# Rheological Properties of Dough Made from Flour Exposed to Gaseous Ammonia

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## ABSTRACT

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Dough made from flour treated with gaseous ammonia showed specific rheological properties similar to those of doughs made from heat-conditioned flour or from flour containing "Kansui," a mixture of alkali carbonates and phosphates, or oxidizing agents. The ammonia-treated dough showed an increased resistance to extension with decreased extensibility. Flour processed with gaseous ammonia offers the benefit of special dough quality without the problems associated with presently used additives.

The globulin fraction of flour is readily oxidized to form high molecular weight proteins on incubation under alkaline conditions (Terada et al 1978a). The oxidation was considered responsible for features of the dough which was manufactured using Kansui (Terada et al 1978b).

Wheat flour usually contains about 13% moisture in equilibrium with atmospheric moisture. The suggestion was made that following exposure of flour to gaseous ammonia, this flour moisture might produce enough alkalinity to change the rheological properties in a way similar to that observed in dough made using Kansui. This article presents the procedure of exposing flour to gaseous ammonia and reports the characteristic rheological properties of dough prepared using the processed flour.

## MATERIALS AND METHODS

The flour was obtained by commercially milling dark hard winter wheat grown in the United States. The general composition of the flour was 11.4% crude protein, 30.8% wet gluten, 0.4% ash, and 12.8% moisture.

### Adjustment of Flour Moisture

Six kilograms of flour was put into a fluid bed spray granulator (model WSG-5, Glatt-Okawara Co., Japan) and fluidified with air and atomized water at 32°C for various periods. The flour was transferred to a polypropylene bag and allowed to equilibrate at room temperature overnight.

### Exposure of Flour to Gaseous Ammonia

Two methods were employed for exposing flour to gaseous ammonia.

*In Desiccator.* Solutions of 2,000, 500, 200, or 50 ml of 0.7, 2.8, 7.0, or 28% aqueous ammonia were placed in the bottom of 7.3-L desiccators. The inside of each desiccator was separated into two levels by wire net racks, and flour was spread 3-cm thick on a cotton cloth on each rack. The desiccator was closed tightly and kept at 25°C for 48 hr. The flour was then taken from the desiccator and spread on a filter paper placed in a fumehood; air was circulated until the odor of ammonia gas was not detectable. The pH of a 10% aqueous flour suspension after removal of ammonia gas was about 6.3.

*In Column.* Three kilograms of flour were packed in a column made from polyacrylic resin, 10 cm in diameter and 76 cm long. A mixture of ammonia and nitrogen gases of various ratios ($NH_3/N_2$ = 1:0, 1:4, 1:10, v/v) was blown up through the packed flour at a rate of 5 L/min. Flour was exposed to the gas mixture for 60 min unless otherwise specified.

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In this test, another kind of flour (crude protein, 10.4%; wet gluten, 29.2%; ash, 0.4%; moisture, 14.0%) commercially manufactured by mixing flours derived from No. 1 Canada western red spring, dark hard winter (U.S.A.) and western white (U.S.A.) wheat was used.

### Rheology of Dough

Extensigraphs and farinographs were performed according to AACC methods 54-10 and 54-21 (AACC 1962), respectively. For the farinographs, the constant flour weight method was used with a 300-g bowl. Amylograph values were determined as follows. The viscosity of a slurry of 65 g of flour in 450 ml of water was recorded as a function of temperature. Temperature was raised at a rate of 1.5°C per min over the range from 50 to 95°C. In this case, to eliminate the effect of pH-difference, the pH of the slurry of untreated flour (pH 5.8) was adjusted to pH 6.3 with 0.94 ml of 0.7% aqueous ammonia.

The structural relaxation test with dough was carried out by the method of Dempster et al (1955).

