# Effect of α-Glucosyl Rutin as Improvers for Wheat Dough and Breadmaking

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#### ABSTRACT

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The effect of water-soluble  $\alpha$ -glucosyl rutin (G-rutin) on some rheological properties of wheat flour dough, on interaction between gluten and starch in dough, and on loaf volume were studied. Addition of G-rutin (200 ppm) increased the loaf volume significantly. Farinograph tests showed that G-rutin decreased the development and stability times. Dough containing a higher amount (1,000 ppm) of G-rutin increased the modulus of elasticity, viscosity coefficient, and relaxation time over that of the control and over that of dough with a lower amount

(50 ppm). Combined additions of G-rutin and L-ascorbic acid (AsA, 50 ppm) gave higher values in all parameters tested. Scanning electron microphotography of doughs showed that G-rutin-treated doughs changed somewhat in viscous appearance when compared with the control. There were no changes in the gelatinization temperature or starch enthalpy with the addition of G-rutin to dough. The sulfhydryl content in dough treated with G-rutin was higher than that of the control. The size of gas cells of crumb baked with G-rutin and AsA decreased slightly.

Rutin is a kind of flavonol glucoside containing rutinose at the C-3 position of quercetin. It is found mainly in buckwheat or Japanese pagoda tree (Fig. 1). Rutin is of pharmaceutical importance as it prevents arteriosclerosis and cerebral hemorrhage (Jing et al 1992). However, it is not very soluble in water (0.01g/100 ml), thereby limiting its application for food. Recently, water-soluble α-glucosyl rutin (G-rutin) has been prepared from rutin and dextrin using glucanotransferase of Bacillus stearothermophilus (Suzuki and Suzuki 1991). It was also reported to be more stable against UV light than the original rutin with a concomitant reduction of odor (Odaka 1990). G-rutin has four conjugated OH groups in its backbone structure and therefore has an important role as radical scavenger or reductant in food materials. In this study, we used G-rutin as a bread improver. Its effect on loaves of baked bread, viscoelastic and internal properties of dough, staling of bread, and gluten-starch interaction are discussed.

## MATERIALS AND METHODS

## Flour and Chemicals

The flour used was the same commercial wheat flour (Hermes, Okumoto Flour Milling Co., Ltd., Osaka, Japan) used previously (Morita et al 1994a). Its protein and ash contents were 11.8 and 0.38%, respectively, on a 13.8% moisture basis.

G-rutin was provided by Hayashibara Biochemical Laboratories Inc., Okayama, Japan. L-Ascorbic acid (AsA) was purchased from Wako Pure Chemical Industries, Ltd., Osaka, Japan. All other chemicals used were of reagent grade.

# **Breadbaking**

Test loaves were baked in five automatic breadmakers (Sharp and Kitchens 1990, Zhang et al 1992) using the same method and ingredients as described previously (Morita et al 1994a). The total time for the entire process was 2 hr, 45 min comprising 25 min of mixing, 5 min of additional mixing after addition of yeast, 90 min of fermentation, and 45 min of baking. Volume of the loaves was

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measured by rapeseed displacement. The data obtained were analyzed to determine significance of difference between means and control (Ichihara 1990).

# **Rheological Tests**

For determination of physicochemical properties of dough, unless otherwise stated, the concentrations of G-rutin were 50 or 1,000 ppm (w/w, flour basis), and concentrations of AsA were 50 ppm.

Farinograph data were obtained in a Brabender Farinograph equipped with the 300-g stainless steel bowl. Mixing was at the standard speed of 63 rpm of the slower blade at 30°C.

Viscoelastic properties of bread were measured using a Fudoh rheometer (Rheotec Co., Ltd., Tokyo, Japan) equipped with a Rikadenki recorder (Tokyo, Japan) as described previously (Morita et al 1994a). Staleness of bread as a measure of compression stress, failure strength, and Young's modulus was determined by rheometer. A  $4 \times 4 \times 3$ -cm³ sample of bread crumb was used. Data were processed by Rheosoft TR-06 (Rheotec Co., Ltd.).

