

# Volumetric PTV For Fermenting Bread Dough Visualized By Micro X-Ray Computed Tomography

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## ABSTRACT

To elucidate the relationship between the formation of internal structures and changes in viscoelastic properties for improving the quality stability of porous and heterogeneous materials, we focused on bread dough that forms an internal bubble structure through yeast metabolism. Specifically, we conducted a time-series measurement of the internal structure and viscoelastic properties during fermentation, particularly in the heterogeneous condition when additives are mixed into the homogeneous dough. We performed a time-lapse imaging using a digital camera to investigate the inflation volume and rate of fermenting bread dough. We used X-ray CT to visualize the internal bubble structure during the fermenting process and evaluated the changes in viscoelastic properties using a dynamic mechanical analysis. The results indicated that the addition of raisins to the bread dough negatively affected its inflation, made the bubble structure of the dough near the raisins larger and coarsened, and altered the viscoelastic properties.

## 1. Introduction

The unique properties of porous and heterogeneous materials have garnered significant interest in recent advanced materials development. The manufacturing processes of these materials are crucial factors that directly affect their quality. However, adequate research on appropriately adjusting these processes to control the final product's quality has not been conducted. On the other hand, bread dough is one of the most familiar porous bodies to us, forming an internal bubble structure during the fermentation process, where carbon dioxide is produced by yeast's alcohol metabolism. This structure affects one of the important qualities of bread dough—its texture. As bread-making books suggest determining the end of fermentation by the softness of a human earlobe, the feasibility of properly adjusting the manufacturing process and controlling the final product's quality is entrusted to the hands of the craftsmen.

Therefore, we conducted experiments on bread dough with added raisins as heterogeneity, using fermentation by yeast to explore improvements in bread quality and to gain insights into the foaming phenomena inside the material and changes in material properties due to heterogeneity. The objective was to perform a parallel time-series evaluation of the inflation amount, inflation rate, internal structure, and viscoelastic properties in heterogeneous bread dough.

Research on the mechanical properties of bread dough has continued since the 1930s, focusing on the bubble structure inside the dough and its inflation during formation. Annalisa Romano et al successfully modeled the volume changes due to the inflation phenomenon of bread dough using the Gompertz function, thereby grasping the trends of the inflation phenomenon. Additionally, M. Stanke and his team devised a carbon dioxide production model based on the Michaelis-Menten equation. Moreover, mathematical modeling of bubble size distribution in addition to dough inflation was also conducted. Guillermo G. Bellido and his colleagues revealed that the bubble diameter distribution is skewed to the left and attempted mathematical modeling using a log-normal distribution. X-ray CT observations indicated different bubble diameter distributions between hard and soft dough. Subsequently, A. Turbin-Orger and others also used X-ray CT to observe the formation of bubble structures during fermentation in various dough compositions and clarified the impact of sugar content on fermentation using mathematical modeling with a log-normal distribution.

On the other hand, there has not been sufficient investigation into the internal structure and inflation phenomena of heterogeneous bread dough when ingredients like raisins are mixed into the dough before the formation of the bubble structure through fermentation. Therefore, we report the results of time-series evaluations, measuring dough volume using a digital camera, visualizing internal structures with X-ray CT, and assessing viscoelastic properties with a rheometer, focusing particularly on samples of dough mixed with raisins.

## 2. Experimental Method

In this study, two types of dough were employed for use as experimental samples: dough containing raisins (hereafter referred to as "w/ raisin") and dough without raisins for comparison

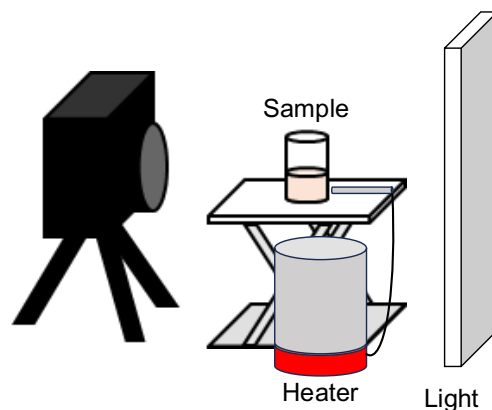
**Table 1** Ingredients of bread dough sample.