### Analysis of Ammonia

Flour-bound ammonia was analyzed by a combination of the micro-diffusion method of Conway and Byrne (1933) and the color reaction with Nessler reagent, using the formula of Wicks (1941). Saturated potassium carbonate (1.5 ml) and 1 ml of a suspension of 500 mg of flour in 2 ml of water were placed in the outer chamber of a 60-mm diameter micro-diffusion vessel, and 1 ml of 0.01 *N* HCl was placed in the inner chamber. After incubating the micro-diffusion vessel at 40°C for 5 hr, the inner chamber was washed several times with water to bring the total volume up to 3 ml. One milliliter was further diluted with water, as necessary, and mixed with 0.5 ml of Nessler reagent to a total reaction volume of 10.5 ml. The color was allowed to develop for 20 min at 25°C and measured at 425 nm. The test was performed in duplicate and its mean value expressed. A solution consisting of 0.5 ml of Nessler reagent and 10 ml of water was used as a blank. Ammonium chloride solutions of known concentrations were used as a standard.

The concentration of ammonia inside the desiccator was determined by gas chromatography.

### Estimation of Wet Gluten and pH

Wet gluten was estimated by AACC method 38-11 (AACC 1962). pH was measured with a 10% (w/v) suspension of flour, using a pH meter.

### Amylase Assay

*Saccharifying Activity.* Three hundred milligrams of flour was suspended in 30 ml of 0.05 *M* acetate buffer, pH 6.0, containing 0.1 *M* NaCl, and allowed to stand for 6 hr at 4.5°C followed by filtration through a filter paper. One milliliter of the filtrate was incubated with 4 ml of 0.5% soluble starch (0.02 *M* acetate buffer, pH 6.0) at 40°C for 5 min. The reducing sugar formed was determined by the Shaffer-Somogyi method (Shaffer and Somogyi 1933). The activity was tentatively expressed as a percentage of that of untreated flour.

*Dextrinizing Activity.* Potato starch (1 g, dry weight) was suspended in 50 ml of 0.02*M* NaOH and gently heated to boiling. After 15 min of boiling, the solution was cooled, neutralized to pH 6.0 with acetic acid, and filled up to 100 ml. One hundred milligrams of flour suspended in 1 ml of 0.05*M* acetate buffer, pH 6.0, containing 0.1*M* NaCl was added to 10 ml of the solubilized potato starch solution at 40°C. After a 10-min incubation, 1 ml of the reaction mixture was taken into 10 ml of 0.1*N* HCl. The mixture was filtered, and 1 ml of the filtrate was added to 10 ml of 0.005% $I_2$-KI solution; the color developed was measured at 660 nm. The dextrinizing activity was expressed as a percentage of that of untreated flour.

**Noodle Texture**

Five hundred grams of a mixed flour (30 parts of ammonia-treated flour [Fig. 1] and 70 parts of untreated flour), 12.5 g of NaCl, and 170 ml of water were mixed at 120 rpm for 15 min in a horizontal ribbon mixer similar to that used in macaroni processing. The resulting dough was rolled six times, made into a 1.4-mm thick sheet through a pair of revolving rolls, cut into 3-mm widths with a cutting roll, steamed under normal pressure for 110 sec, and dried in an air current (170°C) for 23 sec to obtain the so-called instant noodle. As a reference, the texture of a noodle manufactured by adding 730 mg of Kansui and 2.0 g of NaCl or 2.0 g of NaCl alone per 100 g of flour was also studied.

In order to measure the texture of the noodles, 90 g of the noodles was boiled with 500 ml of water under reflux for 5 min. After boiling, the noodles were placed on a wire gauze for a few minutes to remove excess water, cooled to room temperature, and cut into 30-mm lengths. The texture was then determined on a texturometer (GTX-2, Zenken Co., Japan). The plunger of the texturometer was an 18-mm diameter column made of Lucite resin; the aluminum platform was flat; and the clearance between the plunger and platform was 0.2 mm. The texture measurement was performed 20 times for each sample.