Fig. 1. Chemical formula of  $\alpha$ -glucosyl rutin (G-rutin).

## **Differential Scanning Calorimetry (DSC)**

DSC measurement was done with a Shimadzu instrument (model 50, Kyoto) as described previously (Zhang et al 1992, Morita et al 1994b), with liquid paraffin as reference.

## Scanning Electron Microscopy (SEM)

SEM in a Hitachi apparatus (model S-800) was essentially the same method described previously (Nihei et al 1989, Zhang et al 1992).

## Sulfhydryl (SH) Determination

SH content of lyophilized dough samples was determined by the dithiobisnitropyridine (DTNP) method (Obata et al 1989).

## **Image Analysis of Crumb Grain**

The procedure used was a modification of that reported (Gohtani et al 1992, Morita et al 1994b) using a Pias computer

TABLE I

Effects of Varying Concentrations of α-Glucosyl Rutin
on Specific Volume of Baked Bread

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α-Glucosyl Rutin (ppm)	Specific Volume (cm³/g)		
0	$3.43 \pm 0.08$		
20	$3.49 \pm 0.11$		
30	$3.51 \pm 0.09 (1.0)^a$		
40	$3.54 \pm 0.10$ (2.3)		
50	$3.57 \pm 0.07 (1.3)$		
100	$3.65 \pm 0.13 (0.1)$		
200	$3.80 \pm 0.22 (0.1)$		
500	$3.69 \pm 0.15 (0.2)$		
1,000	$3.64 \pm 0.13  (0.6)$		

<sup>&</sup>lt;sup>a</sup> Values in parentheses show significant difference (%) from the control value. n = 4.

TABLE II
Farinograph Data of Doughs Containing α-Glucosyl Rutin

		α-Gluco:	α-Glucosyl Rutin		
	Control	50 ppm	1,000 ppm		
Arrival time (min)	$2.3 \pm 0.8$	$3.5 \pm 0.4$	$2.7 \pm 0.5$		
Development time (min)	$26.0 \pm 2.1$	$8.5 \pm 0.5$	$14.5 \pm 0.7$		
Stability time (min)	$37.0 \pm 2.1$	$26.5 \pm 2.5$	$19.4 \pm 1.5$		
Water absorption (%)	$70.3 \pm 0.3$	$70.3 \pm 0.3$	$70.3 \pm 0.3$		

a n = 2.

image analyzer (PIAS LA555) equipped with a CCD camera PX-380 and a Victor color monitor AV-MI5OS. Xerox photocopies of bread crumb (7  $\times$  7 cm²) were placed under a nonreflective holding mask, and the cell size information was stored in the computer memory. For the analysis of the mean diameter of gas cells, a  $6\times6$  cm² area in the middle part of a slice was used. A group of pixels of more than 0.0308 mm² was counted as the equivalent diameter.

#### RESULTS AND DISCUSSION

## Effect of G-Rutin on Specific Volume of Bread

Results in Table I show that increasing concentrations of Grutin gave corresponding increases in specific volume of loaves of baked bread. Bread baked with 50 ppm, or more, of G-rutin was significantly larger than the control. Concentrations of 200 ppm were the most effective, but higher concentrations (up to 1,000 ppm) showed a decreasing trend in specific volume. However, the volumes were still higher than that of control.

Figure 2 shows the cross-sectional views of baked bread with varying concentrations of G-rutin and 50 ppm of AsA. AsA is added for the improvement of dough for baking (Johannson and Cooke 1971, Zhang et al 1993) because addition of G-rutin makes the dough soft. Crumb grain of G-rutin-treated bread, with or with-out AsA, was fairly good with no distinct differences observed.

