



## Explaining the different textures of commercial processed cheese from fractured structures

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### ABSTRACT

This study aimed to explain the different textures of commercial processed cheeses based on their fractured structures. A creep-meter wedge plunger was used to fracture processed cheese samples for imitating the first bite in the mouth. Four representative processed cheeses (A, B, C, D) were classified based on the load–strain curves obtained. Cracks in fractured cheeses were observed using digital photography and scanning electron microscopy. The largest crack developed in sample D, signifying a substantial decrease in load, whereas the inconspicuous crack that developed in sample A indicated a smaller decrease in load. According to sensory evaluation, sample D had the highest firmness score. Sample C had the highest brittleness, collapsing easily because of its heterogeneous structure. Sample A showed the highest adhesion, and sample B had the highest smoothness score; no cracks were found either in the external or internal structures of the fractured cheese.

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## 1. Introduction

Processed cheese is a popular and traditional gel-like dairy product consumed in many countries. A rapid growth in cheese consumption has developed for its use as a component or ingredient in foods such as pizza, cheeseburgers, and breads (Lucey, Johnson, & Horne, 2003). Recently, processed cheese has also become popular as a snack food, and people pay more attention to the texture of processed cheese than its nutrients (Kapoor & Metzger, 2008; Lu, Shirashoji, & Lucey, 2008; Weiserova et al., 2011). To this end, more versatile commercial processed cheeses have appeared in markets to satisfy this large consumer demand.

Food texture is considered important for the acceptability of food (Wilkinson, Dijksterhuis, & Minekus, 2001). For example, smoothness and elasticity contribute to the palatability of jelly-like foods; moreover, adhesion and melting properties are important to cheese-like foods (Everard et al., 2007; Solowiej,

Cheung, & Li-Chan, 2014). Understanding food texture will depend on research in the fields of physicochemical measurement, structure analysis, sensory analysis, etc (Booth, Earl, & Mobini, 2003; Chen & Opara, 2013; Guinard & Mazzucchelli, 1996; Wilkinson, Dijksterhuis, & Minekus, 2001). Furthermore, the timescale of oral processing is also important in sensorial perception of food texture, attribute changes described in terms of first bite and mastication through to swallowing: the food no longer behaves as an unfractured structure, but is composed of particles or pieces during subsequent oral processing.

When the food structure is fractured via mastication, dispersions or continuous structures of components produce a wealth of food textures. Therefore, it is beneficial to have a better understanding of how a fractured structure can be manipulated for perception of food texture. Techniques used to observe the microstructure of processed cheese have been sufficiently developed using various microscopy techniques, such as confocal laser scanning microscopy and scanning electron

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microscopy (Kapoor & Metzger, 2008); however, definitive techniques for observing fractured structures have not been established. Comparison of the non-fractured structure with the fractured structure has been used to clarify the mechanism of fractured properties. It is considered if the importance of structural changes contributing to the fractured properties can be understood, it will benefit texture control of food systems effectively (Fu & Nakamura, 2017).

Processed cheese mainly consists of natural cheese, emulsifying salts, water and limited amounts of additional ingredients. The demand and consumption of processed cheese has grown for many years in the Japanese market. People prefer to evaluate the texture of processed cheese when consumed as a snack food (Fu & Nakamura, 2018; Saitou, Dousako, & Igoshi, 2010). It is clear that various processed cheese products from different manufacturers have different textures when tasted. The changes in ingredient combination and production conditions (e.g., emulsifying conditions, moisture content, pH, cooling, and ripening) enable manipulation of the texture of processed cheese (Fu et al., 2018b; Guinee & O'Callaghan, 2013; Mizuno & Lucey, 2005). It is essential that the casein network and fat globules in processed cheeses are influenced by the cheese-making process, and any change in the casein network affects the final properties of cheeses (Fu et al., 2018a; Fu & Nakamura, 2018; Lee, Buwalda, Euston, Foegeding, & McKenna, 2003).

The objective of this study was to classify the different textures of commercial processed cheeses using a fracture test and sensory evaluation, and explain the different textural properties from the standpoint of fractured properties and structures at the casein level. Understanding the importance of the contribution of fractured structures to textural properties is beneficial to offering possibilities to desire textural properties of cheese and thereby satisfies customers more effectively.

## 2. Materials and methods

### 2.1. Materials

Four types of commercial processed cheeses were purchased from a Japanese supermarket. It was known that the 4 types of processed cheeses represented different mechanical properties due to the load-strain curves, which were selected from 15 types of commercial processed cheeses (Fig. 1a). The ingredients and component content are presented in Table 1. Pretreatment was performed using 50% glutaraldehyde and 2% osmium tetroxide (TAAB) (Nisshin EM Co., Ltd., Tokyo, Japan). Rhodamine B (Nakarai Tesque, Inc., Kyoto, Japan) and BODIPY 500/510 c4, c9 (Life Technologies Ltd., Tokyo, Japan) were used to stain the samples for confocal laser scanning microscopy.

