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RESEARCH

Storable Cheese Curd—Effect of Milk Homogenization as a Pre-treatment and Freezing and Extrusion of Cheese Curd on Production of Pasta Filata Style Cheese

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Abstract

This study investigates the production of pasta filata style cheese from a storable, frozen intermediate material. Homogenization (2–16 MPa, single-staged) of milk (fat/protein = 0.9) was used as a tool to decrease fat globule size and consequently fat losses. Plasticization was achieved by using a single-screw extruder set up with double-jacketed hot water cycle. Non-frozen and frozen cheese curd as well as the extruded pasta filata style cheese pre-treated with different homogenization pressure was analyzed regarding the thermo-rheological properties. Fat and protein gain/loss during extrusion was evaluated by analyzing fat in dry matter (FDM) and protein in dry matter (PDM) before and after extrusion. Homogenization of cheese milk leads to a reduction of tan δ for thereof produced raw cheese curd material as well as the extruded products. Freezing and extrusion counteract the reduction of tan δ. A homogenization pressure of 8 MPa is sufficient to prevent fat losses during extrusion while still maintaining plasticization of the product for fresh and frozen material, respectively. The FDM after extrusion is 0.8% higher for fresh material and 4.9% higher for frozen material, which means that the fat concentrates during extrusion due to water loss. Moreover, there is no loss of PDM for all samples, regardless of the homogenization pressure. A combination of homogenization pressure, freezing, and extrusion leads to a plasticizable product without losses of fat and protein. Hence, frozen cheese curd can be used as a storable intermediate.

Keywords Extrusion · Cheese · Freezing · Rheology

Introduction

The shelf life of food can be prolonged through various preservation techniques such as heating, freezing, or drying (Zeuthen & Bøgh-Sørensen, 2003). In comparison to ripened cheese, unripened products like mozzarella or pasta filata style products have a limited shelf life at refrigerated temperatures. Recent studies investigated the potential to preserve raw cheese curd as intermediate product through freezing or drying. Thereafter, plasticization and texturization through extrusion technology are realized on demand (Frank et al., 2023). Advantageously, cheese production will become flexible in terms of time and location, and even countries without access to fresh raw milk will be able to produce fresh cheese products. However, upon freezing of cheese curd techno-functional properties change. Due to crystallization of water and fat a shift in plasticization temperature to lower temperatures occurs, this is prevented by homogenizing milk before cheese making. Homogenization involves a mechanical process that reduces the diameter of dispersed liquids and increases their surface area and a denser and more stable secondary fat globule membrane is created (Guinee et al., 2000). Within processing milk, it serves as a means to extend shelf life by reducing creaming through decreasing the fat globule size (Walstra, 1999). In cheese making, homogenization of cheese milk leads to a firmer cheese with less free oil formation compared to non-homogenized cheese milk (Rowney et al., 2003a) and is therefore a promising pre-treatment to produce cheese from storable cheese curd. Homogenized fat globules are covered with caseins and consequently behave like casein micelles (Cano-Ruiz & Richter,

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1997; Walstra & Oortwijn, 1982).Upon rennet-induced gel formation, these casein-covered fat globules are then integrated in the para-casein network which reduces curd contraction and syneresis. Thus, curd yield is increased due to a higher moisture content of the curd (Kelly et al., 2008; Vigneux et al., 2022). Curd firmness is reported to decrease with increased homogenization pressure while curd firming rate increased (Thomann et al., 2008a, b).

High-moisture extrusion (water content > 40%) is a well-suited and widely used technique to create proteinaceous, texturized food structures (Osen et al., 2015). Thereupon, vegetable proteins like soy or pea are processed into textured vegetable protein (TVP) as alternatives to meat (Grossmann & Weiss, 2021) or as fat replacer in dairy products (Tanger et al., 2021a, b). Moreover, animal proteins like casein-based products such as cheese curd are texturized into anisotropic structures. Thereby, extrusion combines thermal treatment with mechanical stress, inducing protein denaturation, plasticization (gel-sol transition), and molecular interactions. Finally, fibrous protein strands that are aligned in shear flow direction are created (Dekkers et al., 2016). The resulting textured cheese can be further processed by, e.g., injection of starter culture to initialize ripening (Kern et al., 2019a, 2020).

Traditionally, cooker-stretcher systems with one or two screws are used, where plasticization is reached by adding hot water (stretch water) or direct steam injection (Ma et al., 2013; Yu & Gunasekaran, 2005). Thereof, fat and protein losses occur, negatively impacting yield and resulting in economic losses (Bähler et al., 2016; Rowney et al., 2003b). In comparison to cooker-stretcher systems, a waterless single-screw extruder is operated without additional stretch water. Double-jacketed hot water cycles transfer heat into the product (plasticization), the screw speed controls the residence time (conveying), while a kneading section within the screw induces anisotropic structures (texturization). Moreover, such a set-up was shown to be a promising approach towards saving economic resources and reducing waste water (Kern et al., 2019b).

We hypothesize that homogenization and freezing are effective tools to create storable intermediate material for pasta filata style cheese production via single-screw extrusion. This enables on demand cheese production while maintaining protein and fat.

Material and Methods

Production of Cheese Curd

Cheese curd was produced according to Schmidt et al. (2022) with slight modifications. Raw milk was obtained from the Research Station Meiereihof (University of Hohenheim, Stuttgart, Germany) and pasteurized in the Dairy for Research and Training (University of Hohenheim, Stuttgart, Germany). The fat-to-protein ratio was standardized to 0.9 for each batch (500 kg). A one-stage homogenization (HL 1–400-TX; HST-Maschinenbau GmbH, Dassow, Germany) between 2, 4, 8, 12, and 16 MPa was performed at 65 °C. Unhomogenized, standardized milk was used as reference. After homogenization the milk was cooled to $\vartheta < 10$ °C and stored overnight. 0.02 g/100 g v/w of calcium chloride solution (40 g/100 g w/w calcium chloride dehydrate, AppliChem GmbH, Gatersleben, Germany) was added, and the pH was adjusted to 5.7 with lactic acid solution (9 g/100 g w/w, AppliChem GmbH, Gatersleben, Germany). The pH was re-adjusted to 5.7 after an equilibration time of 30 min due to the buffering capacity of milk. Coagulation was induced by adding 0.02 g/ g (v/w) chymosin (200 IMCU/mL; CHY-Max Plus, Christian Hansen, Lübeck, Germany) at 35 °C. After 15–17 min, the coagulum was cut into curd grains ($d = 3$–5 mm) and constantly stirred at 35 °C for 1 h before drainage. To avoid self-merging, curd grains were loosened by hand on drainage racks for about 30 min. Half of the curd grains were kept at 4 °C until extrusion (the same week); the other half was evenly spread on stainless steel plates, covered with plastic wrap, and frozen in a freezing chamber at − 18 °C. The frozen half was extruded between 1 day and 3 weeks after freezing.

Extrusion of Cheese Curd

Figure 1 presents the waterless single-screw extruder that was used for plasticization and texturization of cheese curd (Kern et al., 2019b). The extruder was run at a screw speed of 10 rpm while the cheese curd (non-frozen at $\vartheta = 4$ °C; frozen at $\vartheta = -10$ °C) was dosed into the filling funnel. The water temperature in the double-jacket hot water barrel was initially set to 63 °C. After 10 min of equilibration, a sample at the extruder outlet was taken, and the temperature was subsequently increased by 3 K up to a temperature of 78 °C. Samples were cooled and stored at 4 °C until further analysis.

Protein, Fat, and Dry Matter Content

Protein content was analyzed using a nitrogen analyzer (Dumatherm N Pro, Gerhardt GmbH & Co, KG, Königswinter, Germany) according to the Dumas method ISO 14891–2002 (International Dairy Federation, 2002). A protein conversion factor of 6.38 was used. Fat content was analyzed by means of van Guliks' method ISO 3432:2008 (International Dairy Federation, 2008). Dry matter was determined with the drying oven method ISO 5534:2004 (International Dairy Federation, 2004).

