	13	160
7	6.00	30	126
8	6.00	30	160
9	6.00	30	160
10	6.00	30	160
11	6.00	30	160
12	6.00	30	160
13	6.00	30	160
14	6.00	30	194
15	6.00	47	160
16	8.00	20	140
17	8.00	20	180
18	8.00	40	140
19	8.00	40	180
20	9.36	30	160

Structural Integrity

AEMGM-DSF extrudates processed with high DSF content (80%) were more friable than other AEMGM-DSF extrudates (Table V). These extrudates fragmented into small particles upon emerging from the extruder, or upon drying, and disintegrated when immersed in boiling water. Generally, acidic AEMGM-DSF extrudates were more fibrous and pliable than neutral or alkaline AEMGM-DSF extrudates (Fig. 1). These extrudates retained their rigid, fibrous texture when immersed in boiling water. These results are similar to those reported by Smith (1975). AEMGM-DSF extrudates processed at higher extruder exit temperatures (180 or 194° C) retained their structural integrity when exposed to boiling water and exhibited increased cohesiveness (Fig. 1). In contrast, the AEMGM-DSF extrudates processed at lower extruder exit temperatures (126° C) resembled raw dough and fragmented easily upon emerging from the extruder. These extrudates also disintegrated in an aqueous environment (Fig. 1). Smith (1975) observed similar results. The AEMGM-DSF extrudates processed from high-moisture blends (40–47%) also resembled raw cookie dough. Upon drying, these extrudates became tough, but in comparison to the extrudates processed from low moisture blends (13-20%) maintained better structural integrity (Fig. 1). AEMGM-DSF extrudates processed from low moisture blends were brittle and disintegrated easily in boiling water (Fig. 1).

Functional Analysis

The microstructure of these AEMGM-DSF extrudates and their functionality determined by BD, WHC, and shear strength were reported by Harris et al (1988). The AEMGM-DSF extrudates produced in this study exhibited functional characteristics similar to those reported by Cummings et al (1972), Smith (1975), and Bhattacharya et al (1986). All AEMGM-DSF extrudates were retorted to determine their thermal stability. All extrudates remained intact with no evidence of disintegration. Additional research (data not shown) demonstrated that the 50:50 AEMGM-

TABLE IV
HunterLab Color Analysis of AEMGM-DSF[a] Extrudates[b]

Sample	*L*[c]	*a*[c]	*b*[c]
20:80 MGM-DSF[d]	75.6 ± 8.1	3.9 ± 1.5	2.6 ± 1.4
30:70 MGM-DSF[e]	75.1 ± 8.4	3.7 ± 1.7	2.0 ± 2.5
50:50 MGM-DSF[f]	68.6 ± 9.2	3.3 ± 1.8	9.6 ± 7.8

[a]AEMGM-DSF = Acetone-extracted maize gluten meal and defatted soy flour.
[b]Means of triplicate analysis ± standard deviation.
[c]*L* = + Lightness, − darkness; *a* = + red, − green; *b* = + yellow, − blue.
[d]Mean of all samples extruded with 20% AEMGM-80% DSF blend.
[e]Mean of all samples extruded with 30% AEMGM-70% DSF blend.
[f]Mean of all samples extruded with 50% AEMGM-50% DSF blend.

TABLE V
Evaluation of 20:80, 30:70, and 50:50 Blend Extrudates[a]

Blend[a]	Bulk Density (g/L)	Structural Integrity	Water-Holding Capacity	Trypsin Inhibitor Units	Retort[b]
20:80 AEMGM-DSF					
Mean	695	3.3	5.7	0.8	5
SD	100	1.5	1.2	0.3	0
30:70 AEMGM-DSF					
Mean	625	3.9	5.5	0.9	5
SD	140	1.1	1.1	0.4	0
50:50 AEMGM-DSF					
Mean	602	4.5	4.6	0.9	5
SD	127	0.8	1.4	0.3	0

[a]20:80 = 20% Acetone-extracted maize gluten meal (AEMGM), 80% defatted soy flour (DSF); 30:70 = 30% AEMGM, 70% DSF; and 50:50 = 50% AEMGM, 70% DSF.
[b]Retortability 121°C, 18 psi, 15 min.