### 2.2. Fracture test

The samples were shaped into cylinders with a diameter of 10 mm and a height of 5 mm using a straw and stored at 4 °C. In the fracture test, measurement was performed with a creepmeter (RE2-33005s; Yamaden Ltd., Tokyo, Japan) as described by Fu and Nakamura (2017). The samples were measured at 4 °C when taken out from a refrigerator. The samples were fractured using a wedge plunger (W28 × 60°) with a load cell of 20 N at 1.0 mm s<sup>-1</sup>. The samples were measured up to 99% strain. The load-strain curves of samples were drawn using a fracture test analysis Ver. 2.0 software (Yamaden Ltd., Tokyo, Japan). Each type of commercial processed cheese was measured at least 10 times. More than 10 samples for each type were prepared, and the measurement was carried out for one time in a sample. A digital camera (EX-ZR300; CASIO Computer Co., Ltd., Tokyo, Japan) was placed to observe the spread of cracks. The following settings, 640 × 480 pixel and 120 fps, were used for photography. Static pictures were edited with a video editing software (Windows Live Movie Maker Ver. 2012; Microsoft Corporation, Tokyo, Japan).

### 2.3. Confocal laser scanning microscopy

Briefly, 4 mL Rhodamine B solution (50 µL) was used to stain the protein phase, and 4 µL BODIPY (1 mM) was used to stain the fat phase. Sectioned samples were cut into 1 mm in thickness using a blade, and placed on the coverglass and observed using an FV1000D inverted confocal laser scanning microscope (Olympus Corp., Tokyo, Japan). An examination using an Ar laser with an excitation line of 488 nm and a HeNe laser with an excitation line of 559 nm was performed in a manner such that only the fluorescent wavelength bands could reach the fat and protein detections).

### 2.4. Scanning electron microscopy analysis

The samples were cut into thin strips (approximately 2 × 2 × 3 mm) with a blade. Chemical double-steady, dehydration, and critical point drying of samples were performed using the methods described by Moriguchi, Mutoh, and Nakamura (2013). Processed samples were affixed on a microscope stage using a carbon tape. An osmium plasma machine (NL-OPC80N; Filgen Inc., Nagoya, Japan) was used for evaporation coating. A scanning electron microscope (JSM-6700F, JEOL Ltd., Tokyo, Japan) was used to observe the non-fractured structure of samples with an accelerating voltage of 3 kV and an irradiation current of 8 µA. To assess the changes in the casein network and the fat globules contributing

**Table 1**  
Ingredients and component contents of four commercial processed cheeses.

Sample	Ingredients	Component content (%)				
		Protein	Fat	Carbohydrate	Sodium	Calcium
A	Natural cheese, emulsifier	21.0	26.5	1.0	1.0	0.6
B	Natural cheese, processed cheese, emulsifier	13.5	28.0	2.5	1.0	0.4
C	Natural cheese, emulsifier	22.3	29.3	2.2	1.0	0.7
D	Natural cheese, butter, emulsifier	20.6	26.6	0.9	1.1	0.6

to the textural properties, the structures of samples fractured at 20%, 35%, and 70% strain were observed.

### 2.5. Sensory evaluation

A 7-point scale (where 1 and 7 represented very weak and very strong, respectively) was performed for sensory evaluation. The panel of evaluators (seven men and six women aged 21–25 years) comprised individuals from the Food Engineering Laboratory, School of Agriculture, Meiji University, Japan. The panellists were professionally trained for sensory evaluation. The samples were cut into cylinders of 5 mm in height and then stored at 4 °C. To avoid negative effects that could arise from the sequence, the samples were offered in random. For each sample, there were three phases of mastication: the first bite (beginning to chew), several bites (chewing period), and the last bite (before swallowing).

### 2.6. Statistical analysis

According to IBM SPSS Statistics Ver. 21 software (IBM Japan, Ltd., Tokyo, Japan), one-way analyses of variance and Tukey's test were used to test the significant differences of samples (defined as  $p < 0.05$ ). Principal component analysis was performed to show the factor loading of items in sensory evaluation using a two-dimensional scatter diagram.

## 3. Results and discussion

### 3.1. Fracture properties of commercial processed cheeses

Fig. 1a shows the load–strain curves of the 15 types of commercial processed cheeses from different companies. Four groups were created based on the aspects of load–strain curves, and four representative commercial processed cheeses were selected for analysis (Fig. 1b). In sample A, the load did not change even as the strain increased after the fracture point (from 30% strain to 70% strain), but it decreased again under high strain (after 70% strain). Sample B had the lowest initial load, and the load showed no decrease in all strain levels, which meant that there was no fracture in sample B. The load of sample C increased twice in the low strain region and then sharply decreased after showing fracture at 35% strain. Sample D had an increased load under low strain, and the load decreased twice after showing fracture at 25% strain.

The static pictures at 0%, 20%, and 70% strain of four samples when fractured are shown in Fig. 2. None of the samples exhibited cracks at 20% strain. At 70% strain, no cracks could be found in samples A and B, but they could be seen clearly in samples C and D. The cracks in C and D developed from the centre and parted into left and right sections by the plunger. It was considered that the cracks that developed at specific strain levels were in accordance with the load decreasing sharply in the load–strain curves, which implied that the cracks had developed when the load decreased after the fracture point (the fracture point occurred at 35% and 25% strain in samples C and D, respectively). The difference between A and B was observed on the contacting area with the wedge plunger (pictures at 70% strain). In sample B, the transformation of the sample occurred on the surface area beside the plunger, whereas in A the surface beside the plunger remained level in both the left and right sides. It was considered that B presented less firmness than A due to the less protein content (Table 1). The sample A had a fracture point in the load–strain curves, but the obvious crack could not be seen in the static picture of sample A.

