Twin-screw extrusion puffing of non-fat dry milk powder

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Abstract

The use of twin-screw extrusion to produce puffs and crisps for cereals and snacks is widely utilized in the food industry. Soy protein is the leading protein used in this category. Extrusion puffing of caseinates and whey protein concentrates has been researched extensively, but much less has been published using non-fat dry milk (NDM). This study evaluated formula and processing conditions for creating expanded puffs with significant amounts of NDM. Nonfat dry milk (NDM) and a modified cornstarch were extruded at three NDM concentrations (45, 65, 85%), three lactic acid levels (0, 33 and 50% of the added fluid), and two fluid addition rates (6.5, 7.3 kg/h) to alter extrudate characteristics. Process responses (die temperature, die pressure, motor torque and specific mechanical energy) and product responses (color, solubility, expansion ratio, bulk density and peak viscosity) were analyzed and response surface plots were generated of the results. It was demonstrated that NDM provided structural integrity to the extrudates, and formula conditions could be manipulated to alter product characteristics such as density and color to improve its fitness for use in extruded products. The response surfaces generated allow for the selection of formula conditions to produce desired product attributes.

Background Information

The use of twin-screw extrusion to produce puffed products for cereals and snacks is widely utilized in the food industry. Puffs and crisps are generally starch-based, but those with elevated protein contents are produced as nutritious inclusions in bars and cereals, or for forming into snacks. Soy protein is by far the leading protein used in this category. Extrusion puffing of dairy proteins with or without starch has been researched. There is currently a milk protein concentrate puff being marketed by Kerry Ingredients in Ireland, and soy/whey protein isolate puffs being marketed as nutritional supplement snacks (Chef Jay's Protein Puffs and Costa's Foods Muscle Puffs). The majority of research for using dairy proteins in extruded puffs has concentrated on whey protein concentrates (80% protein) and isolates, caseinates, and milk protein concentrates & isolates. The use of non-fat dry milk (NDM) has not been studied as extensively because it has less protein and more lactose than 80% concentrates and isolates. It has also been traditionally difficult to create expanded puffs when it is used at high levels. NDM currently has the advantage of an abundant and inexpensive supply, has the full amount of calcium found in milk, and has a cleaner flavor than whey protein concentrates.

A puffed product is created on a twin-screw extruder by the application of heat and shear to a dough. The water in the dough vaporizes after passing through the die of the extruder where a reduction in pressure back to atmospheric is reached, and a product with low bulk density can be achieved. In order for a puffed, crisp product to be obtained, water in the formula needs to vaporize, and the formula must have the ability

to trap gas and expand. It also must hold this expanded structure through drying (and not shrink excessively), and needs to allow the release of moisture during drying to create a crisp product. To be useful in cereal applications, the finished puff must also be resistant to hydration by milk so the product will have sufficient bowl life. In a high quality extruded protein product, protein will form a textured matrix, starch will be evenly distributed as an inclusion, and steam generated voids will be present (Stanley, 1989). When milk proteins are added to starch ingredients for puffing, the density of the product tends to increase due to the protein affecting the expansion ratio (Onwulata, 2006). Protein also introduces amino acids which can participate in Maillard browning and cause undesirable brown colors and loss of nutritional value. The Maillard reaction also contributes to cross-linking of the proteins, which can be desirable for texturization (Prudencio-Ferriera and Areas, 1993; Stanley). Moisture content during extrusion also contributes to the amount of browning and nutritional loss while extruding milk proteins with lower moisture contents causing the most browning (Singh et al., 2007). The main characteristics of a protein affecting its usefulness for extrusion are:

- Solubility
- Protein-protein interactions
- Protein-starch interactions

The extrusion process introduces heat and shear to protein ingredients and causes them to denature during the process (Onwulata, 2006). This can lead to exposure of amino acids to reducing sugars, exposure of hydrophobic amino acid residues causing a reduction in solubility, and facilitate intermolecular bond formation (Camire, 1991). Factors which can increase bonding between proteins have been found to improve the functional characteristics of extruded products (Liu and Hsieh, 2008, 2007; Prudencio-Ferriera and Areas, 1993). Techniques to increase protein bonding include adding reducing agents, modifying the pH, and pre-denaturing proteins by heat and shear (Onwulata, 2003; Onwulata et al., 2006; Prudencio-Ferriera and Areas, 1993; Tunick and Onwulata, 2006). The pH milk proteins are extruded at will affect their solubility and the amount of protein interaction during extrusion. Onwulata et.al. (Onwulata et al., 2006) investigated the effect of pH on extrusion of whey protein isolate (WPI) pastes. They found that extrusion of WPI at alkaline conditions decreased solubility and resulted in textured protein. One of the advantages of soy and whey proteins is that the heat in the extruder can aggregate the proteins, reducing their solubility. Caseins are highly resistant to heat denaturation due to the disordered structure of the molecule (J. Szpendowski et al., 1994). If the proteins are too soluble and retain large amounts of moisture, problems can be encountered during expansion (water cannot flash-off as steam) and drying (time to dry is extended and a crisp texture might not be achieved). The caseins in NDM will heat aggregate as the pH is reduced from the native milk pH, and at their isoelectric point they will aggregate at relatively low temperatures. This can be used as a technique to induce interactions in the extruder. A recently published US patent application describes the production of caseinate puffs from dry acid casein with

the addition of lime powder to create a pH of extrusion of 5.2 – 5.6 to introduce protein interactions (Bisson, 2008). The technique of pH or enzyme modification of milk proteins prior to extrusion has been used in the production of meat protein analogs from high moisture milk protein solutions (Manski et al., 2008). The effect of reducing the calcium in a casein preparation on the ability to induce interactions and form texture in an extruder was demonstrated by Szpendowski et al. (J. Szpendowski et al., 1994). They found that it was difficult to structure caseins at high levels of calcium, but better expansion was achieved with sodium caseinate and low-calcium paracaseinate.

The moisture content of an extrudate has a significant effect on Maillard browning. When milk proteins were extruded with various sugars, puffs with 16% moisture were much lighter than puffs extruded at approximately 14% moisture (Singh et al., 2007). The objective of this research was to evaluate the effects of pH and moisture on NDM in order to provide industry with guidance that will allow for its incorporation into extruded puffs at high concentrations with minimal browning and a desirable texture. Reducing the pH of the NDM could achieve solubilization of calcium, resulting in less heat stability of the protein. Reducing the pH should also benefit the color of the extruded product by reducing Maillard browning

Objectives

1. Determine the effect of different NDM levels on puff properties
2. Determine the effect of different extrusion pH's on puff structure
3. Determine the effect of different extrusion pH's on browning
4. Determine the effect of moisture level on puff properties

Materials and Methods

Materials

Grade A Low Heat NDM (DairyAmerica, Inc., Fresno, CA), Clearjel S, a modified waxy maize (National Starch, Bridgewater, NJ), Elaine 100, potato starch (National Starch, Bridgewater, NJ) and 88-92% liquid lactic acid (Food Chemicals Codex grade) item A159-500 (Fisher Scientific, Inc., Fairlawn, NJ) were used in all extrusion trials.

Pretrial Experiments to Determine Extrusion Conditions

Five pretrial extrusion runs took place in order to determine the extruder processing conditions (barrel temperatures, screw design, screw speed), along with the experimental parameters (starch type, fluid addition rates, and acid levels in the injected fluid). The processing conditions were chosen to maximize visual appeal and to obtain puffs with a potentially acceptable expansion ratio and bulk density. Selections of the initial barrel temperature profile and screw design were based on prior experience.