# **Effect on Rheological Properties**

Some rheological properties of dough were tested after mixing in a farinograph (Table II). The doughs with 50 and 1,000 ppm of

TABLE III

Effects of α-Glucosyl Rutin (G-rutin) on Viscoelastic Properties of Dough
After Mixing for 30 min in a Home Baker, as Measured by Rheometer

	Viscoelastic Parameters <sup>a</sup>				
Additives	g	γ	τ	η	
Control	104 ± 8	$3.23 \pm 0.2$	19.1 ± 3.3	$6.2 \pm 1.4$	
G-rutin (50 ppm)	$108 \pm 10$	$3.38 \pm 0.3$	$19.7 \pm 1.7$	$6.6 \pm 1.1$	
G-rutin (1,000 ppm) G-rutin (1,000 ppm)	$118 \pm 8$	$3.68 \pm 0.2$	$21.7 \pm 1.2$	$8.0 \pm 0.5$	
+ AsA (50 ppm)	$141 \pm 22$	$4.40 \pm 0.7$	$31.8 \pm 3.3$	$14.0\pm3.2$	

<sup>&</sup>lt;sup>a</sup> g = stress (gram);  $\gamma$  = modulus of elasticity (10<sup>5</sup>dyn/cm<sup>2</sup>);  $\tau$  = relaxation time (sec);  $\eta$  = viscosity coefficient (10<sup>6</sup> poise). Values are averages  $\pm$  standard deviations. n = 3.

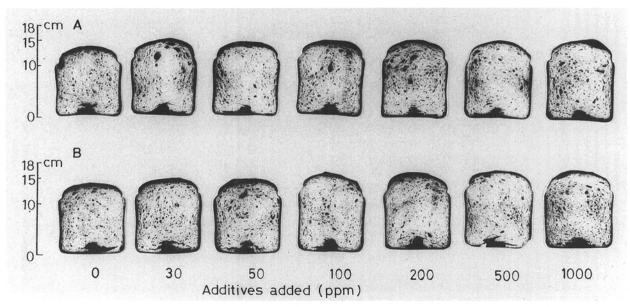


Fig. 2. Cross-sectional views of baked bread added with varying concentrations of α-glucosyl rutin (G-rutin) and ascorbic acid (AsA). A, G-rutin + 50 ppm AsA. B, G-rutin alone.

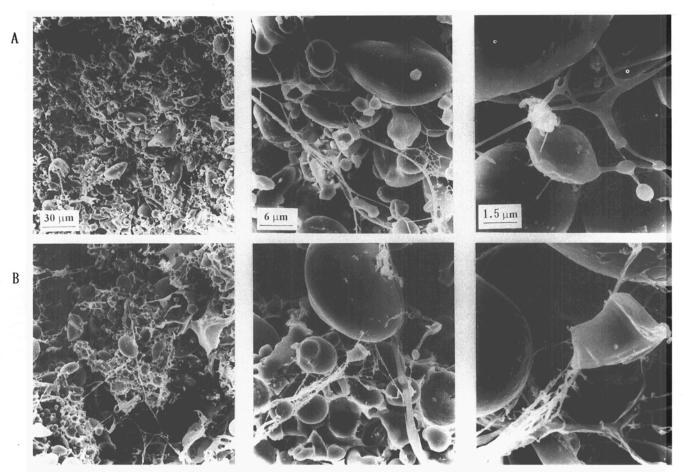


Fig. 3. Scanning electron micrographs of doughs containing 1,000 ppm  $\alpha$ -glucosyl rutin with (A) and without (B) 50 ppm AsA after mixing in a home baker.