In the past literature, Everard et al. (2007) reported that increased protein content resulted in a stronger protein matrix,

giving cheeses more firmness and viscoelasticity. Fu et al. (2018b) demonstrated that the firmness of processed cheese increased in a binary mixture of polyphosphate and sodium polyphosphate as the emulsifier rather than when used separately. In addition, it is known that the emulsifiers of processed cheeses were generally used in a binary mixture of polyphosphate and sodium polyphosphate (Saitou et al., 2010). In this study, it was considered that the different behaviours of four commercial processed cheeses in the load–strain curves were dependent on the microstructure of processed cheeses, especially the fractured structures.

### 3.2. Non-fractured structure

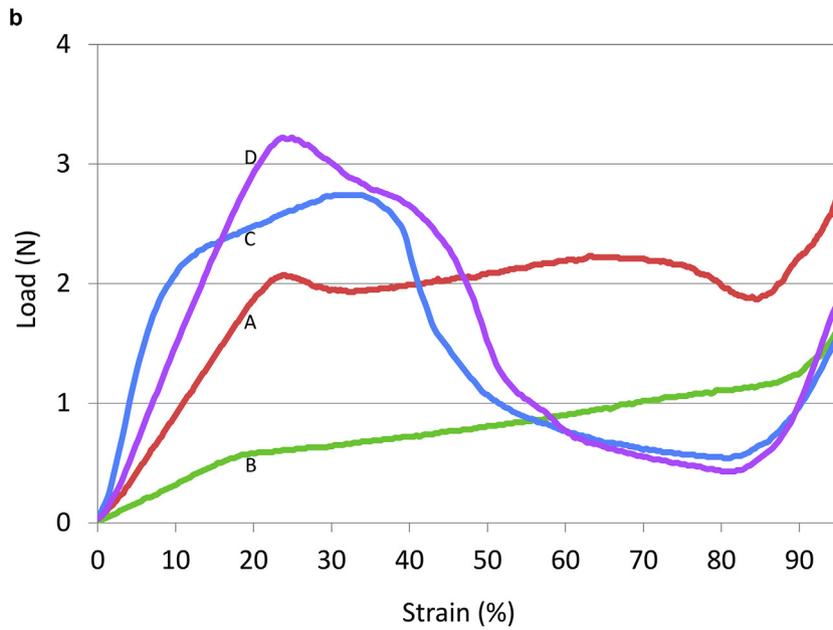
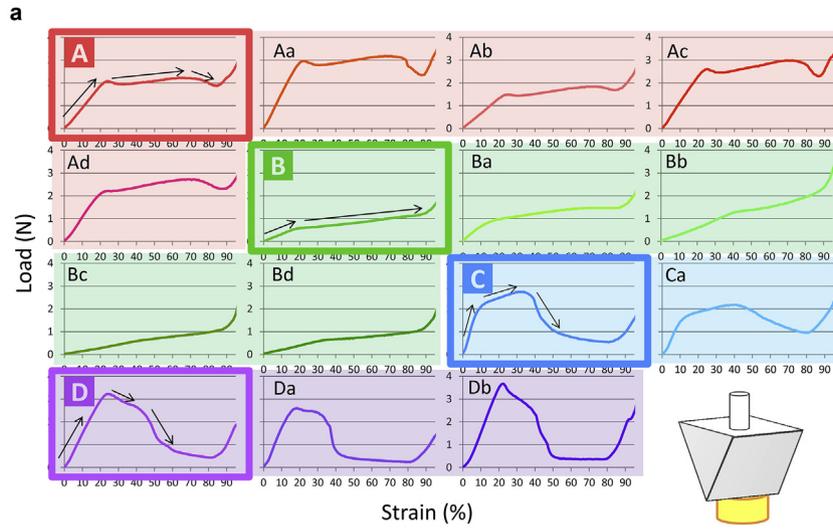
Fig. 3 shows the confocal laser scanning micrographs of non-fractured samples. The red regions represent the protein phase and the green regions represent the fat phase. Sample A, B, and D had small regularly shaped fat globules dispersed homogeneously in the protein phase. In contrast, the fat phase in C was aggregated and heterogeneous, which is largely different from the fat phase in other samples. To make sure the aggregated fat phase in sample C was not because of low levels of coalesced fat at the surface of sample, or during the sample preparation, the scanning electron micrographs of non-fractured structures were observed (Fig. 4a). The photographs showed similar structures to the confocal laser scanning micrographs, in which the white regions show the protein phase and the black regions show the fat globules. It was known that sample C displayed a tattered casein structure in contrast with the other samples, which was considered the reason for forming the aggregated fat phase as viewed by the confocal observation.

In the past literature related to the fat phase in cheese, Lee et al. (2003) proposed that fat in the processed cheese is not essential for the viscosity change, though its presence may influence some aspects of the viscosity curve. The change in the viscosity profile of processed cheese is probably the phenomenon that gives rise to the “creaming effect”. In addition, Dimitreli and Thomareis (2004) also demonstrated that an increase in protein content leads to processed cheese with high values of viscosity. In contrast, fat does not significantly affect the apparent viscosity of processed cheese. Kawasaki (2008) and Fu et al. (2018b) verified the “creaming effect” in processed cheese increased viscosity due to the changes in casein network, and also makes the fat globules become smaller due to fully emulsifying and hence gives more firmness. It was generally recognised that processed cheeses with smaller fat globules brought more firmness (Guinee, Caric, & Kalab, 2004; Shirashoji, Jaeggi, & Lucey, 2006). However, it is not only the fat globule in the complicated system of processed cheese, the interactions between fat globules and protein matrix, between fat globules and polysaccharide such as starches, between fat globules and moisture content had also large influences on the firmness of processed cheese (Guinee & O'Callaghan, 2013; Lee, Klostermeyer, & Anema, 2015; Trivedi et al., 2008; Ye & Hewitt, 2009).