The intermediate NDM level (65%) was used in all the trials, and two starches (Clearjel S and Elaine 100) were evaluated to decide which starch produced a more desirable extrudate. Clearjel S was ultimately chosen for the experimental trials. The screw was designed to promote ingredient hydration and to obtain optimum shear to promote interactions amongst components, yet not result in a jagged appearance due to the application of too much shear. The best product was obtained setting the first two barrel sections at 40°C, and the last two sections at 80°C. The screw speed selected was 350 rpm. Fluid addition rates that could be run on our extruder without reaching maximum torque, and also not resulting in excessively dense product which can occur at high fluid addition rates, were evaluated. The acid levels used at the beginning of the pretrial were based on evaluating NDM/starch blends with acid in a Rapid Visco Analyzer Model 4 (Newport Scientific, Australia) where lactic acid was added and the pH prior to and after RVA analysis was determined, so that we could estimate the pH change with heating. We chose an acid level that resulted in a pH of 5.2. Unfortunately, this laboratory method was not effective in determining the amount of acid to add during extrusion because the resulting samples tasted too acidic and had very little expansion. Ultimately, the acid levels chosen for the experimental trials were based on whether or not a desirable extrudate was obtained. The best conditions determined during these pretrials were used in the experimental design.

Experimental Design and Preparation of Raw Materials

The experimental design consisted of three NDM levels (45, 65, 85%) (dry blend brought to 100% with Clearjel S, a modified corn starch derived from waxy maize), two water addition rates (6.5, 7.3 kg/h) and three lactic acid levels (0, 33, 50% of the added water), totaling 18 treatments. Treatments are defined according to NDM concentration (45NDM, 65NDM, 85NDM), water addition level (low moisture (LM), high moisture (HM)), and lactic acid level (low acid (LA), medium acid (MA), high acid (HA)). One NDM formula was run on each processing day, and each treatment was replicated. The order in which samples were collected was determined by fluid addition rate and acid level; HM conditions preceded LM conditions, and within each water addition level, LA samples were collected first, followed by MA and then HA samples. Due to extrusion operation constraints, the order in which samples were collected could not be randomized.

The NDM and starch were mixed in 50 lb batches with a ribbon blender model IMS-1 (Bepex International LLC, Minneapolis, MN) in forward and reverse directions for 2 minutes each. The mix was processed on a Buhler 44mm co-rotating twin-screw extruder DNDL 44 (Bühler AG, Uzwil, Switzerland) with a L/D ratio of 28, which consisted of 4 barrel zones set at 40°C, 40°C, 80°C, 80°C from inlet to die. The temperatures of the barrels were maintained by a heat transfer control system model H47212DT (Mokon, Buffalo, NY). The same screw configuration (Figure 1) was used throughout the experimental design and was selected to provide medium shear. Dry feed was conveyed at 40 kg/h into the extruder with a K-Tron Soder K-ML-KT20 loss-in-weight feeder (K-

Tron Ltd., Niederlenz, Switzerland). The screw speed was maintained at 350 rpm. The water (and water with lactic acid added) was pumped by metering pumps (type N-P33, Bran Luebbe, Inc., Falo Grove, IL) and computer controlled by measuring the flow rate with a mass flow meter (Micro Motion, Inc., Boulder, CO). The extruded product was collected in ropes after the die temperature, die pressure and extruder shaft torque reached steady state, cut into 5 cm lengths with scissors, and then dried for 10 minutes at 100°C on trays with a screened bottom on a fluidized bed dryer (Model: OTW 05TRR2,Bühler AG, Braunschweig, Germany). Process responses (die temperature, die pressure, motor torque and specific mechanical energy) were collected at the beginning and end of sample collection for each treatment. Extrudates were placed 1 L glass jars, and stored at room temperature until analyzed.

Chemical Analysis

Total protein (nitrogen x 6.38) of the extrudates and the NDM and starch mixtures was determined using a TruSpec® N (Leco Corporation, St. Joseph, MI) based on the Dumas method of combustion. Moisture content was determined by vacuum oven and ash was determined by muffle furnace following Bulletin 916, Standards for Grades of Dry Milk (American Dairy Products Institute 2002). The samples were prepared for moisture analysis by grinding with a mortar and pestle and then passing through a 60 mesh sieve 60BB8H (Seedburo® Equipment Company, Chicago, IL). Fat was determined following method 15.086 from Standard Methods for the Examination of Dairy Products (Wehr and Frank 2004). Carbohydrate content was calculated by difference from the other components.

The pH was measured on an Acorn® pH 6 meter (Oakton Instruments, Vernon Hills, IL) with an Orion 8172BNWP Ross Sure-Flow pH electrode (Thermo Scientific, Inc., Waltham, MA). The pH was measured after combining 10 mL reverse osmosis (RO) treated water with 15 g crushed extrudate.

Physical Analysis

The bulk density of the dried extrudates was determined by filling a 100 cubic inch container with dried extrudates and then measuring the mass of that volume.

The expansion ratio was determined by dividing the diameter of the extrudates by the die orifice diameter (3.0 mm). The diameters of 10 randomly chosen samples within each replicate were measured with an electronic caliper.

The color of uncrushed extrudates was determined using the method described by Wu et al. (2007). Samples were placed in a glass Petri dish, and the color was analyzed with a HunterLab D25 A Optical Sensor Colorimeter (Hunter Associates Laboratory, Inc., Reston, VA). A total of 5 measurements were recorded, with rotating the Petri dish 90° after each reading.

Extent of browning during extrusion was determined by measuring absorbance at 420 nm by the method described by Davies et al. (1998) with a few modifications. Samples were ground and sifted through a 60 mesh sieve. The sample (0.375 g) was dissolved in 12.5 mL RO water, vortexed, and then digested with 0.5 mL of pancreatin solution (50 mg/mL dissolved in RO water), (pancreatin 3X 102558, MP Biomedicals, LLC, Solon, OH) while shaking at 45°C for 2 hours. Samples were vortexed, and 1 mL was transferred into each of three 2 mL microcentrifuge tubes. 40 µL trichloroacetic acid (Sigma-Aldrich®, St. Louis, MO) prepared at 50% weight per volume of RO water was added to each sample, the tubes were centrifuged at 15,850g with a Beckman Microfuge E (Beckman Instruments, Inc., Palo Alto, CA) for 5 minutes, and then filtered through a 32 mm Whatman no. 1 filter paper (Whatman, Inc., Piscataway, NJ) into cuvettes. A blank was prepared as above but without extrudate. Browning was determined by measuring the absorbance at 420 nm with a Beckman DU® 650 Spectrophotometer (Beckman Instruments, Inc., Fullerton, CA), and the results were converted to absorbance per gram of solids.

Water absorption index (WAI) and water solubility index (WSI) were determined by modifying the methods reported by Onwulata et al. (1998). Samples were ground with a commercial spice grinder and sifted through a 212 micron sieve (NC0003425, Thermo Fisher Scientific, Inc., Waltham, MA). 1.010 g (±0.005 g) was placed in a 50 mL centrifuge tube and 10 mL RO water was added. After standing 15 minutes (shaking every 5 minutes), the samples were centrifuged for 15 minutes at 1000 g in a C5-6R Centrifuge (Beckman Instruments, Inc., Palo Alto, CA). The supernatant was decanted into a tared aluminum pan, and the weight of the gelled material remaining in the centrifuge tube was recorded. WAI was calculated as the weight of the gel obtained after the removal of the supernatant per unit weight of original dry solids. To determine the WSI, the liquid from the supernatant was evaporated over boiling water, and then the pans were dried at 100°C in a gravimetric oven (Imperial II Radiant Heat Oven,Lab-Line Instruments, Inc., Melrose Park, IL) for 3 hours as recommended in the Standards for the Examination of Dairy Products for the determination of moisture in milk (Case et al. 1985). The pans were weighed after cooling, and the WSI was determined as the weight of the dried supernatant expressed as a percentage of the original weight of the sample.

