Influence of protein content and storage temperature on the particle morphology and flowability characteristics of milk protein concentrate powders

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ABSTRACT

Milk protein concentrate (MPC) powders are widely used as ingredients for food product formulations due to their nutritional profile and sensory attributes. Processing parameters, storage conditions, and composition influences the flow properties of MPC powders. This study investigated the bulk and shear flow properties of 70.3, 81.5, and 88.1% (wt/wt, protein content) MPC after storage for 12 wk at 25 and 40°C. Additionally, the morphological and functional changes of the MPC powders were investigated and correlated with flowability. After 12 wk of storage at 25°C, the basic flow energy values significantly increased from 510 to 930 mJ as the protein content increased from 70 to 90% (wt/wt). Flow rate index was significantly higher for samples with high protein content. Dynamic flow tests indicated that MPC powders with high protein content displayed higher permeability. Shear tests confirmed that the samples stored at 25°C were more flowable than samples stored at 40°C. Likewise, the higher-protein content samples showed poor shear flow behavior. The results indicated that MPC powders stored at 25°C had less cohesiveness and better flow characteristics than MPC powders stored at 40°C. Overall, the MPC powders had markedly different flow properties due to their difference in composition and morphology. This study delivers insights on the particle morphology and flow behavior of MPC powders.

Key words: high-protein dairy powders, flow properties, and powder rheology

INTRODUCTION

Milk protein concentrate (MPC) powders are high-protein dairy ingredients that are added to a variety of food product formulations to improve their nutritional, functional, and sensory properties. Milk protein concentrate powders contain higher protein content compared with nonfat dry milk and are lower in serum phase components such as lactose and soluble minerals. The MPC powders are manufactured using membrane separation techniques such as ultrafiltration or microfiltration in combination with diafiltration to achieve higher protein content (Agarwal et al., 2015). Subsequently, unit operations such as reverse-osmosis/evaporation and spray drying are employed to manufacture MPC in powder forms. High protein content, low lactose content, high buffering capacity, pleasant milk flavor profile, and functional properties such as water binding have increased the usage of MPC powders as ideal ingredients for a wide range of applications; for example, in beverages, yogurt, cheeses, nutritional formulations, and protein bars (Huffman and Harper, 1999; Agarwal et al., 2015). However, rehydration and flowability characteristics in MPC powders are influenced by intrinsic powder properties, such as surface and bulk composition (Crowley et al., 2014), particle structure (presence of pores and capillaries), and rehydration conditions (Crowley et al., 2015).

Changes in storage conditions (temperature and relative humidity), composition, capillary interactions within particles, and migration of cohesive chemical components to the surface of powder particles affects the flowability of food powders (Teunou et al., 1999; Iqbal and Fitzpatrick, 2006; Siliveru et al., 2017a). Additionally, before processing, dairy powders will be stored in silos and bags; thus, it is important to understand, predict, and control their flow behavior during storage, handling, and processing (Fitzpatrick

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et al., 2004b). The flow properties of powders also depend on its physical characteristics, such as particle size distribution, particle shape, surface structure, and bulk density (Crowley et al., 2014; Kim et al., 2005). Rennie et al. (1999) studied the effect of composition, particle size, moisture, and temperature on the cohesion of whole milk powder and skim milk powder (SMP) and noted that whole milk powder was more cohesive than SMP with increasing temperature, indicating the influence of fat and formation of liquid bridges in the cohesive mechanism. The flow properties of milk powders with different fat contents (Fitzpatrick et al., 2004b), infant milk powders (Szulc et al., 2017), and nonfat dry milk (Abdalla et al., 2017) have been studied previously. As MPC powders are widely used in different food product formulations, the need for high quality in the final product has motivated researchers to carry out studies on its functional properties, such as solubility (Anema et al., 2006; Fyfe et al., 2011; Hauser and Amamcharla, 2016a), improving solubility on storage (Bansal et al., 2017), and heat stability (Eshpari et al., 2014). Most of these studies focused on the technological aspects of MPC powders and their application in food product formulations. However, a lack of fundamental understanding exists on the bulk flow and shear characteristics of MPC powders, especially its effects during storage. Also, there is a need for more insight on the bulk flow and shear characteristics of MPC powders, because these properties are important for handling, formulation, mixing, processing, storage, and packaging (Fitzpatrick et al., 2004a). Moreover, investigating the flow behavior of powders is crucial in process equipment designed to ensure a reliable flow of powders and to avoid the formation of clogs or rat-holes (Fitzpatrick et al., 2004a). Very few researchers have investigated the effect of protein content on the flowability (Crowley et al., 2014; Silva and O'Mahony, 2017) and morphology or microstructure (Mimouni et al., 2010; Fang et al., 2012; Ji et al., 2017; Nasser et al., 2017) of high-protein milk powders. Likewise, no published information is available on the dynamic and shear properties of stored MPC powders.

The Freeman FT4 powder rheometer (Freeman Technology, Worcestershire, UK) is extensively used to characterize powder flow behavior by measuring the resistance offered by a powder bed to a helical blade (Krantz et al., 2009; Bharadwaj et al., 2010; Juliano and Barbosa-Canovas, 2010). The Freeman FT4 powder rheometer consists of blades, pistons, and shear heads that could be rotated and simultaneously moved transversely down into a powder bed while axial force and rotational force are measured (Freeman, 2007). The objective of our research was to measure and com-

pare the flow, dynamic, and shear flow properties of 70.3, 81.5, and 88.1% (wt/wt, protein content) MPC (MPC70, MPC80, and MPC90, respectively) after storage for 12 wk at 25 and 40°C.