**Synthetic Dough**

Wheat starch was of reagent grade (Wako Chemicals Co., Japan). Gluten powder was prepared by the method of Jones et al (1959), but the heating of gluten solution before lyophilization was cut down. Synthetic dough was made by mixing 260 g of wheat starch, 40 g of gluten, and 3 g of NaCl and kneading them with a suitable amount of water (usually 170–200 ml) for the extensigraph test. Ammonia treatment of gluten (moisture, 5.3%) was performed with 50 ml of 28% aqueous ammonia in a desiccator, as described. The pH of the synthetic dough was adjusted to pH 5.1 using 1*M* acetic acid.

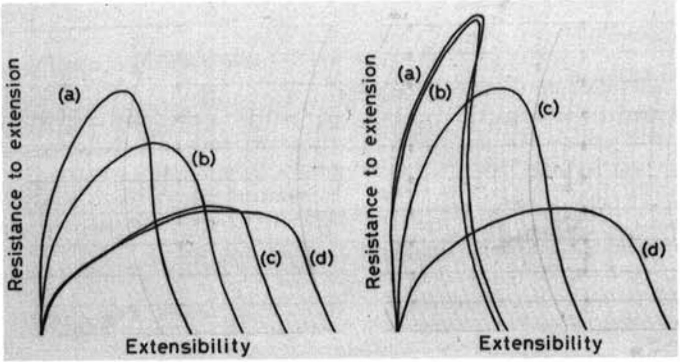


**Fig. 1.** Extensigrams of doughs made from ammonia-treated flour and flour treated with other additives. **a,** ammonia-treated flour; **b–d,** untreated flour. Additives: **b,** Kansui; **c,** $KBrO_3$; **d,** NaCl. Rest period: **left,** 45 min; **right,** 135 min.

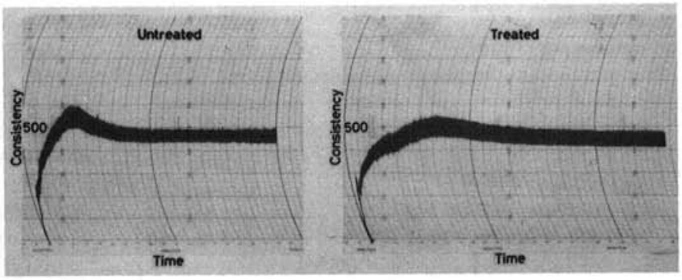


**Fig. 2.** Farinograms of doughs from ammonia-treated and untreated flour. **Left,** untreated; **right,** treated.

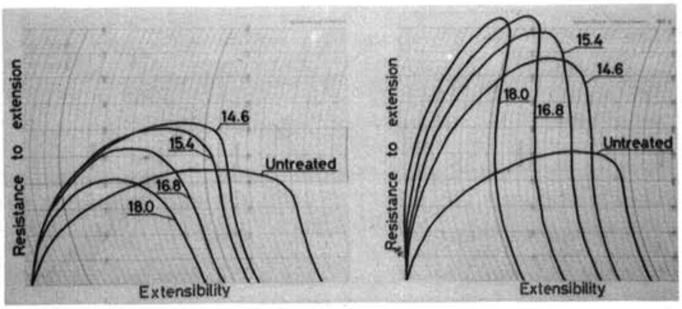


**Fig. 3.** Extensigraphs of doughs made from ammonia-treated flour moistened to various degrees. Numbers indicate percent moisture of flour treated with gaseous ammonia. Rest period: **left,** 45 min; **right,** 135 min.

## RESULTS

**Extensigraph and Farinograph Properties of Dough**

The extensigraph and farinograph properties of dough made from ammonia-treated flour were compared with those of dough

**TABLE I**
**Values of Farinograms of Doughs Prepared from Ammonia-Treated and Untreated Flours**[a]

| | Untreated | Ammonia-Treated |
|---|---|---|
| Water absorption, % | 58.7 | 63.0 |
| Peak time, min | 4.0 | 7.5 |
| Development time, min | 4.5 | 8.3 |
| Stability, min | 1.9 | 5.0 |
| Weakness, BU | 78 | 50 |
| Valorimeter value | 58 | 72 |
| Arrival time, min | 2.7 | 4.7 |
| Departure time, min | 5.8 | 13.0 |
| Mechanical tolerance index, BU | 48 | 45 |

[a] Fig. 2.