To determine the peak viscosity, which can provide an indication of the degree of starch gelatinization that occurred in the extruder, a Rapid Visco Analyzer Model 4 (Newport Scientific, Australia) equipped with Thermocline for Windows software version 2.3 was used. Extrudates were collected after exiting the die and prior to drying, and were frozen until analyzed. Extrudates were ground with a spice mill until a fine powder was obtained, and then 14.0 g of crushed extrudates were standardized to 14% moisture using RO water. The pasting properties were determined by the following conditions: Starting temperature was 25°C, and paddle rpm was 960 for 10 seconds. Speed was reduced to 160 rpm for the remainder of the test. Beginning at 2 minutes, the

temperature was ramped to 95°C over 5 minutes. The sample was held at 95°C for 3 minutes, then cooled to 25°C over 5 minutes.

Cross-sectional images of the extrudates were taken with a Nikon SMZ1500 Zoom Stereomicroscope (Nikon Instruments, Inc., Melville, NY) equipped with an 8 mega pixel Olympus E330 digital single lens reflex camera (Olympus America, Inc., New Hyde Park, NY). Extrudates were chosen at random from each treatment, and a razor blade was used to cut sections.

Statistical Analysis

Statistical analysis was conducted using R version 2.12.0 (R Development Core Team, 2008). Process and product responses were analyzed to assess the effects of the independent variables, and response surfaces were generated. A mixed model was used to obtain estimates for variability between product and sample replicates for select responses. After averaging all subsamples, a standard analysis of variance (ANOVA) was used with a pairwise comparison, corrected using Tukey honestly significant difference.

To estimate the conditional correlation between responses after accounting for NDM concentration, fluid addition rate and acid level, Spearman's correlation was calculated using the mean-adjusted responses for each combination of independent variables, after first averaging over all subsamples.

Results and Discussion

Obj. 1-4 Chemical Analysis

As the percentage of NDM increased, the protein, fat and ash content of the NDM/starch dry blend increased as shown in Table 1. The pH of dried extruded product decreased with an increase in the percent acid in the fluid injected (Table 2). Also, as the amount of NDM increased, there was a concurrent increase in pH, presumably due to buffering from the milk solids. One treatment, 65% NDM, LM and HA, was produced but was not evaluated due to incorrect processing conditions in the initial trial. Two additional trials were carried out in an effort to collect this treatment; however, the product produced in two attempts did not agree with past product trends. Specifically, the puffs did not brown, even with increases in temperature and run time. All ingredients were from the same lot. Differences in the time of year caused the incoming water temperature to be considerably colder at the time of injection, and we had no ability to control the temperature at this point. This may have resulted in the difference observed. Future studies could be carried out to determine the effect of inlet water

temperature on product temperature in different extruder zones, and its effect on puff color and other product characteristics.

Obj 1, 2 and 4 Process Responses

There were numerous differences and interaction effects due to the treatments as illustrated in Table 3, and the average values are shown in Table 4. NDM concentration, fluid addition rate and acid level all affected the motor torque, SME, die pressure and die temperature.

SME can provide an indication of how much energy is transmitted to the extruded material (Ortiz *et al.* 2010). The SME in our study was at a maximum at intermediate NDM concentrations, and generally higher when lower fluid addition rates were used as shown in the response surface in Figure 2. Others have observed similar effects of water addition on SME (Akdogan 1996; Köksel *et al.* 2004). Campanella *et al.* (2002) attributed this effect to increased resistance to screw rotation and flow through the die.

Die temperature can affect the viscosity, with a high die temperature decreasing the viscosity of the material exiting the extruder (Akdogan 1996). The viscosity can also be related to the die pressure, and can often be correlated to finished product characteristics (Colonna *et al.* 1989). Reduced expansion can occur when materials with high viscosities are extruded under low die pressure, or conversely, when materials are extruded with low viscosities and a high die pressure. High viscosity materials extruded at high die pressures favor expanded products. In our study, acid level had variable effects on die temperature (Figure 3), while a decrease in NDM concentration and fluid addition rate increased the die temperature, so likely decreasing the viscosity of the extruded material at the die. The low die temperature, combined with the high die pressure most likely contributed to the structurally sound, expanded extrudates for the treatment that was produced at 85% NDM, 0% acid level in the injected fluid and extruded at the high fluid addition rate (Figure 4).

Monitoring the motor torque enables the operator to determine the resistance of the material to the process and conditions, similar to the SME (Ghebre-Sellassie and Martin 2003). Higher motor torque was observed at intermediate NDM contents and low fluid addition rates in our study. This is in contrast to the study by Faller *et al.* (1999) who saw that all levels of added soy protein decreased motor torque. They attributed this to the protein competing for water with the starch, and delaying starch gelatinization until just before the die, so less gelatinization occurred in the extruder barrel.

Obj 1 – 4 Physical Analysis

NDM, fluid addition rate and acid level in injected fluid had an effect on the physical analysis measurements. NDM concentration affected both the bulk density and the expansion ratio, while acid level and fluid addition rate independently did not (Table

5). Figure 5 shows the effect of NDM concentration and acid level on the bulk density at low and high fluid injection rates. As NDM and acid levels increased, the bulk density increased (meaning the extrudate was denser and not as expanded). Our findings are consistent with research by Limón-Valenzuela *et al.* (2010) who extruded milk protein concentrate (MPC) (0-10%) with a blend of cornstarch and maize, and observed an increase in bulk density as MPC concentration increased. Conversely, Onwulata *et al.* (2001a) observed no significant effect on the bulk density as milk proteins including casein, WPC and WPI were added to corn meal flour at 250 g/kg. It is likely that the product formula (type and amount of protein, starch, fiber, etc.), as well as the processing conditions, impacted the bulk density and differences observed when compared to other studies.

An increase in the NDM concentration was associated with a decrease in the expansion ratio. Chaiykul *et al.* (2009) observed a similar effect as protein content was varied in a high protein glutinous rice-based snack. They attributed the effect to a decrease in the extensibility of the starch polymer at the die. The decreased expansion observed in our study could also be attributed to the lactose in the NDM, as hypothesized in a study with whey by Onwulata *et al.* (2001b). Bulk density has been correlated with the expansion of extrudates, with a high bulk density associated with a less expanded product (Colonna *et al.* 1989). As expected in this study, the bulk density and expansion ratio were negatively correlated with a correlation coefficient of -0.51 ($p = 0.0026$).

All independent variables affected the production of brown pigments as measured by spectroscopy at 420 nm, as well as the *L* value (shift from black to white with increase in *L* value), *a* value (shift from green to red with increase in *a* value) and *b* value (shift from blue to yellow with increase in *b* value) (Table 5). The average values for each treatment are shown in Table 6. In general, as NDM concentration increased and the fluid addition rate decreased, brown pigments increased and extrudates became darker (Figure 6), the *L* and *b* values decreased, and the *a* value increased. Bailey *et al.* (1994) observed similar effects on colorimeter parameters as whey protein concentrate (5-25%) was extruded with corn meal and the feed moisture was varied (18-22%). Increased levels of browning can be explained by higher levels of protein and reducing sugars in NDM, and increased residence time as the fluid addition rate decreased as described by Heldman and Hartel (1997). The observed brown pigments, *L*, *a* and *b* values exhibited no clear trends with respect to the acid level in the injected fluid, and were highly dependent on the fluid addition rate and in some instances, the NDM concentration. As shown in the images in Figure 7, differences in color can be easily observed between treatments.