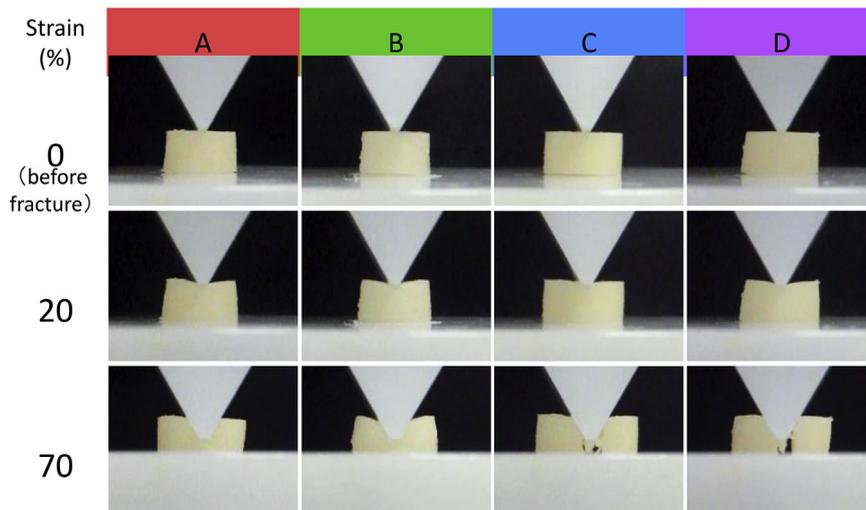
In this study, the sample C with aggregated fat phase had higher firmness than sample A and B with smaller fat globules (Table 2). The casein networks of non-fracture structure in four samples were magnified and observed, the obvious differences were not seen (Fig. 4b). It was expected that a difference in casein network could be found from the fractured structures.

### 3.3. Fractured structure

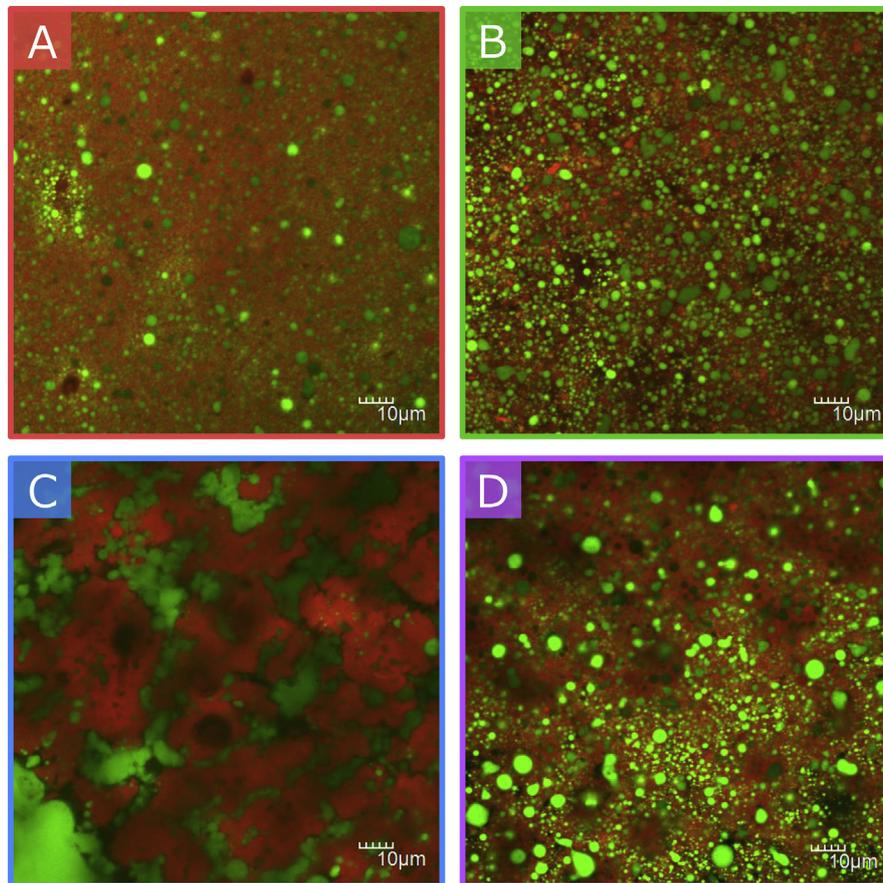
Based on the load–strain curve and the digital photographs, differences in fracture properties and cracks were observed at different strains among the four samples. Although cracks in A or B



**Fig. 1.** Load–strain curves of (a) 15 types of commercial (companies A–D) processed cheeses (lowercase letters represent different products from the different companies; arrows represent aspects of curves) and (b) four representative samples chosen from commercial (companies A–D) processed cheeses.



**Fig. 2.** Static images of four representative samples chosen from commercial (companies A–D) processed cheeses on fracture test at 0%, 20%, and 70% strain.