TABLE VI
Percentage Proximate Analysis
of Optimum 50:50 AEMGM-DSF Extrudate[a]

50:50 AEMGM-DSF	Moisture	Ash	Protein[b] ($N \times 6.25$)	Fat[b]	Fiber[b]	CHO[c]
Wet-basis	1.6	4.12	50.7	0.16	1.60	41.82
Dry-basis	···	4.71	58.0	0.18	1.83	43.42

[a]50% Acetone-extracted maize gluten meal (AEMGM) - 50% defatted soy flour (DSF). Extrusion conditions: pH 6.5, moisture 32%, extruder exit temperature 153°C, screw speed 145 rpm.
[b]Crude.
[c]CHO = Carbohydrate.

TABLE VII
Optimum Variable Values for 50:50 AEMGM-DSF[a] Extrudates
Based on Response-Surface Data

Response	pH	Moisture (%)	Temperature (°C)
Structural integrity	6.4	37	153
Water-holding capacity	6.1	28	155
Trypsin inhibitor units	5.7	33	147
Lab	6.8	30	157
Mean	6.25	32	153

[a]50% Acetone-extracted maize gluten meal and 50% defatted soy flour.

DSF extrudates maintained their structural integrities upon retorting after being immersed in a gravy type sauce.

Chemical Analysis

Trypsin inhibition assays were performed on each AEMGM-DSF extrudate to detect differences in the destruction rate of TIs. As extruder exit temperature increased, the amount of TI activity observed in the resultant AEMGM-DSF extrudates decreased (Fig. 2). As expected, the greatest amount of TI destruction was observed in the 20:80 AEMGM-DSF extrudates processed at an extruder exit temperature of 194°C. AEMGM-DSF extrudates processed under these conditions exhibited a final TI activity of 0.1 TIU. The TI activity of the AEMGM-DSF extrudates was not affected by pH or moisture of the blend (Fig. 2). Similar decreases in TI activity as a result of increasing extruder exit temperatures were reported by Mustakas et al (1970) and Koeppe et al (1987) who worked with extruded full-fat soy flour and MGM-amaranth blends, respectively.

Optimum Product

Sternberg et al (1980) and Neumann et al (1984) reported a 20–30% improvement in calculated protein efficiency ratio and protein efficiency ratio values for maize-soy blends in comparison with maize alone. From a nutritional viewpoint, an optimum w/w ratio mixture of soy-maize is 80:20 (Sternberg et al 1980). At this level, lysine, methionine, and cysteine contents are within 10% of the National Research Council (1980) recommended amino acid profile. Although an 80:20 DSF-MGM blend is nutritionally superior, the 50:50 AEMGM-DSF blend ratio exhibited the best functionality. Nutritional quality could be improved by adding fortifying ingredients.

The proximate analyses of the optimum AEMGM-DSF extrudates obtained in this study are given in Table VI. A relatively high quantity of protein with a low fat content was observed in these extrudates. These results are in agreement with Neumann et al (1984) and Koeppe et al (1987).

Based upon response-surface statistical data, optimum textural properties of these AEMGM-DSF extrudates occurred using a 50:50 AEMGM-DSF blend ratio, 32% moisture, pH of 6.25, and extruder exit temperature of 153°C. These results approximate those used by Neumann et al (1984) (moisture 30–35%, pH 7, extrusion temperature 145°C). The optimum extrusion conditions are based on a product characterized by low WHC and TI activities (3.8×10^{-2} TIU/g of protein) as well as on color and structural integrity scores (Table VII). A low WHC (2.5) (Harris et al 1988) is