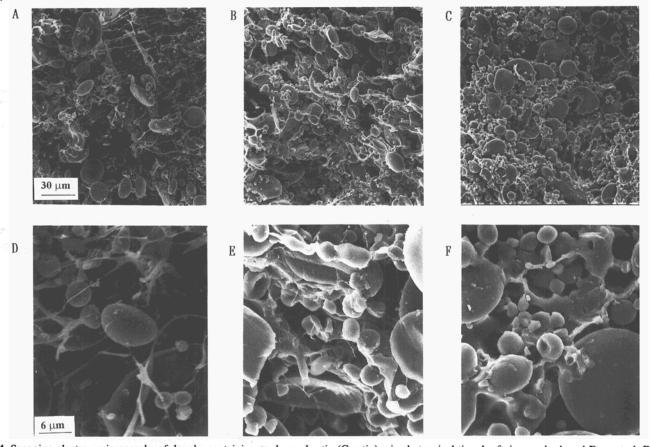


Fig. 4. Scanning electron micrographs of doughs containing  $\alpha$ -glucosyl rutin (G-rutin) mixed at arrival time by farinograph. A and D, control; B and E, 50 ppm G-rutin; C and F, 1,000 ppm G-rutin.

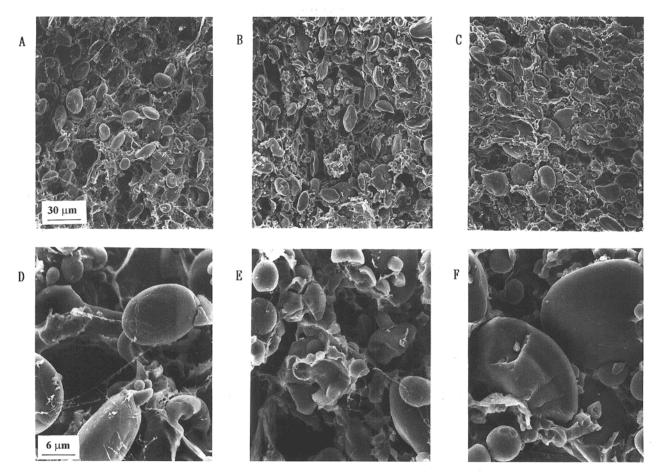


Fig. 5. Scanning electron micrographs of doughs containing α-glucosyl rutin (G-rutin) mixed for 30 min by farinograph. A and D, control; B and E, 50 ppm G-rutin; C and F, 1,000 ppm G-rutin.

TABLE IV Summary of Gelatinization Temperature and Starch Enthalpy in Wheat Flour Dough with α-Glucosyl Rutin (G-rutin) as an Additive, After Mixing for 30 min in a Home Baker

	Gelatiniza			
Additive (ppm)	Onset	Peak	Final	Enthalpy (J/g)
Control	$51.4 \pm 0.9$	$62.7 \pm 0.4$	$97.9 \pm 0.3$	$3.79 \pm 0.07$
G-rutin (50)	$50.9 \pm 0.4$	$63.2 \pm 0.6$	$97.9 \pm 0.6$	$3.99 \pm 0.17$
G-rutin (1,000)	$51.2 \pm 0.4$	$62.8 \pm 0.2$	$97.1 \pm 0.6$	$4.01 \pm 0.23$

<sup>&</sup>lt;sup>a</sup> Values are averages  $\pm$  standard deviation. n = 4.

TABLE V
Effect of α-Glucosyl Rutin (G-rutin) on the
Sulfhydryl (SH) Level of Dough After Mixing for 30 min

	SH Content (10 <sup>-7</sup> eq/g of flour)			
Additive	Home Baker	Farinogragh		
Control	$6.9 \pm 0.1$			
G-rutin (50 ppm)	$7.7 \pm 0.1$	$6.8 \pm 0.3$		
G-rutin (1,000 ppm)	$8.3 \pm 0.1$	$7.2 \pm 0.2$		

 $<sup>^{</sup>a} n = 2.$ 

G-rutin added gave lower arrival times than that of the control. The development time of dough at 50 ppm showed lowest value at 8.5 min, which increased further to 14.5 when 1,000 ppm of Grutin was added. Both values, however, were still lower than that of the control (26 min). On the other hand, the stability time decreased when either 50 or 1,000 ppm of G-rutin was added. This may be caused by degradation of the gluten matrix due to its