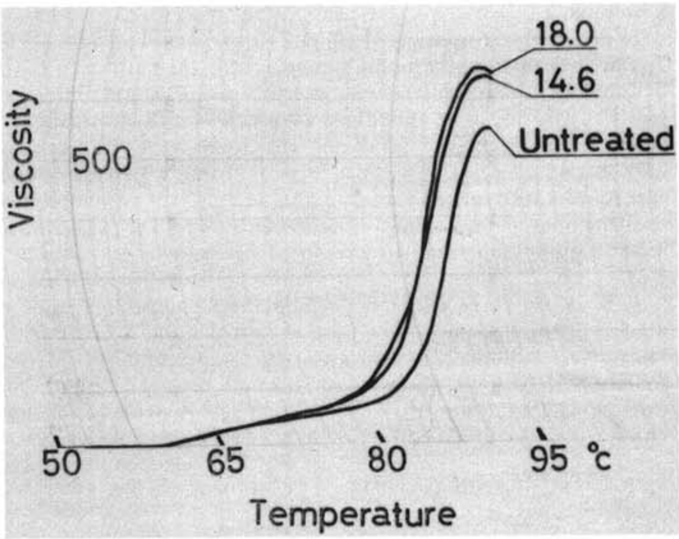


**Fig. 4.** Amylographs of doughs made from ammonia-treated flour at different moisture levels. Numbers indicate percent moisture of flour treated with gaseous ammonia.

made from untreated flour containing 6.0 g of NaCl (control), or 6.0 g of NaCl plus either 2.2 g of Kansui or 10 mg of $KBrO_3$ per 300 g of flour. Flour (moisture, 16.6%) had been treated with 50 ml of 28% aqueous ammonia in the desiccator. As shown in Fig. 1, the resistance to extension of the dough made from ammonia-treated flour was greater and the extensibility was significantly smaller than those of the control. The pH of the dough with Kansui was 9.5 and that of ammonia-treated flour was 6.3. The latter value was similar to doughs treated with $KBrO_3$ (pH 6.0) or with NaCl alone (pH 5.8).

Figure 2 and Table I show farinograph test data for the doughs of ammonia-treated and untreated flours. The flours were the same as those used for the extensigraph test. The dough of ammonia-treated flour had larger values for peak time, development time, stability, and several other items.

### Effect of Flour Moisture

Tests performed on flour adjusted to various moisture contents before ammonia treatment and on doughs made from that flour showed (Figs. 3 and 4, Tables II and III) that the higher the flour moisture, the lower was the extensibility and the higher was the resistance to extension and the area under the curve after the 135-min rest period. The effect of ammonia-treatment was more pronounced as the moisture content of the flour increased. The intensification of the effect of ammonia treatment was always accompanied by an increase of bound ammonia and a decrease of amylase activities in the flour.

Flour with moisture contents over 18%, however, produced dough with less area and less resistance to extension. This was especially true for the dough examined after a 45-min rest period. The gaseous ammonia treatment of higher moisture flour may cause destruction of the flour's dough-forming capacity.

The ammonia remaining in the flour after ammonia-treatment was not removed even on allowing the flour to stand over concentrated $H_2SO_4$ in vacuo. Therefore, this remaining ammonia was considered to exist in a bound form.

The maximum viscosity of ammonia-treated flour shown on the amylogram, expressed as Brabender units, was about 17% higher than that of untreated flour. This effect may be attributed partly to the partial inactivation of amylolytic enzymes in the flour.

The wet-gluten content decreased slightly as the flour moisture increased.

### Effect of Concentration of Gaseous Ammonia

As shown in Fig. 5 and Table II, resistance to extension and area under the curve of dough derived from the ammonia-treated flour were increased with an increase in the concentration of gaseous ammonia to which the flour was exposed. Conversely, the extensibility was decreased with increase in concentration of gaseous ammonia.