MATERIALS AND METHODS

Experimental Design

The MPC powders (1 lot each) with 3 different protein contents 70, 80, and 90% (wt/wt), were collected from a commercial manufacturer within the United States. The MPC powders were sealed in Whirl-Pak bags (Nasco, Fort Atkinson, WI) and were stored at 2 different temperatures (25 and 40°C) in an incubator (Percival Scientific, Perry, IA) for 12 wk. Each measurement was carried out in duplicate. To mark and correlate the storage changes with morphology and flow measurements, the MPC powders were analyzed for microstructure, solubility index, and dissolution characteristics.

Microstructure

The microstructure of MPC powders were examined using a scanning electron microscope according to the method described by Mimouni et al. (2010). The MPC powders were directly mounted onto a carbon double-sided adhesive tape on microscopy stubs and sputter coated with palladium using a Denton Vacuum Desk II sputter coater (Denton Vacuum, Moorestown, NJ) for 15 min to avoid the charge buildup under the electron beam. The imaging was conducted using a S-3500N (Hitachi Science Systems Ltd., Tokyo, Japan) and examined by a secondary electron detector operating at 10 kV.

Morphological Analysis

Morphological characteristics of MPC powders were analyzed by Malvern Morphologi G3ID (Malvern Instruments, Worcestershire, UK). The circle equivalent diameter (CED), high sensitivity circularity (HSC), elongation, and convexity were calculated from the 2-dimensional images. Circularity (range 0 to 1) describes how close the shape of the particle to a perfect circle, whereas convexity (range 0 to 1) is a measure of the surface roughness of a particle. A smooth particle has a convexity of 1, whereas an irregularly shaped particle or very spiky has a convexity closer to 0. Circle or square has an elongation value of 0, whereas shapes with large aspect ratios have an elongation closer to 1 (Li et al., 2016). For each MPC sample, the mea-

surements were carried out in triplicate and the mean values were obtained.

Flow Properties

The FT4 powder rheometer was used to evaluate the flow properties of the stored MPC powders. A detailed description of FT4 and its application methods in powder characterization were previously reported by Freeman (2007) and Leturia et al. (2014). The FT4 powder rheometer comprises a vertical glass sample container (120 mm in height and 50 mm internal diameter) and a rotating blade (10 mm in height and 48 mm diameter) that navigates across the sample bed. Flow measurements using FT4 were calculated by continuously recording the forces causing deformation and flow of the powder particles executed by moving blade (Leturia et al., 2014). The FT4 rheometer has an built-in preconditioning step that helps uniformly pack the powder particles before the flow property measurement and is followed by splitting (to remove excess powder particles); the mass of the MPC powders were automatically recorded. Splitting the conditioning run performed on powders helped to decrease variability between trials due to filling (Bharadwaj et al., 2010). The properties of MPC powders measured using FT4 powder rheometer are described below.

Basic Flowability Energy. The basic flowability energy (**BFE**) is the basic essential energy required to start a specific flow pattern for an exact volume of conditioned MPC powders during the downward movement of the blade (Leturia et al., 2014). During downward displacement through the MPC powders, the BFE was calculated from work done in downward traverse of the blade at a constant tip speed of 100 mm/s.

Stability Index. Agglomeration and segregation of MPC powders were evaluated by a stability test. The test cycles were conducted at 100 mm/s of blade tip speed with the blade traversing through the MPC powder bed (Leturia et al., 2014). The stability index (**SI**) was calculated by using Equation 1:

$$SI = \frac{\text{total energy consumed at test 7 (mJ)}}{\text{total energy consumed at test 1 (mJ)}} \quad [1]$$

Flow Rate Index. To evaluate the flow rates of MPC powders, the flow energy was measured at 4 different blade tip speeds (100, 70, 40, and 10 mm/s) during downward traversing of the blade (Freeman, 2007; Leturia et al., 2014). Equation 2 was used to calculate the flow rate index (**FRI**):

$$FRI = \frac{\text{flow energy at test 4/}}{\text{flow energy at test 1.}} \quad [2]$$

Specific Energy. The specific energy was estimated during the upward navigation of the blade through the MPC powder bed using a very slight shearing and elevating mode of displacement. This gives an indication of the flow properties of the MPC powder in a loosely packed and unconfined state (Mitra et al., 2017). The value of specific energy (**SpE**) was then determined using Equation 3:

$$SpE = \frac{(\text{upward energy at cycle 6} + \text{upward energy at cycle 7})}{(2 \times \text{split mass})} \quad [3]$$

Compressibility

Compressibility evaluates the change in density as a function of applied normal stress. The MPC powder bed was conditioned and, subsequently, after splitting, the vented piston assembly was used to compress the MPC powder samples under increasing normal stress from 0.5 to 15 kPa (0.5, 1, 2, 4, 6, 8, 10, 12, and 15 kPa). After reaching equilibrium at the target stress, the distance traveled by the vented piston was measured and the compressibility was calculated as a percent change in volume (Freeman, 2007; Bian et al., 2015c).

Permeability

A vented piston assembly was used to measure the resistance to air flow across the powder bed and was quantified as the air pressure drop at each applied normal stress from 0.5 to 15 kPa (0.5, 1, 2, 4, 6, 8, 10, 12, and 15 kPa). The airflow velocity through the MPC powder bed was maintained at 2 mm/s throughout the test period (Freeman, 2007; Bian et al., 2015c).