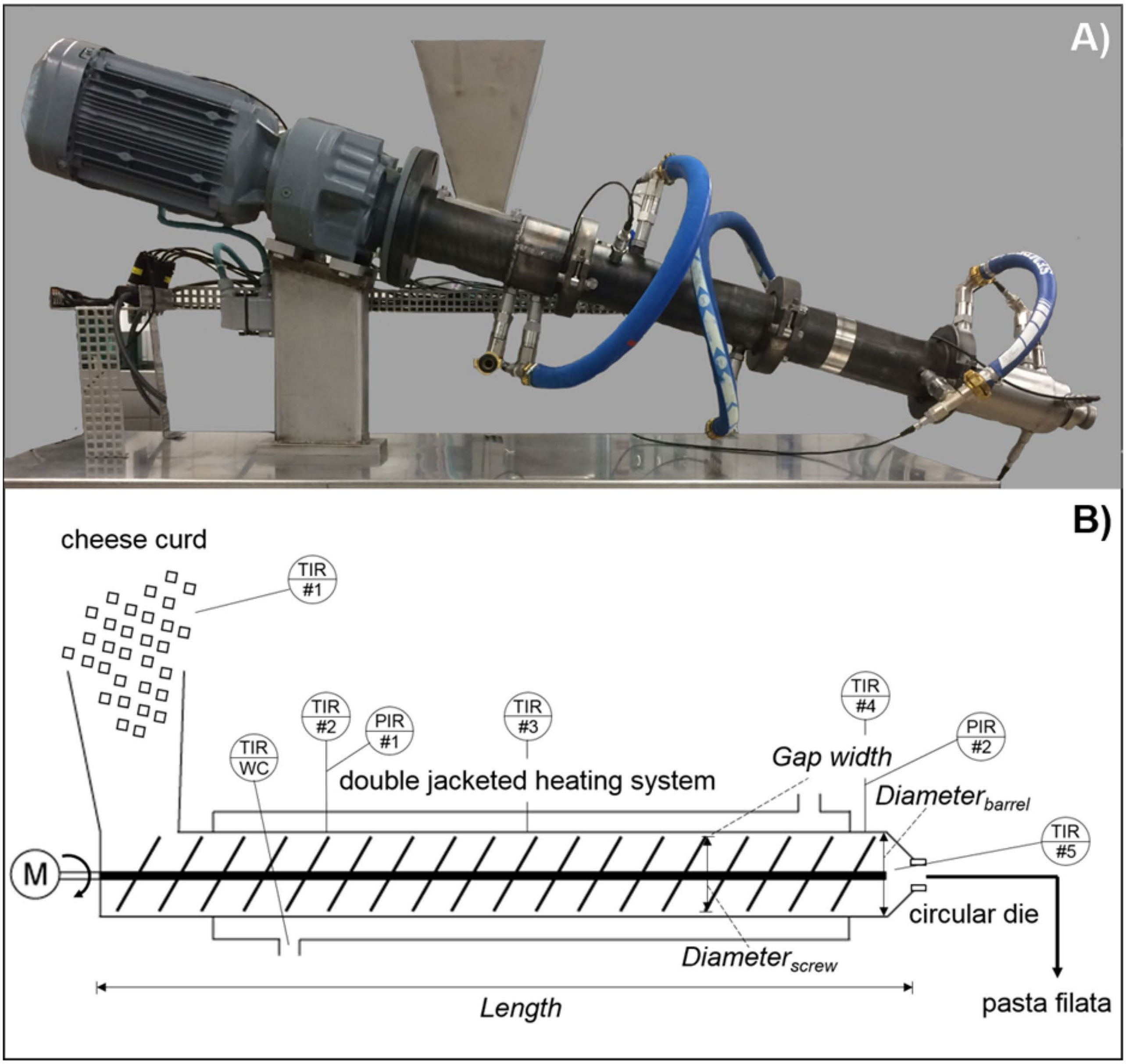


Fig. 1 Prototype of a waterless single screw extruder (Helix GmbH, Winnenden, Germany). **A** Setup including all temperature and pressure probes (not used in experiments), and heating cycle. **B** Schematic drawing with equipment dimensions, product flow, process (TIR-WC), and product temperature probes (TIR_WC) and pressure probes (PIR#1–2) (not used in experiments). Taken from Kern et al. (2019b) with permission from the authors

Moreover, the change in fat in dry matter (ΔFDM in g/100 g) after extrusion compared to fat in dry matter before extrusion was calculated with Eq. 1. The change of protein in dry matter (ΔPDM) was calculated analogously.

$$\Delta \mathrm{FDM}\,(\mathrm{g}/100\ \mathrm{g}) = \frac{\mathrm{FDM\ before\ extrusion\ -\ FDM\ after\ extrusion}}{\mathrm{FDM\ before\ extrusion}} \cdot 100(g/100g) \tag{1}$$

Thermo-Rheological Properties

By using a stress-controlled rheometer (MCR502, Anton Paar GmbH, Ostfildern, Germany), small amplitude oscillatory strain (SAOS) measurements with a temperature sweep were performed to analyze thermo-rheological properties of renneted casein-based gel particles before and after extrusion (Appendix) (Kern et al., 2018). The rheometer was equipped with a serrated plate-plate geometry (PP25/P2, $D = 25$ mm) and a Peltier temperature hood (H-PTD). The temperature was increased from 20 to 80 °C with a heating rate of 3 K/min at constant oscillation. Frequency was set to 1.5 Hz and the amplitude to 0.02%. The normal force before the start of the measurement was 1 N. Circular samples with a diameter of 25 mm and height of 2.5 mm were taken from renneted casein-based gel particles (self-pressure merged) or samples after extrusion and placed between the plate-plate geometry. Drying out of the samples during measurement was prevented by coating the samples' edge with silicone oil. Elastic (G') and viscous (G'') modulus were measured. Plasticization temperature is obtained as soon as G' equals G'', $(G'G'', \tan\ \delta = \frac{G''}{G'} = 1)$. Moreover, the maximum loss tangent $\tan \delta$ was measured which expresses the flowability (Guinee et al., 1999).

Fat Particle Size Measurement

The particle size distribution was analyzed by static light scattering with a LS13320 (Beckmann Coulter, Brea, California, USA). Samples were diluted 1:1 with deionized water. 70–100 µL of the sample was filled in the measuring chamber. The pump speed was set to 20%. The refractive index for distilled water as dispersing medium was set to 1.33 and to 1.46 for the sample representing fat globules (Michalski et al., 2001). All samples were measured in duplicate.

Confocal Laser Scanning Microscopy (CLSM)

The dye Fat Red (V03-01136) (Dyomics GmbH, Jena, Germany) for visualization of fat was used to stain the samples. Therefore, 15 µL of Fat Red was pipetted on a microscope slide, and fresh cut samples were placed on top. After staining, the samples were left for at least 15 min before measurement. Microscopic images were taken by using a confocal laser scanning microscope (CLSM, Zeiss LSM 900, Carl Zeiss AG, Oberkochen, Germany) with a laser at 645–700 nm at room temperature. ZEN microscopy software (Carl Zeiss AG, Oberkochen, Germany) was used for image analysis.

Statistical Analyses

Experiments without homogenization, at 8 MPa and 16 MPa, were performed in double determination as they represent the lowest, middle, and highest homogenization pressure. The remaining experiments (2, 4, and 12 MPa) were performed in single determination. Analytical analyses have been performed in duplicate (thermo-rheological measurements and fat content) or triplicate (protein content and dry matter). For all analyses, the arithmetic mean and standard deviation (triplicate) or range (duplicate) were calculated.

OriginPro 2023 (OriginLab Corporation, Northampton, MA, USA) and Microsoft Power Point 2019 (Microsoft, Redmond, Washington, USA) were used to create figures.

Results and Discussion

Homogenization of Cheese Milk: Effect on Fat Globule Size

Fat particle Size Measurement

In this study, homogenization is used as a tool to reduce fat globule size as these casein-covered fat globules are less susceptible to fat losses.