**Fig. 3.** Confocal laser scanning micrographs of representative samples chosen from commercial (companies A–D) processed cheeses. Green and red fields highlight the fat and protein regions, respectively; magnification  $1000\times$ ; scale bars,  $10\mu\text{m}$ . (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

were not observed in the digital photographs, the difference in load–strain curves was assessed. It was expected that the difference might be found in the microstructure. Fig. 5a shows the fractured structure when the sample was compressed up to 70% strain. It was found that a small crack developed in the centre of compressed sample A. Magnification of the structure near the crack showed that the casein network was torn off, which contributed to the crack (Fig. 5b). In contrast, no obvious crack was found in B. The cross-linking of the casein network was maintained in the magnified fractured structure (Fig. 5c).

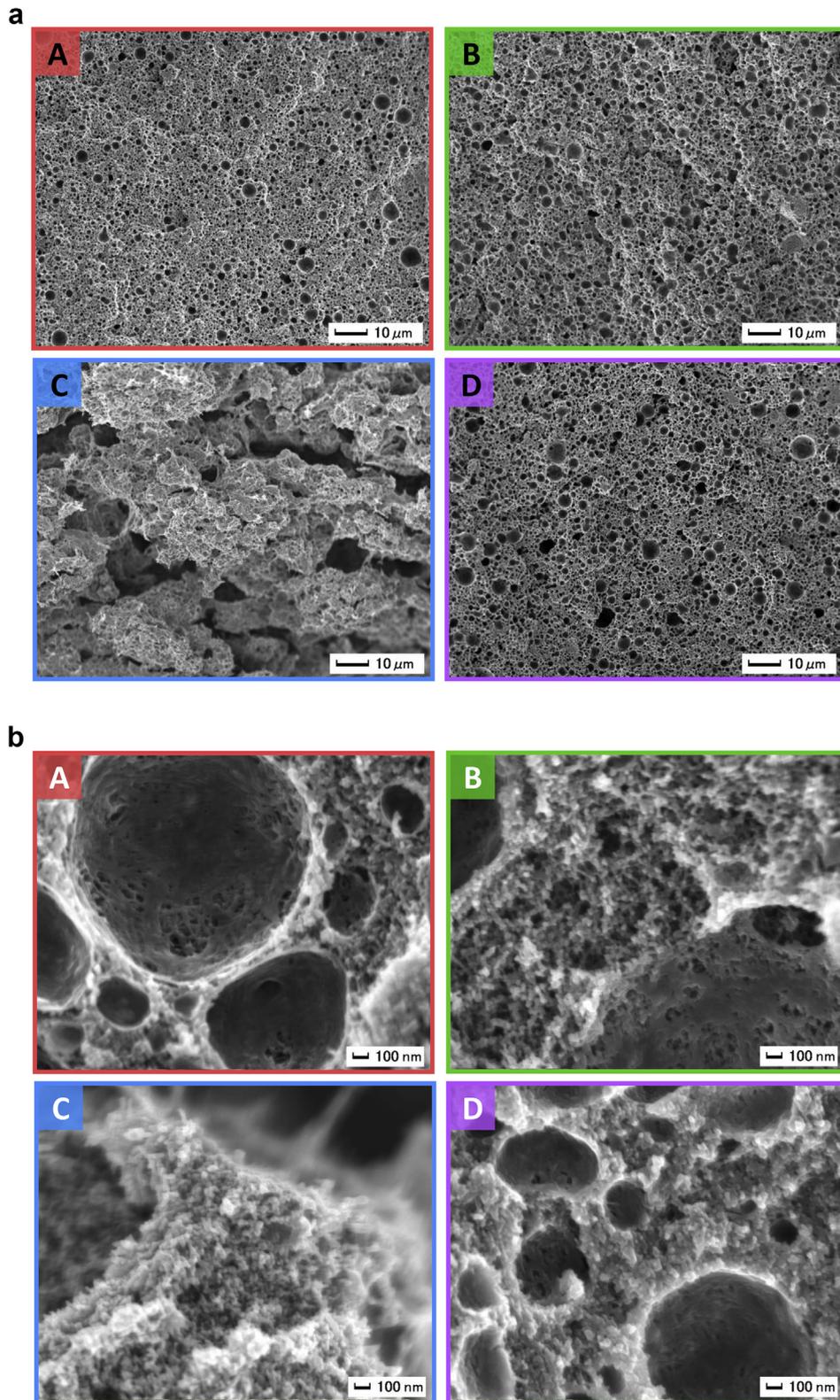
In the previous study (Fu et al., 2018b), none of the samples presented the fracture point in the stress–strain curves of processed cheeses produced by different emulsifying conditions; furthermore, the stress slowly increased even under high strain. It was shown that the mechanical properties of processed cheeses were largely dependent on the status of casein network structures. In this study, although cracks in sample A were not observed in the digital photographs, the cracks in the network structures were considered the reason for the decreased load under high strain, as described in section 3.1.

To investigate the differences between samples C and D, the structures were observed at a strain before the fracture point (20% strain) (Fig. 6), which was the strain before that at which the crack developed, according to the digital photos. The heterogeneous structure of sample C was similar to that of the non-fracture structure, and the roughness in structure was

considered to have easily caused the collapse in sample C. The crack was not found in the microstructure of sample D, and this observation was in accordance with the results of the load–strain curve (the strain occurs before the fracture point is attained) and the digital photographs. Moreover, the fractural structures at 35% strain (after the fracture point in the load–strain curve) were compared between samples A and D (Fig. 7a). It was observed that the casein network was torn off, thereby causing the crack to develop in sample A (Fig. 7b), which was similar to the fractural structure at 70% strain. The crack in the casein network of sample D was larger than that in A (Fig. 7c). This could explain why the load in sample D sharply decreased after the fracture.