The WAI is commonly used to provide an indication of the extent of gelatinization of starch, but it can also indicate if the puffs will be compatible in a higher water activity matrix and maintain a crisp texture (Cristina da Silva *et al.* 2009). All independent variables affected the WAI (Table 5). Average values for each treatment are shown in Table 6. As shown in the response surface in Figure 8, a decrease in the

WAI was generally associated with a decrease in fluid addition rate, a decrease in NDM, and an increase in acid level in the injected fluid. The opposite was observed by Fernández-Gutiérrez *et al.* (2004) when they increased the protein level in a starch-acid casein blend. The authors attributed this effect to protein denaturation. In their study, an increase in moisture (18-29%) was associated with an increase in the WAI. This was hypothesized to be due to less starch damage and less protein denaturation.

The WSI provides an indication of the amount of soluble solids, and can be used to indicate the extent of starch molecule degradation and dextrinization during extrusion (Cristina da Silva *et al.* 2009; Lazou and Krokida 2010). All independent variables affected the WSI; however, the three-way interaction between these variables was not significant. An increase in the WSI was associated with a decrease in NDM concentration as depicted in the response surfaces for WSI in Figure 9. Also, an increase in acid in the injected fluid and a decrease in fluid addition rate increased the WSI. This trend was also observed as milk proteins were added to buckwheat, barley and maize starch extrusion formulations (Smietana *et al.* 1988). The authors attributed this effect to the formation of a protein-starchcomplex. The results from Amaya-Llano *et al.* (2007) also provide evidence that a protein-starch complex may have formed in our study. They extruded WPC with cornstarch at pH 3 to 8 and observed a decrease in pH to be associated with an increase in the WSI. They attributed this effect to the hydrolysis and fragmentation of starch, and the subsequent interaction with protein, which prevented denaturation of the protein and promoted water solubility.

NDM concentration and acid level affected the peak viscosity measured by RVA, while fluid addition rate did not (Table 5). As the NDM concentration increased, the peak viscosity also increased. A higher peak viscosity in the RVA, when comparing one treatment to another, can indicate that less starch was gelatinized during extrusion (and instead is gelatinized during the RVA analysis). The presence of substances other than starch can affect the degree of starch gelatinization if they compete with starch for available water (Núñez *et al.* 2009). Acid addition resulted in a decrease in the peak viscosity in our study. These results are consistent with results obtained by Ohishi *et al.* (2007). They used acetic acid to adjust the pH of a rice starch solution, and found that the non-pH adjusted samples had the highest maximum viscosity. The effect was attributed to a shear thinning, whereby the starch granules became increasingly fragile and more prone to breakdown during extrusion. Also, added acid would have reduced protein solubility in the NDM, thereby reducing the competition for water binding with starch. As mentioned previously, fluid addition rate did not affect the peak viscosity in this study. However, Köksel *et al.* (2003) increased the water feed rate during the extrusion of whole-grain durum wheat from 367 g/kg to 455 g/kg, which led to an increase in the peak viscosity as measured by RVA. They attributed the effect to the reduction of shear applied to the starch because water acted as a lubricant, as well as a reduced retention time during extrusion.

Differences in cellular structure as well as overall shape are apparent in stereomicroscope images of extruded puffs as shown in Figure 10. As NDM

concentration increased, the size of the pores decreased, the extrudates gained more structural integrity and were less prone to collapsing, which was observed as the product exited the extruder die. Similar effects on porosity have been reported by others as protein-containing ingredients have been added to extrusion formulations at high concentrations (Ghorpade *et al.* 1997; Lazou *et al.* 2007). In our study, a decrease in fluid addition rate resulted in an irregular extrudate structure and an observed increase in fragility and glassiness in the dried extrudates. Larger air cells were also more common at low fluid addition rates, and in the case of the 45% NDM extrudates, the structure was unable to withstand the extent of expansion at the die and subsequently collapsed prior to drying. The amount of fluid injected affects the rheological properties by reducing the viscosity, and has been reported to cause product shrinkage after expansion as well as increased fragility in some formulas (Miller, 2006; Barrett, 2003; Lazou *et al.* 2007). The acid addition level had variable effects in our study. At low concentrations of NDM (45%), high levels of acid increased the fragility of the puff, and resulted in a less regular structure, whereas at high concentrations of NDM (85%), acid addition increased the structural integrity of the extrudate. This effect was also observed by Barrett and Peleg (1992). They extruded citric acid and cornmeal and observed a reduction in density and average cell size with acid addition. Starch hydrolysis was seen asa probable explanation, with a reduction in molecular weight and subsequent decrease in cell wall strength. The effects observed at high NDM concentrations in our study are most likely due to the higher protein content, and protein-protein interactions as a result of the heat and acid, thus strengthening the extrudate structure.

Conclusions

In order for puffs and crisps to be used for snack and ingredient uses, the crisps must have good expansion, not be too dense, be light in color (not be too brown), and, if used in a high or intermediate water activity matrix (cereal bars, for example), must have a low WAI. Based on these criteria, as well as the responses examined, several conclusions can be drawn in regards to the use of NDM-starch puffs in product applications. First, puffs with higher percentages of NDM have higher bulk densities and lower expansion ratios. Sensory studies would need to be conducted in order to conclude if puffs have an acceptable texture. Also, the percentage of NDM, fluid addition rate and acid level affected puff color, with high concentrations of NDM and low moistures adversely affecting puff color, and acid level having a variable effect, depending on the fluid addition rate. Additionally, the WAI values observed in this study are similar to many breakfast cereals (Tan and Chinnaswamy 1993). It is clear that NDM provided structural integrity to the extrudates and can be used as a protein source for extruded products.

Benefit to Minnesota Economic Development

This study allows producers of extruded protein ingredients for use as inclusions in bars or in cereal to use NDM in their products as a protein source. This could lead to increased utilization of this milk commodity. Also, two different starches were evaluated in pretrial experiments and modified waxy maize starch was preferable to the potato starch evaluated and would be recommend for use with NDM.

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Tables and Figures

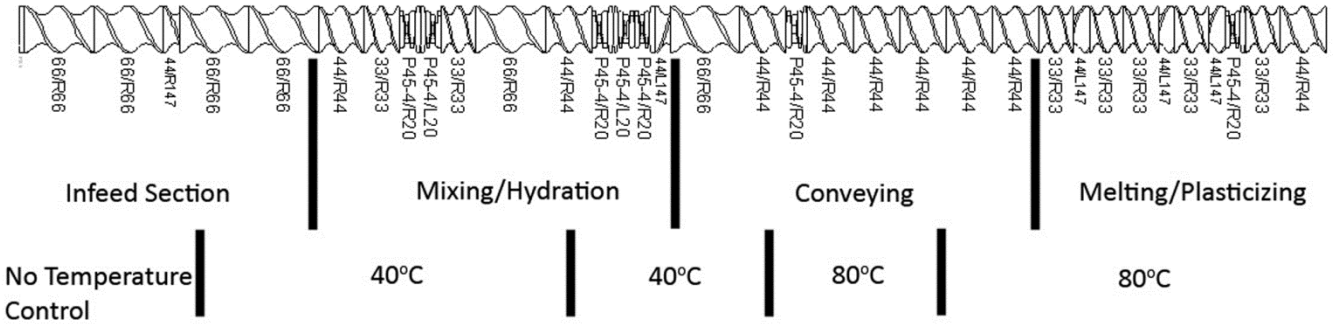

Figure 1. Screw design used on a Buhler 44mm co-rotating twin-screw extruder for the manufacture of nonfat dry milk and starch puffs