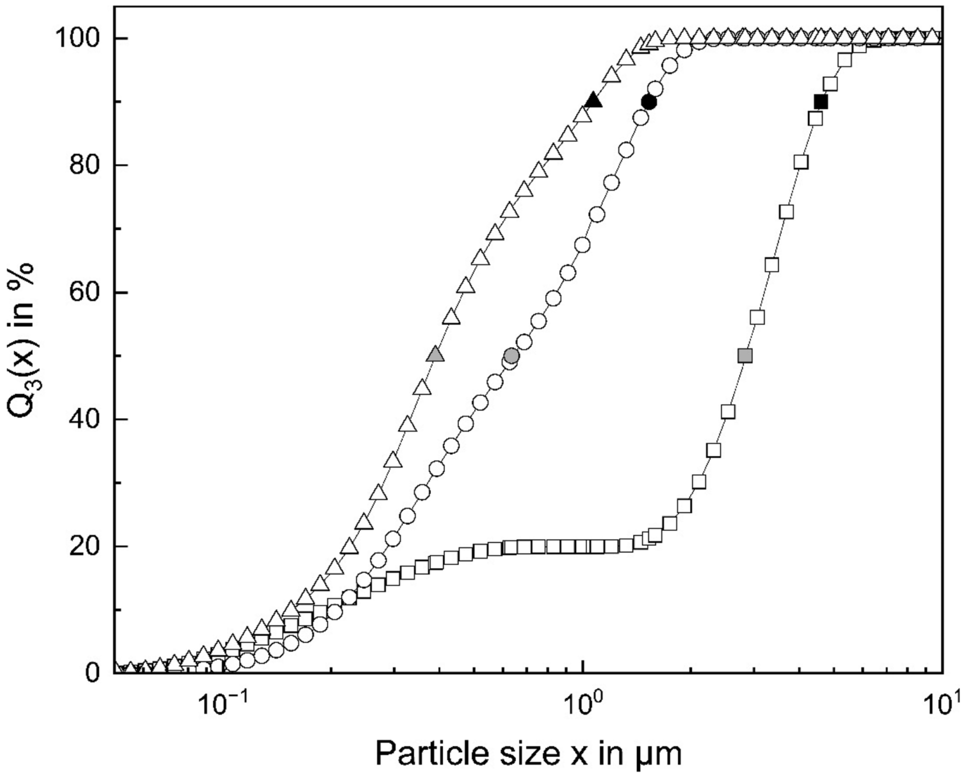
Figure 2 shows the cumulative fat globule size distribution of homogenized cheese milk at certain homogenizing pressures. For simplicity and clarity, only the fat globule size distributions at homogenizing pressures of 8 and 16 MPa as well as unhomogenized milk are shown. However, the authors are aware that equal homogenization pressures at different homogenizers may lead to different results. The unhomogenized sample had a $d_{90,3}$ of 4.6 µm (black symbols) and a $d_{50,3}$ of 2.8 µm (gray symbols). The sample homogenized at 8 MPa had a $d_{90,3}$ of 1.5 µm and a $d_{50,3}$ of 0.6 µm and at 16 MPa a $d_{90,3}$ of 1.1 µm and a $d_{50,3}$ of 0.4 µm.

Microscopic Images of Cheese Curd

CLSM images from fresh and frozen cheese curd, unhomogenized and at 8 and 16 MPa homogenized, are shown in Fig. 3 at a 20 × magnification to illustrate the microstructure of the cheese curd. The fluorophore fat red enabled to stain the fat phase of the samples.

The fat globule size in cheese curd decreases with increasing homogenization pressure which is in line with the results from the cumulative particle size distribution of unhomogenized and homogenized milk visualized in Fig. 2. Moreover, in the unhomogenized cheese curd, fat aggregates and clusters are visible while the fat is evenly distributed when homogenized. However, when the cheese curd was frozen and thawed, it appears that larger fat globules are formed again due to breaking of structure and reformation of fat globules. Concerning the performed trials, the denser network of fat globules in the samples made with homogenized milk indicates that fat globules now act as active fillers indicating they could influence protein plasticization. This is in contrast to cheese curd made with unhomogenized milk, where fat globules act as passive fillers (Schenkel et al., 2013). Thus, the role of fat and therefore extrusion of cheese curd may be different.

Fig. 2 Cumulative particle size distribution $Q_3(x)$ of non-homogenized (squares), homogenized at 8 MPa (circles), and homogenized at 16 MPa (triangles) cheese milk ($f/p = 0.9$) in dependency of the particle size x. $d_{50,3}$ and $d_{90,3}$ are marked with gray and black colored symbols, respectively



Single-Screw Extrusion of Differently Pre-treated Cheese Curd

Influence of Freezing on Extruder Outlet Temperature

In order to achieve a homogeneous and textured pasta filata style cheese product, two conditions must be fulfilled: i) the material temperature must exceed the gel-sol transition temperature to enable product plasticization, and ii) mechanical stress is necessary to create anisotropic structures (Kern et al., 2019b). The latter is fulfilled within the utilized single-screw extruder setup, as demonstrated in previous studies (Kern et al., 2020; Schmidt et al., 2022).

To obtain gel-sol transition of the cheese curd material, a temperature of approximately 60 °C and higher, depending on the pre-treatment during cheese curd production, must be obtained (Kern et al., 2019b). To verify this, the temperature of the extruded cheese curd product at the end of the extruder is shown in Fig. 4. It is measured with a PT100 temperature sensor as depicted in Fig. 1.

Since the temperature of the frozen cheese curd before extrusion is approximately $\vartheta_{\text{frozen}} = -10$ °C (compare: $\vartheta_{\text{fresh}} = 4$ °C for fresh curd), the heating profile of fresh and frozen cheese curd differs. Additionally, there is a phase transition during heating from frozen to non-frozen which requires energy (Kessler, 1981). However, the end temperatures of both, the fresh and frozen material, are approximately equal. It can be concluded that the length of the extruder and consequently the residence time of 7.3 min (Kern et al., 2019b) are sufficient to heat up the material to the desired temperature (above gel-sol transition temperature) regardless of its starting condition.

Calculation of Heat Transfer Coefficient with Fresh and Frozen Material

With knowledge of the heat transfer coefficient k in W/(m^{2}K) between the barrel and the extruded material extruder processes are assessed and designed. This becomes essential within considerations for scaling up (Levine & Rockwood, 1986; Mohamed et al., 1988). Moreover, phase transition, i.e., from ice to water plays an important role as latent heat of fusion is required. In the following part k is calculated for unhomogenized fresh and frozen material at the lowest and highest extruder barrel temperature (63 and 78 °C), respectively.

Following assumptions were made:

- Transient heating of temperature-equalized liquid goods
- Water inlet (ϑ_{E1} in °C) and outlet (ϑ_{A1} in °C) temperature at the extruder barrel are approximately equal ($\vartheta_{E1} \approx \vartheta_{A1}$) due to the high volume flow in the hot water cycle
- The latent heat of melting $r_{\text{sch},w}$ in kJ/kg of fat is neglected
- At a material temperature of cheese curd $\vartheta_{CC} \leq 0$ °C all water is frozen; at $\vartheta_{CC} > 0$ °C all water is liquid