The amount of bound ammonia in the flour and the degree of the maximum viscosity on amylograms also increased with an increase in the concentration of gaseous ammonia (Table III).

Rheological properties of the dough made from mixed flour exposed to a mixture of ammonia and nitrogen gases of various ratios by the column method are shown in Fig. 6. With the doughs

**TABLE II**
**Extensigram Values at Two Rest Times (45 and 135 min) of Doughs from Ammonia-Treated Flours of Varying Moisture Content and Exposure Concentration**

| | Water Absorption (%) | Area ($cm^2$) | | Resistance to Extension ($\times 10^{-2}$ g) | | Extensibility (cm) | |
|---|---|---|---|---|---|---|---|
| | | 45 | 135 | 45 | 135 | 45 | 135 |
| Flour moisture content before treatment (%) | | | | | | | |
| 12.8, control | 60.3 | 123.4 | 116.3 | 6.7 | 7.6 | 46.7 | 40.8 |
| 14.6 | 58.3 | 128.8 | 169.6 | 9.2 | 13.0 | 35.0 | 32.2 |
| 15.4 | 58.0 | 130.1 | 169.6 | 9.0 | 14.6 | 35.0 | 32.2 |
| 16.8 | 57.5 | 91.7 | 140.3 | 7.9 | 15.5 | 31.3 | 26.2 |
| 18.0 | 57.3 | 64.2 | 103.3 | 6.1 | 15.4 | 28.0 | 19.5 |
| Flour exposed to concentration of gaseous ammonia (ppm) | | | | | | | |
| 0, control | 60.3 | 123.4 | 116.3 | 6.7 | 7.6 | 46.7 | 40.8 |
| 300 | 56.5 | 117.0 | 145.0 | 8.6 | 11.6 | 35.7 | 32.8 |
| 540 | 56.0 | 130.8 | 164.3 | 8.8 | 14.3 | 38.6 | 31.0 |
| 720 | 56.0 | 108.9 | 140.2 | 8.4 | 15.2 | 33.8 | 25.9 |
| 1,610 | 56.0 | 113.9 | 139.6 | 10.2 | 15.0 | 30.0 | 25.9 |

**TABLE III**
**Properties of Doughs from Ammonia-Treated Flours of Varying Moisture Content and Exposure Concentration**

| | Moisture After Treatment (%) | pH | Bound Ammonia (ppm) | Wet Gluten (%) | Amylase Activity (%) | | Minimum Viscosity (BU) |
|---|---|---|---|---|---|---|---|
| | | | | | Saccharifying | Dextrinizing | |
| Flour moisture content before treatment (%) | | | | | | | |
| 12.8, control | ··· | 5.80 | 34 | 30.8 | 100 | 100 | 550 |
| 14.6 | 14.3 | 6.45 | 257 | 30.7 | 89.0 | ··· | 640 |
| 15.4 | 14.2 | 6.45 | 262 | 30.7 | 84.8 | ··· | ··· |
| 16.8 | 14.4 | 6.45 | 275 | 28.6 | 81.5 | ··· | ··· |
| 18.0 | 14.4 | 6.50 | 309 | 27.5 | 74.5 | 24.0 | 650 |
| Flour exposed to concentration of gaseous ammonia (ppm) | | | | | | | |
| 0, control | ··· | 5.80 | 34 | 30.8 | 100 | 100 | 550 |
| 300 | 13.2 | 6.20 | 191 | 30.9 | 88.0 | ··· | 630 |
| 540 | 13.5 | 6.37 | 202 | 29.6 | 82.0 | ··· | ··· |
| 720 | 13.3 | 6.30 | 216 | 29.5 | 84.6 | ··· | ··· |
| 1,610 | 13.4 | 6.35 | 247 | 29.5 | 81.1 | 27.0 | 620 |

made from the flour exposed to gas mixtures of 1:4 and 1:10 ($NH_3/N_2$, v/v), the extensibility was decreased and the resistance to extension was increased as compared with these characteristics in dough made from untreated flour. The resistance to extension was decreased, however, by exposure to gaseous ammonia alone. A similar tendency was also observed for the area under the curve for doughs.

