USE OF HYDROGEN PEROXIDE TREATED NONFAT DRY MILK IN THE CONTINUOUS DOUGH MIXING PROCESS

by

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INTRODUCTION

The baking industry is one of the major domestic, non-governmental buyers of nonfat dry milk. For several years, the baking industry has purchased more than one-fourth of the total production of nonfat dry milk available in the United States. A considerable portion of the nonfat dry milk used by the baking industry has gone into the production of white pan bread. Six percent nonfat dry milk, based on the flour weight, is commonly used in the bread formula made by the sponge dough method. From a nutritional standpoint, this is approximately equivalent to using liquid skim milk in the formula in place of water.

Skim milk, or the equivalent amount of nonfat dry milk, was originally added to bread to enhance its nutritional value. Several secondary benefits also were noted; namely, increased water absorption, greater mixing tolerance, improved shelf life, fermentation or oxidation stability, dough strengthening, a softer texture, more uniform grain, a creamy crumb color, and golden brown crust color (15, 36, 51). However, the initial use of skim milk did pose a serious problem with respect to loaf volume and dough softening. It was demonstrated in 1927 (14) that the loaf volume depressing factor could be destroyed by scalding the skim milk prior to its incorporation into bread dough. It was noted that the subsequent use of nonfat dry milk in bread also had a loaf volume depressing effect unless the milk had been sufficiently heated prior to drying. A considerable amount of research effort has gone into the study of the loaf volume depressing factor during the last three decades. It is generally accepted that the bakery grade nonfat dry milk must receive a minimum heat treatment of 80 C for 20 min (3).

In 1954, a continuous dough mixing process was developed to replace, in part, the conventional sponge dough process. Many problems were encountered when 6% nonfat dry milk was used in the formula with the continuous dough mixing process. It is reported that the use of more than 1% nonfat dry milk in the bread formula in the continuous dough mixing process resulted in a bread with decreased loaf volume, coarse grain, dull crumb, and harsh texture (8). Reports from industry also indicate that the power requirement for dough development increased significantly when 6% nonfat dry milk was used in a formula.

Approximately 30% of the white bread produced in this country is made by the continuous dough mixing process. The inability to use nonfat dry milk in the continuous dough mixing process in amounts commonly used in the conventional sponge dough process has posed some problems for the dairy industry (9).

Several investigations have been undertaken to study the incompatibility of nonfat dry milk with the continuous dough mixing process (4, 9, 51, 55). New knowledge in the field of cereal chemistry has shown that the sulhydryl compounds play an important role in dough formation, development and fermentation (32). Also of significance in attempting to interpret the fundamental problem is the recent knowledge of milk protein interaction upon heating (56) and the possible further interaction between the milk protein and wheat flour protein during dough development and fermentation (6).

This investigation was undertaken to study the following: (A) effect of oxidation of sulfhydryl compounds of milk by hydrogen peroxide before drying; (B) compatibility of nonfat dry milk produce as in (A) with the continuous dough mixing process when used in the bread formula at 3% and 6% of the flour weight.

The compatibility of nonfat dry milk with the continuous dough mixing process was studied with respect to volume, texture, grain and total score of bread and the power requirement for dough development. The information obtained from this study should help to open new avenues in understanding the problems of continuous dough mixing process using levels of nonfat dry milk equivalent to those used in the conventional sponge dough process.

LITERATURE REVIEW

Problems related to the baking quality of nonfat dry milk (NFDM) involves the chemistry of milk and wheat flour proteins. It is widely known that skim milk must be heat-treated prior to drying in order to produce NFDM with a good baking quality. In 1954, Jenness (18) reviewed the effect of heat treatment on serum proteins and its relations to the baking quality of NFDM. Since then, the literature has been enriched with new knowledge of the interactions of milk proteins on heating (31, 43, 56).

Sullivan (47) reviewed the literature pertaining to the reactive groups involved in the oxidation-reduction systems of dough. Considerable information has been reported in recent years regarding the chemical changes occuring during dough mixing, formation and relaxation (32, 34, 54).

Heat Induced Changes In The Milk

Greenbank et al. (14) were among the first to report the beneficial effect of heat treatment on skim milk before condensing. The heat treatment removed the problem of a loaf volume depressing factor and it also increased the water-holding capacity of NFDM. Grewe and Holm (15), Shovholt and Bailey (44), and Johnson and Ward (21) verified and extended this work.

The preheat treatment of 73 C for 30 min was found to be adequate for producing NFDM of a good baking quality. Ashworth and Krueger (3) indicated that 80 C for 30 min yielded NFDM capable of improving loaf volume. The latter treatment was accepted by industry for producing "high heat" or "bakery grade" NFDM.

Rowland (40, 41) reported the denaturation of milk serum proteins on heating the milk between 75 C and 100 C for 30 to 60 min. The coincidence of preheat treatment given to bakery grade NFDM with this wide heat treatment range, led to the conclusion that the serum protein denaturation itself was responsible for the improvement of baking quality. The serum protein denaturation was found to be proportional to the amount of heat treatment given to the skim milk. Harland and Ashworth (17) developed a test to measure the undenatured whey protein nitrogen as an index of heat treatment. This led to a development of standards specifying the amount of undenatured whey protein nitrogen permissible per gm of NFDM. The "high heat" classification allows not more than 1.5 mg undenatured whey protein nitrogen (WPN) per g of NFDM (1).

Skovholt and Bailey (45) observed that the unheated milk solids caused a slackness and extreme extensibility of the dough. It was thought that milk contained a factor which breaks down the gluten matrix. They further noticed that casein did not carry this factor. Larsen et al. (25) confirmed that this factor remained in the serum protein after removal of casein by acid or rennet. Harland et al. (16) proved that this factor was associated with the protein fraction as it was nondializable. Jenness (18) reviewed the findings of the fractionation studies, and concluded that serum protein carried the loaf volume depressing factor. B-lactoglobulin, alpha lactalbumin, serum albumin, euglobulin and pseudoglobulin were reported as devoid of this factor. The lactoglobulin precipitated by the classical method at 43% saturation with ammonium sulfate was found to carry this factor (13). The fraction contained about 40% euglobulin and pseudoglobulins did not depress the loaf volume. Thus, they indicated that the loaf volume depressing factor resided in the remaining 60% globulin.

Larsen et al. (24) studied this problem by fractionating skim milk into casein, lactose and serum protein. All the unheated fractions when incorporated in the formula depressed the loaf volume, but the effects of serum proteins were more pronounced. Proper heat treatment of these fractions did not alter the loaf volume depressing effect except in the case of the serum protein fraction. They also reported that the heating reduced the sulfhydryl content of the skim milk. The decrease in sulfhydryl content by heating paralled the improvement in the baking properties of NFDM.

Larson et al. (27) fractionated serum proteins and studied euglobulin, pseudoglobulins, serum albumin, and B-lactoglobulin individually for the loaf volume depressing factor. The B-lactoglobulin fraction contained most of the sulfhydryl groups of milk and was more intensely studied. None of the individual fractions was deleterious. The entire serum protein fraction depressed the loaf volume but it was overcome by heating. Heat treatment of pure B-lactoglobulin exposed the previously unreactive sulfhydryl groups which were deleterious to the baking quality.

Larson and Rolleri (28) identified a fraction from acid whey of heated milk by electrophoretic techniques. This component appeared as the fifth peak and so it was called "component 5". Jenness (19) isolated "component 5" from unheated milk by sodium chloride saturation and acid precipitation. Fractions rich in this component were found to reduce the loaf volume. The harmful effect on loaf volume was reduced by heating.

McGugan et al. (31) reported the possibility of a heat induced interaction between B-lactoglobulin and alpha casein. Trautman and Swanson (52) and Zittel et al. (56) added further support to this concept. Sawyer and Coulter (43) indicated that the sulfhydryl groups of B-lactoglobulin are exposed during heating. This facilitated the reaction of the disulfide bonds of K-casein during heating as K-casein lost its ability to stabilize $alpha_s$-casein against rennin. Barrett (5) reported that K-casein itself was not harmful to the dough properties on loaf volume.

Dough Properties

Sullivan (47) reviewed the literature regarding oxidation reactions and the changes in the dough structure during mixing and fermentation. New information relating to oxidation and dough structure, has been reported in the last decade. The studies on the role of soluble proteins, sulfhydryl groups and sulfhydryl-disulfide interchange, and other flour components in the dough structure led to a better understanding of basic dough properties (11, 32, 35, 54).

The flour proteins are fairly unreactive in a dry state. However, the soluble proteins may be removed by water extraction. With the mechanical mixing action, the insoluble proteins are brought into proximity to form a coherent dough mass. The hydrated gluten protein structure possesses considerable elasticity (7). Like other elastic and visco elastic substances, the gluten structure consists of long protein chains with occasional cross-bonding. The disulfide cross-linkages are predominant in the gluten structure. The other cross-linkages are thiol ester, amide, and hydrogen bonds (48).

The soluble protein contains a larger number of sulfhydryl groups than the gluten. Incorporation of soluble protein into the gluten structure during mixing causes an equilibrium between the two types of proteins. This equilibrium is very important for visco elastic properties of gluten. Oxidation reduction reactions of dough have a direct bearing to this equilibrium and so oxidation reduction balance is of importance for a stable structure of dough.

The sulfhydryl-disulfide interchange, through reactions of disulfide linkage of gluten and sulfhydryl groups of non-gluten, acid, and water-soluble constituents, is important to dough formation (46). Sullivan et al. (47) reported that the intra-molecular disulfide bonds are likely changed to intermolecular disulfide bonds and that this change was thought to be capable of conferring toughness and greater resistance to extension. Lee and Reynolds (23) reported that this interchange is important to gas retention of dough and hence it affected the loaf volume of bread. Hlynka (7) reported that this interchange may take place as follows:

$$R_1\ SSR_2 + XSH \rightleftharpoons R_1\ SSX + R_2SH$$

$$R_1\ SSX + R_3SH \rightleftharpoons R_1\ SSR_3 + XSH$$

In the above reactions, XSH acts as a catalyst. If XSH is a reducing agent, this reaction may be detrimental, but if it is a water-soluble -SH containing protein, the reaction may be beneficial. If water-soluble protein mediates the incorporation of R_3, the reaction may be beneficial. Sullivan et al. (48) demonstrated a rapid loss of sulfhydryl groups when dough was mixed. They also reported the maximum improvement in the rheological properties of dough on addition of specific sulfhydryl blocking reagents equivalent to half of the titrable sulfhydryl groups. This type of interchange is also reported for other proteins (20).

In dough mixing, considerable stress is imparted by straining the intermolecular disulfide bonds and creating new ones. The dough must retain a stable structure in order to produce a good bread. The dough stress is relaxed through sulfhydryl-disulfide interchange. The absence of sulfhydryl groups and thus, a reduced amount of interchange, will result in the undesirable "tight" dough properties. There will be possibly a limited amount of interchange through some other reducing groups. This interchange is detrimental to dough properties, and will lead to a break down of dough resistance to mixing. On the other extreme, if too many sulfhydryl groups are available for the interchange, it will produce a highly extensible dough. The dough will be slack and gas retention in the dough will be poor. This will cause a loss of loaf volume and grain structure (49). Mechan (32) observed that the incorporation of sulfhydryl blocking agents in the dough causes a slight reduction in time to maximum resistance, a slight increase in maximum resistance, and a great increase in the break down after the maximum resistance is reached.

Water-soluble proteins are important in the dough rheological properties of dough and in final baking performance of the dough. Pence et al. (38) reported a favorable effect of the albumin fraction of soluble protein on loaf volume. This was confirmed by Mattern and Sandstedt (30), and Finney (10). The exact mechanism of the reactions of soluble proteins is not well known, but it is assumed that the soluble proteins are involved in dough properties through the sulfhydryl groups and participate in the sulfhydryl disulfide interchange.

Flour lipids also influence in the dough properties. Tsen and Hylnka (53) observed that normal flour dough lost its sulfhydryl groups faster than did defatted flour dough when mixed in air. They attributed the difference to the oxidation of the lipids in dough

Mecham and Kanpp (33) observed that the inclusion of NFDM at 3% and 6% levels, based on flour weight, protected the loss of the sulfhydryl groups during mixing. They speculated that this was due to some interaction between NFDM protein and flour protein, which protected the loss of sulfhydryl groups during mixing or perhaps, that the sulfhydryl groups of the milk might be preferentially oxidized. Swanson et al. (50) observed that when two different flours were used to calculate the water absorption of the same NFDM samples, different results were obtained. They assumed that the flour proteins exerted some effect on NFDM. In 1965, Bernardin et al. (6) reported that wheat flour has a proteolytic action on NFDM.