Ingredients	weight	w/w flour
flour (Super King, Nissin, Japan)	60 g	-
dry yeast (Super Camellia Dry Yeast, Nissin, Japan)	1.2 g	2%
salt	1.2 g	2%
water	34.8 g	58%

(hereafter referred to as "w/o raisin"). The standard materials and quantities used in the creation of the dough are shown in Table 1 (e.g., Sandoval, A. J et al., 2017). The dough was mixed using a mixer (Stand Auto Mixer DL7523, Kai, Japan) at a speed of 60 rpm for 10 minutes. The dough with 3.0 grams were measured by an electric valance, and when creating the w/ raisin dough, one raisin ( $\approx 0.5$  g) was embedded at the center of the dough. For observation of the inflation phenomenon and internal structure, the prepared test samples were filled at the bottom of tubes (Laboran Styrol Rod Bottle 9-850-01, As One, Japan), and the top of the tube was covered with a wetted paper (Kimwipe, Nippon Paper Crecia, Japan) to prevent drying of the dough. Additionally, the end of mixing was considered as the start of fermentation (0 min) for all experiments.

### 2-1. Measuring inflation by digital camera

To examine the effect of raisin addition on the inflation phenomenon of bread dough, bread dough samples that are with/without a raisin undergoing fermentation were captured in a time-lapse image from the side by a digital camera. The equipment was assembled in a clean bench with a constant temperature of 30°C using a thermal controller as shown in Fig. 1. LED light (SLT-A4C, Mutoh Kogyo, Japan) was illuminated the sample from backside, and a digital camera (D5200, Nikon, Japan) was taken a time-lapse imaging for 180 minutes at 5-minute intervals. To estimate the volume of bread dough was performed using an image analysis software (ImageJ, NIH) by measuring the 2-dimensional projected area of bread dough, which are assumed to be an axial symmetric shape.



**Figure 1** Settings for measuring inflation.

### 2-2. Visualizing inner bubble structure by X-ray CT

During the fermentation of bread dough, the volume of the dough was inflated by bubble generation by a yeast anerobic metabolism. To visualize the formation of bubble structures, imaging and reconstruction were performed using a micro-focus X-ray CT scanner (ScanXmate-E090, Comscantecno, Japan), reconstruction software (coneCTexpress, Comscantecno, Japan). The experimental setup is illustrated in Fig. 2. X-ray irradiation was performed with a current of 90  $\mu$ A, voltage of 90 kV, and measurements taken at 10-minute intervals for 60 minutes. The scan

duration was 5 minutes, spatial resolution was  $27.38 \mu\text{m}/\text{pixel}$ , and voxel data was  $1024 \times 1024 \times 1008$ . In this study, the inflation of dough samples was geometrically restricted by the sample tube, and their thickness remained constant over time, so the brightness values of the final 3D data reflect the contrast between the dough and air. To analyze the effect of adding raisins on the inflation of the bread dough during fermentation, the internal bubble structure analysis was performed with auto bubble shape segmentation and individual bubble labeling, using the ImageJ plugin tool (MorphoLibJ) to obtain a bubble distribution of bread dough samples.

### 2.3 Evaluation of viscoelasticity by dynamic rheometer

To investigate the effect of raisin addition on the changes in viscoelastic properties accompanying the formation of internal bubble structures, a dynamic rheometer (PZ-RHEO NDS-1000, Syscom, Japan) was applied for measuring a viscoelasticity of the bread dough during fermentation as

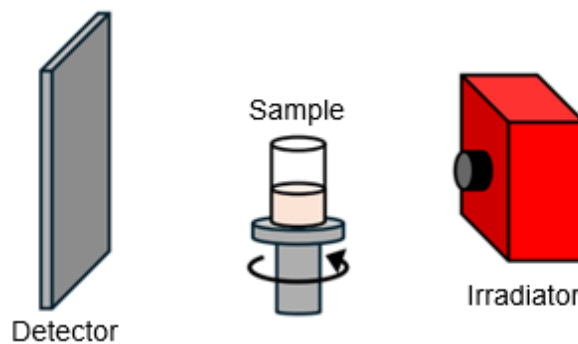


Figure 2 Settings for visualizing inner bubble structure.

shown in Fig. 3. The temperature during fermentation was controlled with  $30^\circ\text{C}$  using the built-in heater of the sample stage, and the measurement frequency was set to 1.5 Hz. This study conducted a two-step experiment. First, the elastic deformation region of the bread dough samples

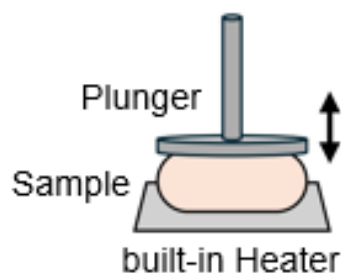


Figure 3 Settings for evaluation of viscoelasticity.

was investigated based on the criterion that the load is proportional to the displacement amplitude. Next, the viscoelasticity of the bread dough samples was measured using the amplitude within the elastic deformation region obtained in the previous step:  $15 \mu\text{m}$ . Three individual samples of 3 grams each were prepared and placed on separate plates. One sample was immediately used for measurement, while the remaining two were fermented in the same environment as the observation experiment of the inflation phenomenon and then used for measurement.