#### 3.4. Sensory evaluation

Sensory evaluation was performed, which focused on six evaluation items: firmness from the first bite, brittleness from the first bite, adhesion from the chewing period, smoothness of surface, spread in mouth before swallowing, and lightness during the whole period. The sensory properties of four representative samples of commercial processed cheeses are shown in Table 2, and Fig. 8 shows a principal component analysis on the factor load of the evaluation items. The first principal component was relative to firmness, and the second principal component was relative to fractural structure. As a result, adhesion was obvious in sample A.



**Fig. 4.** Scanning electron micrographs of representative samples chosen from commercial (companies A–D) processed cheeses with (a) low magnification ( $1000\times$ ; scale bars,  $10\ \mu\text{m}$ ) and (b) high magnification ( $50,000\times$ ; scale bars,  $100\ \text{nm}$ ).

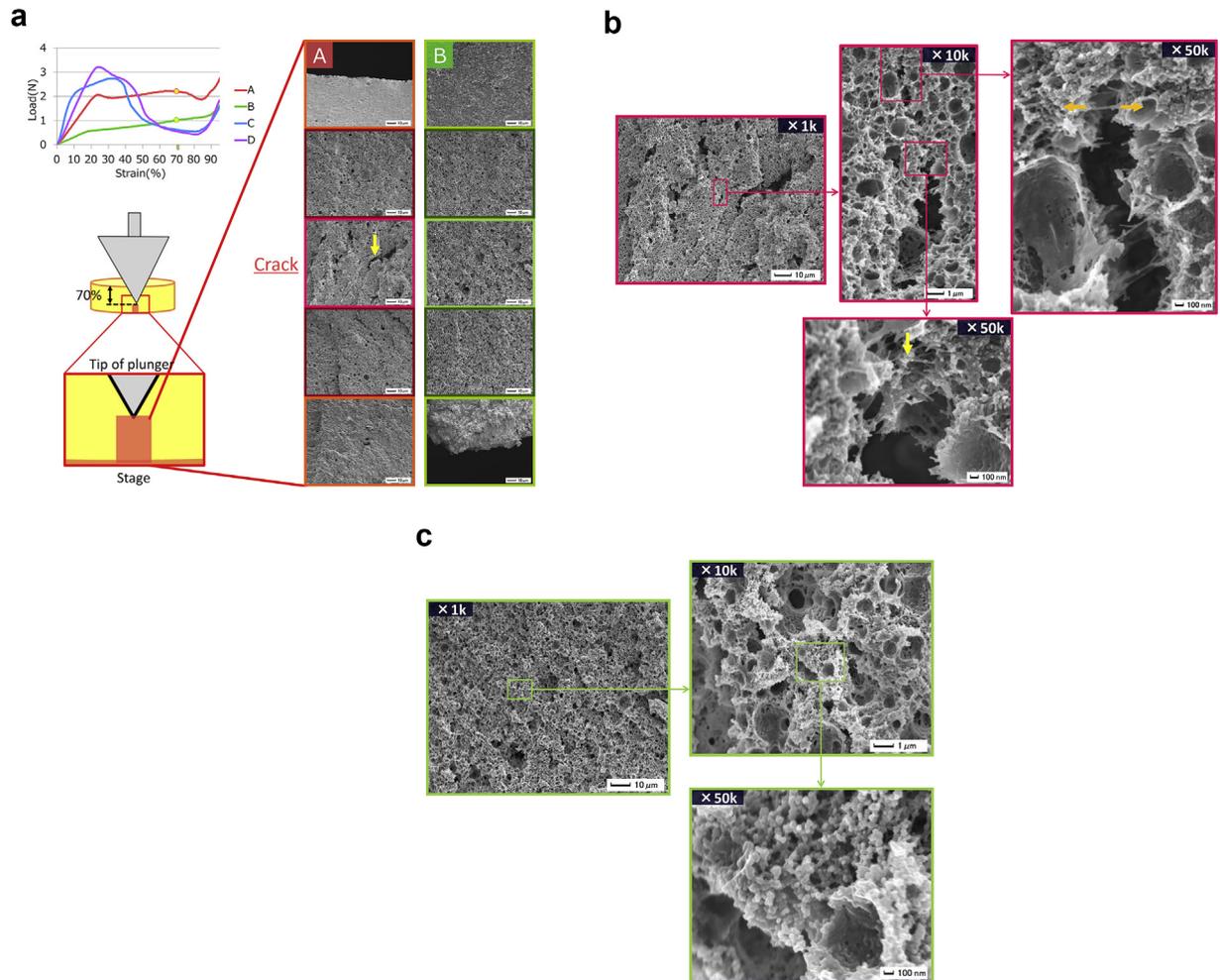
The smoothness, brittleness, and firmness were obvious in samples B, C, and D, respectively. No relationship was found between the spread and the lightness in any sample.