They demonstrated by starch-gel electrophoresis that alpha$_s$ casein was altered. It was also reported that the prolonged digestion caused the alpha$_s$ casein component to disappear completely.

THE PROBLEM

The concept of the sulfhydryl-disulfide interchange has led to the belief that the sulfhydryl groups of the milk protein may be important to dough fermentation. Most of the studies of the baking quality of NFDM and the milk protein fractions have been done with the sponge dough method. However, with the continuous dough mixing method, the total fermentation time, after the dough is mixed, is short as compared with the sponge dough method. It is possible, with the continuous dough mixing process, that all the reducing systems of milk are not oxidized as they may be in the sponge dough method. This may be the cause of the difficulties, such as loss of volume, loss of fine grain structure, dull crumb color, and soft texture, (8) which are thought to originate with the use of NFDM in the continuous dough mixing process.

MATERIALS AND METHODS

The experimental procedure used in the study can be classified under seven categories as follows:

Preparation of NFDM.

Determination of sulfhydryl content of reconstituted NFDM.

Determination of residual peroxide in the reconstituted NFDM.

Determination of undenatured whey protein nitrogen in NFDM.

Determination of flour and flour plus NFDM absorption for 500 Brabender Units (BU) consistency.

Baking Procedure.

Evaluation of Bread.

Preparation of NFDM

The milk used in preparation of NFDM, was obtained from the Kansas State University Dairy. NFDM samples were prepared from winter and spring milk. The winter milk was designated as milk No 1. Two series of NFDM samples were prepared from winter milk and were designated as series A and B to distinguish NFDM prepared on different days. The spring milk was designated as milk No 2, and one series only of NFDM samples was prepared.

For milk No 1, series A, two hundred gal of unheated skim milk were divided into four equal batches. Batch one was condensed at 21 - 24 C to approximately 40% solids in a single effect Mojonnier Lo-Temp evaporator. The concentrated skim milk was introduced into a Mojonnier spray dryer at 450 psi pressure. The inlet temperature on the dryer was 205 C and the outlet temperature was 105 C. This sample was designated as control.

The second batch was subjected to a preheat treatment of 88 C, for 30 min in a 200 gal Creamery Package Multi Process vat, and cooled to 21 C, condensed and dried as described above. This sample was designated as Heated 190 F/30 min.

To the third 50 gal batch, 643.5 ml of 30% hydrogen peroxide were added and allowed to react with the milk at 4 C for 10 min. During this time, the milk was stirred by a vat agitator. At the conclusion of the timed reaction period, steam was turned on and the milk was preheated, cooled, concentrated and dried as described for batch two. This NFDM sample was designated 190 F/30 min, 0.1% H_2O_2.

For milk No 1, series B, 250 gal of unheated skim milk were divided into five equal batches. The first four batches were subjected to the same treatment as described for series A samples, and the samples produced were designated as before. The fifth batch of milk was subjected to the same treatment as described for batch three except that the hydrogen peroxide concentration was reduced to one half that of the third batch. This sample was designated as 190 F/30 min, 0.05% H_2O_2.

For milk No 2, produced during the spring when the cows were on pasture, 200 gal of skim milk were divided into four equal batches. The unheated control sample described for milk No 1, series A and B, was not prepared. The preheat treatment, hydrogen peroxide addition, concentration, and drying treatments were the same, with the samples being designated 190 F/30 min, 190 F/30 min, 0.1% H_2O_2, 190 F/30 min, 0.2% H_2O_2, and 190 F/30 min, 0.05% H_2O_2.

All the NFDM samples thus prepared were bagged and stored at 4 C.

Determination of Sulfhydryl Content of Reconstituted NFDM

Analysis of sulfhydryl content of milk varies depending on methodology. Most of the methods have one or more limitations. Sasago et al. (42) modification of Fridovich and Handler's (12) p-chloromercuribenzoate-dithiazone (PCMB-D) method which was further modified by Nakai et al. (37) provides a simple, accurate method for the determination of sulfhydryl content in milk. This method was applied, with a further minor modification involving filteration, for determination of sulfhydryl content of reconstituted NFDM samples. The stock reagents used were as follows:

<u>PCMB solution</u>; A 0.5 mM solution of the sodium salt of p-chloromercuribenzoic acid (Nutritional Biochemical Corporation) was prepared fresh daily.

<u>Dithiazone solution</u>; 2.4 mg of diphenyl carbazone (dithiazone, Fisher Scientific Company) was dissolved in carbon tetrachloride and the volume made to 100 ml. Thirty-six to 38 ml of this solution were further diluted to 200 ml with carbon tetrachloride. The stock and diluted solution were made fresh on the day of the experiment.

Urea buffer solution; 36.0354 g of urea, 10 ml of 1M phosphate buffer pH 7.0, 10 ml of 0.5M sodium sulfate and 37.2 mg of disodium ethylenediamine-tetraacetate (EDTA) were mixed and the volume made to 100 ml with distilled water. The pH of this buffer solution was adjusted to 7.0 with 50% v/v sulfuric acid.

Reconstitution of NFDM; Ten g of NFDM were dissolved in 100 ml of glass distilled water. The reconstituted samples were allowed to equilibrate overnight at 4 C before the determination of sulfhydryl content was begun.

Standard cysteine solution; A 0.5 mM solution of cysteine monohydrochloride was prepared for the establishment of the standard curve.

The final preparation of the stock solutions for purposes of establishing the standard curve and determining sulfhydryl content of NFDM samples are summarized in table 1.

Table 1. Standard curve of sulfhydryl content in NFDM

Tube	0.5 mM cysteine solution ml	10% reconstituted NFDM ml	Distilled water ml	0.5 mM PCMB sol. ml	Urea buffer ml
1	0.00	0.00	0.25	0.25	1.00
2	0.04	0.00	0.21	0.25	1.00
3	0.08	0.00	0.17	0.25	1.00
4	0.12	0.00	0.13	0.25	1.00
5	0.16	0.00	0.09	0.25	1.00
6	0.20	0.00	0.05	0.25	1.00
7	0.25	0.00	0.00	0.25	1.00
8	0.00	0.25	0.00	0.25	1.00
9	0.00	0.25	0.00	0.25	1.00

Ten ml of diluted dithiazone solution were added to each tube. The solutions were shaken vigorously by hand for 1 min and then allowed to stand for 30 min in an ice-water bath at 1-2 C. At the end of 30 min, the carbon tetrachloride layer was drawn off with a pipette and was filtered through Whatman No 40 filter paper. The color intensity was measured at 620 mμ wavelength using a Beckman Spectrophotometer, Model DU.. Carbon tetrachloride was used as the blank.

A standard curve was plotted (figure 1), and the sulfhydryl content of reconstituted NFDM samples was calculated from this curve. A standard curve was established for each experimental determination.

Determination of Residual Peroxide in the Reconstituted NFDM

Barry et al. described a method using titanium sulfate for determination of hydrogen peroxide in biological material. Amin and Olson (2) modified this method for its application to milk. This modified method was used for determination of residual peroxide. The following reagents were used:

Seventy percent (w/v) trichloroacetic acid. (TCA)

Titanium dioxide solution; 2 to 3 g of titanium dioxide were dissolved in 100 ml of concentrated sulfuric acid by boiling in a Kjeldahl flask over a hot plate. During slow heating, the solution was shaken continuously by hand to avoid settling of undissolved residue. The solution then was cooled to room temperature, and the clear liquid decanted and the volume measured. An equal volume of distilled water was added to the clear acidic solution. The diluted solution was allowed to equilibrate overnight at room temperature and centrifuged at 3000 x g for 30 min to remove the undissolved residue. This solution was very stable.

Reconstitution of NFDM; NFDM samples were reconstituted as described in the determination of sulfhydryl content.

Standard hydrogen peroxide solution; a solution containing 3750 ug/ml of 30% hydrogen peroxide was prepared. This solution was further siluted with reconstituted milk to give a range of concentration of hydrogen peroxide for establishing the standard curve.

Two ml of 70% TCA solution were added to 3 ml of reconstituted milk to precipitate the milk proteins. After 2 min and not more than 3 min, the solution was diluted to 100 ml with distilled water and filtered through two thicknesses of Whatman No. 42 paper.

Two ml of titanium dioxide reagent were added to 5 ml of TCA-milk filtrate and allowed to stand for 5 min. The samples containing hydrogen peroxide developed a yellow color, the intensity of which was directly related to the concentration of hydrogen peroxide present. The intensity of yellow color was determined at 400 mμ in a "Spectronic 20" colorimeter.

The concentration of residual peroxide in NFDM samples was calculated from a standard curve (figure 2).

Whey Protein Nitrogen

The whey protein nitrogen content was determined by the Harland and Ashworth (17) method as modified by Kuramoto et al. (22).

Two g of NFDM were reconstituted in 20 ml of distilled water. Eight g of sodium chloride then were added to the reconstituted NFDM. The tube was stoppered with a rubber stopper, and placed in a water bath at 37 $\pm$ 0.1 C for 30 min. The tubes were shaken 8-10 times within the first 15 min of incubation. At the end of the incubation period, without further cooling, the contents of the tubes were filtered through S and S No 602 (diameter 9 cm) filter paper to remove casein and denatured whey protein. Approximately 5 ml of the filtrate were collected.

One ml of filtrate was pipetted into 50 ml spectrophotometrically-matched tubes, and diluted with 10 ml of saturated sodium chloride solution. The undenatured whey protein was precipitated by 2 drops of hydrochloric acid (23 ml of concentrated hydrochloric acid diluted with distilled water to make a volume of 100 ml). The turbidity thus formed was measured at 420 mμ wavelength. The blank was prepared and adjusted to 100 percent transmittance by diluting a solution composed of 1 ml of the casein free filtrate and 10 ml of saturated sodium chloride solution.

For the preparation of a standard curve, 20 g of high heat and low heat NFDM, obtained from the American Dry Milk Institute, were each reconstituted with 200 ml of water. These samples were subjected to the whey protein nitrogen test described above. The following samples were prepared using different aliquots of the filtrate from low and high heat reconstituted NFDM:

Sample No.	Low heat filtrate ml	High heat filtrate ml
1	10	0
2	8	2
3	6	4
4	4	6
5	2	8

The tubes containing combined filtrated were stoppered and mixed by inverting the tube twice. The turbidity was developed and measured as described above. The serum protein nitrogen values were determined by the Kjeldahl method. A standard curve (figure 3) was plotted.

Determination of Absorption of Flour and Flour Plus NFDM

A Brabender farinograph unit with a 300 g brass mixing bowl was used for this purpose. The farinograph thermostat was turned on 1 hour prior to the start of the experiment so that the circulating water and jacketed mixer were equilibrated at 30 C. The amount of water, in percentage, required to develop dough consistency of 500 BU was designated as the absorption of the sample.

For flour absorption, 300 g of flour at room temperature, were transferred to the mixing bowl. The mixing unit was turned on after adjusting the recording pen on the chart. The flour was mixed for 1 min so that it reached 30 C. At the end of 1 min, the amount of distilled water necessary to obtain 500 BU at the peak consistency, was added to the flour. The traces of flour, water and dough were removed from the sidewalls of the mixing bowl with the aid of a plastic scraper, and were mixed with the forming dough. The bowl was covered with a glass plate and the dough was allowed to mix for 20 min. The farinograph thus obtained was labeled as flour standard.

The absorption for flour plus NFDM was determined using 3 and 6% levels of NFDM based on flour weight. Three hundred g of flour were mixed with 9 g of NFDM for the 3% level, and with 18 g of NFDM for 6% level. The flour and NFDM samples were mixed with a spatula before transferring to the mixing bowl. The farinograms were obtained as described before.

The peak time in min, valorimeter value and mixing tolerance index (MTI) in BU were measured from the farinograms of the samples (7). The farinograms were mounted, labeled and photographed.

The farinograms were obtained from the flours used in baking, and flour plus all NFDM samples of milk No 1, series A and B, and milk No 2.

Baking Procedure

The sponge dough and the continuous dough mixing process were used. The NFDM samples of milk No 1, series A and B, were used for the sponge dough process, while the NFDM samples of milk No 1, series B, and milk No 2 were used with the continuous dough mixing process. The control sample of milk No 1, series B, was not used with the continuous dough mixing process.

For the sponge dough method, the flour contained 11.2% moisture, 0.42% ash, and 12.3% protein. The formula used is shown in table 2.

Table 2. The bread formula used for sponge dough process

Ingredient	Percentage on flour weight	Total weight g	70% sponge g	30% dough g
Flour	100.00	700	490	210
Yeast	2.0	14	14	---
Arkady	0.5	3.5	3.5	---
Malt	0.5	3.5	3.5	---
Sugar	5.0	34	--	35
Salt	2.0	14	--	14
Shortening	3.0	21	--	21
NFDM	var 3.0 or 6.0	21 or 42	--	21 or 42
Water	var			

The variations of water used in the formula were obtained from the farinograms of NFDM using the same flour as in the dough mixing process.