TABLE VI Effect of Additives on Size of Gas Cells as Counted with Image Analyzer<sup>a,b</sup>

Amount of Additive (ppm)	G-Rutin	G-Rutin + AsA (50 ppm)
0	$0.15 \pm 0.14$	$0.16 \pm 0.13$
30	$0.16 \pm 0.11$	$0.15 \pm 0.19$
50	$0.16 \pm 0.23$	$0.14 \pm 0.20$
100	$0.17 \pm 0.19$	$0.14 \pm 0.22$
200	$0.16 \pm 0.18$	$0.15 \pm 0.15$
500	$0.17 \pm 0.30$	$0.17 \pm 0.17$
1,000	$0.15 \pm 0.13$	$0.17 \pm 0.20$

 $<sup>^</sup>a$  Gas cell size was counted in a  $6\times 6~\text{cm}^2$  area in the middle part of the bread crumb.

interaction with G-rutin. However, the ratio of water absorption seems to be unaffected.

# Viscoelastic Parameters of Dough

Doughs containing 50 or 1,000 ppm of G-rutin were mixed in a home baker and measured for viscoelastic properties by rheometer. Table III shows that viscoelastic properties of dough increased upon addition of 50 and 1,000 ppm of G-rutin. Also, viscoelastic parameters increased with the combination of G-rutin at 1,000 and AsA at 50 ppm. Previously, dough samples containing 50 and 1,000 ppm of G-rutin consistently showed lower values when mixed in a farinograph. The reasons why the doughs appeared weaker when measured by farinograph and showed increased viscoelasticity when measured by rheometer are not clear. This could be due to the reduction of disulfide

b Data show the mean diameter (cm) of gas cells ± standard deviation.

TABLE VII
Effect of Additives on the Firmness of Bread Crumb During Storage

Additives		Amount of G-Rutin (ppm)				
	Storage (days)	0	100	200	500	1,000
G-rutin	0	56.4 ± 7.1	$35.9 \pm 4.0$	$41.2 \pm 9.4$	$43.0 \pm 6.7$	$31.7 \pm 7.1$
	1	$101.7 \pm 13.4$	$58.9 \pm 6.4$	$75.8 \pm 15.2$	$74.2 \pm 12.0$	$59.0 \pm 13.0$
	2	$155.0 \pm 20.3$	$96.5 \pm 9.7$	$113.4 \pm 24.3$	$112.4 \pm 14.3$	$89.9 \pm 11.9$
	3	$179.5 \pm 10.8$	$112.6 \pm 12.9$	$149.9 \pm 31.5$	$142.7 \pm 14.9$	$127.1 \pm 13.9$
G-rutin + AsA (50 ppm)	0	$50.1 \pm 8.0$	$43.8 \pm 8.4$	$36.1 \pm 9.0$	$45.2 \pm 2.7$	$36.0 \pm 11.9$
	1	$90.3 \pm 15.1$	$76.6 \pm 12.1$	$73.6 \pm 16.6$	$79.7 \pm 5.4$	$70.5 \pm 21.3$
	2	$144.9 \pm 18.3$	$123.0 \pm 18.8$	$120.1 \pm 25.9$	$135.4 \pm 10.8$	$122.3 \pm 31.8$
	3	$157.6 \pm 22.3$	$139.5 \pm 15.6$	$143.9 \pm 27.0$	$154.2 \pm 13.2$	$133.4 \pm 25.8$

<sup>&</sup>lt;sup>a</sup> Values are stress (gram)  $\pm$  standard deviation. n = 4.

(SS) cross-linkage or softening of dough that subsequently degraded the dough as a result of continued mixing.