Wall Friction Test

This test measures the ability of MPC powders to flow continuously (steady-state flow) across a container wall material. In our study, wall friction of MPC powders was measured against stainless steel, considering its use as the common processing equipment material. The rotational wall friction module used to analyze the MPC samples consisted of a serrated base assembly and a wall friction head to induce both normal and shear stresses for wall friction angle (**WFA**, Φ) measurement (Freeman, 2007). As the powder bed resisted the rota-

tion of the wall friction head, the torque increased until the resistance was overcome. The torque required to maintain this rotational momentum was measured as the shear stress. Equation 4 was used to calculate the WFA using the relationship between shear stress (τ_w) and normal stress (σ_w ; Leturia et al., 2014):

$$\Phi = \tan^{-1} \left(\frac{\tau_w}{\sigma_w} \right). \quad [4]$$

Shear Properties

The shear tests were carried out using the rotational shear cell accessory of the FT4 powder rheometer. The shear properties provide information on the flow of powders at rest and are commonly used to evaluate the flowability of powders during discharge in a process line (Mitra et al., 2017). Shear flow property data are important to design hoppers and select hopper construction material. The rotational shear cell part of the FT4 powder rheometer consists of a vessel with serrated base with a column containing the MPC sample and a FT4 shear head to achieve both vertical and rotational stresses. The powder shear properties, such as unconfined yield strength (**UYS**), cohesion, angle of internal friction (**AIF**), and flow function (**FF**), were measured using the standard shear testing program of FT4 rheometer. The normal stress was maintained constant at 9 kPa throughout the measurements.

Evaluation of Solubility and Dissolution Behavior

The MPC powders were reconstituted at 5% (wt/wt) powder concentration in distilled water and the solubility of the MPC powders stored at 25 and 40°C for 12 wk were estimated based on the TS in the supernatant obtained by centrifugation at $700 \times g$ for 10 min at 25°C, as described by Anema et al. (2006). The amount of soluble material (σ) in the MPC was calculated using Equation 5:

$$\sigma = (\text{weight of dry material/weight of solution}) \times 100. \quad [5]$$

The dissolution characteristics of the MPC powders stored at 25 and 40°C were evaluated using focused beam reflectance measurement (**FBRM**) method described by Hauser and Amamcharla (2016b).

Statistical Analysis

The flow and morphological characteristics were analyzed using PROC MIXED in SAS (version 9.4, SAS Institute Inc., Cary, NC) by Tukey's test at a significance P -value of 0.05. The Pearson correlation analysis that summarizes the strength of linear relationships between selected variables was also performed.

RESULTS AND DISCUSSION

As per the certificate of analysis provided by the manufacturer, the composition of MPC powders used in our study is shown in Table 1. As expected, the protein and lactose contents were significantly different ($P < 0.05$) for all the MPC powders used in our study. As the protein content increased from 70.3 to 88.1% (wt/wt), the lactose content decreased from 16.1 to 0.5% (wt/wt). On the other hand, the fat content in the MPC powders did not exhibit any significant difference ($P > 0.05$). Differences in physical properties, as influenced by the storage temperature and protein content, were the key factors evaluated in our study. The strength of linear relationships between selected variables is provided in Table 2.

Microstructure

Figure 1 illustrates the scanning electron micrographs of MPC powders after 12 wk of storage at 40°C. The scanning electron micrograph of MPC70 (stored for 12 wk at 40°C) revealed smoother surfaces of milk protein particles when compared with MPC90 and MPC80 stored at the same temperature. Additionally, no major microstructural changes were observed in the state of lactose (due to collapse of the particle structure) in the MPC powders, demonstrating that higher lactose contents can only cause such structural changes (Fyfe et al., 2011). Figures 2B and 2C illustrate that

Table 1. Compositional analysis (% wt/wt; means $\pm$ SD) of milk protein concentrate (MPC) powders used in this study

Type of MPC	Protein (% wt/wt)	Fat (% wt/wt)	Moisture (% wt/wt)	Lactose (% wt/wt)	Ash (%)
MPC70	70.3 $\pm$ 0.25	1.3 $\pm$ 0.05	4.7 $\pm$ 0.13	16.1 $\pm$ 0.51	6.4 $\pm$ 0.06
MPC80	81.5 $\pm$ 0.41	1.1 $\pm$ 0.06	4.9 $\pm$ 0.11	6.6 $\pm$ 0.62	6.3 $\pm$ 0.13
MPC90	88.1 $\pm$ 0.52	1.1 $\pm$ 0.07	4.6 $\pm$ 0.14	0.5 $\pm$ 0.66	6.3 $\pm$ 0.11

Table 2. Correlation coefficients between selected functional, morphological, and flowability properties of milk protein concentrate (MPC) powders¹

Variable	σ	CED	BFE	SI	FRI	SpE	UYS	Ch	AIF	FF
σ	1.00									
CED	-0.27	1.00								
BFE	-0.18	0.97*	1.00							
SI	0.23	-0.86*	-0.92*	1.00						
FRI	-0.42*	0.93*	0.89*	-0.88*	1.00					
SpE	-0.18	0.83*	0.86*	-0.97*	0.83*	1.00				
UYS	-0.78*	0.73*	0.65*	-0.65*	0.88*	0.58*	1.00			
Ch	-0.72*	0.74*	0.67*	-0.74*	0.91*	0.71*	0.97*	1.00		
AIF	-0.73*	0.79*	0.70*	-0.64*	0.90*	0.59*	0.99*	0.95*	1.00	
FF	0.86*	-0.59*	-0.53*	0.60*	-0.79*	-0.54*	-0.97*	-0.96*	-0.93*	1.00

¹ σ = solubility index; CED = circle equivalent diameter; BFE = basic flow energy; SI = stability index; FRI = flow rate index; SpE = specific energy; UYS = unconfined yield strength; Ch = cohesion; AIF = angle of internal friction; FF = flow function coefficient.