The sponge part of the ingredients was mixed for 2 min using No 2 speed on Hobart, Model A-200, dough mixer. Mixed sponges were fermented for 4 hours at 30 C and 90% relative humidity. Upon completion of a predetermined fermentation, the sponges were remixed with the remaining ingredients to optimum development, and then given additional 30 min fermentation. The dough was then divided and rounded into 200 g pieces in duplicate and allowed 15 min intermediate proof at room temperature and atmospheric humidity. After intermediate proof, the dough was molded and placed in the dough pan with identification tags. The final proofing was done in an Anetsberger proof cabinet at 35-38 C and 90% relative humidity, to a constant height of three-quarters of an inch above the pan's top edige. If dough failed to rise to predetermined height, it was proof for 65 min maximum. The proofed dough was baked at 205 C for 30 min. The baked bread was allowed to cool for 1 hour at room temperature before the volume was measured by seed displacement. One sample of each duplicate was saved for photographic purposes (Appendix figure 1 and 2). The evaluation of the baked bread was done as described later.

For the continuous dough mixing process, the AMF laboratory continuous dough mixing unit was used. The flour with 11.2% moisture, 0.42% ash, and 12.3% protein was used in baking the bread with NFDM samples of milk No 1, series B. The flour used, in baking bread with NFDM samples of milk No 2, contained 11.9% moisture, 0.41% ash, and 12.2% protein. The formula and proportions used in the sponge and Amflo process appear in table 3:

Table 3. The formula and proportions used in the sponge and Amflo process.

Ingredient	Percentage on flour weight	Sponge phase I	Sponge phase II	Amflo phase
Flour	100	var	4	100
Water	var	3		var
Yeast	3	0.5		
Arkady	0.5	0.25		
Malt	0.25			
Salt	2		2	
Sugar	7	2		5
Milk	var 3 or 6			var 3-6
Shortening	3			3
Oxidation (ppm)	var 50, 70, or 90			var 50, 70, or 9(

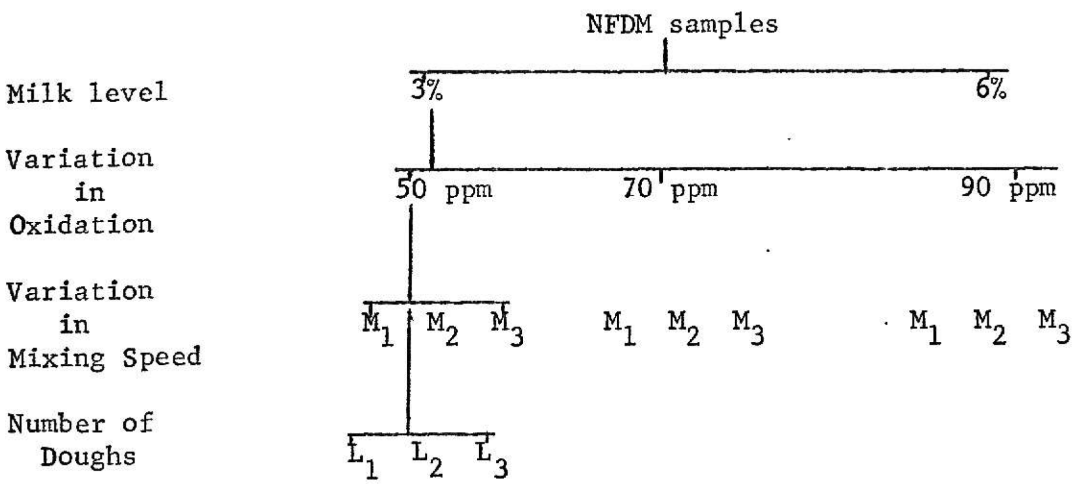
The oxidant used was a mixture of potassium bromate and potassium iodate. The potassium bromate contributed 75% of total oxidation while potassium iodate contributed the remaining 25%. The dry mixture of the oxidant was dissolved in water 1 hour prior to its use in the Amflo process.

The water absorptions used in the baking of bread containing NFDM samples of milk No 1, series B were arbitrarily fixed at 70% when NFDM was used at 3% level and 73% when used at the 6% level. The variations in water absorptions used in baking the bread containing NFDM samples of milk No 2, were obtained from the farinograms.

The sugar in the sponge phase or the Amflo process was reduced from 2% to 1% in baking the bread with NFDM samples of milk No 2. In the Amflo process, the sugar was increased to 6%.

The ingredients of sponge phase I of the Amflo process were mixed and were allowed to ferment for 150 min. After 60 min of fermentation, the ingredients of sponge phase II were mixed with the sponge phase I ingredients. At the end of the predetermined sponge fermentation, it was incorporated in the dough.

The following statistical design was used in evaluating the baking quality of NFDM samples:



M_1, M_2, and M_3 designate three different mixing speeds. L_1, L_2, and L_3 designate triplicate loaves.

The same design was used for the 6% milk level. The mixing speeds used were predetermined. The dough was allowed to develop to a maximum. Three doughs were cut off and panned. The fermentation, proofing and baking of doughs were done as described in the sponge dough. The pannary fermentation was done at 98% relative humidity and baked at 232 C for 25 min.

Evaluation of Bread. The volume of a 1 pound loaf was calculated for evaluating volume score using a 0-20 scale. Two experienced judges scored the bread for crust color, symmetry, break and shred, and crumb color using a 0-10 scale for all observations except grain and texture which were evaluated using a 0-20 scale.

The power requirement for dough development was calculated from the total through-put per hour and kilowatt reading using the following formula:

$$\text{Power requirement (Hp/lb/min)} = \frac{Kw \times 60 \times 1000}{746 \times T.P.}$$

Kw = Kilowatt reading

TP = Through-put in lbs/hour

The bread made by the continuous dough mixing process was statistically evaluated for power requirement, total score, specific loaf volume, grain and texture score.

RESULTS AND DISCUSSION

Skim milks produced during the winter and spring were oxidized with various concentrations of hydrogen peroxide prior to being heated and manufactured into NFDM. Sulfhydryl content, residual peroxide content, undenatured whey protein nitrogen content, and water absorption characteristics were determined on each variable of NFDM produced. NFDM samples were incorporated into sponge and continuous mix doughs in amounts equal to 3 and 6% of the flour weight. The power required to develop the doughs with the continuous mix process was calculated. The baked breads made with the continuous mix process were statistically analyzed for loaf volume, grain, texture and total score. The data have been assembled to show the effect of treating the skim milk with hydrogen peroxide prior to drying on loaf volume, grain, texture, and total score of baked bread and the power requirement for the proper development of the dough.

Determination of Sulfhydryl Content

The sulfhydryl content of different NFDM samples was determined from a standard curve (figure 1) and the data are reported in table 4. Figure 1 is a representation of a typical standard curve. Separate standard curves were prepared for each determination.

Table 4. Sulfhydryl content of different reconstituted NFDM samples[a]. (mg of cysteine/100 ml of reconstituted NFDM - 10% TS)

Samples	Milk No 1 Series A	Milk No 1 Series B	Milk No 2
Unheated control	2.494	2.459	
Heated 190 F/30 min	1.592	1.128	1.215
Heated 190 F/30 min, 0.05% H_2O_2		0.850	0.983
Heated 190 F/30 min, 0.10% H_2O_2	0.843	0.829	0.878
Heated 190 F/30 min, 0.20% H_2O_2	0.485	0.457	0.323

[a] Duplicate analysis

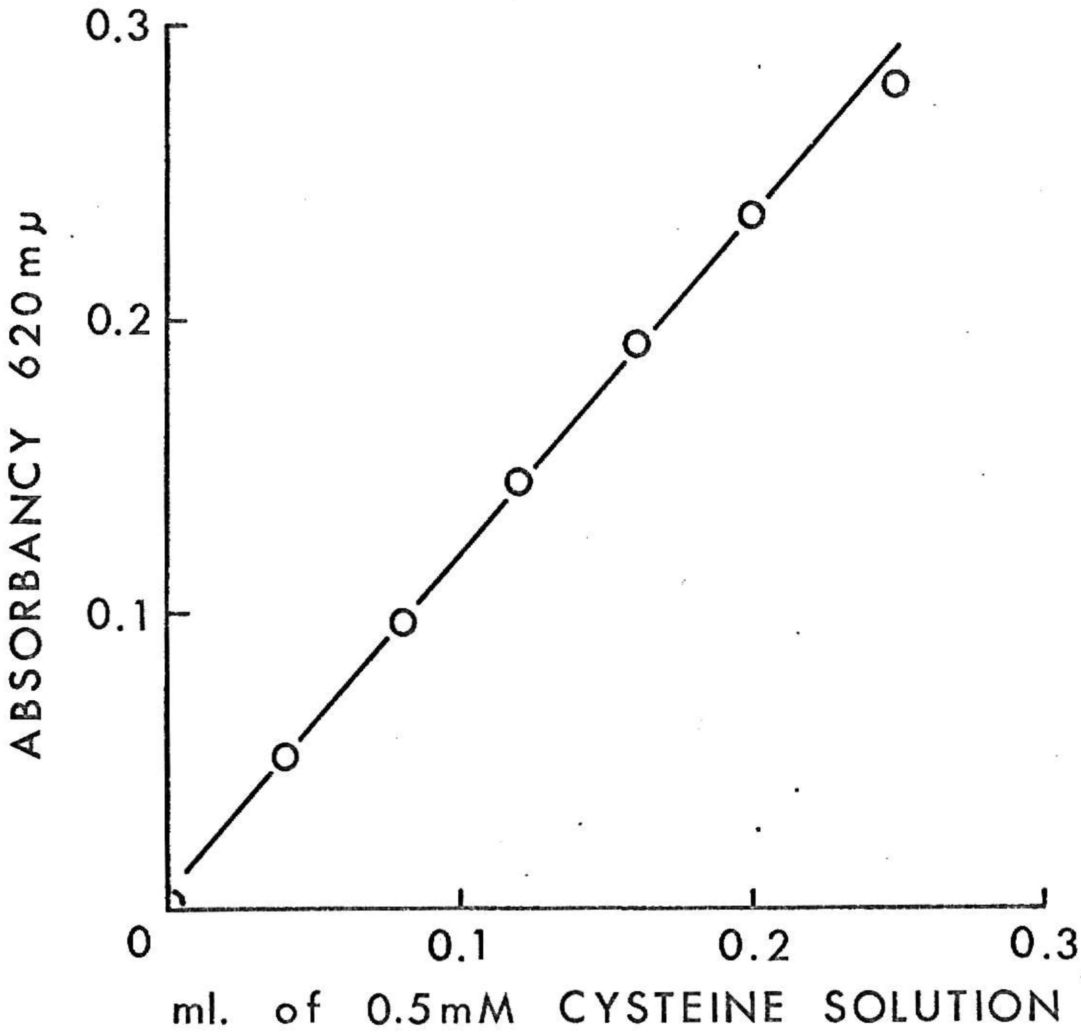


Figure 1. Typical standard curve for sulfhydryl determination.

The sulfhydryl content of the skim milk decreased on heating. These data agree with the findings of Larsen et al. (25). The sulfhydryl content of milk was decreased further by treating the skim milk with hydrogen peroxide. Decrease in the sulfhydryl content was assumed due to oxidation of sulfhydryl groups.

Residual Peroxide

The standard curve for known concentrations of hydrogen peroxide in reconstituted NFDM is shown in figure 2. The residual peroxide, contained in the hydrogen peroxide treated NFDM samples used in the continuous dough mixing experiment, was determined from the standard curve and is summarized in table 5.

These data show that the hydrogen peroxide remaining after the preheating and the drying operation varied from 7.86 to 12.31% of that originally added. When these NFDM samples were incorporated in the bread dough formula at the 3 and 6% levels, the potential oxidizing power due to the residual hydrogen peroxide in the NFDM samples was calculated and is summarized in table 6.

The data show that the potential oxidizing power due to residual peroxide in the NFDM is high in relation to that contributed by the added bromate-iodate oxidant. Undoubtedly this resulted in over-oxidation of certain doughs.

Undenatured Whey Protein Nitrogen

A standard curve (figure 3) was established by using low heat and high heat NFDM samples obtained from the American Dry Milk Institute. The undenatured whey protein nitrogen values for all the NFDM samples prepared were calculated from this curve and are presented in table 7.