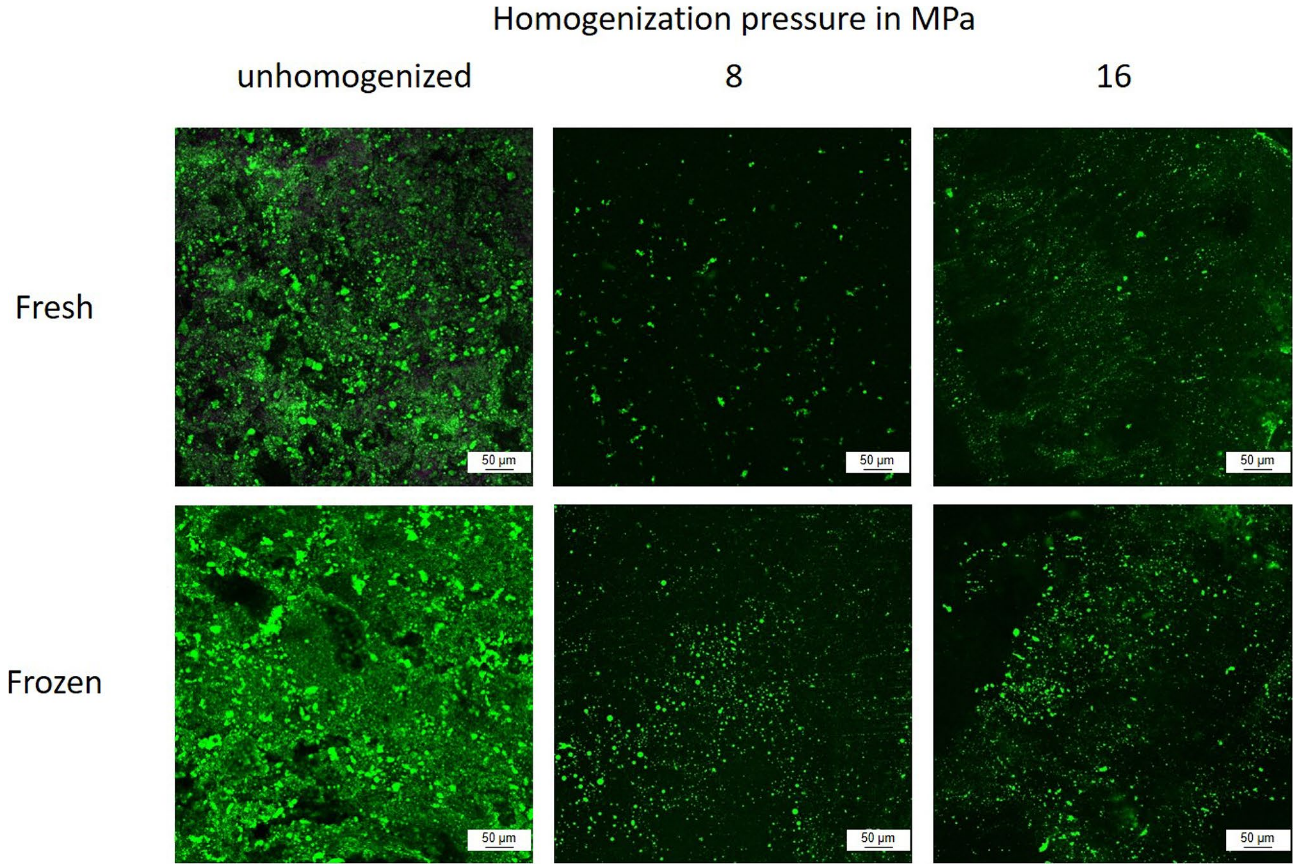


Fig. 3 CLSM images of cheese curd (before extrusion) either unhomogenized or homogenized at 8 or 16 MPa, fresh and frozen. Fat was dyed with Fat Red (green color)

The general formula for the heat flow $\dot{Q}$ in kJ/h is given in Eq. 2 (Kessler, 1981).

$$\dot{Q} = \dot{m} \cdot c_p \cdot \Delta\vartheta = k \cdot A \cdot \Delta\vartheta_m \tag{2}$$

with $\dot{m}$ the mass flow in kg/h, c_p the specific heat capacity in kJ/(kgK), $\Delta\vartheta$ the temperature difference between the incoming material and the outcoming product in K, k the heat transfer coefficient W/(m^{2}K), A the heat transfer area in m^2 and $\Delta\vartheta_m$ the mean logarithmic temperature difference between two mass flows for parallel flow in K.

The values for c_p, $r_{\mathrm{sch},w}$, A, and $\dot{m}$ that were used for the calculations are given in Table 1.

The mean logarithmic temperature difference between the mass flows "water" in the extruder barrel and "material" in the extruder $\Delta\vartheta_m$ in K for concurrent flow is calculated according to Eq. 3 (Kessler, 1981).

$$\Delta\vartheta_m = \frac{\left(\vartheta_{E1} - \vartheta_{E2}\right) - \left(\vartheta_{A1} - \vartheta_{A2}\right)}{ln\frac{\vartheta_{E1} - \vartheta_{E2}}{\vartheta_{A1} - \vartheta_{A2}}} \tag{3}$$

where ϑ_{E1} is the inlet water temperature in the extruder barrel in K, ϑ_{A1} is the outlet water temperature in the extruder barrel in K, ϑ_{E2} is the inlet material temperature in K, and ϑ_{A2} is the outlet product temperature in K.

By inserting Eq. 3 into Eq. 2 k for extrusion of fresh cheese curd is calculated and displayed in Table 2.

For the calculations of k for extrusion of frozen cheese curd three steps have to be considered. (1) warming the frozen material (dry matter plus ice) to 0 °C, (2) melting the ice (by considering $r_{\mathrm{sch,w}}$) and (3) warming the non-frozen material to end product temperature. Thereof Eq. 4 was used and the results are displayed in Table 2.

$$k = \frac{\dot{m}_i \cdot c_{p,i} \cdot \Delta\vartheta_i + \dot{m}_{dm} \cdot c_{p,dm} \cdot \Delta\vartheta_{dm} + \dot{m}_w \cdot r_{\mathrm{sch},w} + \dot{m}_{\mathrm{prod}} \cdot c_{p,\mathrm{prod}} \cdot \Delta\vartheta_{\mathrm{thawed}}}{A \cdot \Delta\vartheta_m} \tag{4}$$

where k is the heat transfer coefficient W/(m^{2}K), $\dot{m}_i$ is the mass flow of the amount of ice in the product in kg/h, $c_{p,i}$ is the specific heat capacity of ice in kJ/(kgK), $\Delta\vartheta_i$ is the temperature difference between the ice in the incoming material and 0 °C in K, $\dot{m}_{\mathrm{dm}}$ is the mass flow of the amount of

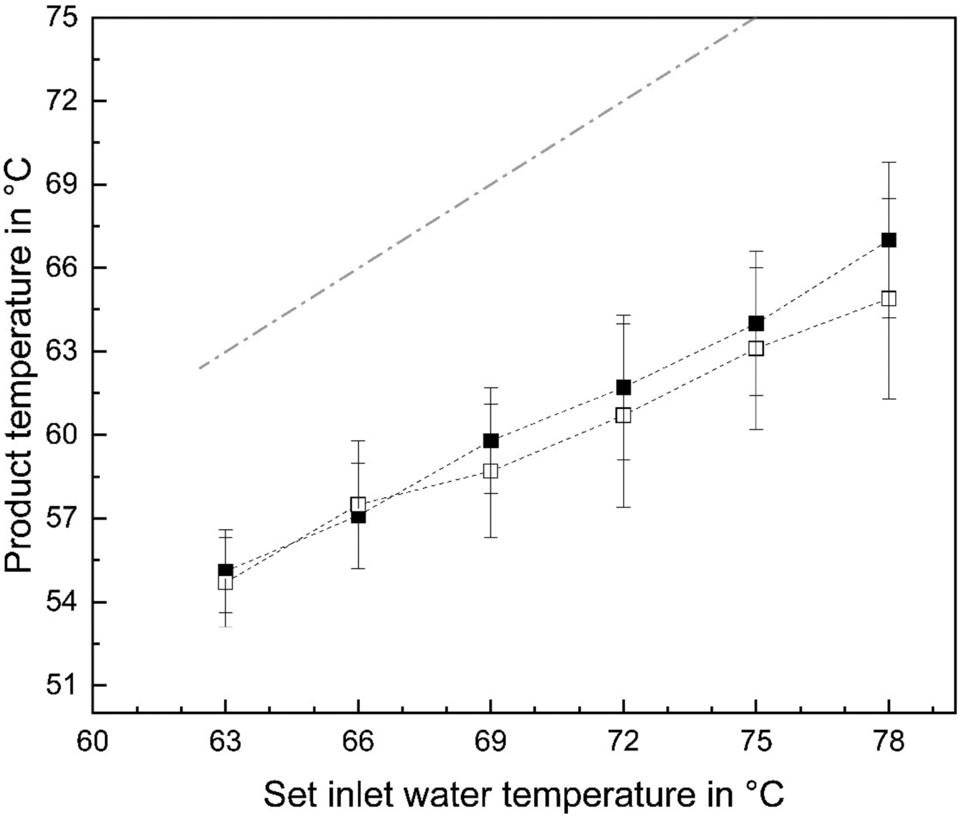


Fig. 4 Product temperature of cheese measured at the outlet of the single-screw extruder plotted over the set inlet water temperature realized by a double-jacketed water cycle. Black symbols represent the fresh cheese curd ($\vartheta_{\text{inlet}}=4$ °C) while white symbols represent frozen cheese curd ($\vartheta_{\text{inlet}}=-10$ °C). The dashed line represents ideal conditions where set inlet water temperature and product temperature are equal