The relationship between sensory evaluation and fractured structures contributing to fracture properties was summarised. In the present study, the fracture test and the method for

**Table 2**  
Sensory properties of four representative commercial processed cheeses.<sup>a</sup>

Sample	Firmness	Brittleness	Adhesion	Smoothness	Spread in mouth	Lightness
A	4.6 ± 1.0 <sup>b</sup>	4.4 ± 1.6 <sup>a</sup>	4.4 ± 1.6 <sup>a</sup>	3.9 ± 1.7 <sup>b</sup>	3.0 ± 1.2 <sup>b</sup>	3.5 ± 1.4 <sup>ab</sup>
B	1.3 ± 0.5 <sup>c</sup>	2.6 ± 1.9 <sup>b</sup>	4.2 ± 1.9 <sup>a</sup>	6.4 ± 0.5 <sup>a</sup>	5.3 ± 1.4 <sup>a</sup>	4.9 ± 2.0 <sup>a</sup>
C	5.5 ± 1.5 <sup>b</sup>	5.2 ± 1.4 <sup>a</sup>	4.2 ± 1.7 <sup>a</sup>	3.5 ± 2.2 <sup>b</sup>	5.9 ± 1.3 <sup>a</sup>	4.8 ± 2.1 <sup>a</sup>
D	6.3 ± 0.9 <sup>a</sup>	5.0 ± 1.7 <sup>a</sup>	4.2 ± 1.6 <sup>a</sup>	2.9 ± 1.1 <sup>b</sup>	2.8 ± 1.5 <sup>b</sup>	2.9 ± 1.6 <sup>b</sup>

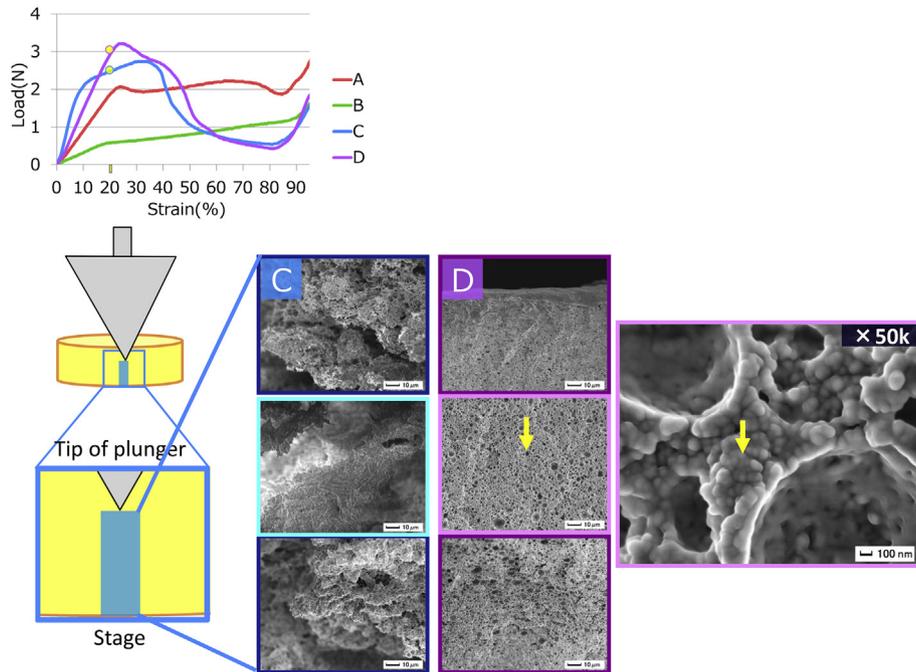
<sup>a</sup> Superscript letters indicate significant differences between mean values in the same column for that sensory property ( $P < 0.05$ ).