*Pearson's *r* values (positive or negative) are found to be significant ($P < 0.05$).

MPC80 and MPC90 after 12 wk of storage at 40°C had size alterations in powder particles, indicating more wrinkled particle surfaces. A similar surface appearance (MPC85) was demonstrated in a previous study (Fang et al., 2012), and such surfaces indicated shrinkage of the protein material (Mimouni et al., 2010). After 12 wk of storage at 40°C (Figure 1D), MPC90 exhibited

more holes, broken particles, and roughness on the surface of the particles. These findings agreed with previous studies in MPC powders stored for 30 (Fang et al., 2012) and 60 d (Gaiani et al., 2006). Overall, the results from scanning electron micrographs suggested that MPC powder particles were affected by the protein content.

Figure 1. Scanning electron micrographs ($\times 500$) of spray-dried milk protein concentrate powder (MPC) particles, (A) MPC70, (B) MPC80, (C) MPC90, and (D) MPC90 ($\times 5,000$), after 12 wk of storage at 40°C. MPC70, MPC80, and MPC90 indicate protein content in MPC powder.

Figure 2. Compressibility of the milk protein concentrate (MPC) powders, MPC70 (triangle), MPC80 (circle), and MPC90 (square), after 12 wk of storage at 25°C (solid) and 40°C (open). Values are means $\pm$ SD from duplicate analysis. MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively.

Morphology

The morphological properties of MPC powders after 12 wk of storage at 25 and 40°C were investigated and the results are summarized in Table 3. The CED of MPC powder particles increased with the increase in protein content at both storage temperatures (Table 3). The HSC results confirmed that morphology of MPC90 powder particles after storage at 25°C exhibited less round-shaped or more irregularly shaped particles (Table 3) when compared with the MPC70 and MPC80. For all the MPC powders, HSC increased as the storage temperature increased. At both storage temperatures, the elongation was higher in MPC90 when compared with MPC70, indicating more irregularly shaped particles. Interestingly, after storage for 12 wk at 40°C, the CED and elongation of MPC90 was $\sim$ 16 and $\sim$ 25% lower, respectively, when compared with MPC90 stored at 25°C. At both storage temperatures, the convexity was higher in MPC70 when compared with MPC90, indicating higher regular-shaped particles in the MPC70. Previously, Li et al. (2016) studied different lactose and milk protein isolate model systems produced in a pilot-scale spray dryer and reported that as the protein content increased the circularity and convexity increased, whereas elongation decreased for the resultant powders. However, in the present study, when comparing MPC powders stored at 25°C, circularity, convexity, and elongation did not follow the trend reported by Li et al.

(2016) and could be attributed to the variations in the spray dryer configurations between the 2 studies.

BFE

The BFE of MPC powders ranged from 510 to 930 mJ (Table 4). For the powders stored at 25°C, the BFE increased significantly ($P < 0.05$) with an increase in protein content from 70 to 90%. This indicated that the energy required to initiate the flow for MPC70 was less compared with the MPC80 and MPC90. A lower BFE requirement for MPC70 at both storage temperatures could be attributed to its particle morphology in terms of lower CED, higher HSC (less irregular shaped particles), and higher convexity. In addition to the morphological characteristics, chemical characteristics such as protein-protein interactions are less in MPC70 compared with MPC80 and MPC90, and consequently resulted in a lower BFE in MPC70. Moreover, the lactose content of MPC70 was 16.1%, and lactose can act as a molecular spacer and potentially limit the protein-protein interactions, thus improving flowability. On the other hand, higher BFE requirements in MPC80 and MPC90 are attributed to the morphology (CED, HSC, and convexity) of its powder particles and greater protein-protein interactions on the surface of the powder particles in MPC80 and MPC90. The MPC80 and MPC90 contained 6.6 and 0.5% lactose, respectively, and more protein-protein interactions on the surface of

Table 3. Morphological characteristics of milk protein concentrate (MPC) powders after 12 wk of storage at 25 and 40°C

Type	Circle equivalent diameter (μm)		High sensitivity circularity ²		Elongation ³		Convexity ⁴	
	25°C	40°C	25°C	40°C	25°C	40°C	25°C	40°C
MPC70	13.51 $\pm$ 0.06 ^{a,x}	14.12 $\pm$ 0.01 ^{a,y}	0.874 $\pm$ 0.002 ^{a,x}	0.890 $\pm$ 0.002 ^{a,y}	0.196 $\pm$ 0.004 ^{a,x}	0.177 $\pm$ 0.001 ^{a,y}	0.992 $\pm$ 0.001 ^{a,x}	0.991 $\pm$ 0.000 ^{a,y}
MPC80	17.06 $\pm$ 0.12 ^{b,x}	15.06 $\pm$ 0.35 ^{b,y}	0.874 $\pm$ 0.001 ^{a,x}	0.892 $\pm$ 0.001 ^{a,y}	0.186 $\pm$ 0.001 ^{b,x}	0.171 $\pm$ 0.001 ^{b,y}	0.988 $\pm$ 0.000 ^{b,x}	0.991 $\pm$ 0.001 ^{a,y}
MPC90	22.85 $\pm$ 0.02 ^{c,x}	19.68 $\pm$ 0.05 ^{c,y}	0.831 $\pm$ 0.001 ^{b,x}	0.879 $\pm$ 0.002 ^{b,y}	0.227 $\pm$ 0.001 ^{c,x}	0.181 $\pm$ 0.003 ^{c,y}	0.981 $\pm$ 0.000 ^{c,x}	0.988 $\pm$ 0.000 ^{b,y}

^{a-c}Mean values for different protein contents within a column with a different superscript differ ($P < 0.05$).