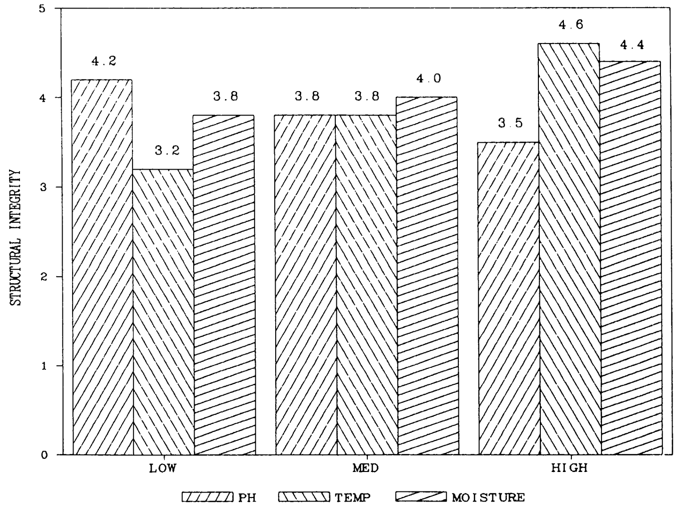

Fig. 1. Structural integrity of extrudates of acetone-extracted maize gluten corn meal and defatted soy flour. **Low** = pH 2.64 and 4, 128 and 140°C, and 13 and 20% moisture. **Medium** = pH 6, 160°C, and 30% moisture. **High** = pH 8 and 9.36, 180 and 194°C, and 40 and 47% moisture.

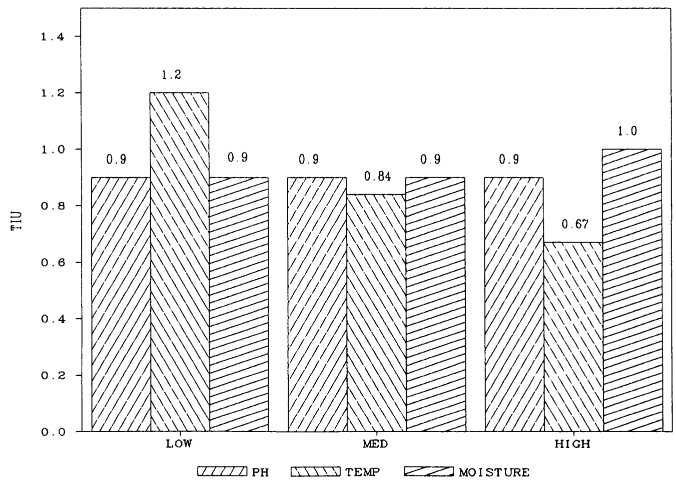

Fig. 2. Trypsin inhibitor activity of extrudates of acetone-extracted maize gluten corn meal and defatted soy flour. **Low** = pH 2.64 and 4, 128 and 140°C, and 13 and 20% moisture. **Medium** = pH 6, 160°C, and 30% moisture. **High** = pH 8 and 9.36, 180 and 194°C, and 40 and 47% moisture.

desirable in an extrusion product that is subsequently thermally processed in an aqueous medium.

Conclusion

Response-surface methodology was used to optimize ingredient variables (initial pH and moisture levels) and extruder exit temperatures. Optimization of the structural integrity, WHC, color, and inactivation of TIs of these extrudates occurred with a blend moisture of 32%, pH of 6.25, and extruder exit temperature of 153°C.

Extrudates processed with equal amounts of AEMGM and DSF demonstrated greater structural integrity values than those processed from blends containing greater amounts of DSF (70 and 80%). AEMGM-DSF extrudates processed at high extruder exit temperatures (180 and 194°C) or with high blend moisture levels (40 and 47%), as well as acidic blends, demonstrated greater cohesiveness and structural integrity. As extruder exit temperature increased, TI activity in the resultant AEMGM-DSF extrudates decreased.

LITERATURE CITED

AMERICAN ASSOCIATION OF CEREAL CHEMISTS. 1983. Approved Methods of the AACC. Methods 08-01, 44-19, and 46-10, approved April 1961. The Association: St. Paul, MN.