The undenatured whey protein nitrogen (WPN) values were decreased by the heat treatment used in this study. However, they did not in any case meet the minimum requirement of not more than 1.5 mg WPN/g NFDM, for high heat classification which is desirable for use in bread. Ashworth and Krueger (3) reported that the optimum preheat treatment for NFDM used in bread is 82 F for 30 min. Harland et al. (16) showed

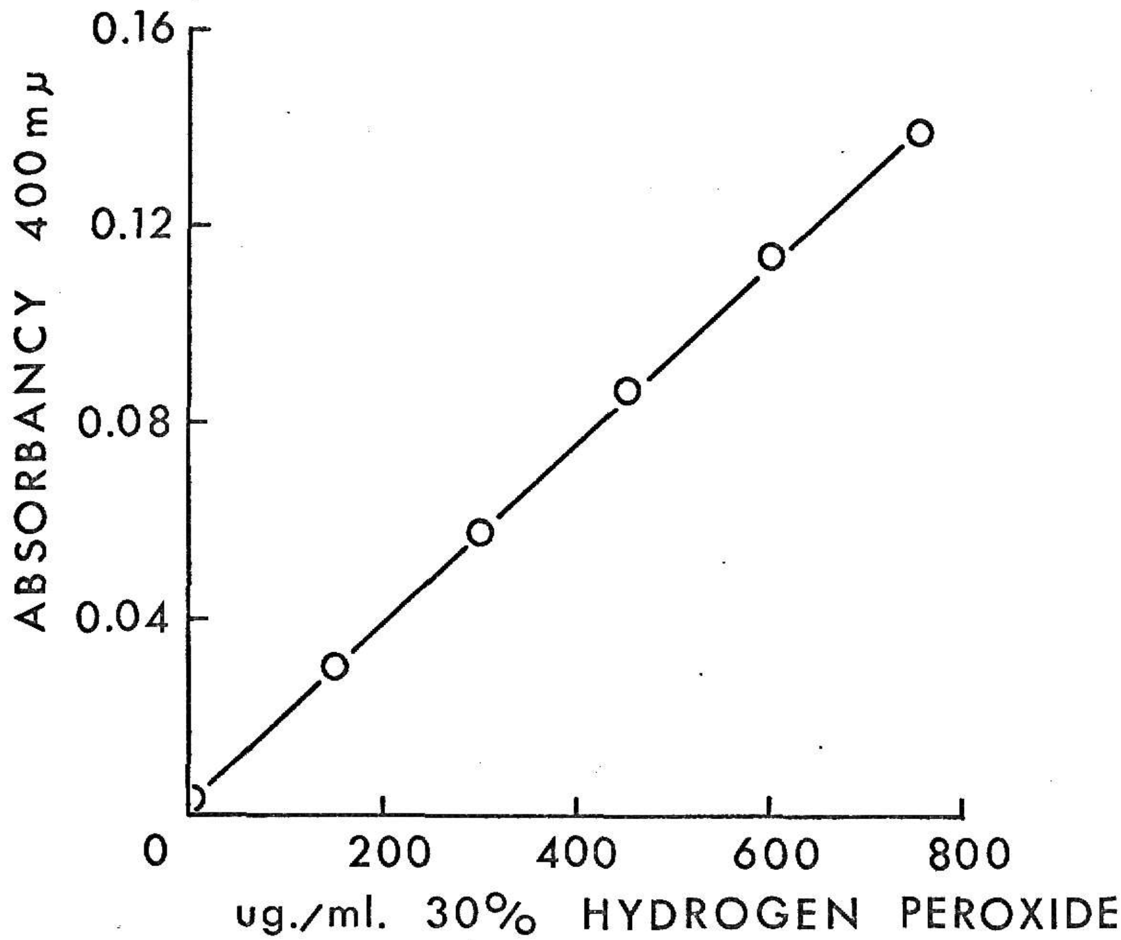


Figure 2. Standard curve for determination of residual hydrogen peroxide in reconstituted NFDM.

Table 5. Summary of residual peroxide in reconstituted NFDM samples used in continuous dough mixing experiments.

Sample	Amount of 30% hydrogen peroxide added	Residual peroxide[a]		Hydrogen peroxide remaining	
		Milk No 1 Series B	Milk No 2	Milk No 1 Series B	Milk Nc
	µg/ml	µg/ml	µg/ml	%	%
Heated 190 F/ 30 min, 0.05% H_2O_2	1,665	131	131	7.86	7.86
Heated 190 F/ 30 min, 0.10% H_2O_2	3,330	394	410	11.83	12.31
Heated 190 F/ 30 min, 0.20% H_2O_2	6,660	542	633	8.14	9.50

[a] 30% hydrogen peroxide/ml of reconstituted NFDM.

Table 6. Percentage of total potential oxidizing power contributed by the residual peroxide.

	Oxidant level ppm					
	50		70		90	
NFDM in sample	3%	6%	3%	6%	3%	6%
Milk No 1, series B						
Heated 190 F/30 min, 0.05% H_2O_2	53.43	70.79	46.15	63.38	39.03	57.38
Heated 190 F/30 min, 0.10% H_2O_2	77.34	87.26	70.92	82.74	65.48	79.20
Heated 190 F/30 min, 0.20% H_2O_2	82.44	90.46	77.03	87.14	72.28	84.05
Milk No 2						
Heated 190 F/30 min, 0.05% H_2O_2	52.75	69.75	45.36	62.22	38.28	58.29
Heated 190 F/30 min, 0.10% H_2O_2	78.37	87.82	72.13	83.75	66.81	80.08
Heated 190 F/30 min, 0.20% H_2O_2	84.74	91.76	79.87	88.83	75.52	86.08

Table 7. Undenatured whey protein nitrogen values on different NFDM samples.

Samples	Milk No 1 Series A	Milk No 1 Series B	Milk No 2
	mg nitrogen/g of NFDM		
Unheated Control	9.25	9.68	----
Heated 190 F/30 min	3.42	2.97	3.23
Heated 190 F/30 min, 0.05% H_2O_2	----	4.87	5.18
Heated 190 F/30 min, 0.10% H_2O_2	5.55	4.99	5.25
Heated 190 F/30 min, 0.20% H_2O_2	6.71	5.95	6.34

that considerable variation exists in the concentration and denaturability of serum proteins. It would seem while undenatured whey protein nitrogen values in this study did not fall below the desired level of 1.5 mg WPN/g NFDM, that the NFDM had been given sufficient heat treatment since 63.0 and 69.3 percent of serum protein had been denatured by heating to 190 F/30 min for Milk No 1, Series A and B, respectively.

It was observed also that the undenatured whey protein nitrogen values increased beyond those which were noted by heating along with the addition of hydrogen peroxide to milk prior to preheating. The increase in undenatured WPN changed with increments of hydrogen peroxide.

Swanson et al. (50) observed an increase in WPN value in their study of the effect of high heat treatment of the concentrated milk after an initial forewarming of the skim milk. They postulated that the increase was due to casein fragmentation which was not precipitated by a saturated salt solution. The oxidation by hydrogen peroxide and subsequent heating may have a similar effect on casein. It may be possible that the oxidation of sulfhydryl groups of B-lactoglobulin

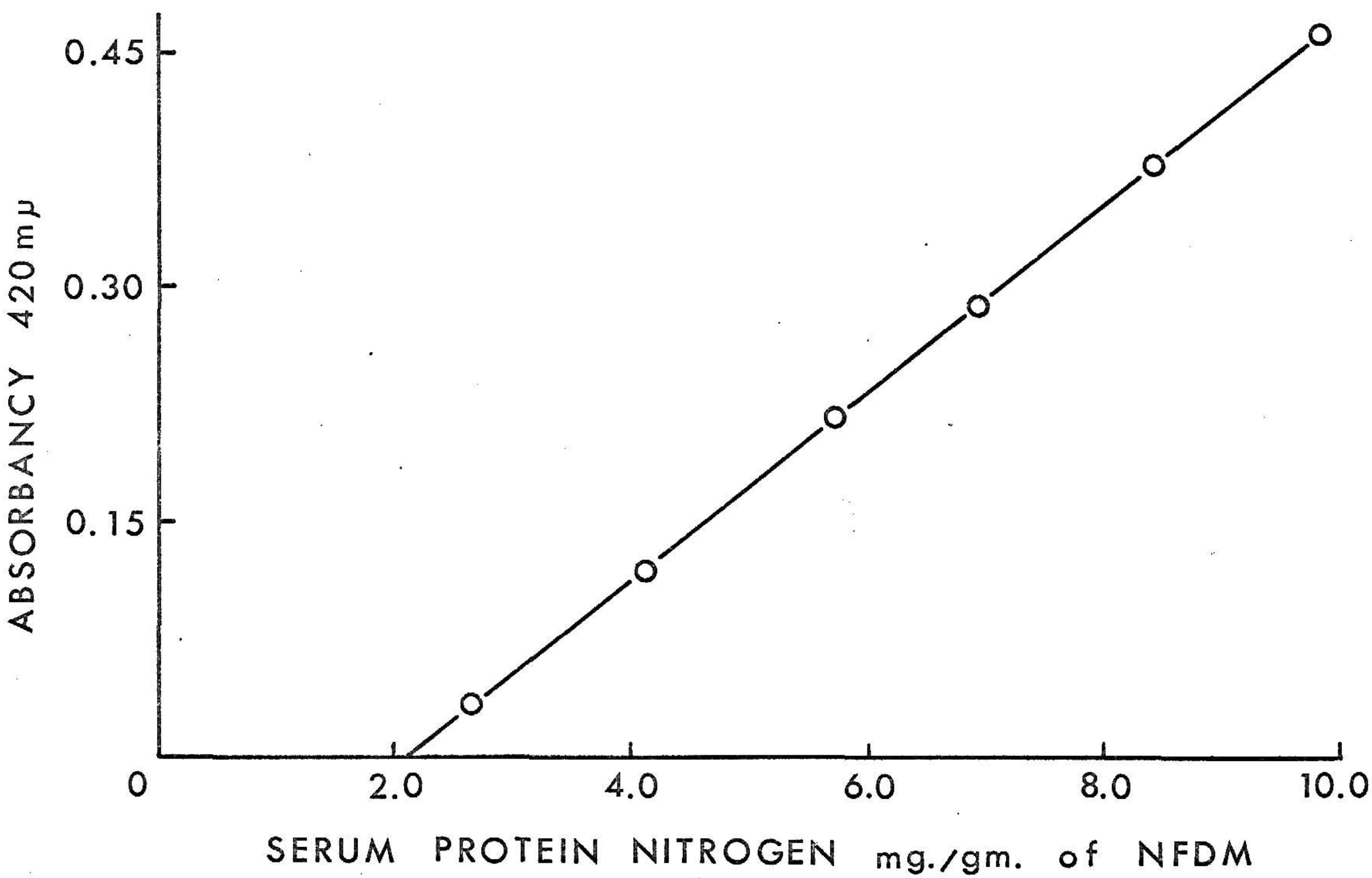


Figure 3. Standard curve for whey protein nitrogen estimation.

by hydrogen peroxide prevented the formation of the well known complex between B-lactoglobulin and K-casein. The uncomplexed B-lactoglobulin may not be precipitated by saturated salt solution in the Hasland-Ashworth WPN test and this may account for the increase in the undenatured whey protein nitrogen value in the case of the hydrogen peroxide treated milks.

Absorbance Study

The amount of water required to develop a dough consistency of 500 BU was measured when different NFDM samples were incorporated at 3 and 6% levels based on flour weight. The peak time, mixing tolerance index (MTI) and valorimeter value were measured from the farinograms of each sample. The data are summarized in tables 8, 9, 10, and 11. The farinograms of each sample are represented in figures 4 and 5.

The flours used in the absorbence study were the same as those used in the bread baking by the sponge dough and continuous mix dough processes. The absorption was increased when the NFDM samples were incorporated which agrees with the observation of Larson et al. (26). This is due to an increase in absorbance sites by the added milk proteins.

Peak time is a measure of the time required for the maximum dough development. The peak time was increased when the NFDM samples were incorporated in the dough. Markely and Bailey (29) observed an increase in the peak time with increases in the water absorption. The increase in the peak time observed may be due to an increase in the water absorption by NFDM.

Mixing tolerance index is a measure of the dough strength. A low MTI reading indicates a high tolerance to mixing and a high MTI reading indicates a low tolerance to mixing. When NFDM heat treated samples were incorporated into the dough at 3 and 6% levels, the MTI reading decreased as compared to the flour standard. This also was observed in the heated 190 F/30 min, 0.05% hydrogen peroxide treated NFDM sample of Milk No 2. In all other samples, an increase in MTI value was observed. The increase ranged from 0 to 15 BU.