The effect of the contact-period of flour with gaseous ammonia was also studied. A mixture of ammonia and nitrogen gases (1:1, v/v) was blown through the packed-flour column for 60–90 min. Both the extensibility and resistance to extension of the dough made from the flour exposed for 90 min were significantly lower than those from the flour exposed for 60 min (Fig. 7). A similar result was observed in the dough made from flour treated with gaseous ammonia alone ($NH_3/N_2$, 1:0; Fig. 6). Treatment of flour with gaseous ammonia at high concentrations or for long times thus led to a loss of viscoelasticity of dough.

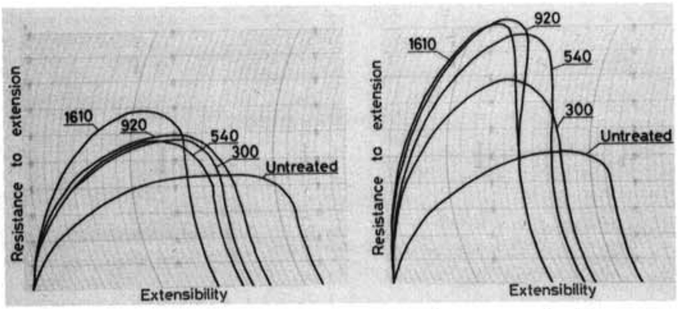


**Fig. 5.** Extensigraphs of doughs made from flour treated with varying concentrations of ammonia. Numbers indicate concentration of gaseous ammonia, ppm, in the flour-treament desiccator. Rest period: **left,** 45 min; **right,** 135 min.

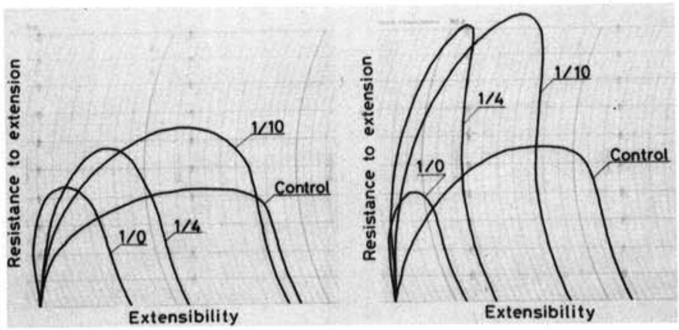


**Fig. 6.** Extensigraphs of doughs made from flour treated with various ratios of gaseous ammonia to nitrogen gas as gases were passed through a flour column. Numbers indicate the mixing ratio of ammonia to nitrogen gas. Rest period: **left,** 45 min; **right,** 135 min.

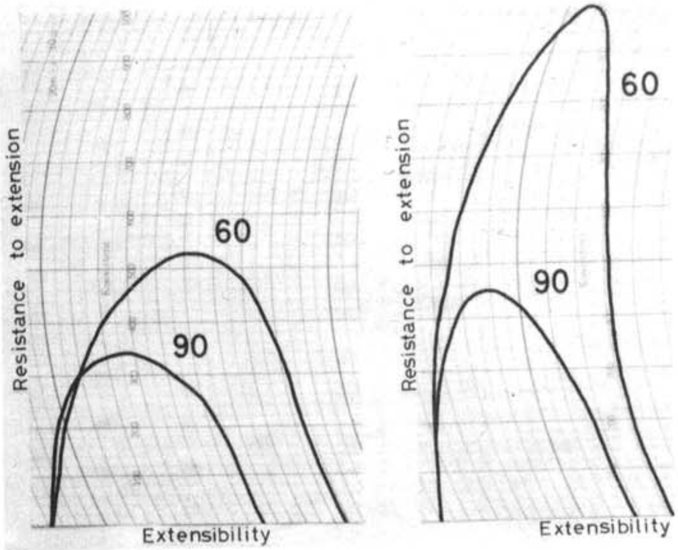


**Fig. 7.** Effect of period of exposure on extensigraphs of the doughs prepared from flour exposed for various lengths of time to a mixture of ammonia and nitrogen gases. Numbers indicate the exposure period, in minutes, of a mixture of ammonia and nitrogen gases passed through a flour column. Rest period: **left,** 45 min; **right,** 135 min.