Investigations were conducted on bread dough samples at 10 minutes, 30 minutes, and 60 minutes after the start of fermentation.

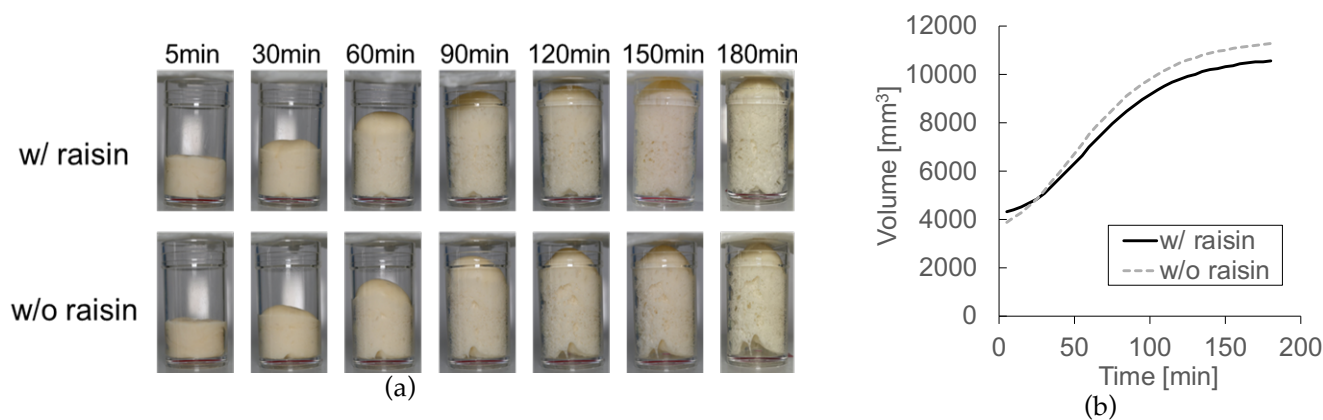
### 3. Experimental Results

#### 3-1. Inflation phenomenon

The time lapse images of inflating bread dough were obtained from observations of fermenting bread dough for both w/ raisin and w/o raisin as shown in Fig. 4(a). The volume of the dough was calculated from the projected area of bread dough. Based on these results, the volume changes due to the inflation phenomenon of the dough were modeled using the following equation, which includes a correction term added to the equation used in previous studies (Romano, A et al, 2007).

$$V = \alpha \exp\left(-\exp\left(\frac{\mu e}{\alpha}(t_{\text{lag}} - t) + 1\right)\right) + \beta$$

where  $\alpha$  is the maximum increased volume of inflated bread dough,  $\mu$  is maximum volume increase rate,  $e$  is the Nepier number,  $t$  is the time,  $t_{\text{lag}}$  is the time lag of fermentation, and  $\beta$  is the initial volume of the bread dough. All volume data obtained for w/ raisin and w/o raisin were fitted to the above equation using the least squares method, and the results as shown in Fig. 4(b). The values of the fitting parameters obtained for w/ raisin and w/o raisin are shown in Table 2. The results indicate that the maximum increased volume  $\alpha$  and the maximum volume increase rate  $\mu$  are smaller for w/ raisin compared to w/o raisin, and the time lag  $t_{\text{lag}}$  before the start of inflation is larger. This suggests that in actual phenomena, the amount and speed of volume change due to sample inflation are smaller, and the time until a certain proportion of volume change progresses is longer. From these findings, it was concluded that the addition of raisins to the dough negatively affects the inflation of the bread dough.