The valorimeter value is an empirical quality score and is dependent upon two characteristics of farinograms; the peak time and rate of dough

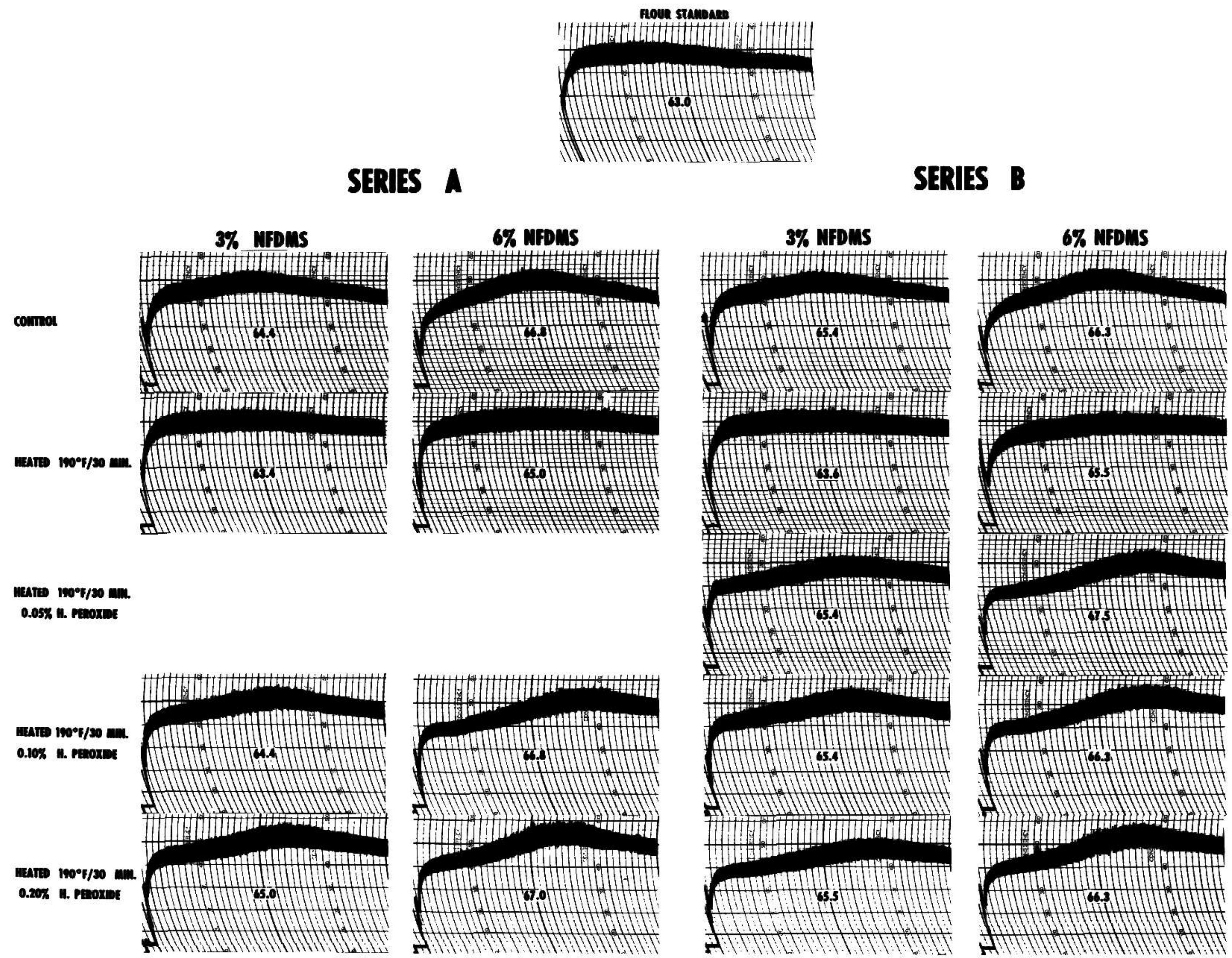


Figure 4. Farinograms of different NFDM samples of milk no. 1, series A and B.

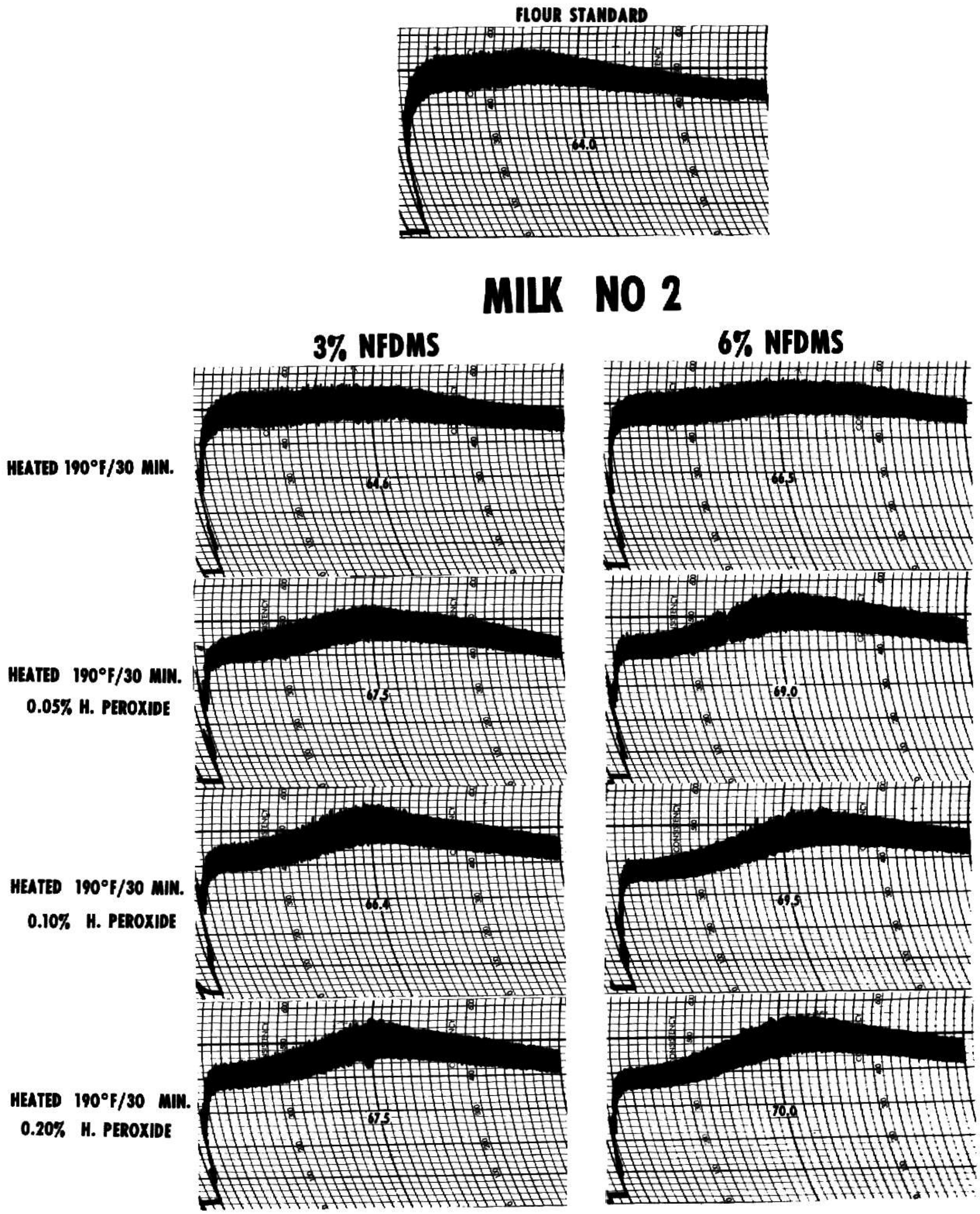


Figure 5. Farinograms of different NFDM samples of milk no. 2.

breakdown after peak time. The valorimeter value indicates the overall mixing strength of the dough. These values increased when NFDM samples were incorporated into the dough.

Bread Baking

Sponge dough method; all NFDM samples of Milk No 1, Series A and B, were incorporated in the sponge dough formula at 3 and 6% levels based on flour weight. The characteristics including loaf volume, grain, and texture were evaluated using a 0-20 scale while symmetry, break and shred, crust color and the crumb color were evaluated using a 0-10 scale. The data are summarized in table 9. The cross section view and side view of representative bread samples are shown in the Appendix figure 1 and 2.

Table 8. Water absorption of dough with NFDM samples.[a]

NFDM Samples	Milk No 1				Milk No 2	
	Series A		Series B			
	3%	6%	3%	6%	3%	6%
Unheated control	64.4	66.8	65.4	66.3	----	----
Heated 190 F/30 min	63.4	65.0	63.6	65.5	64.6	66.5
Heated 190 F/30 min, 0.05% H_2O_2	----	----	65.4	67.5	67.5	69.0
Heated 190 F/30 min, 0.10% H_2O_2	64.4	66.8	65.4	66.3	66.4	69.5
Heated 190 F/30 min, 0.20% H_2O_2	65.0	67.0	65.5	66.3	67.5	70.0

[a] Flour absorption for Milk No 1 and Milk No 2 was 63.0% and 64.0% respectively.

Table 9. Peak time for doughs with NFDM samples.[a]

NFDM Samples	Milk No 1 Series A 3%	Milk No 1 Series A 6%	Milk No 1 Series B 3%	Milk No 1 Series B 6%	Milk No 2 3%	Milk No 2 6%
Unheated control	10.0	10.0	10.0	11.0	----	----
Heated 190 F/30 min	9.0	11.0	10.5	10.5	9.0	11.0
Heated 190 F/30 min, 0.05% H_2O_2	----	----	12.5	14.5	10.0	10.5
Heated 190 F/30 min, 0.10% H_2O_2	11.5	15.0	12.0	14.0	10.0	12.0
Heated 190 F/30 min, 0.20% H_2O_2	12.0	13.0	15.0	13.5	10.0	11.0

[a] Peak time for flour standard doughs. Milk No 1 and No 2 were 8.5 and 7.0 min, respectively.

Table 10. Mixing tolerance index for doughs with NFDM samples.[a]

NFDM Samples	Milk No 1 Series A 3%	Milk No 1 Series A 6%	Milk No 1 Series B 3%	Milk No 1 Series B 6%	Milk No 2 3%	Milk No 2 6%
Unheated control	40	40	35	40	--	--
Heated 190 F/30 min	20	5	25	10	30	25
Heated 190 F/30 min, 0.05% H_2O_2	--	--	30	45	35	40
Heated 190 F/30 min, 0.10% H_2O_2	40	40	40	35	65	40
Heated 190 F/30 min, 0.20% H_2O_2	30	45	35	40	50	45

[a] MTI for flour standard doughs for Milk No 1 and 2 were 30 and 50 BU respectively.

Table 11. Valorimeter value on the absorption farinograph with NFDM samples.[a]

NFDM Samples	Milk No 1 Series A 3%	Milk No 1 Series A 6%	Milk No 1 Series B 3%	Milk No 1 Series B 6%	Milk No 2 3%	Milk No 2 6%
Unheated control	74.0	74.0	73.0	77.0	----	----
Heated 190 F/30 min	70.0	76.0	75.0	80.0	70.0	77.0
Heated 190 F/30 min, 0.05% H_2O_2	----	----	81.0	86.0	73.5	73.0
Heated 190 F/30 min, 0.10% H_2O_2	78.0	86.0	80.0	84.0	73.5	80.0
Heated 190 F/30 min, 0.20% H_2O_2	80.0	82.0	86.0	84.0	73.0	76.5

[a] Valorimeter values on flour standard absorption farinograms for Milk No 1, Series B, and Milk No 2 were 68.0 and 61.5 respectively.

Table 12. The scores of bread made with 3 and 6% NFDM samples of Milk No 1, Series A and B by sponge dough process.[a]

FDM eries	Samples	% Used	Vol	Crust color	Symmetry	Break & shred	Tex-ture	Grain	Crumb color	Total
	Maximum		20	10	10	10	20	20	10	100
A	Unheated control	3	17	9	8	7	17	15	9	82
		6	16	9	7	7	15	14	8	76
	Heated 190 F/30 min	3	18	9	9	9	18	17	9	89
		6	16	9	9	9	18	16	8	85
	Heated 190 F/30 min, 0.10% H_2O_2	3	18	9	9	8	17	17	9	87
		6	17	9	9	8	17	17	8	85
	Heated 190 F/30 min, 0.20% H_2O_2	3	19	9	9	9	18	17	8	89
		6	19	9	9	9	18	17	8	89
B	Unheated Control	3	17	9	8	7	16	15	9	81
		6	17	9	7	7	15	14	8	77
	Heated 190 F/30 min	3	18	9	9	10	18	17	9	90
		6	18	9	9	8	17	15	8	84
	Heated 190 F/30 min, 0.05% H_2O_2	3	19	9	9	10	18	17	9	91
		6	19	9	9	10	18	17	8	90
	Heated 190 F/30 min, 0.10% H_2O_2	3	19	9	9	8	18	17	9	89
		6	19	9	9	10	18	17	8	90
	Heated 190 F/30 min, 0.10% H_2O_2	3	19	9	8	7	18	17	9	87
		6	18	9	9	10	18	17	8	89

[a] Scoring range: 0-10 for crust color, crumb color, symmetry, and bread and shred.
0-20 for volume, texture, and grain.