^{x,y}Mean values for the storage temperatures within a morphological parameter with a different superscript differ ($P < 0.05$).

¹MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively.

²High sensitivity circularity or HSC (range 0–1; a powder particle with perfect circle has a circularity of 1, whereas an irregularly shaped powder particle has a circularity value closer to 0).

³Elongation (range 0–1; shapes with large aspect ratios have an elongation closer to 1, whereas a circle or square-shaped powder particle have an elongation value of 0).

⁴Convexity (range 0–1; a smooth-shaped powder particle has a convexity of 1, whereas an irregularly shaped powder particle has a convexity closer to 0).

Table 4. Dynamic flow properties of milk protein concentrate (MPC) powders after 12 wk of storage at 25 and 40°C

Type ¹	Basic flow energy (mJ)		Stability index		Flow rate index		Specific energy (mJ/g)	
	25°C	40°C	25°C	40°C	25°C	40°C	25°C	40°C
MPC70	510.09 $\pm$ 12.32 ^{a,x}	512.53 $\pm$ 9.60 ^{a,x}	1.04 $\pm$ 0.02 ^{a,x}	1.05 $\pm$ 0.02 ^{a,x}	1.52 $\pm$ 0.01 ^{a,x}	1.49 $\pm$ 0.05 ^{a,x}	18.92 $\pm$ 0.01 ^{a,x}	18.34 $\pm$ 0.05 ^{a,x}
MPC80	695.09 $\pm$ 5.01 ^{b,x}	617.37 $\pm$ 10.01 ^{b,y}	0.97 $\pm$ 0.01 ^{a,x}	0.98 $\pm$ 0.02 ^{a,x}	1.57 $\pm$ 0.04 ^{b,x}	1.60 $\pm$ 0.02 ^{b,x}	24.14 $\pm$ 0.04 ^{b,x}	22.01 $\pm$ 0.02 ^{b,x}
MPC90	930.75 $\pm$ 1.54 ^{c,x}	722.46 $\pm$ 2.15 ^{c,y}	0.94 $\pm$ 0.01 ^{a,x}	0.97 $\pm$ 0.01 ^{a,x}	1.73 $\pm$ 0.01 ^{c,x}	1.71 $\pm$ 0.01 ^{c,x}	24.43 $\pm$ 0.03 ^{b,x}	23.92 $\pm$ 0.04 ^{b,x}

^{a-c}Mean values for different protein contents within a column with a different superscript differ ($P < 0.05$).

^{x,y}Mean values for the storage temperatures within a dynamic flow parameter with a different superscript differ ($P < 0.05$).

¹MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively.

the powder particles (Havea, 2006). The Pearson correlation (Table 2) revealed a positive correlation between CED and BFE ($r = 0.97$).

The MPC90 stored at 25°C showed a BFE of 930 mJ, whereas MPC90 stored at 40°C had a BFE of 722 mJ, which was ~22% lower when compared with MPC90 stored at 25°C. Similarly, MPC80 stored at 25°C had a BFE of 695 mJ, which was ~11% higher when compared with MPC80 stored at 40°C. Therefore, after storage for 12 wk at 40°C, less energy was required to initiate the flow in MPC80 and MPC90. This could be because coarse powders, in general, have better flow properties than fine powders (Li et al., 2016). Lapčík et al. (2015) reported low BFE values ranging from 127 to 157 mJ for SMP, demineralized whey powder, and whey powders. Mitra et al. (2017) observed BFE >750 mJ for *basundi* (heat-desiccated Indian dairy sweet) dry mix. Overall, the results indicated that MPC80 and MPC90 required more energy to initiate the flow when compared with MPC70 and may require more energy during bulk handling of MPC powders.

SI, FRI, and SpE

The SI values showed no significant differences ($P > 0.05$) with increase in protein content and storage temperature (Table 4). Bian et al. (2015a) reported that the powder is stable if the SI values falls in the range of 0.9 to 1.1, indicating no segregation and disintegration during flow. On the other hand, the powders with SI values less than 0.9 and more than 1.1 would be considered unstable (Bian et al., 2015a). After storage for 12 wk at 25°C, SI was highest for MPC70 (1.04), suggesting MPC70 is a stable powder (better flowability) with less segregation during flow. At both the storage temperature (25 and 40°C) the MPC powders were found to be stable and did not agglomerate during flow testing.

The FRI values were less than 1.73, indicating the MPC powders exhibited average flow rate sensitivities and showed less cohesiveness (Leturia et al., 2014). The FRI of MPC90 and MPC80 were significantly ($P < 0.05$) higher than that of MPC70 (Table 4). As expected, the MPC70 had the lowest FRI value because of its low cohesiveness (Table 5). The low FRI values in MPC70 compared with MPC80 and MPC90 indicated less interlocking of the powder particles (Jan et al., 2017) and suggested less irregularly shaped particles (Table 3). The increase in storage temperature showed no significant difference ($P > 0.05$) on the FRI of MPC powders. Indeed, in MPC powders, due to the presence of entrapped air, the powders were slightly influenced by flow rate (Mitra et al., 2017).