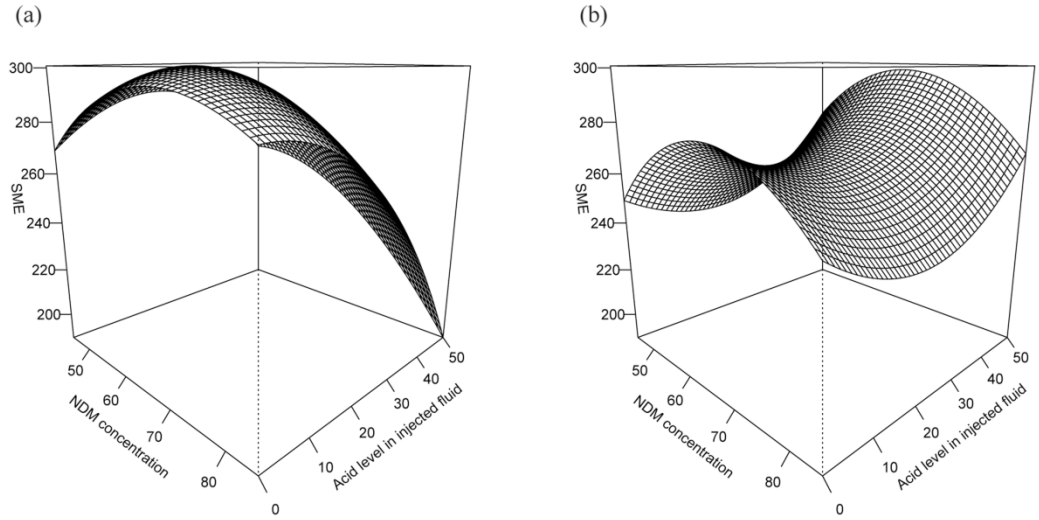

Figure 2. The effect of nonfat dry milk concentration (%) and acid level (% of the added fluid) at (a) low fluid addition rate (6.5 kg/h) and (b) high fluid addition rate (7.3 kg/h) on SME (specific mechanical energy, W/kg) during product extrusion on a Buhler 44mm co-rotating twin-screw extruder

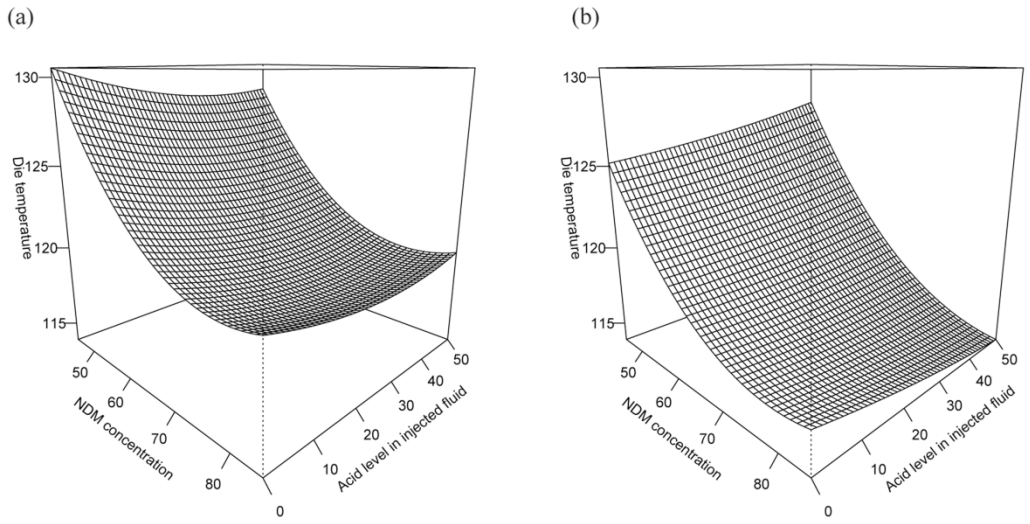

Figure 3. The effect of nonfat dry milk concentration (%) and acid level (% of the added fluid) at (a) low fluid addition rate (6.5 kg/h) and (b) high fluid addition rate (7.3 kg/h) on die temperature (°C) during product extrusion on a Buhler 44mm co-rotating twin-screw extruder

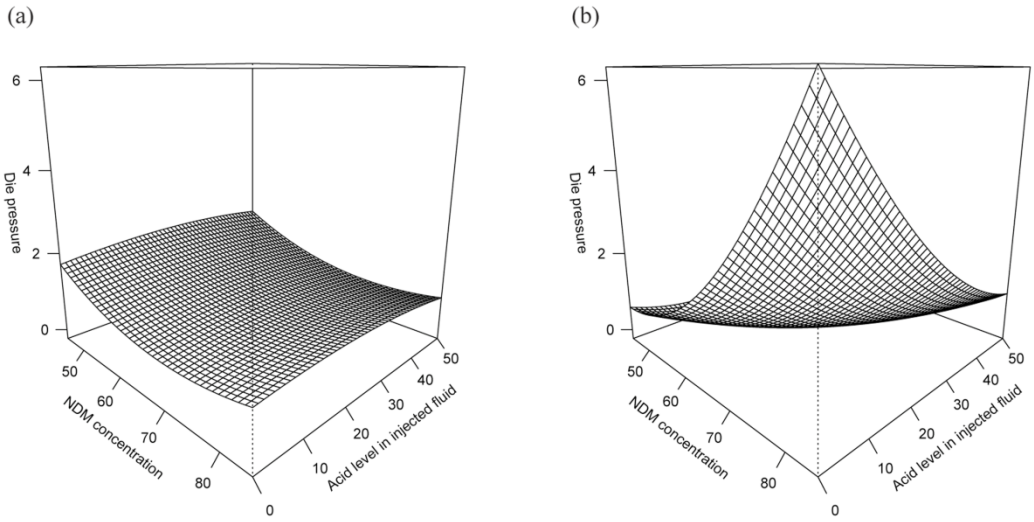

Figure 4. The effect of nonfat dry milk concentration (%) and acid level (% of the added fluid) at (a) low fluid addition rate (6.5 kg/h) and (b) high fluid addition rate (7.3 kg/h) on die pressure (bar) during product extrusion on a Buhler 44mm co-rotating twin-screw extruder

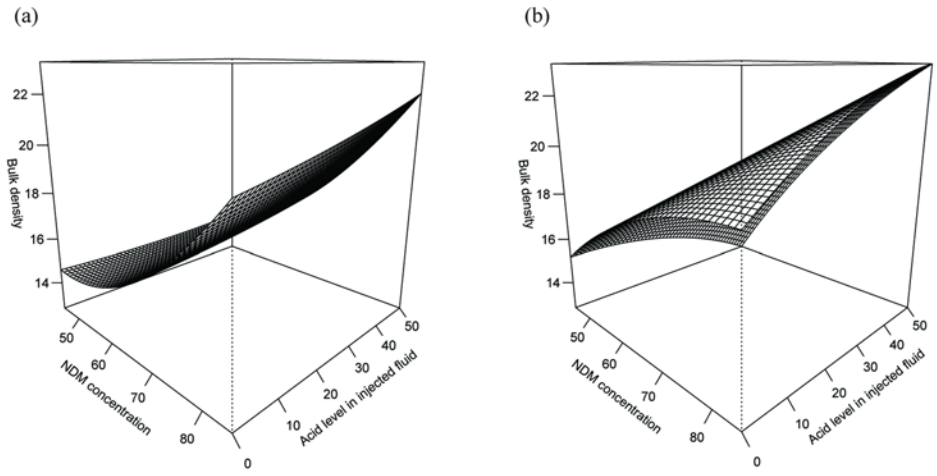

Figure 5. The effect of nonfat dry milk concentration (%) and acid level (% of the added fluid) at (a) low fluid addition rate (6.5 kg/h) and (b) high fluid addition rate (7.3 kg/h) on bulk density (g/100 cm³) of 5cm ropes of dried extrudate