The overall characteristics of baked bread were improved by using 190 F/30 min, 0.05% hydrogen peroxide treated NFDM samples at 3 and 6% levels based on flour weight. The loaf volume was depressed when unheated NFDM samples were incorporated in the dough. The loss of loaf volume impaired the baked bread characteristics such as break and shred, grain and symmetry.

Continuous dough mixing process; All NFDM samples of Milk No 1, series B and Milk No 2 were incorporated into the dough at 3 and 6% levels based on flour weight except the unheated NFDM sample of Milk No 1, series B. The baked bread samples were evaluated for different characteristics as described previously. The specific volume (volume in cc/454 g of baked bread) was calculated for each loaf. The power required for dough development was also calculated for variables studied. The power requirement, specific loaf volume, texture, grain and total score were analyzed statistically. The cross sectional and side views of representative bread samples are shown in Appendix figures 3 to 34.

When NFDM samples of Milk No 1, series B, were incorporated in the dough mixed by this process, the water absorption values were arbitrarily fixed to 70% when NFDM was added at 3% of flour weight, and 73% when 6% NFDM was used in relation to flour weight. The water absorption values for NFDM No 2 samples were determined from the farinograms.

The statistical analysis of grain, texture, and total score for breads made with the NFDM samples of Milk No 1, series B, revealed that the interaction between skim milk treatment, percentage of NFDM used in the formula and the mixing speed was significant ($P < .05$). The individual score for these characteristics and their mean values for the above interaction are presented in tables 13, 14, 15, 16 and 17. The least significant differences (LSD) were calculated for the interactions and are shown in tables 15 and 17.

It was shown by statistical analysis that at the 3% NFDM level, all milk treatments except 190 F/30 min, 0.20% H_2O_2 milk treatment were capable of producing good grain structure, with all three mixing speeds. When 6% NFDM samples were incorporated into the dough, only the 190 F/30 min, 0.10% H_2O_2 milk treatment produced good grain structure at all mixing speeds.

The texture score analysis showed that when 3% NFDM was used, 190 F/30 min, 0.05% H_2O_2 and 190 F/30 min, 0.10% H_2O_2 milk treatments produced a good texture at 171 and 218 rpm mixing speeds. However, when 6% NFDM was used in the formula, only 190 F/30 min, 0.10% H_2O_2 treatment at all mixing speeds produced a good bread texture.

The total score of baked bread indicates the overall quality of the bread. The statistical analysis of the total score showed that bread with a good overall quality was produced at all mixing speeds studied by incorporating 3% of all NFDM samples except that 190 F/30 min, 0.20% H_2O_2 treated NFDM. At the 6% level of NFDM in the formula, only 190 F/30 min, 0.10% H_2O_2 milk treatment produced a good overall quality of bread at all three mixing speeds used.

The statistical analysis of power required for dough development, when NFDM samples of Milk No 1, Series B, were used in the formula, showed that along with the interactions of milk treatments, percentage of NFDM used in the formula, and mixing speeds; the interaction between milk treatment, percentage of NFDM used in the formula and oxidant level also was significant ($P < .05$). Table 18 and 19 show the power requirement for the representative doughs and the mean value of power requirement for the above two interactions along with LSD, respectively.

The statistical analysis for the power requirement disclosed that at 3% NFDM in the formula, 190 F/30 min, 0.20% H_2O_2 treatment, required the least power for dough development at all three oxidant levels. This also was true with the three mixing speeds used in dough development. When 6% NFDM samples were incorporated into the dough, the 190 F/30 min, 0.05% H_2O_2 treatment required the least power at the three different dough oxidant levels used. The same treatment required the least power at 171 rpm mixing speed when NFDM was used at the 6% level.

The interactions between milk treatment, the amount of NFDM used in the formula; milk treatment, and mixing speed were found to be significant ($P < .05$). The specific volume of individual breads and the means of specific volume for the above two interactions along with LSD are given in table 20 and 21, respectively.

The analysis of milk treatment and percentage NFDM used in the formula showed that at 3% NFDM levels, 190 F/30 min and 190 F/30 min

Table 13. Grain score for the baked bread containing NFDM samples of Milk No 1, Series B, made by continuous dough mixing process.[a]

		RPM								
		171			218			260		
		Oxidant level used in the formula, ppm								
Milk Samples	% NFDM used	50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	14	14	15	12	14	14	13	14	12
Heated 190 F/30 min, +0.05% H_2O_2	3	15	15	17	17	17	15	12	17	11
Heated 190 F/30 min, +0.10% H_2O_2	3	15	14	13	16	14	14	14	13	13
Heated 190 F/30 min, +0.20% H_2O_2	3	--	10	6	6	6	6	--	6	6
Heated 190 F/30 min	6	8	17	11	8	15	16	10	10	10
Heated 190 F/30 min, +0.05% H_2O_2	6	6	8	10	6	4	15	8	4	14
Heated 190 F/30 min, +0.10% H_2O_2	6	13	13	13	14	14	14	16	14	13
Heated 190 F/30 min, +0.20% H_2O_2	6	10	10	8	12	10	8	16	15	10

[a] Maximum, 20 points.

Table 14. Texture scores for the baked bread containing NFDM samples of Milk No 1, Series B, made by continuous dough mixing process.[a]

		RPM								
		171			218			260		
		Oxidant level used in the formula, ppm								
Milk Samples	% NFDM used	50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	15	14	16	11	14	14	11	13	10
Heated 190 F/30 min, +0.05% H_2O_2	3	15	16	16	18	18	17	11	15	11
Heated 190 F/30 min, +0.10% H_2O_2	3	16	15	13	17	15	16	16	14	15
Heated 190 F/30 min, +0.20% H_2O_2	3	--	10	6	6	6	6	--	6	6
Heated 190 F/30 min	6	8	16	12	4	13	17	4	4	10
Heated 190 F/30 min, +0.05% H_2O_2	6	7	8	11	7	2	16	8	2	16
Heated 190 F/30 min, +0.10% H_2O_2	6	14	14	14	15	15	14	16	15	13
Heated 190 F/30 min, +0.20% H_2O_2	6	12	12	8	13	12	8	16	15	10

[a] Maximum, 20 points.

Table 15. The means of grain and texture scores as influenced by interactions of milk treatment, mixing speed and levels used of Milk No 1, Series B.

	% NFDM used	Grain score[a] Mixing speed, rpm 171	218	260	Texture score[a] Mixing speed, rpm 171	218	260
Heated 190 F/30 min	3	14.3	13.3	13.0	15.0	13.0	11.3
Heated 190 F/30 min, 0.05% H_2O_2	3	15.7	16.3	13.3	15.7	* 17.7*	12.3
Heated 190 F/30 min, 0.10% H_2O_2	3	14.0	14.7	13.3	14.7	16.0	15.0
Heated 190 F/30 min, 0.20% H_2O_2	3	* 7.3	* 6.0	* 6.0	* 7.3	* 6.0	* 6.0
Heated 190 F/30 min	6	12.0	13.0	10.0	12.0	11.3*	6.0
Heated 190 F/30 min, 0.05% H_2O_2	6	* 8.0 *	* 8.3 *	8.7	8.7 *	8.3 *	8.7 *
Heated 190 F/30 min, 0.10% H_2O_2	6	13.0	14.0	14.3	14.0	14.7	14.7
Heated 190 F/30 min, 0.20% H_2O_2	6	* 9.3	* 10.0*	13.7	10.7	11.0	13.7
LSD			3.56			3.90	

* Denotes statistically significant difference between means adjacent ($P < .05$).

[a] Maximum, 20 points.

Table 16. Total score for the baked bread containing NFDM samples of Milk No 1, Series B, made by continuous dough mixing process.[a]

Milk Samples	% NFDM used	RPM 171, Oxidant 50 ppm	171, 70	171, 90	218, 50	218, 70	218, 90	260, 50	260, 70	260, 90
Heated 190 F/30 min	3	73	70	76	60	69	70	63	69	62
		73	71	74	59	71	70	63	69	59
Heated 190 F/30 min,		65	68	76	77	79	72	58	76	55
0.05% H_2O_2	3	65	68	75	79	80	73	57	75	53
Heated 190 F/30 min,		76	72	64	78	72	72	73	68	68
0.10% H_2O_2	3	75	73	63	79	72	73	75	68	68
Heated 190 F/30 min,		--	46	29	31	28	28	--	29	31
0.20% H_2O_2	3	--	43	30	31	29	28	--	28	34
Heated 190 F/30 min	6	46	74	55	29	71	77	30	35	54
		44	73	56	30	69	77	30	37	54
Heated 190 F/30 min,		40	47	50	38	23	67	42	23	66
+0.05% H_2O_2	6	39	45	50	39	22	68	45	22	65
Heated 190 F/30 min,		62	62	62	66	68	66	76	68	59
+0.10% H_2O_2	6	60	61	63	66	68	66	76	69	62
Heated 190 F/30 min,		50	46	33	57	48	35	73	67	47
+0.20% H_2O_2	6	51	45	33	58	48	35	72	68	49

[a] Maximum, 100 points.

Table 17. The mean of total score as influenced by the interactions of milk treatment, mixing speed and levels used of Milk No 1, Series B.

	NFDM in formula	Total score[a] Mixing speed in RPM 171	218		260
	%				
Heated 190 F/30 min	3	72.8	66.5		64.2
Heated 190 F/30 min, 0.05% H_2O_2	3	69.5	76.7		62.3
Heated 190 F/30 min, 0.10% H_2O_2	3	70.5	74.3		70.0
		*	*		*
Heated 190 F/30 min, 0.20% H_2O_2	3	37.0	29.2		30.5
Heated 190 F/30 min	6	58.0	58.8	*	40.0
			*		
Heated 190 F/30 min, 0.05% H_2O_2	6	45.2	42.8		43.8
		*	*		*
Heated 190 F/30 min, 0.10% H_2O_2	6	61.7	66.7		68.3
		*	*		
Heated 190 F/30 min, 0.20% H_2O_2	6	43.0	46.8	*	62.7
LSD			15.82		

* Denotes statistically significant difference between adjacent means ($P < .05$).

[a] Maximum, 100 points.

Table 18. The power requirement for dough development using NFDM samples of Milk No 1, Series B, with the continuous dough mixing process.[a]

Milk Samples	% NFDM used	171			218			260		
		Oxidant level used in the formula, ppm								
		50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	0.5143	0.6545	0.4909	0.7013	0.6311	0.7480	0.7480	0.7246	0.9350
Heated 190 F/30 min, +0.05% H_2O_2	3	0.4675	0.4441	0.5143	0.7948	0.6545	0.8181	0.9350	0.7948	0.9350
Heated 190 F/30 min, +0.10% H_2O_2	3	0.5253	0.5658	0.5698	0.8072	0.7597	0.7597	0.9496	0.9496	0.9496
Heated 190 F/30 min, +0.20% H_2O_2	3	------	0.2611	0.2849	0.2137	0.3086	0.3324	0.2611	0.3798	0.4511
Heated 190 F/30 min	6	0.6303	0.4502	0.4052	0.9004	0.7203	0.6528	0.9004	0.9004	0.7879
Heated 190 F/30 min, +0.05% H_2O_2	6	0.3377	0.2701	0.3827	0.5628	-----	0.7203	0.7203	0.4277	0.9004
Heated 190 F/30 min, +0.10% H_2O_2	6	0.4138	0.5058	0.5748	0.6667	0.6897	0.7814	0.7127	0.8736	0.9196
Heated 190 F/30 min, +0.20% H_2O_2	6	0.4828	0.3449	0.2759	0.5748	0.5058	0.3449	0.9196	0.8276	0.6667

[a] Hp/lb/min.

Table 19. The means of power requirement for developing doughs as influenced by interactions of milk treatment and level used of Milk No 1, Series B, with oxidation level and mixing speed, respectively.