Table 5. Shear flow properties of milk protein concentrate (MPC) powders after 12 wk of storage at 25 and 40°C

Type ¹	Unconfined yield stress (kPa)		Cohesion (kPa)		Angle of internal friction (°)		Flow function coefficient	
	25°C	40°C	25°C	40°C	25°C	40°C	25°C	40°C
MPC70	2.57 ± 0.01 ^{a,x}	2.73 ± 0.05 ^{a,y}	0.58 ± 0.02 ^{a,x}	0.61 ± 0.01 ^{a,y}	34.55 ± 0.04 ^{a,x}	36.28 ± 0.01 ^{a,y}	6.06 ± 0.01 ^{a,x}	5.72 ± 0.01 ^{a,y}
MPC80	2.61 ± 0.07 ^{a,x}	2.97 ± 0.01 ^{a,y}	0.63 ± 0.02 ^{b,x}	0.72 ± 0.01 ^{b,y}	34.96 ± 0.01 ^{b,x}	37.63 ± 0.01 ^{b,y}	5.92 ± 0.01 ^{b,x}	5.15 ± 0.01 ^{b,y}
MPC90	3.12 ± 0.01 ^{b,x}	3.24 ± 0.11 ^{b,y}	0.75 ± 0.03 ^{c,x}	0.82 ± 0.01 ^{c,y}	40.38 ± 0.01 ^{c,x}	41.35 ± 0.01 ^{c,y}	5.24 ± 0.05 ^{c,x}	4.91 ± 0.02 ^{c,y}

^{a,c}Mean values for different protein contents within a column with a different superscript differ ($P < 0.05$).

^{x,y}Mean values for the storage temperatures within a shear flow parameter with a different superscript differ ($P < 0.05$).

¹MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively.

Specific energy indicated how easily MPC powders will flow in an unconfined environment, signifying relative cohesion of the MPC powders under low-stress conditions; SpE demonstrates the energy needed to establish a specific flow pattern in a preconditioned and precise volume of MPC powders (Freeman, 2007; Freeman and Fu, 2008; Jan et al., 2017). The SpE increased significantly ($P < 0.05$) with the increase in protein content (Table 4). The SpE values ranged from 18 to 24 mJ/g, and higher values indicated that MPC80 and MPC90 were less flowable when compared with MPC70. Morphological results showed that MPC90 has higher CED, and therefore higher particle interlocking could occur during the transition of the blade through the MPC powder bed (Bharadwaj et al., 2010). The MPC70 had the lowest SpE value, indicating that it was the least cohesive sample at controlled condition state, and suggested that it flows readily in a low stress and conditioned state. The lower SpE values in MPC70 could be further explained with less particle interlocking because of comparatively smoother surfaces in MPC70. However, MPC90 showed highest (24.4 mJ/g) SpE value, indicating it to be more cohesive.

Overall, the bulk flow results were correlated (Table 2) with the CED, indicating that MPC powders composed of more regular-shaped particles (high HSC) flow better than those with irregular-shaped particles. Increment of BFE and SpE with increasing protein content and storage temperature could be attributed to the lower flowability, higher cohesion, and increased packing of finer particles in void spaces (Mitra et al., 2017). Scanning electron micrographs (Figure 1D) confirmed the presence and packing of such smaller particles in the MPC90 samples stored at 40°C.

Compressibility

Compressibility results showed that the percent change in volume increased with normal stress applied for all the MPC powders (Figure 2). Indeed, the increase in percent change in volume with increase in normal stress applied for the MPC powders would be due to the closer particle packing and increase in inter-particle surface contact (Bian et al., 2015b; Jan et al., 2017). The difference in protein contents and storage temperatures did not exhibit any noticeable effect on the compressibility. Compressibility at 15 kPa pressure suggested that MPC90 (both at 25 and 40°C), when compared with MPC70 and MPC80, was less compressible, indicating coarser and irregular particles. Compressibility results could be related to the difference in composition, particle morphology, particle interlocking, and inter-particle interactions in MPC powders after storage. Furthermore, fine particles exhibit higher com-

pressibility than the coarser ones because of the greater surface area and fewer voids (Jan et al., 2017). Overall, particle morphology and particle interlocking after storage for 12 wk at 20 and 40°C marginally influenced the compressibility, indicating less compressibility-related constraints in handling of stored MPC powders.

Permeability

Permeability is a measure of the powder particles resistance to airflow. Evaluating permeability of MPC powders is important to understand their flow during handling and processing. Knowledge of powder permeability is also important in developing efficient unloading strategies (Bian et al., 2015c). The MPC90 showed the lowest pressure drop, indicating higher permeability (Figure 3). Above a normal stress of 8 kPa, the increase in pressure drop for MPC80 and MPC70 stored at 40°C was low when compared with MPC80 and MPC70 stored at 25°C, indicating higher permeability for the powders stored at 40°C. Higher permeability of MPC90 could be related to its higher CED and irregular shape (lower HSC), indicating a larger void structure. However, in MPC70 after storage for 12 wk, the rearrangement of particles might have reduced the inter-particle void spaces (Bian et al., 2015c), resulting in an increased pressure drop and reduced permeability. Compared with MPC80, MPC90 has a lower pressure drop and is therefore more permeable (regardless of its comparable compressibility results), indicating the influence of particle shape on packing structure when particle size distributions of the powders are similar (Siliveru et al., 2016, 2017a).