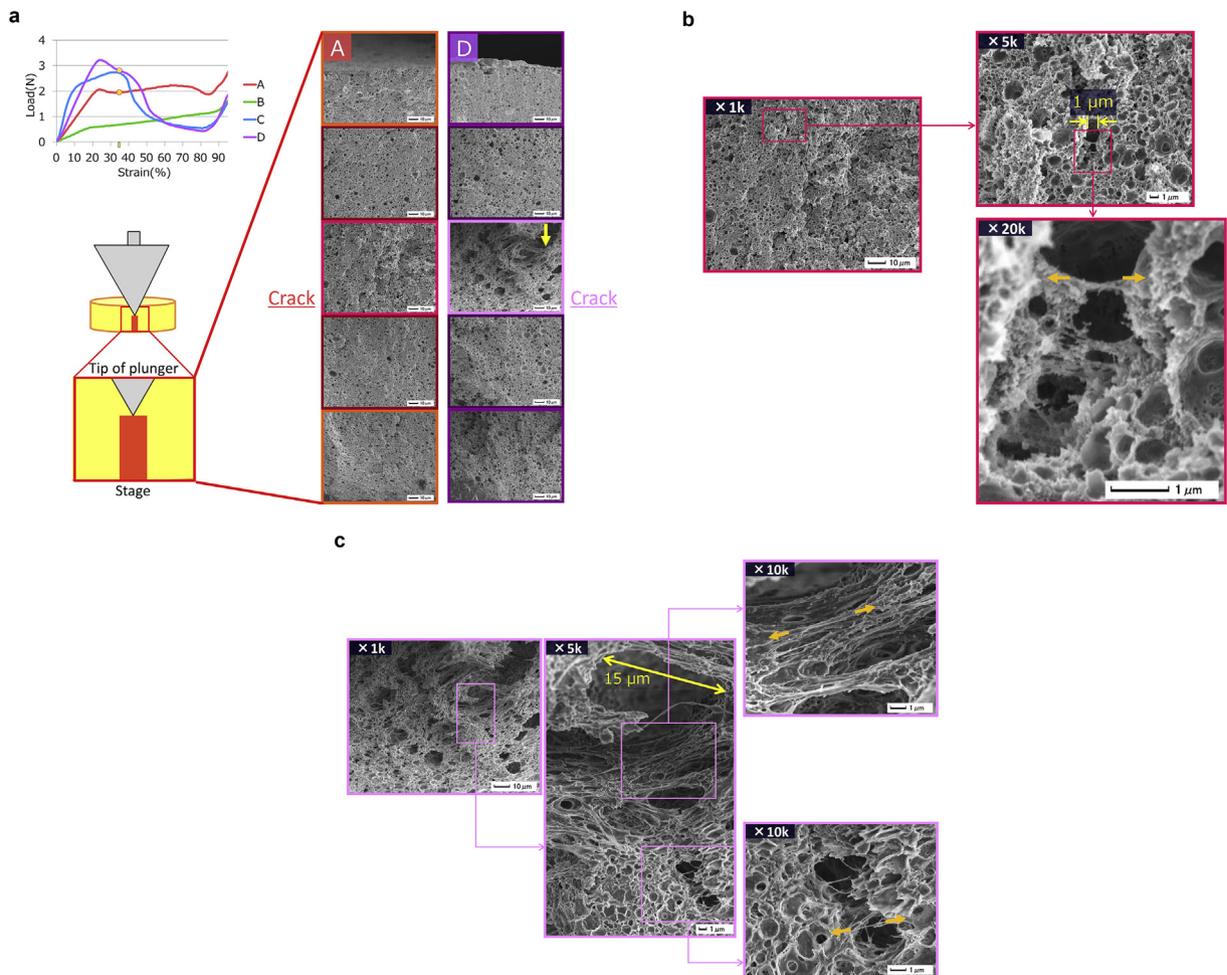
**Fig. 5.** Scanning electron micrographs of (a) fractured structures at 70% strain between samples A and B (arrow indicates developed crack; magnification, 1000 × ; scale bars, 10 μm) and fractured structure in (b) sample A and (c) sample B (arrows indicate aspects of cracks; magnification, 1000 × , 10,000 × , and 50,000 × ; scale bars, 10 μm, 1 μm, and 100 nm, respectively).

observing the fractured structure centred on imaging the first bite. Although the result may be not relative to the sensory evaluation after the first bite, a possible relationship between fractured properties and sensory evaluation was offered as a reference. The lowest score in sample B and the highest score in sample D for firmness were in accordance with the results of the fracture test. Moreover, sample B had the lowest score on brittleness, which could explain why the load did not show any decrease when the sample was compressed. Meanwhile, sample

C had the highest score for brittleness because of its heterogeneous structure, which explains why it collapsed easily. Although significant differences on adhesion were not found in all four samples, the third principal component showed 18.7% relative to adhesion (data not shown). A showed a relationship with adhesion in the principle component analysis and had a higher score than the other samples, which might explain why sample A exhibited no transformation despite the involvement of the plunger, as shown in Fig. 2.



**Fig. 6.** Scanning electron micrographs of fractured structures at 20% strain between samples C and D (arrow indicates aspect of casein network; magnification, 1000 × and 50,000 × ; scale bars, 10 μm and 100 nm, respectively).



**Fig. 7.** Scanning electron micrographs of (a) fractured structures at 35% strain between samples A and D (arrows indicate developed cracks; magnification, 1000 × ; scale bars, 10 μm) and of (b and c) fractured structure in samples A and D, respectively (arrows indicate aspects of cracks; magnification, 1000 × , 5000 × , and 20,000 × ; scale bars, 10 μm, 1 μm, and 1 μm, respectively).

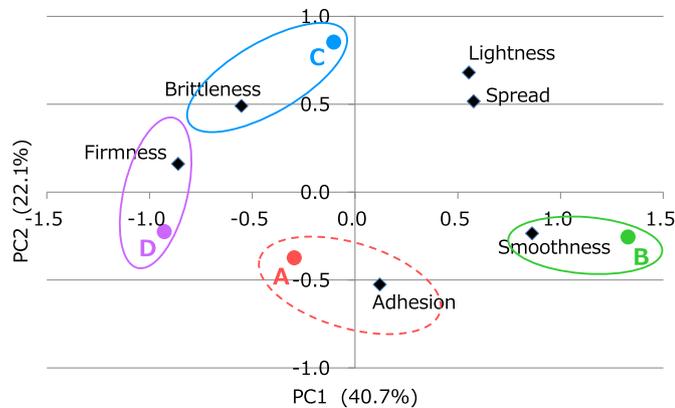


Fig. 8. Principal component analysis of sensory evaluation.

#### 4. Conclusion

Different textures of commercial processed cheeses were explained from the standpoint of fracture structure. The behaviour of the load–strain curve was largely affected by the crack spread and transformation of the casein network. The fracture point was dependent on cracks occurred in the microstructures, and the roughness in structure was considered to have easily caused the collapse of sample. Besides, the relationship between fracture structures and sensory evaluation was obtained using an inference. Sample with no crack expressed less on fragility. The heterogeneous structure contributed to brittleness when fractured. These cases may provide an approach that understands the importance of fracture structure contributing to fracture properties and benefits to offer a possibility to desire the textural properties of processed cheese.

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