ASSOCIATION OF OFFICIAL ANALYTICAL CHEMISTS. 1980. Official Methods of Analysis, 13th ed. The Association: Washington, DC.
BHATTACHARYA, M., HANNA, M. A., and KAUFMAN, R. E. 1986. Textural properties of extruded plant protein blends. J. Food Sci. 51:988.
CONWAY, H. F., and ANDERSON, P. A. 1973. Protein-fortified extruded food products. Cereal Sci. Today 18:94.
CUMMINGS, D. B., STANLEY, J. M., and DeMAN, J. M. 1972. Textural properties and ultrastructure of an extruded soybean product. J. Inst. Can. Technol. Aliment. 5:124.
EL-DASH, A. A. 1981. Application and control of thermoplastic extrusion of cereals for food and industrial uses. Pages 165-208 in: Cereals: A Renewable Resource. Y. Pomeranz and Lars Munck, eds. Am. Assoc. Cereal Chem.: St. Paul, MN.
FELDBERG, C. 1965. Corn gluten, a new ingredient for bakery products. Cereal Sci. Today 10(1):18.
FELDBRUGGE, A., RANKOWITZ, M. W., and TRAVERS, C. K. 1978. Vegetable protein texturization by low pressure extrusion. AIChE Symp. Ser. 73(163):134.
HARRIS, P. L., CUPPETT, S. L., WALKER, C. E., and RUPNOW, J. H. 1987. Note: Lipid and color evaluations of solvent-extracted maize gluten meal. Cereal Chem. 64:283.
HARRIS, P. L., CUPPETT, S. L., and LEE, K. W. 1988. A scanning electron microscope study of maize gluten meal and soy coextrudates. Cereal Chem. 65:228-232.
KAKADE, M. L., RACKIS, J. J., McGHEE, J. E., and PUSKI, G. 1974. Determination of trypsin inhibitor activity of soy products: A collaborative analysis of an improved procedure. Cereal Chem. 51:376-382.
KINSELLA, J. E. 1978. Textured proteins; fabrication, flavoring and nutrition. CRC Crit. Rev. Food Sci. Nutr. 11:137.
KOEPPE, S. J., HARRIS, P. L., HANNA, M. A., RUPNOW, J. H., WALKER, C. E., and CUPPETT, S. L. 1987. Physical properties and some nutritional characteristics of an extrusion product with defatted amaranth seeds and defatted maize gluten meal (80:20 ratio). Cereal Chem. 64:332.
MERCIER, C., and FEILLET, P. 1975. Modification of carbohydrate components by extrusion cooking of cereal products. Cereal Chem. 52:283.
MUSTAKAS, G. C., ALBRECHT, W. J., BOOKWALTER, J. E., and McGHEE, W. F. 1970. Extruder-process to improve nutritional quality, flavor and keeping quality of full-fat soy flour. Food Technol. 24:1290.
MYERS, R. H. 1976. Response Surface Methodology. Virginia Polytechnic Institute and State University: Blacksburg, VA.
NATIONAL RESEARCH COUNCIL. 1980. Protein and amino acids. In: Recommended Dietary Allowance. National Academy of Sciences: Washington, DC.
NEUMANN, P. L., JASBERG, B. K., and WALL, J. S. 1984. Uniquely textured products obtained by coextrusion of corn gluten meal and soy flour. Cereal Chem. 61:439.
PERI, C., BARBIERI, R., and CASERAGI, E. M. 1983. Physical, chemical and nutritional quality of extruded corn germ flour and milk protein blends. J. Food Technol. 24:1290.
REINERS, R. A., HUMMEL, J. B., PRESSICK, J. C., and MORGAN, R. E. 1973. Composition of feed products from wet milling of corn. Cereal Sci. Today 18:372.
SHRODER, J. D., and HEIMAN, V. 1979. Feed products from corn processing. Page 223 in: Preparation of Edible Proteins from By-Products of Corn Wet-Milling. CRC Press: Boca Raton, FL.
SMITH, O. B. 1975. Extrusion and forming; creating new foods. Food Eng. 7:48.
STERNBERG, M., PHILLIPS, R. D., and DALEY, L. H. 1980. Maize Protein Concentrate. Page 275 in: Cereal for Food and Beverages. Academic Press: New York.

[Received August 12, 1987. Revision received January 13, 1988. Accepted January 17, 1988.]