	% NFDM used	Power requirement[a] Oxidant level, ppm 50	70	90	Mixing Speed, rpm 171	218	260
Heated 190 F/30 min	3	.6535	.6701	.7246	.5532*	.6935*	.8025
Heated 190 F/30 min, 0.05% H_2O_2	3	.7324	.6311*	.7558	.4753*	.7558*	.8883
Heated 190 F/30 min, 0.10% H_2O_2	3	.7597	* .7597	.7597	.5540*	.7755*	.9496
Heated 190 F/30 min, 0.20% H_2O_2	3	* .2492	* .3165	* .3561	* .2730	* .2849	* .3640
Heated 190 F/30 min	6	.8104*	.6903	.6153	.4952*	.7578	.8629
Heated 190 F/30 min, 0.05% H_2O_2	6	* .5403	* .4465*	.6678	* .3302*	.6416	* .6828
Heated 190 F/30 min, 0.10% H_2O_2	6	.5977	* .6897	.7587	* .4981*	.7127*	.8353
Heated 190 F/30 min, 0.20% H_2O_2	6	.6591	* .5594	* .4292	* .3679	* .4752*	.8046
LSD			.123			.123	

[a] Hp/lb/min

* Denotes statistically significant difference between adjacent mean (P < .05).

at 0.05% H_2O_2 treatment, produced the breads with the largest specific volume. When 6% NFDM was used in the formula, there was no significant difference observed between the milk treatments. All milk treatments except the 190 F/30 min, 0.20% H_2O_2, produced significantly larger specific volumes at the 3% NFDM level than the 6% level. The 190 F/30 min, 0.20% H_2O_2, was the only treatment resulting in a larger specific volume at the 6% NFDM level than that at the 3% level. This also was statistically significant ($P < .05$).

The statistical analysis of milk treatment and mixing speed interaction, showed no significant difference between the first three milk treatments used at all three mixing speeds. The 190 F/30 min, 0.20% H_2O_2 treatment produced bread with lowest specific volume at all three mixing speeds.

In the statistical analysis of the grain score of breads containing NFDM No 2, the milk treatment, percentage of NFDM used in the formula, dough oxidant level, and mixing speed interactions were found to be significant ($P < .05$). Individual grain scores and mean grain scores for the interactions are shown in table 22 and 23, respectively. The calculated LSD values for these interactions are given in table 23.

The breads with a good grain structure were produced at 70 and 90 ppm oxidant levels when 190 F/30 min and 190 F/30 min, 0.05% H_2O_2 treated NFDM samples were incorporated in the dough at 3% level. At 6% NFDM, the 190 F/30 min, 0.10% H_2O_2 milk treatment was the only combination that resulted in a good grain structure with 50 and 70 ppm oxidant.

Milk treatment and mixing speed interactions revealed that only the 190 F/30 min, 0.05% H_2O_2 treatment produced a good grain structure at 171 and 218 rpm. The 190 F/30 min, 0.10% H_2O_2 treated NFDM produced a good grain structure at 171 rpm only.

The statistical analysis of texture score of breads made with NFDM samples of Milk No 2 revealed that the interactions between milk treatment and percentage NFDM used in the formula were significant ($P < .05$). The individual texture scores on the representative breads and the mean of texture score for the interactions are given in the tables 24 and 25.

The statistical analysis of milk treatment and percentage of NFDM used in the formula showed that at 3% NFDM level, the milk treatments of 190 F/30 min and 190 F/30 min, 0.05% H_2O_2 produced a good texture.

Table 20. Specific volume of the bread made with NFDM samples of Milk No 1, Series B, using continuous dough mixing process.[a]

Milk Samples	% NFDM used	RPM 171, Oxidant level used in the formula, ppm: 50	171: 70	171: 90	218: 50	218: 70	218: 90	260: 50	260: 70	260: 90
Heated 190 F/30 min	3	2654	2666	2869	2455	2621	2651	2373	2730	2628
		2727	2760	2724	2432	2754	2687	2442	2735	2341
Heated 190 F/30 min,		2623	2575	2865	2609	2739	2627	2415	2753	2410
+0.05% H_2O_2	3	2565	2583	2823	2817	2833	2746	2275	2735	2260
Heated 190 F/30 min,		2595	2348	2153	2506	2339	2388	2338	2284	2305
+0.10% H_2O_2	3	2490	2353	2203	2623	2330	2353	2662	2270	2479
		2500	2360	2137	2576	2327	2516	2492	2260	2324
Heated 190 F/30 min,		----	1891	1637	1550	1488	1465	----	1608	1718
+0.20% H_2O_2	3	----	1681	1598	1533	1475	1586	----	1615	1681
		----	1632	1730	1519	1560	1477	----	1453	2037
Heated 190 F/30 min	6	2188	2717	2342	1621	2550	2631	1671	1604	2349
		2032	2636	2380	1684	2373	2624	1748	1800	2295
Heated 190 F/30 min,		2097	2294	2259	1929	1717	2392	2047	1805	2503
+0.05% H_2O_2	6	2028	2077	2154	1998	1618	2519	2296	1654	2360
Heated 190 F/30 min,		2053	2102	2048	2108	2197	2175	2373	2213	1948
+0.10% H_2O_2	6	1993	2073	2097	1976	2193	2155	2289	2200	1972
		1884	2044	2083	2072	2251	2173	2374	2270	2168
Heated 190 F/30 min,		2041	1756	1483	2105	1973	1675	2494	2336	2049
+0.20% H_2O_2	6	2001	1734	1475	2343	2101	1559	2355	2456	1882
		2144	1740	1453	2207	2048	1692	2391	2390	2163

[a] cc/454 g of bread loaf.

Table 21. The mean value of specific volume of breads as influenced by the interactions of milk treatment, level of NFDM No 1, Series B, in the formula and mixing speeds, respectively.

	Specific volume[a] cc/454 g						
	NFDM percent in dough formula			Mixing speed, rpm			
	3%		6%	171	218		260
Heated 190 F/30 min	2625	*	2180	2558	2424		2226
Heated 190 F/30 min, 0.05% H_2O_2	2625	*	2097	2412	2379		2293
	*						
Heated 190 F/30 min, 0.10% H_2O_2	2373	*	2141	2217	2302		2279
	*			*	*		
Heated 190 F/30 min, 0.20% H_2O_2	1645	*	2008	1746	1730	*	2004
LSD	205.69			251.92			

* Denotes statistically significant difference between adjacent mean ($P < .05$).

[a] cc/454 g.

Table 22. Grain score for the baked bread containing NFDM samples of Milk No 2, made by continuous dough mixing process.[a]

		RPM								
		125			171			218		
		Oxidant level used in the formula, ppm								
Milk Samples	% NFDM used	50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	14	15	15	12	14	15	10	12	16
Heated 190 F/30 min, +0.05% H_2O_2	3	6	10	8	16	17	17	15	16	17
Heated 190 F/30 min, +0.10% H_2O_2	3	9	5	6	17	12	15	12	11	10
Heated 190 F/30 min, +0.20% H_2O_2	3	6	4	3	8	7	4	10	12	10
Heated 190 F/30 min	6	6	4	7	4	7	8	5	8	7
Heated 190 F/30 min +0.05% H_2O_2	6	9	6	4	17	10	18	15	16	17
Heated 190 F/30 min +0.10% H_2O_2	6	15	13	9	17	15	13	16	17	15
Heated 190 F/30 min +0.20% H_2O_2	6	4	2	2	11	9	7	8	7	6

[a] Maximum, 20 points.

Table 23. The mean grain score of bread as influenced by the interaction of the milk treatments, levels used of NFDM No 2 and oxidant level; also shown are data for interactions of milk treatment and mixing speed.

	% NFDM used	Grain score[a] Oxidant level, ppm 50		70		90
Heated 190 F/30 min	3	12.0		13.7		15.3
Heated 190 F/30 min, 0.05% H_2O_2	3	12.3		14.3		14.0
				*		*
Heated 190 F/30 min, 0.10% H_2O_2	3	12.7	*	9.3		10.3
		*				*
Heated 190 F/30 min, 0.20% H_2O_2	3	8.0		7.7		5.7
Heated 190 F/30 min	6	5.0		6.3		7.3
		*		*		*
Heated 190 F/30 min, 0.05% H_2O_2	6	13.7	*	10.7		13.0
				*		
Heated 190 F/30 min, 0.10% H_2O_2	6	16.0		15.0	*	12.3
		*		*		*
Heated 190 F/30 min, 0.20% H_2O_2	6	7.7		6.0		5.0
LSD				2.76		

	Mixing Speed, rpm 125		171	218
Heated 190 F/30 min	10.2		10.0	9.7
	*		*	*
Heated 190 F/30 min, 0.05% H_2O_2	7.2		15.8	16.0
	*			*
Heated 190 F/30 min, 0.10% H_2O_2	9.5	*	14.8	13.5
	*		*	*
Heated 190 F/30 min, 0.20% H_2O_2	3.5	*	7.7	8.8
LSD			1.95	

* Denotes statistically significant differences between adjacent means ($P < .05$).

[a] Maximum, 20 points.

When 6% NFDM samples were incorporated in the formula, the best texture resulted by using 190 F/30 min, 0.10% H_2O_2 treated NFDM sample.

The milk treatment and oxidant level interactions showed that the 190 F/30 min, 0.05% H_2O_2 and 190 F/30 min, 0.10% H_2O_2 milk treatments produced good texture at all dough oxidant levels used.

The following three interactions were found to be statistically significant ($P < .05$) with respect to total score analysis: milk treatment and oxidant level; milk treatment and mixing speed; and milk treatment and percentage NFDM used in the formula.

The individual total score of all representative breads, and the mean of total score for these three interactions along with their LSD values are given in tables 26 and 27, respectively.

At 50 and 70 ppm oxidant levels fairly good overall characteristics of bread were obtained using NFDM treated when 190 F/30 min, 0.05% H_2O_2 and 190 F/30 min, 0.10% H_2O_2. At 90 ppm oxidant level, the 190 F/30 min and 190 F/30 min, 0.05% H_2O_2 milk treatments produced good overall quality bread.

With the dough mixing speed of 171 rpm the 190 F/30 min, 0.05% H_2O_2 and the 190 F/30 min, 0.10% H_2O_2 milk treatment improved significantly the overall quality of bread produced. At the dough mixing speed of 218 rpm, the 190 F/30 min, 0.05% H_2O_2 treatment produced bread with good overall quality.

The overall bread quality was found to be affected by percentage of NFDM used in the formula and milk treatment. With 3% NFDM level, the 190 F/30 min, and 190 F/30 min, 0.05% H_2O_2 milk treatment produced good quality bread. At the 6% NFDM level, milk treatments 190 F/30 min, 0.05% H_2O_2 and 190 F/30 min, 0.10% H_2O_2 produced good quality bread. All the milk treatments except 190 F/30 min, 0.10% H_2O_2, produced better quality breads at 3% usage level compared to the mean total score of breads with 6% NFDM. In case of the latter treatment, the effect was reversed.

The statistical analysis of the power requirement for dough development using NFDM sample No 2 indicated that the interactions between milk treatment, percentage of NFDM used in the formula, mixing speed, milk treatment and dough oxidation level were statistically significant ($P < .05$).

Table 24. Texture score for the baked bread containing NFDM samples of Milk No 2 made by continuous dough mixing process.[a]

Milk Samples	% NFDM used	RPM 125			RPM 171			RPM 218		
		Oxidant level used in the formula, ppm								
		50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	15	17	16	12	13	14	10	12	15
Heated 190 F/30 min, +0.05% H_2O_2	3	6	12	9	17	17	18	16	17	18
Heated 190 F/30 min, +0.10% H_2O_2	3	10	8	9	17	12	16	12	10	8
Heated 190 F/30 min, +0.20% H_2O_2	3	9	7	5	14	15	9	13	14	14
Heated 190 F/30 min	6	6	5	8	5	6	7	5	7	9
Heated 190 F/30 min, +0.05% H_2O_2	6	8	6	9	17	13	18	16	17	15
Heated 190 F/30 min, +0.10% H_2O_2	6	16	16	10	15	16	17	15	16	17
Heated 190 F/30 min, +0.20% H_2O_2	6	9	7	6	12	11	10	10	9	6

[a] Maximum, 20 points.

Table 25. The means of texture scores of bread as influenced by interactions of milk treatments with levels of NFDM used and dough oxidant levels, respectively.

	Texture score[a]					
	% NFDM used			Oxidant level, ppm		
	3		6	50	70	90
Heated 190 F/30 min	13.8	*	6.4	8.8	10.0	11.5
			*	*	*	*
Heated 190 F/30 min, 0.05% H_2O_2	14.4		13.2	13.3	13.7	14.5
	*		*			
Heated 190 F/30 min, 0.10% H_2O_2	11.3	*	15.3	14.2	14.2	12.8
			*	*	*	*
Heated 190 F/30 min, 0.20% H_2O_2	11.1	*	8.9	11.2	10.5	8.3
LSD		1.92			2.35	

* Denotes statistically significance between adjacent means (P < .0

[a] Maximum, 20 points.