## Structural Relaxation of Dough

Figure 8 shows the result of the structural relaxation test on doughs made from flour of various moisture contents and treated with several concentrations of gaseous ammonia. The dough made from ammonia-treated flour was greater in both relaxation constant and asymptotic load than was the control dough. A similar result was observed between the control and the dough prepared using Kansui (Terada et al 1978a) and oxidizing agents (Dempster et al 1955, Hlynka 1955, Hlynka and Matsuo 1959), suggesting that the three-dimensional structure of dough was highly developed by treating flour with gaseous ammonia or by adding Kansui or $KBrO_3$.

## Noodle Texture

The cohesiveness and hardness of noodles prepared from ammonia-treated flour was greater than those of noodles prepared using NaCl alone, and comparable to those of noodles manufactured using Kansui and NaCl (Fig. 9).

Noodles produced from ammonia-treated flour showed less variation in quality than did those manufactured using NaCl alone, judging from the area surrounded by the lines.

## Stability of Treated Flour

The flour treated with gaseous ammonia and stored for nine months in a kraft bag at room temperature ranging from 10 to 30°C was shown to be completely stable, as judged from the rheological properties of dough made from the flour.

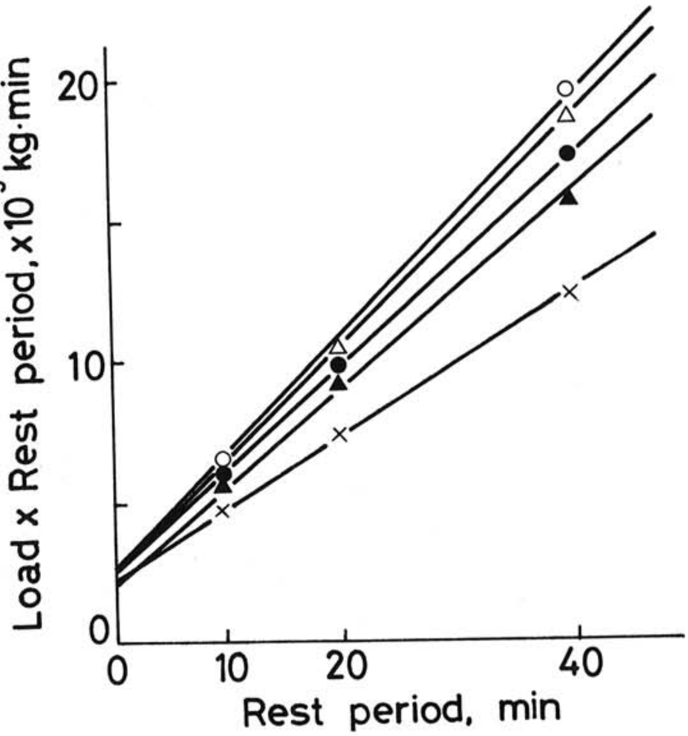


**Fig. 8.** Linear transformations of structural relaxation curves of doughs made from flour moistened in various degrees and treated with two different concentrations of gaseous ammonia. Concentrations of gaseous ammonia (ppm) and moisture (%), respectively: ---o--- = 1610, 16.4; ---•--- = 920, 16.4; ---×--- = 0, 12.8; ---Δ--- = 1610, 15.4; ---▲--- = 1610, 18.0.