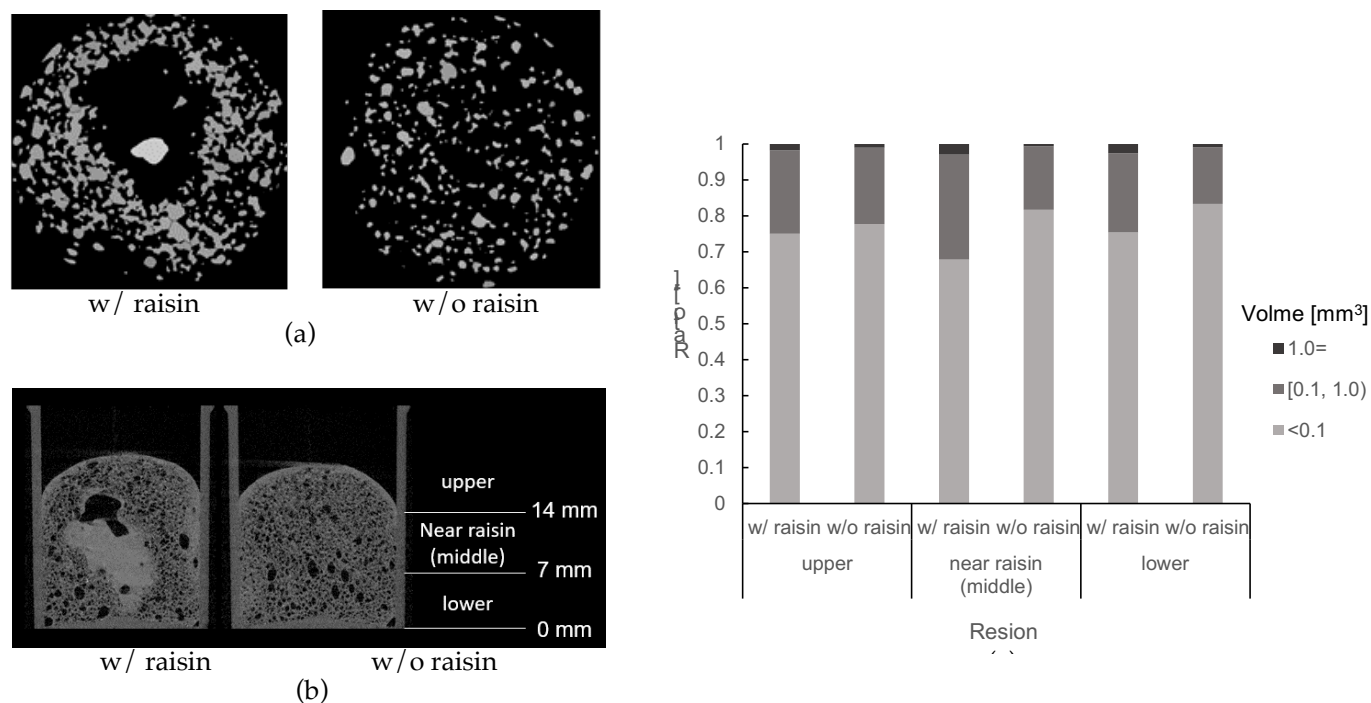
**Figure 4** (a) Shooting results of dough inflation. (b) Fitting results of volumetric change of dough using Gompertz function.

**Table 2** Fitting parameters of volumetric change of dough using Gompertz function.

	$\alpha$	$\mu$	$t_{\text{lag}}$	$\beta$
w/ raisin	$6567.4 \pm 539.7$	$67.8 \pm 9.4$	$18.5 \pm 6.6$	$4171.8 \pm 98.5$
w/o raisin	$7954.6 \pm 522.9$	$79.3 \pm 7.8$	$9.6 \pm 6.4$	$3514.3 \pm 250.8$

### 3-2. Internal bubble structure in fermenting bread dough

The result of visualization of the internal bubble structure of the fermenting bread dough that is 60 minutes after the start of fermentation as shown in Fig. 5 (a). From these results, the distribution of bubbles inside the samples was analyzed by segmentations of bubble structure. When the region of interest was divided vertically into three sections as shown in Fig. 5 (b), the distribution of bubble volumes in each region is shown in Fig. 5 (c). The proportion of bubbles in each class relative to the total number of bubbles in the region is shown when the bubbles are divided into three classes based on their volume: less than 0.1 mm<sup>3</sup>, 0.1 mm<sup>3</sup> or more but less than 1.0 mm<sup>3</sup>, and 1.0 mm<sup>3</sup> or more. From the results, it was found that for both samples, small bubbles with a volume of less than 0.1 mm<sup>3</sup> account for about 70% of the total. The proportion of bubbles with a volume of 0.1 mm<sup>3</sup> or more, however, is larger in w/ raisin compared to w/o raisin, especially near the raisins (middle layer) and in the lower layer. The reason for the increase in bubble volume inside the samples is believed to be due to the accelerated fermentation inside the dough, leading to increased production of carbon dioxide, which results in individual bubbles growing larger or bonding to form larger bubbles.



**Figure 5** (a) Horizontal and (b) Vertical cross-sectional images of dough visualized by XRT and region setting. (c) Volume distribution of bubble in divided region.