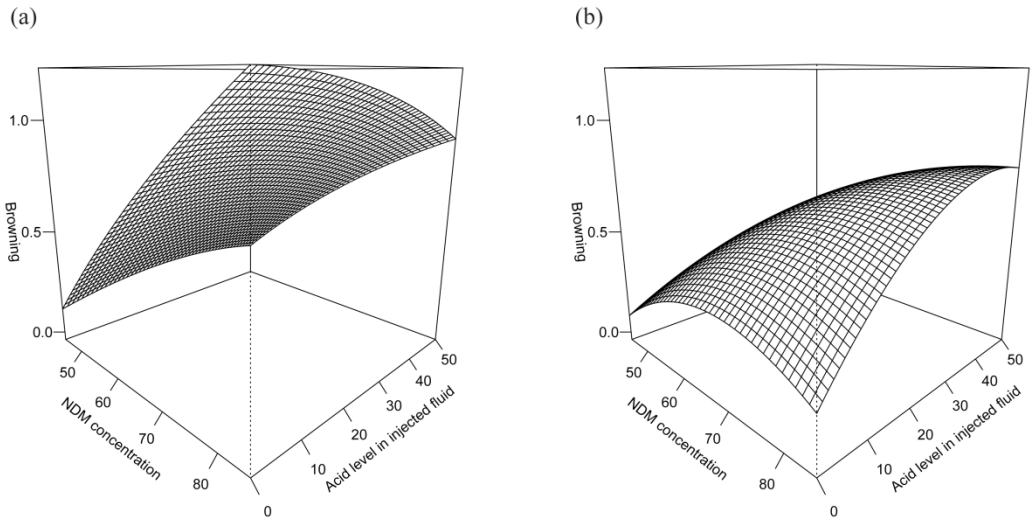

Figure 6. The effect of nonfat dry milk concentration (%) and acid level (% of the added fluid) at (a) low fluid addition rate (6.5 kg/h) and (b) high fluid addition rate (7.3 kg/h) rate on browning (Absorbance420/g solids as per the method of Davies et al. 2008) of dried extruded samples

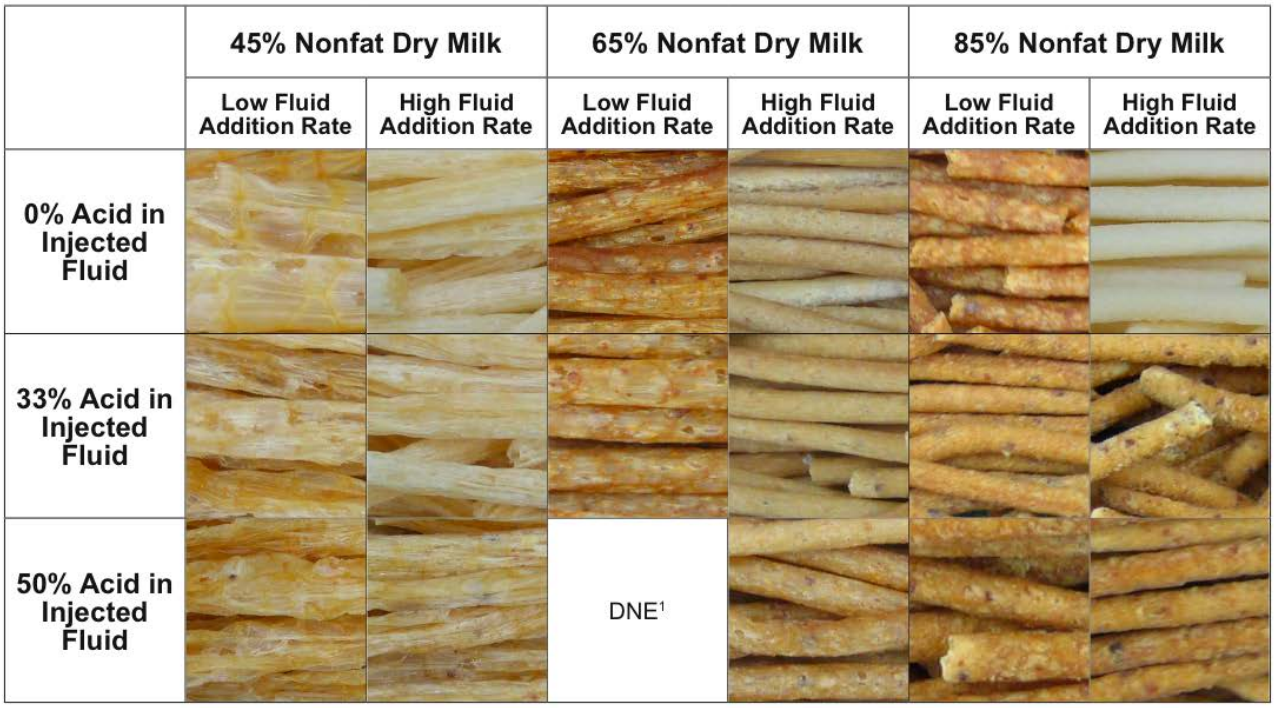

Figure 7. The effect of nonfat dry milk concentration, fluid addition rate and acid level in injected fluid on dried extrudate appearance

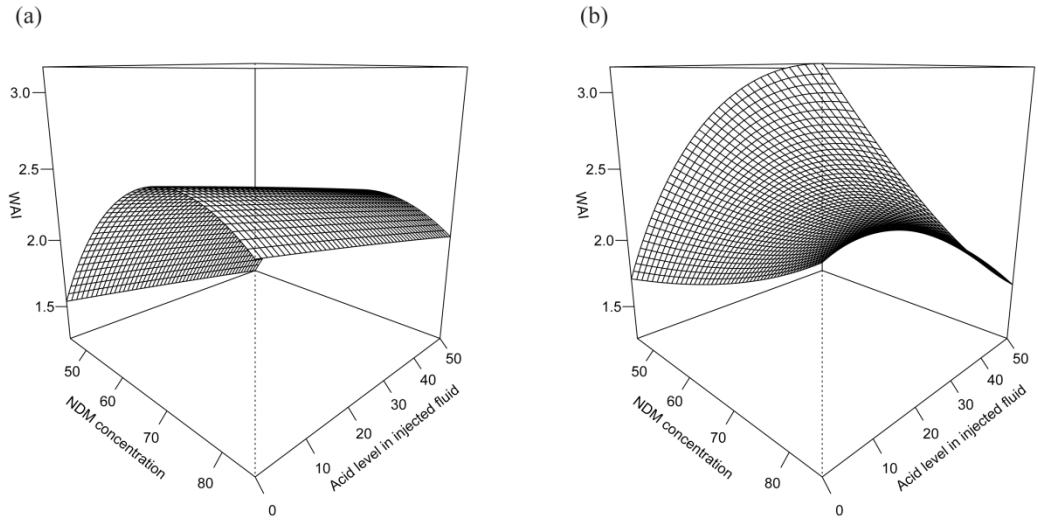

Figure 8. The effect of nonfat dry milk concentration (%) and acid level (% of the added fluid) at (a) low fluid addition rate (6.5 kg/h) and (b) high fluid addition rate (7.3 kg/h) on water absorption index (WAI) of dried extruded samples. WAI = the weight of gel/weight of dry solids per the method reported in Onwulata et al. 1998.