dry matter in the product in kg/h, $c_{p,\text{dm}}$ is the specific heat capacity of the dry matter of the product in kJ/(kgK), $\Delta\vartheta_{\text{dm}}$ is the temperature difference between the dry matter in the incoming material and 0 °C in K, $\dot{m}_w$ is the mass flow of the amount of water in the product in kg/h, $r_{\text{sch},w}$ the latent heat of fusion in kJ/kg, $\dot{m}_{\text{prod}}$ is the mass flow of the product in kg/h, $c_{p,\text{prod}}$ the specific heat capacity of the product in kJ/(kgK), $\Delta\vartheta_{\text{thawed}}$ is the temperature difference between 0 °C and the outcoming product in K, A is the area in m^2, and $\Delta\vartheta_{\text{m}}$ is the mean logarithmic temperature difference between the mass flows "water" in the extruder barrel and "material" in the extruder for parallel flow in K.

For fresh cheese curd extruded at 63 °C and at 78 °C k is approximately equal (132 W/(m^2 K) and 126 W/(m^2 K), respectively). In comparison, k for frozen cheese curd is almost double the value with 252 W/(m^2 K) at 63 °C and 231 W/(m^2 K) at 78 °C. This is due to the latent heat of the phase transition from ice to water which explains the still approximately equal product temperatures at the outlet of the single-screw extruder as shown in Fig. 4 and discussed in the "Influence of Freezing on Extruder Outlet Temperature" section.

Literature data regarding k in extruder systems, especially in the food sector, are scarce (Chiruvella et al., 1995; Karwe & Godavarti, 1997). Mohamed and Ofoli (1989) used a twin-screw extruder where screw speed and mass flow were higher than in this study (150 1/min to 450 1/min and 33 to 60 kg/h, respectively). Moreover, soy polysaccharide

Table 1 Values for specific heat capacity c_p, latent heat of fusion $r_{\text{sch,w}}$, area A, and mass flow $\dot{m}$ that are used to calculate the heat transfer coefficient k

Description	Variable	Value	Unit	Reference
Specific heat capacity product	$c_{p,\text{prod}}$	2.90	kJ/(kg K)	Kern et al. (2020)
Specific heat capacity water	$c_{p,w}$	4.20	kJ/(kg K)	Kessler (1981)
Specific heat capacity ice	$c_{p,i}$	2.00	kJ/(kg K)	Kessler (1981)
Specific heat capacity dry matter	$c_{p,\text{dm}}$	1.57	kJ/(kg K)	Calculated[a]
Latent heat of fusion ice	$r_{\text{sch},w}$	334	kJ/kg	Kumano et al. (2007)
Area extruder barrel	A	0.18	m^2	Kern et al. (2020)
Mass flow product	$\dot{m}_{\text{prod}}$	15	kg/h	Schmidt et al. (2022)

[a] Calculated according to Kern et al. (2020), the specific heat capacity of the dry matter has been calculated by multiplying the specific heat capacity of the product with the dry matter content of the material

Table 2 Heat transfer coefficient k calculated for fresh and frozen starting material at extrusion temperatures of 63 and 78 °C

Material state	Extruder barrel temperature ϑ_{E1} in °C	k in W/(m^2 K)
Fresh	63	132
	78	126
Frozen	63	252
	78	231

was used as food material which results in a different specific heat capacity. However, the obtained values ($k = 191$ to 768 W/(m^2 K)) are in the same magnitude as in our study. Thus, although two different systems were used, the values are still comparable and confirm our results.

Rheological Behavior of Cheese Curd

The thermo-rheological behavior of the cheese curd in dependency of the homogenization pressure and pretreatment is shown in Fig. 5. When exceeding a value of 1 ($\tan\delta \geq 1$), gel-sol transition and thus plasticization of the cheese is obtained. Temperatures where $\tan\delta = 1$ as well as trend lines for $\tan\delta = 0.9$ and $\tan\delta = 1.1$ are given in dependency of the homogenization pressure. Moreover, the temperature at $\tan\delta_{max}$ at different homogenization pressures is shown. In addition, $\tan\delta_{80\,°C}$ at the first homogenization