#### **Effect on Physicochemical Properties**

SEM. Figure 3 shows the SEM results for dough containing 1,000 ppm of G-rutin, with and without 50 ppm of AsA, after 30 min of mixing in a home baker. Addition of G-rutin to the dough made it soft, with the starch granules uniformly covered. When combined with AsA, the dough hardened slightly with some portion of gluten aggregates becoming fibrous. AsA probably altered the properties of the dough. Oxidation of the SH group in the dough followed by the formation of SS cross-linkage is likely (Sullivan 1965).

SEM results for doughs mixed by the farinograph (Fig. 4) showed that the dough supplemented with 50 ppm of G-rutin appeared smoother, with no fibrous gluten, as seen in the control dough. When an excess amount of G-rutin (1,000 ppm) was added, the doughs disintegrated and discontinuous gluten particles were observed (Fig. 5). This suggests that G-rutin makes the dough fragile and susceptible to breakage by mixing. Formation of cross-linkage of gluten in the dough could have been enhanced by the intermittent mixing in a home baker.

DSC results. The gelatinization temperature of starch granules in the dough samples mixed for 30 min in a home baker was tested by DSC (Table IV). The gelatinization temperature did not change significantly upon addition of G-rutin. However, addition of G-rutin increased the enthalpy slightly more than that of the control. This result suggests that G-rutin affects the three-dimensional structure of the dough, as the degree of adherence of gluten to the surface of starch granules appears to be weaker.

The doughs with 1,000 ppm of G-rutin were also tested by DSC at arrival time and after 30 min of mixing in the farinograph. The gelatinization temperature of starch did not change for all mixing stages tested. However, the enthalpy value was slightly lower for the dough mixed for 30 min, suggesting degradation or breakdown of the dough matrix during mixing. This suggests a change in the covering of the surface of starch granules by the gluten after addition of G-rutin.

SH content. The SH level of the dough mixed in a home baker and farinograph after addition of 50 or 1,000 ppm of G-rutin increased slightly when compared with that of the control dough (Table V). The increase for dough with 1,000 ppm was lower than that for 50 ppm. This result indicates that G-rutin is not a strong reducing agent. The reason why SH content is lower in doughs mixed by farinograph is that the newly exposed SH groups tend to be oxidized by atmospheric oxygen during continuous mixing.

Size of crumb grain. Mean diameter of gas cells of crumb grain of baked bread supplemented with 50 or 500 ppm of G-rutin was ≈1.6–1.7 mm, indicating a rather coarse structure of the crumb (Table VI). However, when 50 ppm of AsA was combined with various amounts of G-rutin, the size of gas cells decreased considerably. Referring to the mean values of the size of gas cells, the standard deviation values were high, because relatively large gas

cells exist in the bread crumb. As the specific volume of bread increased, the size of gas cells also increased. However, for the addition of combined G-rutin and AsA, gas cells tend to become small, suggesting that the distribution of gas cells were improved slightly.

Staleness of bread. Staleness or firmness of bread crumb was tested daily during storage (Table VII). The bread baked with a single addition of 100 ppm of G-rutin showed lower stress values. Thus, the bread crumb appeared softer and the staleness progressed more slowly when compared with bread crumb with higher concentrations, except at 1,000 ppm. Moreover, G-rutin had only a slight effect on the change in softness of bread crumb during storage.

## **CONCLUSION**

Water-soluble G-rutin in bread formula increased loaf volume and improved the rheological properties of dough. Gelatinization temperature and enthalpy of the starch of the wheat dough did not change significantly, but a viscous appearance of dough was observed when using SEM. The SH content in dough supplemented with G-rutin and mixed for 30 min in a home baker was higher than that of the control. The oxidation of SH groups thus formed in the dough, and subsequent SS cross-linkage formation during mixing, might have strengthened its viscoelastic properties and improved baking performance and softness of bread. From these results, G-rutin could be recommended for practical application as bread improver considering its pharmaceutical value.

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