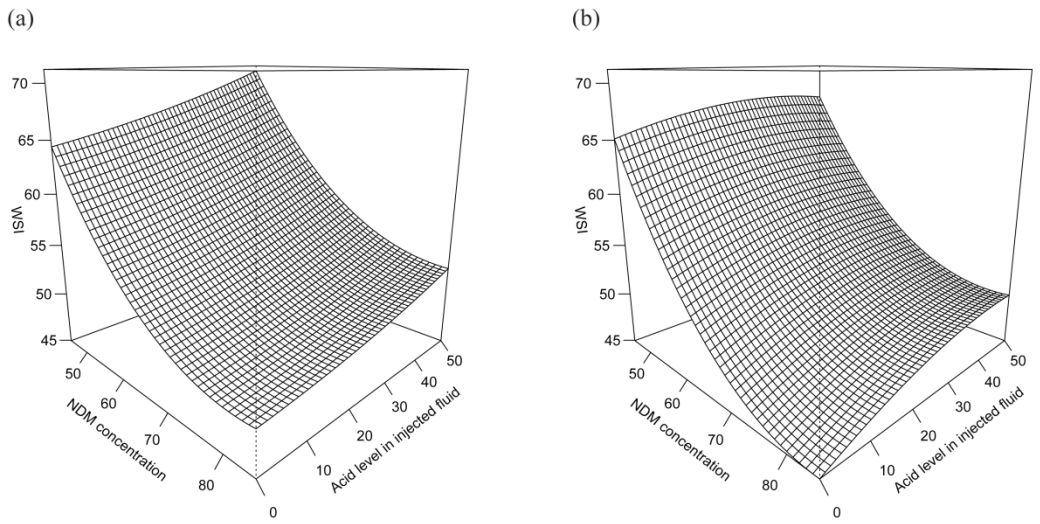

Figure 9. The effect of nonfat dry milk concentration (%) and acid level (% of the added water) at (a) low fluid addition rate and (b) high fluid addition rate on water solubility index (WSI) of dried extruded samples. WSI = weight dried supernatant/weight sample) x 100, per the method per the method reported in Onwulata et al. 1998.

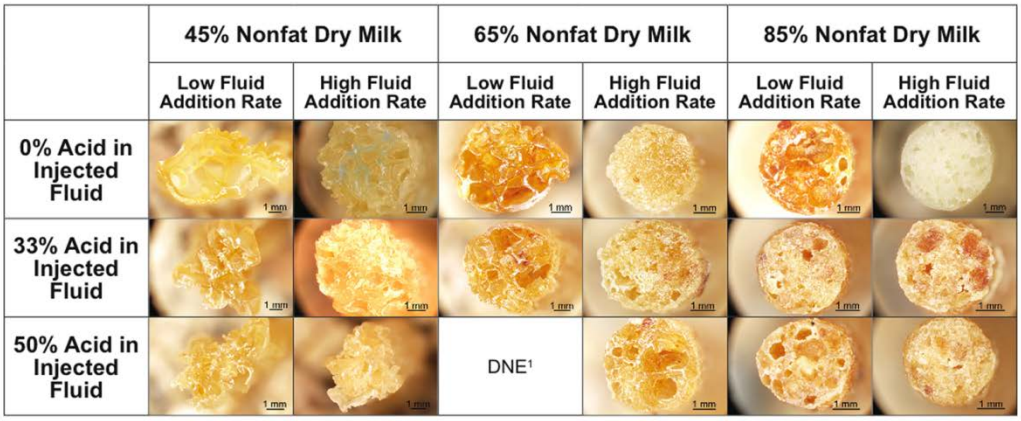

Figure 10. Stereomicroscopic images of the extruded samples as nonfat dry milk concentration, fluid addition rate, and the acid level in injected fluid were varied

TABLE1. PROXIMATE COMPOSITION OF THE DRY BLENDED NONFAT DRY MILK AND STARCH MIXTURES[1]

	% Nonfat Dry Milk/% Starch		
Measurement[2]	45/55	65/35	85/15
Moisture, % (SE[3])	6.33 (0.07)	5.38 (0.04)	4.14 (0.07)
Protein, % (SE)	15.78 (0.07)	22.80 (0.04)	30.00 (0.04)
Fat, % (SE)	0.348(0.022)	0.455 (0.019)	0.602 (0.036)
Ash, % (SE)	3.69 (0.01)	5.22 (0.01)	6.77 (0.01)
Carbohydrates, % (SE)	73.85 (0.05)	66.15 (0.05)	58.50 (0.10)

[1]Determined on a wet basis, g/100 g

[2]Number of observations per dry blend formula: Moisture= 2; Protein= 3; Fat= 2; Ash=2; Carbohydrates= not applicable because calculated by difference

[3]Standard error

TABLE 2. pH OF THE DRIED EXTRUDATES[1]

Nonfat dry milk concentration (% of dry mix) [2]	Fluid addition rate[3]	Acid level in injected fluid[4]		
		LA	MA	HA
45	LM	6.08	4.75	4.45
45	HM	6.11	4.77	4.37
65	LM	5.99	5.02	DNC[5]
65	HM	6.06	4.95	4.61
85	LM	5.99	5.31	4.97
85	HM	6.16	5.25	4.95

[1]SD between product replicates=0.05 (n= 2); SD between sample replicates=0.02 (n=2)

[2]NDM = low-heat nonfat dry milk; mix fed at 40 kg/h

[3]Fluid addition rate: LM = 6.5 kg/h; HM = 7.3 kg/h

[4] Liquid lactic acid was added to water, and the combined fluid was injected into the extruder at two different fluid addition rates. Acid level was the percent of the liquid acid, by weight, in the injected fluid: LA = 0%; MA = 33%; HA = 50%

[5]Did not collect

TABLE 3. THE EFFECT OF INDEPENDENT VARIABLES, AND INTERACTIONS BETWEEN THE INDEPENDENT VARIABLES ON PROCESS RESPONSES

Variable	Process responses[1]			
	SME[2] (W/kg)	Motor torque (Nm)	Die temperature (ºC)	Die pressure (bar)
NDM[3]	*[4]	**[5]	**	**
Acid level in injected fluid	*	**	**	**
Fluid addition rate	*	**	**	**
NDM x acid level in injected fluid	*	**	**	**
NDM x fluid addition rate	NSD[6]	**	**	**
Acid level in injected fluid x fluid addition rate	*	**	**	**
NDM x acid level in injected fluid x fluid addition rate	NSD	**	**	**