### 3-3. Viscoelasticity of fermenting bread dough

The time variations of viscoelastic properties are shown in Fig. 6 (a). The storage modulus is represented by circles (●), and the loss modulus is represented by squares (■). Error bars indicate standard errors, with  $n=5$ . For both samples the storage modulus decreased, and the loss modulus increased in 60 minutes after the start of fermentation compared to 10 minutes after the start of fermentation. The decrease in storage modulus indicates softening of the samples, while the increase in loss modulus indicates a decrease in the samples' fluidity. Previous studies have reported that in elastic materials with bubble structures, a decrease in the Young's modulus is observed with an increase in porosity (e.g., Gibson, I. J., & Ashby, M. F., 1982). Since the Young's modulus corresponds to the storage modulus measured in this experiment, the decrease in storage modulus of each sample over time, indicating softening of the samples due to the formation of internal bubble structures during fermentation, can be attributed to an increase in the porosity of the samples. Figure 6 (b) can be obtained by considering the relationship between porosity and viscoelastic properties obtained from the Fig. 4(b) and Fig. 6(a). As the porosity of each sample increases, the storage modulus decreases, and the loss modulus increases. There are differences in the values of the elastic modulus of each sample even at the same porosity level, which indicates the influence of raisin addition.

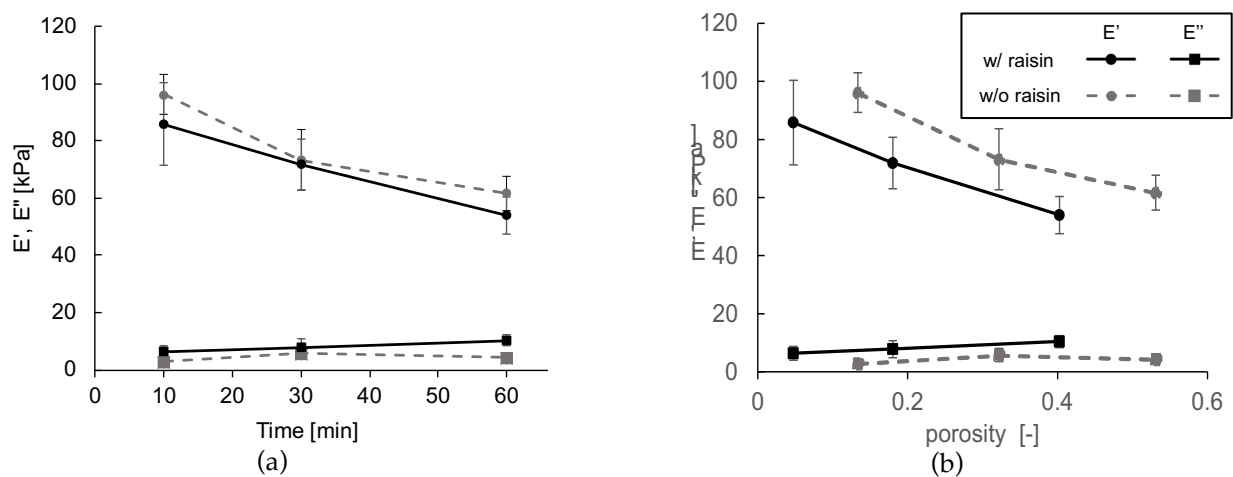


Figure 6 Viscoelasticity change due to (a) fermentation time and (b) porosity.

## 4. Conclusions

In this study, the relationship between the internal structure and mechanical properties of porous bodies and heterogeneous materials was investigated by visualization of fermenting bread dough and viscoelasticity measurements. Raisins were added to bread dough, a familiar porous material, to introduce heterogeneity. We focused on the bubble structure and mechanical properties of bread dough when raisins were added for understanding of their relationship in terms of



mechanical properties. First, the inflation phenomenon of the bread dough was observed using a digital camera. The volume of fermenting bread dough calculated from the image was mathematically modelled using the Gompertz function. It found that the addition of raisins negatively affects the fermentation of the bread dough. Next, observations of the internal bubble structure of the bread dough were conducted using X-ray CT. The volume and spatial distribution of bubbles within the dough were investigated that confirming that the bubble structure near the raisins becomes larger and coarsened due to raisin addition. Finally, the viscoelastic properties of the bread dough were evaluated using a dynamic mechanical analysis. It was confirmed that the addition of raisins affects the relationship between porosity and viscoelastic properties as fermentation progresses.

The addition of raisins alters the inflation phenomenon of bread dough and the bubble structure formed internally, thereby affecting the viscoelastic properties of the dough. In the future, developing models that extract specific characteristics of raisins and conducting comparative experiments using those models, observing the internal bubble structure with higher spatial and temporal resolutions, as well as using MRI methods to investigate into the proton quantity in the samples will be conducted to gain a deeper understanding of the results obtained in this study.

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