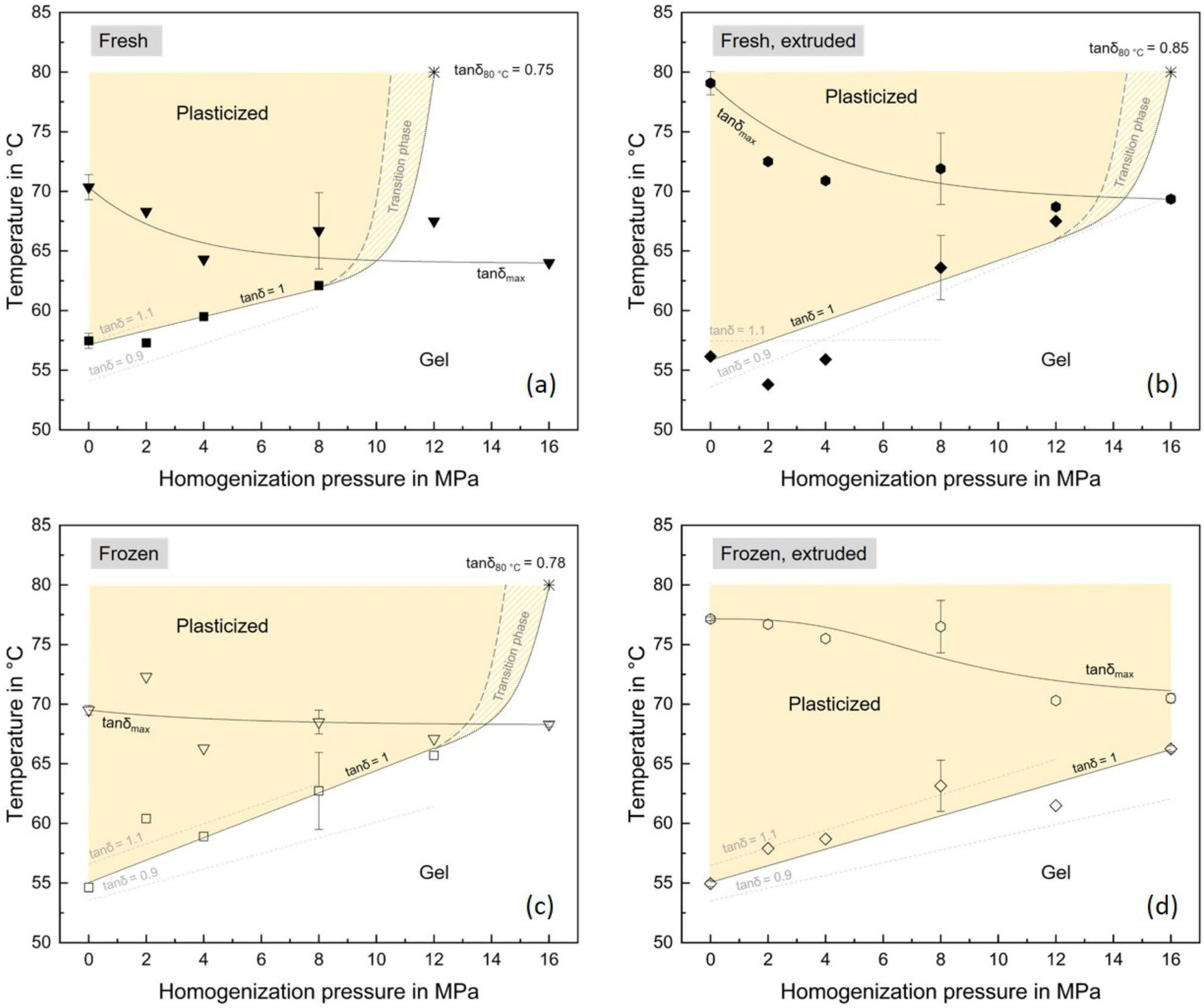


Fig. 5 Rheological behavior of cheese curd **a** fresh, **b** fresh and extruded at 69 °C, **c** frozen, and **d** frozen and extruded at 69 °C. The temperatures at which $\tan\delta_{max}$ as well as $\tan\delta = 0.9$, $\tan\delta = 1.0$, and $\tan\delta = 1.1$ are reached in dependency of the homogenization pressure are shown. $\tan\delta$ at 80 °C is given to better assess the transition phase between gel and plasticized product. The region of plasticized product is shown in yellow. The transition phase is estimated and shows the region where plasticization is supposed to occur

pressure where plasticization did not take place is shown to assess the region of transition phase for separation of the region of gel and plasticized product better. It is important to note that the transition phase is only a prediction of where the plasticization happens.

As displayed in Fig. 5 the plasticization temperature increases with increasing homogenization pressure and eventually levels out with the curve of tan δ_{max} (ϑ). Above a certain homogenization pressure plasticization and thus texturization of the cheese curd does not occur anymore. This is caused by a denser network where fat globules act as active fillers due to homogenization and thus hinder the plasticization of protein. Contrarily, fat globules in unhomogenized milk act as passive fillers (Schenkel et al., 2013) and do not influence plasticization of protein. The highest homogenization pressure where plasticization still occurs is at 8 and 12 MPa for fresh and frozen cheese curd before extrusion, respectively (see Fig. 5a, c). In comparison to fresh cheese curd tan $\delta = 1$ of frozen cheese curd is generally measurable at higher homogenization pressures. This is explained due to structural damage induced by ice and fat crystals (Everett & Auty, 2008; Kuo et al., 2003). When producing storable frozen cheese curd, higher homogenization pressures have to be applied to obtain comparable techno-functional properties. However, it has to be considered that the cheese yield decreases with increasing homogenization pressure (Escobar et al., 2011) and at high pressure renneting is even hindered.

However, we assume that extrusion partly counteracts the effect of homogenization and tan δ_{max} increases again (Fig. 5b, d). After extrusion, plasticization is still achieved when homogenized at 12 MPa (fresh material) while the frozen material is still plasticizable at 16 MPa. Since the screw speed is set to 10 rpm, the possibility of exceeding the critical shear rate which could lead to an increase in tan δ_{max} is negated in this setup (Kern et al., 2019b). One possibility could be that fat melts, re-aggregates into larger clusters, and is therefore no longer available as an active filler and the protein can plasticize again. However, more studies are needed in this regard. In the following chapter, the loss or gain of FDM and PDM is investigated and it is discussed which homogenization pressures are necessary to decrease fat losses after extrusion.

Effect of Extrusion on Fat and Protein in Dry Matter

The gain/loss of fat in dry matter (FDM) and protein in dry matter (PDM) due to extrusion of differently pre-treated fresh and frozen cheese curd are shown in Fig. 6. The data shown in the figure were measured with extruded cheese at 69 °C and were exemplarily selected. The values for the other temperatures show similar trends. Positive values indicate a loss of FDM or PDM and negative values a gain in FDM or PDM.

It can be seen that PDM increases independently upon homogenization of milk. Moreover, no trend in the gain of PDM over homogenization pressure is discernible. Therefore, it can be concluded that protein is not present in the leaking fluid and is concentrated upon extrusion. However, there is a loss of FDM when unhomogenized or homogenized at pressures up to 4 MPa. Consequently,

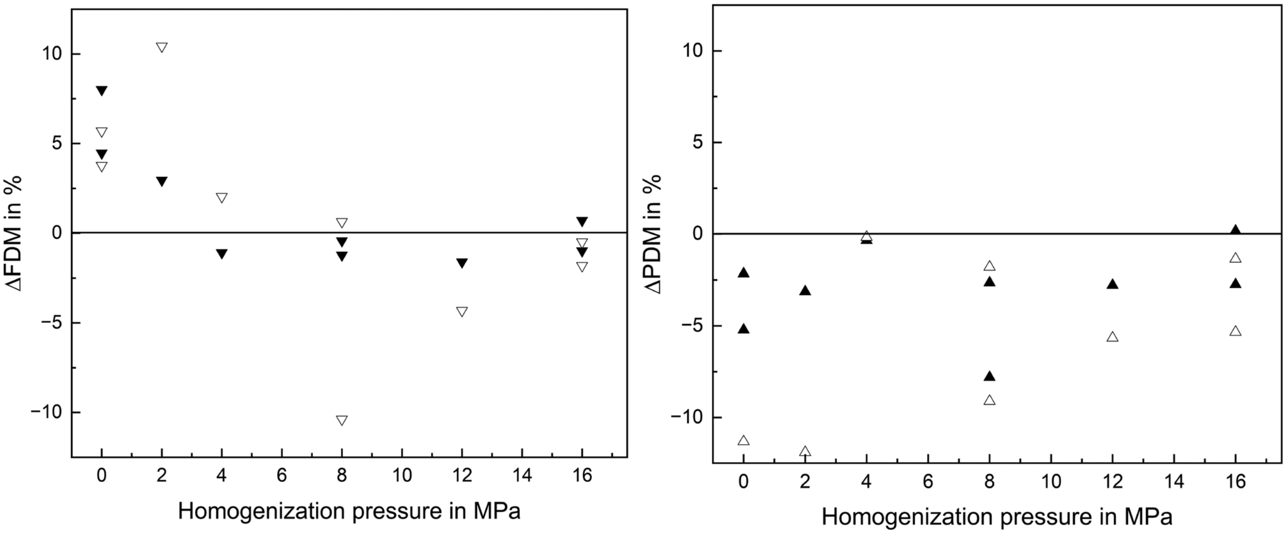


Fig. 6 Left: difference of FDM of extruded cheese curd (at 69 °C) in dependency of the homogenization pressure. Positive values indicate fat decrease during extrusion, negative values indicate fat increase during extrusion. Right: difference of PDM of extruded cheese curd (at 69 °C) in dependency of the homogenization pressure. Positive values indicate protein decrease during extrusion, negative values indicate protein increase during extrusion. Black squares: fresh cheese curd; white squares: frozen cheese curd. Note: 0 MPa means unhomogenized. Two identical symbols at one homogenization pressure represent a double determination

homogenization ≥ 8 MPa avoids FDM loss and fat concentrates in the dry matter upon extrusion. Independently of freezing the cheese curd, results were similar.

In general, the extent of protein and/or fat loss depends on various factors such as renneting temperature (Hussain et al., 2012), concentration of protein and dry matter content by means of ultrafiltration (Farkye & Yim, 2003), or stretching temperature and mixing conditions (Banville et al., 2016). During the production of traditional pasta filata cheese, the addition of hot stretch water to cooker-stretcher systems causes fat and protein losses (Bähler et al., 2016; Mayes & Sutherland, 2002; Rowney et al., 2003b). Additionally, the stretch water needs to be recycled at great expense. One method to reduce costs is recycling the stretch water by means of nanofiltration. The resulting permeate can be reused as stretch water (Faccia et al., 2021). Costs are nevertheless incurred, which are circumvented with the extruder system presented, as no stretch water is added in the process and FDM losses are comparably low even without homogenization. Mild homogenization further decreases losses and even results in concentration of fat due to evaporation of water.