[1]Number of observations for each process response, per treatment, per rep = 2

[2]Specific mechanical energy

[3]Nonfat dry milk

[4]Significant difference p <0.05

[5]Significant difference p <0.001

[6]No significant difference

TABLE 4. SME[1], MOTOR TORQUE, DIE PRESSURE AND DIE TEMPERATURE RESPONSES FOR EACH TREATMENT

Nonfat dry milk concentration (% by weight of dry mix)[2]	Fluid addition rate[3]	Acid level in injected fluid[4]	SME (W/kg)	Motor torque (Nm)	Die pressure (bar)	Die temperature (ºC)
45	LM	LA	272^{ab}	293^{a}	1.78^{a}	131^{c}
45	LM	MA	271^{ab}	308^{a}	1.84^{a}	129^{bc}
45	LM	HA	272^{ab}	317^{a}	1.87^{a}	129^{bc}
45	HM	LA	260^{a}	281^{a}	1.21^{a}	125^{a}
45	HM	MA	260^{a}	299^{a}	0.64^{a}	127^{ab}
45	HM	HA	255^{ac}	300^{a}	1.23^{a}	128^{abc}
65	LM	LA	301^{ab}	324^{a}	0.80^{a}	122^{h}
65	LM	MA	276^{ab}	314^{a}	1.04^{a}	120^{dfh}
65	LM	HA	DNC[5]	DNC	DNC	DNC
65	HM	LA	255^{a}	281^{a}	0.68^{a}	118^{def}
65	HM	MA	254^{ac}	310^{a}	0.14^{a}	117^{deg}
65	HM	HA	340^{b}	298^{a}	1.03^{a}	118^{def}
85	LM	LA	272^{ab}	311^{a}	1.07^{a}	121^{fh}
85	LM	MA	266^{ab}	307^{a}	1.16^{a}	119^{defh}
85	LM	HA	177^{c}	307^{a}	0.80^{a}	120^{dfh}
85	HM	LA	266^{ab}	190^{b}	7.02^{b}	116^{egi}
85	HM	MA	249^{ac}	304^{a}	1.60^{a}	114^{gi}
85	HM	HA	249^{ac}	291^{a}	0.57^{a}	114^{i}
SD between product replicates (n= 2)			19	11	0.35	0.402
SD between sample replicates (n= 2)			5	11	0.74	0.970

[1]SME = specific mechanical energy

[2]Dry mix fed at 40 kg/h

[3]Fluid addition rate: LM = 6.5 kg/h; HM = 7.3 kg/h

[4]Liquid lactic acid was added to water, and the combined fluid was injected into the extruder at two different fluid addition rates. Acid level was the percent of the liquid acid, by weight, in the injected fluid: LA = 0%; MA = 33%; HA = 50%

[5]Did not collect

[a-i]Means without a common superscript letter within the same column are significantly different ($p < 0.05$)

TABLE 5.THE EFFECT OF THE INDEPENDENT VARIABLES AND INTERACTIONS BETWEEN THE INDEPENDENT VARIABLES ON PRODUCT RESPONSES

Variable	Product responses[1]								
				Color[2]					
	Bulk density[3]	ER[4]	Browning[5]	*L* value	*a* value	*b* value	WAI[6]	WSI[7]	RVA Peak viscosity[8]
NDM[9]	**[10]	**	**	**	**	**	**	**	**
Acid level in injected fluid	NSD[11]	NSD	**	**	**	**	**	**	**
Fluid addition rate	NSD	NSD	**	**	**	**	**	*	NSD
NDM x Acid level in injected fluid	*[12]	NSD	**	**	**	**	**	NSD	**
NDM x fluid addition rate	NSD	NSD	**	**	**	**	**	NSD	NSD
Acid level in injected fluid x fluid addition rate	NSD	NSD	**	**	**	**	**	NSD	NSD
NDM x Acid level in injected fluid x fluid addition rate	NSD	NSD	**	**	**	**	**	NSD	NSD

[1] Number of observations for each product response, per treatment, per rep: Bulk density= 1; ER= 10; Browning= 6; Color= 5; WAI= 2; WSI= 2; RVA profile= 2

[2]Color determined by measuring *L*, *a* and *b* values of uncrushed extrudates on a HunterLab colorimeter

[3]Measured in g/100 cm^3

[4]ER=Expansion ratio = diameter of extrudate/diameter die orifice

[5]Browning was determined by measuring absorbance at 420 nm after treating samples as per the method of Davies *et al.* 2008

[6]WAI=Water absorption index determined by the method of Onwulata *et al.* 1998 (weight of gel/weight of dry solids)

[7]WSI=Water solubility index was determined by the method of Onwulata *et al.* 1998 ((weight of dried supernatant/weight of sample) x 100)

[8]Peak viscosity (cP) was measured on hydrated extrudate (pre-dried) using a pasting method on a Rapid Visco Analyzer Model 4 (Newport Scientific, Australia)

[9]Nonfat dry milk

[10]Significant difference $p < 0.001$

[11]NSD = No significant difference

[12]Significant difference $p < 0.05$

TABLE 6. BROWNING, *L* VALUE, *a* VALUE, *b* VALUE AND WAI[1] RESPONSES FOR EACH TREATMENT

Nonfat dry milk	Fluid additio	Acid level in	Browning (A_{420}/g	*L* value	*a* value	*b* value	WAI (wt of

concentration (% by wt of dry mix)[2]	n rate[3]	injected fluid[4]	solids)				gel/wt of dry solids)
45	LM	LA	0.162^{ab}	44.8^{c}	8.5^{cd}	19.9^{ac}	1.53^{abe}
45	LM	MA	0.160^{ab}	40.7^{cdef}	8.4^{cd}	16.8^{cde}	1.41^{bde}
45	LM	HA	0.127^{a}	43.3^{cde}	8.5^{cd}	18.2^{cd}	1.20^{d}
45	HM	LA	0.0819^{a}	60.5^{ab}	5.1^{a}	23.0^{ab}	1.66^{abc}
45	HM	MA	0.0542^{a}	55.9^{a}	5.9^{ab}	19.6^{ac}	1.49^{abde}
45	HM	HA	0.104^{a}	44.0^{ce}	7.9^{bc}	17.7^{cde}	1.24^{de}
65	LM	LA	0.640^{cde}	37.5^{cdefg}	9.5^{cd}	15.7^{def}	2.48^{k}
65	LM	MA	0.796^{ef}	34.9^{defg}	9.4^{cd}	14.4^{ef}	2.32^{ijk}
65	LM	HA	DNC[5]	DNC	DNC	DNC	DNC
65	HM	LA	0.435^{bc}	44.7^{c}	7.9^{bc}	17.7^{cde}	2.79^{h}
65	HM	MA	0.678^{cde}	39.7^{cdef}	8.8^{cd}	16.2^{cde}	2.12^{fij}
65	HM	HA	0.465^{cd}	34.2^{dfg}	9.6^{cd}	14.3^{ef}	1.84^{cfg}
85	LM	LA	1.27^{g}	29.8^{g}	10.1^{d}	12.2^{f}	2.39^{jk}
85	LM	MA	1.02^{fg}	35.0^{defg}	9.8^{cd}	14.9^{def}	2.06^{fgi}
85	LM	HA	0.930^{ef}	34.7^{defg}	10.0^{cd}	14.4^{def}	2.07^{fgi}

85	HM	LA	0.0971[a]	69.0[b]	2.0[e]	24.9[b]	3.22[l]
85	HM	MA	0.919[ef]	38.2[cdefg]	9.5[cd]	16.3[cde]	1.89[cfg]
85	HM	HA	0.754[def]	33.8[fg]	9.4[cd]	14.3[ef]	1.79[acg]
SD between product replicates (n=2)			0.0736	2.3	0.4	0.9	0.0747
SD between sample replicates[6]			0.0248	0.9	0.7	0.3	0.0192

[1]Water absorption index

[2]Dry mix fed at 40 kg/h

[3]Fluid addition rate: LM = 6.5 kg/h; HM = 7.3 kg/h

[4]Liquid lactic acid was added to water, and the combined fluid was injected into the extruder at two different fluid addition rates. Acid level was the percent of the liquid acid, by weight, in the injected fluid: LA = 0%; MA = 33%; HA = 50%

[5]Did not collect

[6]n = 2 for WAI; n = 5 for *L*, *a* and *b* values; n = 6 for browning

[a-i]Means without a common superscript letter within the same column are significantly different ($p < 0.05$)