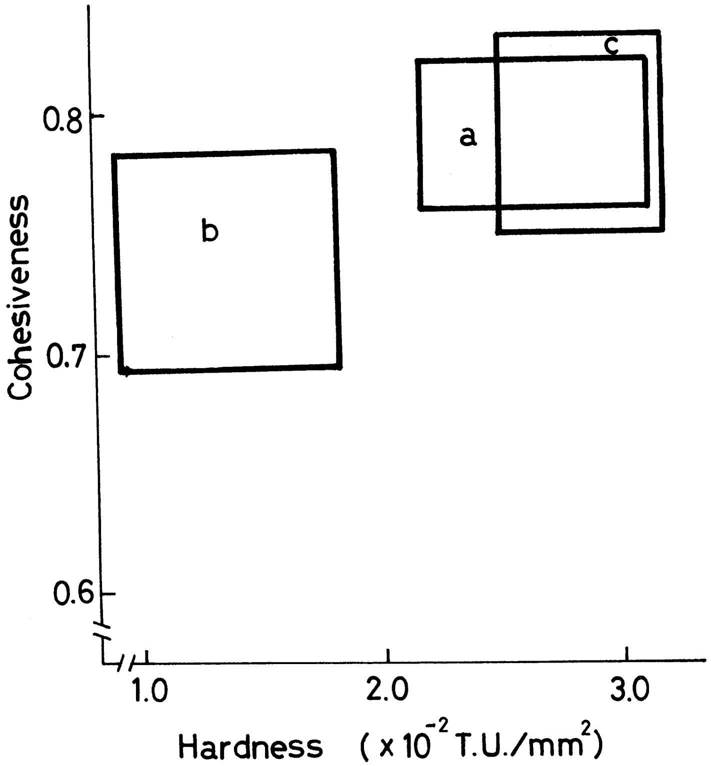


**Fig. 9.** Texturograms of air-dried noodles made from ammonia-treated flour. Contents of noodles: **a,** 30 parts of ammonia-treated flour and 70 parts of untreated flour; **b,** untreated flour; **c,** flour containing Kansui.

## DISCUSSION

In the manufacture of noodles and breads, the addition of Kansui or some oxidizing agents to dough has been widely used to increase the resistance to extension and area under the curve and to decrease the extensibility. These additives present problems, however. The dough with added Kansui is alkaline (pH $> 9$), and thus the use of Kansui is at present limited to "Chinese noodle," "Alpus-steamed-bread," etc. The alkaline reaction of dough may induce Maillard and lysino-alanine reactions. The formation of lysino-alanine complex (De Groot and Slump 1969, De Groot et al 1976, Woodard and Short 1973) and the presence of oxidizing agents (Hall 1976) in dough are undesirable from the viewpoints of hygiene and nutrition. The treatment of flour with gaseous ammonia, of course, requires further investigation regarding such problems as lysino-alanine complex formation and the nature of bound ammonia in flour. For example, the bound ammonia may be metabolically consumed if the dough is fermented by adding yeast.

A mixed flour (30% of ammonia-treated flour) produced a noodle similar in texture to that manufactured using Kansui. This may indicate that the mixed flour alone can yield the desired properties for dough without the use of strong alkali or oxidizing agents.

In addition, the characteristic properties induced in the flour by exposure to gaseous ammonia were preserved for at least nine months of storage in a kraft paper bag at room temperature.

This method may thus be useful for flour produced, for example, in Brazil and Argentina for manufacturing certain specific products (eg, oriental noodles and breads) because the flours from the wheat grown in these countries usually have lower resistance to extension but large extensibility.

A synthesized dough made from a mixture of wheat starch and wheat gluten isolated and treated with gaseous ammonia showed rheological properties similar to those of the mixed dough. However, the synthetic dough manufactured using the gluten without treatment with gaseous ammonia showed none of these characteristic properties. This fact may indicate that the principal component modified by exposing flour to gaseous ammonia is protein. Mechanisms involved in modification of flour protein by gaseous ammonia and the relation of treatment to the rheological properties of the dough obtained is a subject for future investigation.

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