Conclusion

This study demonstrated that raw frozen cheese curd serves as storable intermediate material upon producing pasta filata style cheese. Plasticization and texturization are realized through single-screw extrusion technology on demand. Thus, pasta filata style cheese production is more flexible regarding time, place, and production. Temperature measurements of the product at the end of the extruder showed that frozen starting material is heated up sufficiently and comparable to fresh starting material. This is explained with the calculated higher heat transfer coefficient for frozen goods.

In detail, homogenization of cheese milk leads to a reduction of tan δ for thereof produced raw cheese curd material as well as the extruded products. Calculation of the heat transfer coefficient k ($k = 126$–252 W/(m^2 K) for fresh and frozen material, respectively) helps to examine and compare different extrusion processes. However, freezing and extrusion counteract the reduction of tan δ and influence the plasticization temperature of the cheese curd. Plasticization of cheese is obtained until 12 and 16 MPa for extruded fresh and frozen product, respectively. There was no loss of FDM at a homogenizing pressure of 8 MPa for both, frozen and non-frozen product. Moreover, the extrusion process does not lead to losses in PDM even for unhomogenized material.

In future work, microwave extrusion as a new processing method will be focused. Microwave heating has potential as it heats up quickly and volumetrically. The length of the extruder could be decreased, and thereof, it might be a viable technology to scale the extrusion process up.

Appendix

Table 3 tan δ_{max} of fresh cheese curd before and after extrusion at different temperatures. Experiments with standard deviation (range) were performed in duplicate

$\vartheta_{\text{extruder, set}}$ in °C	tan δ_{max} (homogenization pressure in MPa)					
	0	2	4	8	12	16
Before extrusion	1.77 ± 0.03	1.49	1.11	0.99 ± 0.07	0.86	0.81 ± 0.04
63	2.10 ± 0.19	1.88	1.53	1.12 ± 0.08	1.01	0.87 ± 0.05
66	2.00 ± 0.15	1.90	1.52	1.13 ± 0.1	1.00	0.85 ± 0.05
69	2.07 ± 0.25	1.87	1.50	1.13 ± 0.06	1.01	0.87 ± 0.06
72	2.07 ± 0.19	1.91	1.44	1.13 ± 0.14	1.00	0.86 ± 0.09
75	2.10 ± 0.23	1.86	1.32	1.10 ± 0.07	0.99	0.86 ± 0.07
78	2.10 ± 0.33	1.78	1.37	1.10 ± 0.04	0.95	0.85 ± 0.1

Table 4 tan δ_{max} of frozen cheese curd before and after extrusion at different temperatures. Experiments with standard deviation (range) were performed in duplicate

$\vartheta_{\text{extruder, set}}$ in °C	tan δ_{max} (homogenization pressure in MPa)					
	0	2	4	8	12	16
Before extrusion	1.84 ± 0.01	1.51	1.31	1.12 ± 0.16	1.01	0.88 ± 0.03
63	2.20 ± 0.16	1.65	1.64	1.25 ± 0.07	1.23	1.05 ± 0.01
66	2.15 ± 0.09	1.65	1.69	1.35 ± 0.23	1.20	1.07 ± 0.04
69	2.17 ± 0.01	1.78	1.66	1.32 ± 0.22	1.20	1.06 ± 0.01
72	2.27 ± 0.02	1.76	1.63	1.30 ± 0.30	1.21	1.05 ± 0.01
75	2.19 ± 0.20	1.81	1.13	1.27 ± 0.06	1.16	1.08 ± 0.05
78	2.25 ± 0.02	1.83	1.13	1.30 ± 0.20	1.15	1.07 ± 0.01

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Author Contribution Florian Schmidt and Britta Graf defined the general objective and the topics to be discussed. Florian Schmidt and Britta Graf conducted the experiments. Florian Schmidt, Britta Graf, and Jörg Hinrichs discussed the results. Florian Schmidt, Britta Graf and Jörg Hinrichs wrote the manuscript. All authors contributed to the final review of the manuscript.

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Data Availability No datasets were generated or analyzed during the current study.

Declarations

Conflict of Interest The authors have no relevant financial or non-financial interests to disclose.

References

Bähler, B., Ruf, T., Samudrala, R., Schenkel, P., & Hinrichs, J. (2016). Systematic approach to study temperature and time effects on yield of pasta filata cheese. *International Journal of Dairy Technology, 69*(2), 184–190. https://doi.org/10.1111/1471-0307.12248

Banville, V., Chabot, D., Power, N., Pouliot, Y., & Britten, M. (2016). Impact of thermo-mechanical treatments on composition, solids loss, microstructure, and rheological properties of pasta filata–type cheese. *International Dairy Journal, 61*, 155–165. https://doi.org/10.1016/j.idairyj.2016.05.004

Cano-Ruiz, M. E., & Richter, R. L. (1997). Effect of homogenization pressure on the milk fat globule membrane proteins. *Journal of Dairy Science, 80*(11), 2732–2739. https://doi.org/10.3168/jds.S0022-0302(97)76235-0

Chiruvella, R. V., Jaluria, Y., & Abib, A. H. (1995). Numerical simulation of fluid flow and heat transfer in a single-screw extruder with different dies. *Polymer Engineering & Science, 35*(3), 261–273.

Dekkers, B. L., Nikiforidis, C. V., & Van Der Goot, A. J. (2016). Shear-induced fibrous structure formation from a pectin/SPI blend. *Innovative Food Science & Emerging Technologies, 36*, 193–200. https://doi.org/10.1016/j.ifset.2016.07.003

Escobar, D., Clark, S., Ganesan, V., Repiso, L., Waller, J., & Harte, F. (2011). High-pressure homogenization of raw and pasteurized milk modifies the yield, composition, and texture of queso fresco cheese. *Journal of Dairy Science, 94*(3), 1201–1210. https://doi.org/10.3168/jds.2010-3870

Everett, D. W., & Auty, M. A. (2008). Cheese structure and current methods of analysis. *International Dairy Journal, 18*(7), 759–773. https://doi.org/10.1016/j.idairyj.2008.03.012

Faccia, M., Natrella, G., Loperfido, P. P., Gambacorta, G., & Cicco, G. (2021). Quality characteristics of mozzarella cheese manufactured with recycled stretchwater. *LWT, 147*, 111554. https://doi.org/10.1016/j.lwt.2021.111554

Farkye, N. Y., & Yim, B. (2003). Use of dry milk protein concentrate in pizza cheese manufactured by culture or direct acidification. *Journal of dairy science, 86*(12), 3841–3848. https://doi.org/10.3168/jds.S0022-0302(03)73991-5

Frank, J., Schlitter, M., Hinrichs, J., & Kohlus, R. (2023). Fluidized bed drying of dairy gel granules supported by in-line monitoring of the water content. *Drying Technology*. https://doi.org/10.1080/07373937.2023.2216774

Grossmann, L., & Weiss, J. (2021). Alternative protein sources as technofunctional food ingredients. *Annual Review of Food Science and Technology, 12*, 93–117.

Guinee, T. P., Auty, M. A., & Mullins, C. (1999). Observations on the microstructure and heat-induced changes in the viscoelasticity of commercial cheeses. *Australian Journal of Dairy Technology, 54*(2), 84.

Guinee, T. P., Auty, M. A. E., Mullins, C., Corcoran, M. O., & Mulholland, E. O. (2000). Preliminary observations on effects of fat content and degree of fat emulsification on the structure-functional relationship of Cheddar-type cheese. *Journal of Texture Studies, 31*(6), 645–663. https://doi.org/10.1111/j.1745-4603.2000.tb01026.x

Hussain, I., Yan, J., Grandison, A. S., & Bell, A. E. (2012). Effects of gelation temperature on Mozzarella-type curd made from buffalo and cows' milk: 2. Curd yield, overall quality and casein fractions. *Food chemistry, 135*(3), 1404–1410. https://doi.org/10.1016/j.foodchem.2012.05.110

International Dairy Federation. (2002). Milk and milk products — Determination of nitrogen content (ISO, 14891:2002).