Wall Friction

Figure 4 shows the WFA values of MPC powders stored at 25 and 40°C for 12 wk and illustrates possible flow constraints due to frictional resistance in MPC powder particles on bin or hopper wall material. The influence of protein content and storage temperature on WFA was notable, as observed from Figure 4. The increase in storage temperature had a significant effect on the WFA ($P < 0.05$) in MPC powders, indicating an associated increase in wall-particle interactions. It is difficult to interpret the exact reasons for the observed increase in wall friction with temperature and protein content for some powders and a decrease for other powders. Iqbal and Fitzpatrick (2006) also reported the effect of storage conditions on the wall friction characteristics of whey permeate powder and concluded that it is difficult to interpret why wall friction increases with temperature for some powders and decreases for other. Higher values of WFA in MPC90 stored (12 wk) at 25

Figure 3. Effect of applied normal stress on pressure drop across the milk protein concentrate (MPC) powders, MPC70 (triangle), MPC80 (circle), and MPC90 (square), after 12 wk of storage at 25°C (solid) and 40°C (open). Values are the means of data from duplicate analysis. MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively. 1 mBar = 100 Pa.

and 40°C were in accordance with increasing cohesivity (Table 5), which was within the range of values (11.8 to 27.3°) reported by Fitzpatrick et al. (2004a) for various food powders. The larger WFA in MPC powders indicated higher wall friction and greater deposition or segregation on wall (Iqbal and Fitzpatrick, 2006; Crowley et al., 2014). The WFA was higher in MPC90 when compared with MPC80, suggesting particle shape influenced WFA. Higher WFA values indicated that MPC90 have a greater chance of adhesion in a hopper or bin wall material, suggesting that steeper hopper angle is required to obtain consistent and reliable bulk flow in MPC90. Previously, Fitzpatrick et al. (2007) found that whey permeate powder stored at 30°C had less WFA when compared with the powder stored at 15°C. For comparison, Crowley et al. (2014) reported lower WFA values (18°) for MPC70 and comparatively higher WFA values (19.6°) for MPC90.

Shear Properties

Protein content and storage temperature have influenced the shear flow properties and the results are shown in Table 5. The UYS values of the MPC powders increased with the increase in protein content and storage temperature (Table 5), indicating that more cohesive interactions between the particles of MPC powders. The UYS increased from 2.57 to 3.12

kPa with an increase in protein content from 70 to 90% for samples stored for 12 wk at 25°C. For MPC70, the UYS significantly increased ($P < 0.05$) from 2.57 to 2.73 kPa with an increase in storage temperature from 25 to 40°C. At both storage temperatures, the particles of MPC90 powders were more resistant to flow than MPC70 powder particles. Irregular particle shape

Figure 4. Wall friction angle of the milk protein concentrate (MPC) powders after 12 wk of storage at 25°C (gray) and 40°C (white). Values are the means of data from duplicate analysis. Error bars indicate SD. Mean values with different letters for different protein content (a–c) and for the storage temperature (x,y) differ ($P < 0.05$). MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively.

could be a possible reason for the higher UYS values of MPC90 than MPC70. Teunou et al. (1999) reported that, for whey permeate powders, the UYS increased after storage for 1 wk at 20°C at a maximum consolidating stress of 40 kPa. Previously, Lapčik et al. (2015) observed the UYS of SMP and whey powders to be 4.48 and 4.69 kPa, respectively.

A significant increase ($P < 0.05$) in cohesion values were observed with the increase in protein content and storage temperature. Possible reasons for this could be differences in particle arrangement and particle interlocking in MPC powders after storage for 12 wk at 25 and 40°C. Although, the lactose content was higher in MPC70, the increased storage temperature did not increase the cohesion because the glass transition temperature of MPC was higher than the storage temperatures used in our study (Li et al., 2016). We noted a positive correlation (Table 2) of cohesion to FRI ($r = 0.91$), indicating cohesive powders were more sensitive to flow rate because of the presence of entrained air (Mitra et al., 2017). We found a positive correlation (Table 2) of CED to cohesion ($r = 0.74$). This suggests that particle morphology imparts significant flow changes. The MPC90 showed cohesion values ~34% higher than MPC70 due to larger CED in MPC90. Similarly, MPC80 showed cohesion values ~15% higher than MPC70, suggesting less particle-particle interlocking and resistance to flow in MPC70. The higher cohesion in MPC90 could be due to its higher protein content, differences in particle shape, particle interlocking, packing of smaller particles in void spaces, and surface irregularity (Figure 1D), enabling higher cohesive interactions (Teunou and Fitzpatrick, 2000; Siliveru et al., 2016, 2017b). The poor flow of MPC80 and MPC90 was probably due to high CED (Table 3), which may be due to increased particle-particle interactions and particle interlocking. Previously, Fitzpatrick et al. (2007) reported that rennet casein and sodium caseinate powders were also cohesive. Although the flowability of MPC powders after storage has not been previously characterized, Crowley et al. (2014) reported that more cohesive interactions occurred between particles in MPC powders with higher protein content.

A significant increase ($P < 0.05$) in AIF values was observed with the increase in protein content and storage temperature, indicating flow constraints. The AIF values increased from 40.38° to 41.35° and 34.55° to 36.28° with an increase in temperature from 25 to 40°C for MPC90 and MPC70, respectively. In comparison, Lapčik et al. (2015) observed AIF of 26.5, 36.4 and 40.4° for whey, deproteinized whey powder, and SMP, respectively. Scanning electron micrographs and morphology of MPC powder particles showed that elevated storage temperature increased the particle-to-particle

interlocking due to higher intermolecular attractions among MPC powder particles (Scoville and Peleg, 1981; Anema et al., 2006; Nasser et al., 2017).