International Dairy Federation. (2004). Cheese and processed cheese — Determination of the total solids content (ISO, 5534:2004).

International Dairy Federation. (2008). Cheese — Determination of fat content (ISO, 3432: 2008).

Karwe, M. V., & Godavarti, S. (1997). Accurate measurement of extrudate temperature and heat loss on a twin-screw extruder. *Journal of Food Science, 62*(2), 367–372. https://doi.org/10.1111/j.1365-2621.1997.tb04003.x

Kelly, Alan L., Huppertz, Thom, & Sheehan, Jeremiah J. (2008). Pretreatment of cheese milk: principles and developments. *Dairy Science and Technology, 88*(4–5), 549–572. https://doi.org/10.1051/dst:2008017

Kern, C., Stefan, T., Kügler, P., & Hinrichs, J. (2019a). New technology platform with injection of starter cultures for the production of sliced cheese. *DMW die Milchwirtschaft, 10*(2), 59–62.

Kern, C., Scharfe, M., & Hinrichs, J. (2020). Texturization of renneted casein-based gel particles by sheet die extrusion: Mechanical properties and numerical analysis of flow characteristics. *Journal of Food Engineering, 278*, 109938. https://doi.org/10.1016/j.jfoodeng.2020.109938

Kern, C., Bähler, B., Hinrichs, J., & Nöbel, S. (2019b). Waterless single screw extrusion of pasta-filata cheese: Process design based on thermo-rheological material properties. *Journal of Food Engineering, 260*, 58–69. https://doi.org/10.1016/j.jfoodeng.2019.04.015

Kern, C., Weiss, J., & Hinrichs, J. (2018). Additive layer manufacturing of semi-hard model cheese: Effect of calcium levels on thermo-rheological properties and shear behavior. *Journal of Food Engineering, 235*, 89–97. https://doi.org/10.1016/j.jfoodeng.2018.04.029

Kessler, H. G. (1981). Food engineering and dairy technology. *Food Engineering and Dairy Technology*.

Kumano, H., Asaoka, T., Saito, A., & Okawa, S. (2007). Study on latent heat of fusion of ice in aqueous solutions. *International Journal of Refrigeration, 30*(2), 267–273. https://doi.org/10.1016/j.ijrefrig.2006.07.008

Kuo, M.-I., Anderson, M. E., & Gunasekaran, S. (2003). Determining effects of freezing on pasta filata and non-pasta filata Mozzarella cheeses by nuclear magnetic resonance imaging. *Journal of Dairy Science, 86*(8), 2525–2536. https://doi.org/10.3168/jds.S0022-0302(03)73847-8

Levine, L., & Rockwood, J. (1986). A correlation for heat transfer coefficients in food extruders. *Biotechnology Progress, 2*(2), 105–108. https://doi.org/10.1002/btpr.5420020210

Ma, X., James, B., Zhang, L., & Emanuelsson-Patterson, E. A. (2013). Correlating mozzarella cheese properties to its production processes and microstructure quantification. *Journal of Food*

Engineering, 115(2), 154–163. https://doi.org/10.1016/j.jfoodeng.2012.10.002

Mayes, J. J., & Sutherland, B. J. (2002). Effect of high stretch temperature onthe properties of mozzarella. *Australian Journal of Dairy Technology, 57*(2), 178.

Michalski, M.-C., Briard, V., & Michel, F. (2001). Optical parameters of milk fat globules for laser light scattering measurements. *Le Lait, 81*(6), 787–796. https://doi.org/10.1051/lait:2001105

Mohamed, I. O., Morgan, R. G., & Ofoli, R. Y. (1988). Average convective heat transfer coefficients in single screw extrusion of non-Newtonian food materials. *Biotechnology Progress, 4*(2), 68–75. https://doi.org/10.1002/btpr.5420040204

Mohamed, I. O., & Ofoli, R. Y. (1989). Average heat transfer coefficients in twin screw extruders. *Biotechnology Progress, 5*(4), 158–163. https://doi.org/10.1002/btpr.5420050405

Osen, R., Toelstede, S., Eisner, P., & Schweiggert-Weisz, U. (2015). Effect of high moisture extrusion cooking on protein–protein interactions of pea (Pisum sativum L.) protein isolates. *International Journal of Food Science & Technology, 50*(6), 1390–1396. https://doi.org/10.1111/ijfs.12783

Rowney, M. K., Hickey, M. W., Roupas, P., & Everett, D. W. (2003a). The effect of homogenization and milk fat fractions on the functionality of Mozzarella cheese. *Journal of Dairy Science, 86*(3), 712–718. https://doi.org/10.3168/jds.S0022-0302(03)73651-0

Rowney, M. K., Roupas, P., Hickey, M. W., & Everett, D. W. (2003b). The effect of compression, stretching, and cooking temperature on free oil formation in Mozzarella curd. *Journal of dairy science, 86*(2), 449–456. https://doi.org/10.3168/jds.S0022-0302(03)73623-6

Schenkel, P., Samudrala, R., & Hinrichs, J. (2013). The effect of adding whey protein particles as inert filler on thermophysical properties of fat-reduced semihard cheese type Gouda. *International Journal of Dairy Technology, 66*(2), 220–230. https://doi.org/10.1111/1471-0307.12036

Schmidt, F., Graf, B., Hinrichs, J., & Kern, C. (2022). Continuous microwave-assisted extrusion for high moisture texturized foods: A feasibility study. *Innovative Food Science & Emerging Technologies,* (78), 103020. https://doi.org/10.1016/j.ifset.2022.103020

Tanger, C., Schmidt, F., Utz, F., Kreissl, J., Dawid, C., & Kulozik, U. (2021a). Pea protein microparticulation using extrusion cooking: Influence of extrusion parameters and drying on microparticle characteristics and sensory by application in a model milk dessert. *Innovative Food Science & Emerging Technologies, 74*, 102851. https://doi.org/10.1016/j.ifset.2021.102851

Tanger, C., Utz, F., Spaccasassi, A., Kreissl, J., Dombrowski, J., Dawid, C., & Kulozik, U. (2021b). Influence of pea and potato protein microparticles on texture and sensory properties in a fat-reduced model milk dessert. *ACS Food Science & Technology, 2*(1), 169–179. https://doi.org/10.1021/acsfoodscitech.1c00394

Thomann, S., Schenkel, P., & Hinrichs, J. (2008a). The impact of homogenization and microfiltration on rennet-induced gel formation. *Journal of Texture Studies, 39*(4), 326–344. https://doi.org/10.1111/j.1745-4603.2008.00146.x

Thomann, S., Schenkel, P., & Hinrichs, J. (2008b). Effect of homogenization, microfiltration and pH on curd firmness and syneresis of curd grains. *LWT, 41*(5), 826–835. https://doi.org/10.1016/j.lwt.2007.05.023

Vigneux, M.-P.B., Villeneuve, W., Pouliot, Y., & Britten, M. (2022). Increasing the proportion of homogenised fat in cheese milk: Effect on cheese-making properties. *International Dairy Journal, 126*, 105254. https://doi.org/10.1016/j.idairyj.2021.105254

Walstra, P., & Oortwijn, H. (1982). The membranes of recombined fat globules. 3. Hode of formation. *Netherlands Milk and Dairy Journal, 36*, 103–113.

Walstra, P. (1999). Dairy technology: principles of milk properties and processes. CRC Press. https://doi.org/10.1201/9780824746414

Yu, C., & Gunasekaran, S. (2005). A systems analysis of pasta filata process during Mozzarella cheese making. *Journal of Food Engineering, 69*(4), 399–408. https://doi.org/10.1016/j.jfoodeng.2004.08.031

Zeuthen, P., & Bøgh-Sørensen, L. (2003). *Food preservation techniques.* Woodhead Publishing Limited. ISBN: 1 85573 530 X.