According to the Jenike's flow classification (Jenike, 1964), a powder is cohesive if its flow function is <4 and easy flowing if its flow function is within 4 to 10. All the MPC powders stored at 25 and 40°C showed a flow function of >4 . A significant decrease ($P < 0.05$) in FF values were observed with the increase in protein content and storage temperature, indicating potential flow issues. Significantly higher ($P < 0.05$) FF values were observed for MPC70 compared with MPC80 and MPC90, indicating that the powders will tend to become comparatively more cohesive with the increase in the protein content. However, with increase in storage temperature, the FF values slightly decreased, indicating that the MPC powders tend to become cohesive at higher temperature. Furthermore, the microstructure and morphological results showed that MPC70 had a higher proportion of regular-shaped particles and a comparatively smoother surface (Table 3), which confirms that MPC70 was more easy flowing or less cohesive than MPC80 and MPC90.

Solubility Index and Dissolution Behavior After Storage

The solubility index of MPC70, MPC80, and MPC90 after storage for 12 wk at 25°C was 88.2, 78.2, and 51.3%, respectively; after storage for 12 wk at 40°C the solubility index of MPC70, MPC80, and MPC90 was 38.8, 31.6, and 24.6%, respectively. The MPC powders stored at 25°C exhibited a higher solubility index as compared with powders stored at 40°C. Additionally, the solubility decreased with the increase in the protein content from 70 to 90%. Previous studies reported that the solubility of MPC powders is higher immediately after production and decreases with the increase in storage time and temperature (Anema et al., 2006; Fyfe et al., 2011), which were in agreement with the results from our study. The increase in protein content also negatively affected the solubility (Gazi and Huppertz, 2015). In addition to solubility index, the dissolution characteristics observed from FBRM results also confirmed that the solubilities of MPC powders were influenced by the protein content and storage temperature. The changes in the fine particle counts ($<10 \mu\text{m}$ chord length) for the MPC powders obtained from the FBRM are provided in Figure 5. It was observed that fine particle counts (Figure 5) increased at a higher rate for MPC powders stored at 25°C compared with the MPC powders stored at 40°C. Additionally, the slow disintegration of large powder particles into fine particles for the powders stored at 40°C further confirms the nega-

Figure 5. Changes in counts of fine particles ($<10\ \mu\text{m}$) after 12 wk of storage as obtained from focused beam reflectance measurement for the milk protein concentrate (MPC) powders (a) MPC70, (b) MPC80, (c) MPC90 stored at 25°C ; and (d) MPC70, (e) MPC80, (f) MPC90 stored at 40°C . MPC70, MPC80, and MPC90 are MPC with 70.3, 81.5 and 88.1% (wt/wt) protein content, respectively.

tive effect of storage temperature on the MPC powders. Similar observations were obtained in previous studies (Hauser and Amamcharla, 2016b; Gandhi et al., 2017). Storing the MPC powders for 12 wk at 40°C resulted in crosslinking networks at the surface of the MPC powders (Anema et al., 2006). These crosslinking networks include interactions between hydrophobic caseins and whey proteins, which hinders the hydration in the MPC powders (Anema et al., 2006; Uluko et al., 2016). As the protein content increased in MPC powders from 70 to 90% (wt/wt), the MPC90 showed more primary particle aggregates and exhibited more resistance to dispersing in water (Crowley et al., 2015). Likewise, with the increase in storage temperature, the protein-protein aggregation or association increased, as shown by lesser counts of fine particles as observed using the FBRM.

Compared with MPC90 and MPC80, MPC70 had better solubility, indicating the powder particles were less closely packed and thereby decreasing the chances for intermolecular reactions (Anema et al., 2006). Indeed, higher protein contents and higher storage temperature increased these intermolecular interactions leading to higher protein linkages, which consequently adversely affected the shear behavior of MPC powders. Interestingly, we have observed that changes in solubility index could be decently correlated with the shear test results (Table 2). We noted a positive correlation of UYS, cohesion, and FF to solubility index ($r = 0.78$,

0.72, and 0.86, respectively). The MPC powders with poor dissolution characteristics also exhibited poor shear behavior. Moreover, storage for 12 wk has induced a substantial rearrangement at the MPC powder particle surface (Nasser et al., 2017). Scanning electron micrographs (Figure 1) also showed the presence of surface roughness and packing of smaller particles in void spaces in the powder with 90% (wt/wt) protein content, corresponding to storage for 12 wk at 40°C . Overall, cohesion, AIF, and FF were significantly different ($P < 0.05$) for MPC70, MPC80, and MPC90 (after storage at both 25 and 40°C) samples, and results indicated that the flow behavior of MPC70 under consolidation would be better than MPC80 and MPC90. Thus, on prolonged storage, the cohesion forces developed between the MPC powder particles subsequently lead to a higher bulk cohesion (Fitzpatrick et al., 2007). The results also suggested that modification of MPC powder particle shape could improve the flow of MPC powders.

CONCLUSIONS

This study investigated the effect of protein content and storage temperature on particle morphology and flow properties of stored MPC powders (12 wk). Processing and subsequent storage resulted in MPC powders with varying physical and functional characteristics, which sequentially influenced the flowability.

The BFE and SpE of the MPC powders increased with an increase in protein content, indicating bulk flow challenges and higher energy requirement for making the powder flow at unconfined conditions. The inter-particle interactions and particle interlocking has influenced the flow behavior of MPC powders after prolonged storage at 25 and 40°C. Shear tests showed that the MPC powders were more cohesive with the increase in protein content and storage temperature. The shear flow properties of MPC powders were influenced by particle morphology and particle interlocking. Overall, the results indicated that differences in protein content and storage temperature affected the particle morphology and flow behavior of stored MPC powders.

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