Table 26. Total score of baked bread, containing NFDM No 2, made by continuous dough mixing process.[a]

Milk Samples	% NFDM used	RPM 125			RPM 171			RPM 218		
		Oxidant level used in the formula, ppm								
		50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	73	77	76	64	68	73	59	60	74
		73	77	75	65	68	73	57	61	75
Heated 190 F/30 min, +0.05% H_2O_2	3	32	57	48	71	81	82	74	76	68
		33	57	45	73	80	80	73	75	77
Heated 190 F/30 min, +0.10% H_2O_2	3	52	42	39	75	62	71	62	59	56
		50	41	39	75	62	72	62	60	55
Heated 190 F/30 min, +0.20% H_2O_2	3	40	28	22	58	55	34	60	65	61
		39	26	21	57	54	35	61	65	60
Heated 190 F/30 min	6	40	38	51	32	46	50	29	40	46
		40	38	51	32	46	50	29	38	45
Heated 190 F/30 min, +0.05% H_2O_2	6	38	35	40	75	53	81	69	74	74
		38	34	39	75	54	80	68	74	73
Heated 190 F/30 min, +0.10% H_2O_2	6	57	51	41	76	62	71	72	72	73
		56	51	41	77	64	71	71	72	73
Heated 190 F/30 min, +0.20% H_2O_2	6	36	28	27	55	48	47	50	45	41
		34	28	28	54	48	47	50	45	41

[a] Maximum, 100 points.

Table 27. The means of total scores of breads as influenced by the interaction of milk treatments with oxidant level, mixing speed and levels used of NFDM No 2, respectively.

	Total score[a]					
	Oxidant level, ppm			Mixing speed, rpm		
	50	70	90	125	171	218
Heated 190 F/30 min	49.4	54.7	61.6	59.1	55.6	51.1
Heated 190 F/30 min,	*	*		*	*	*
0.05% H_2O_2	59.9	62.5	66.4	41.3	73.7	73.7
Heated 190 F/30 min,			*			*
0.10% H_2O_2	65.4 *	58.2	58.5	46.7 *	69.8	65.6
Heated 190 F/30 min,	*	*	*	*	*	
0.20% H_2O_2	49.5	44.6	38.7	29.7 *	49.3	53.7
LSD		6.66			6.66	

	Total score		
	Percent NFDM in formula		
	3%		6%
Heated 190 F/30 min	69.3	*	41.2
Heated 190 F/30 min, 0.05% H_2O_2	66.2	*	59.7
Heated 190 F/30 min,	*		
0.10% H_2O_2	57.4	*	63.9
Heated 190 F/30 min,	*		*
0.20% H_2O_2	46.7		41.8
LSD		5.44	

* Denotes statistically significant differences between adjacent means ($P < .05$).

[a] Maximum, 100 points.

The power requirement for the individual representative doughs and mean values of the power requirement for the above interactions are given in tables 28 and 29, respectively.

When 3% NFDM was incorporated in the dough, the 190 F/30 min, 0.05% H_2O_2 and 190 F/30 min, 0.20% H_2O_2 treatment required less power for dough development at 125 rpm mixing speed than at other mixing speeds studied. When used at the 6% level the same two milk treatments along with the 190 F/30 min, 0.10% H_2O_2 milk treatment required less power for dough development at 125 rpm mixing speed than at the other two mixing speeds.

The milk treatment and oxidant level interactions in the power requirement analysis revealed that at 50 ppm oxidant level, the 190 F/30 min, 0.05% H_2O_2 and the 190 F/30 min, 0.20% H_2O_2 milk treatments required the least power for dough development when compared with usage of the other two milks at this particular dough oxidant level. Using 70 ppm dough oxidant, the same two milk treatments along with the 190 F/30 min, 0.10% H_2O_2 treatment required essentially the same power for dough development. The 190 F/30 min, 0.20% H_2O_2 treatment required the least power for dough development at 90 ppm oxidant level.

The analysis of specific volume of breads made with NFDM No 2, revealed that the interactions between milk treatment and percentage of NFDM used in the formula; milk treatment and oxidant level; and mixing speed and percentage of NFDM used in the formula, were significantly different ($P < .05$). The individual specific volume of each bread and the mean value of specific volume of breads are given in tables 30 and 31.

It was observed that there was no significant difference in the mean values of specific volume when the 190 F/30 min, the 190 F/30 min, 0.05% H_2O_2, and the 190 F/30 min, 0.10% H_2O_2 treated NFDM samples were incorporated at 3% level. The specific volume was lowered, however, by using 6% NFDM.

There were no significant differences in the specific volume between three oxidant levels when all NFDM samples were used at both 3 and 6% level, except for two cases. Significant difference was found between the 190 F/30 min, 0.05% H_2O_2 treatment and the 190 F/30 min, 0.10% H_2O_2 treatment at 70 ppm oxidant level. A significant difference also was

Table 28. The power requirement for dough development using NFDM No 2 with the continuous dough mixing process.[a]

Milk Samples	% NFDM used	RPM 125			RPM 171			RPM 218		
		Oxidant level used in the formula, ppm								
		50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	0.3647	0.5105	0.5348	0.7293	0.7050	0.7779	0.9238	0.9238	0.9724
Heated 190 F/30 min, +0.05% H_2O_2	3	0.2874	0.3593	0.3354	0.5270	0.6228	0.6468	0.8624	0.8624	0.9342
Heated 190 F/30 min, +0.10% H_2O_2	3	0.5299	0.4817	0.4094	0.7226	0.6744	0.7226	>0.9634	0.9152	0.9393
Heated 190 F/30 min, +0.20% H_2O_2	3	0.3833	0.2875	0.2396	0.6228	0.5270	0.3833	0.8145	0.7426	0.6707
Heated 190 F/30 min	6	0.5920	0.5920	0.5920	0.8998	0.8762	0.7578	>0.9472	>0.9472	>0.9474
Heated 190 F/30 min, +0.05% H_2O_2	6	0.3043	0.2809	0.3277	0.5351	0.4447	0.6085	0.8660	0.8426	0.8192
Heated 190 F/30 min, +0.10% H_2O_2	6	0.2801	0.2335	0.2801	0.5836	0.3969	0.5603	0.8404	0.8638	0.8171
Heated 190 F/30 min, +0.20% H_2O_2	6	0.3260	0.3027	0.3493	0.5123	0.6753	0.5588	0.6986	0.7218	0.6986

[a] Hp/lb/min.

Table 29. The mean values of power requirements for developing dough as influenced by the interactions of milk treatments, levels of NFDM No 2, used and mixing speed. Also shown are data for interactions of milk treatment and dough oxidant levels.

	% NFDM used	Power requirement[a] Mixing speed, rpm 125		171		218
Heated 190 F/30 min	3	.4700	*	.7374	*	.9400
Heated 190 F/30 min, 0.05% H_2O_2	3	* .3274	*	* .5989	*	.8863
Heated 190 F/30 min, 0.10% H_2O_2	3	* .4737	*	* .7065	*	.9393
Heated 190 F/30 min, 0.20% H_2O_2	3	* .3035	*	* .5110	*	* .7426
Heated 190 F/30 min	6	.5920	*	.8446	*	.9472
Heated 190 F/30 min, 0.05% H_2O_2	6	* .3043	*	* .5461	*	* .8426
Heated 190 F/30 min, 0.10% H_2O_2	6	.2646	*	.5136	*	.8404
Heated 190 F/30 min, 0.20% H_2O_2	6	.3260	*	.5821	*	* .7063
LSD				0.082		

	Oxidant level, ppm 50		70		90
Heated 190 F/30 min	.7428		.7591		.7637
Heated 190 F/30 min, 0.05% H_2O_2	* .5720		* .5688		* .6120
Heated 190 F/30 min, 0.10% H_2O_2	* .6533	*	.5942		.6215
Heated 190 F/30 min, 0.20% H_2O_2	* .5596		.5428	*	* .4834
LSD			0.058		

[*] Denotes statistically significant difference between adjacent

found between the 190 F/30 min, 0.10% H_2O_2 treatment and 190 F/30 min, 0.20% H_2O_2 treatment at 90 ppm oxidant level.

The interaction between mixing speed and percentage of NFDM in formula showed that 175 rpm dough mixing speed produced the largest specific volume of the bread when NFDM samples were used at 3 and 6% level.

Table 30. Specific volume for baked breads containing NFDM No 2 made by continuous dough mixing process.[a]

Milk Samples	% NFDM used	RPM 125			RPM 171			RPM 218		
		Oxidant level used in the formula, ppm								
		50	70	90	50	70	90	50	70	90
Heated 190 F/30 min	3	2643	2689	2862	2519	2724	2726	2650	2511	2557
		2654	2753	2838	2546	2714	2788	2552	2536	2517
		2642	2728	2813	2571	2663	2678	2528	2559	2506
Heated 190 F/30 min, +0.05% H_2O_2	3	1933	2536	2484	2477	2951	2768	2648	2603	2634
		1997	2538	2362	2694	2952	2470	2601	2521	2533
		2011	2522	2206	2708	2864	2631	2544	2497	2467
Heated 190 F/30 min, +0.10% H_2O_2	3	2470	2471	2206	2616	2566	2650	2369	2430	2653
		2425	2398	2144	2644	2604	2770	2392	2329	2632
		2338	2375	2157	2552	2607	2757	2372	2468	2550
Heated 190 F/30 min, +0.20% H_2O_2	3	2420	2081	1947	2559	2571	2072	2469	2564	2661
		2284	1961	1814	2446	2590	2059	2506	2628	2551
		2261	1892	1829	2480	2519	2184	2570	2647	2569
Heated 190 F/30 min	6	2388	2308	2442	2186	2289	2374	1907	2125	2256
		2398	2195	2466	2184	2256	2327	1907	1954	2190
		2408	2345	2442	2166	2256	2351	1868	1907	2190
Heated 190 F/30 min, +0.05% H_2O_2	6	1929	2026	2160	2509	2223	2861	2390	2409	2453
		1893	1826	2105	2429	2317	2725	2307	2354	2366
		1889	1896	2057	2503	2349	2722	2317	2400	2440
Heated 190 F/30 min, +0.10% H_2O_2	6	1916	1650	2032	2325	2094	2458	2279	2270	2424
		1878	1664	1984	2381	2130	2399	2284	2191	2314
		1779	1651	2045	2370	2270	2468	2238	2297	2373
Heated 190 F/30 min, +0.20%	6	2162	1939	2026	2474	2302	2391	2320	2265	2331
		2151	1838	2021	2444	2372	2392	2270	2311	2306
		2035	1895	2099	2409	2338	2396	2288	2312	2265

[a] cc/454 g.

Table 31. The mean values of specific volumes of breads as influenced by the interactions of milk treatments with level of NFDM No 2, used and oxidant level, respectively. Also, data are shown of the interactions between mixing speed and levels of NFDM used.

	Specific volume[a]					
	Percent NFDM used in formula			Oxidant level		
	3%		6%	50	70	90
Heated 190 F/30 min	2647	*	2225	2372	2417	2518
Heated 190 F/30 min, 0.05% H_2O_2	2524	*	2291	2321	2432	2469
			*		*	
Heated 190 F/30 min, 0.10% H_2O_2	2479	*	2154	2313	2248	2390
	*					*
Heated 190 F/30 min, 0.20% H_2O_2	2338		2235	2364	2279	2217
LSD	130.38			159.68		

Mixing Speed	Specific volume Percent NFDM used in formula 3%		6%
125	2352	*	2054
	*		*
175	2602	*	2374
			*
191	2537	*	2252
LSD		112.91	

* Denotes statistically significant differences between adjacent means ($P < .05$).

[a] cc/454 g.

SUMMARY AND CONCLUSIONS

This investigation was undertaken to study the feasibility of incorporating NFDM, treated with various levels of hydrogen peroxide during its preparation, into bread made by the continuous dough mixing process. Milks produced during the winter and spring were used in the preparation of NFDM. The following treatments of the milk were used: (a) unheated control; (b) 190 F/30 min; (c) 190 F/30 min, +0.05% H_2O_2; (d) 190 F/30 min, +0.10% H_2O_2; and (e) 190 F/30 min, +0.20% H_2O_2.

The NFDM samples were analyzed for sulfhydryl content, whey protein nitrogen value and residual peroxide values. Their effects on dough water absorption, in sponge dough and in continuous dough mixing processes were studied.

The sulfhydryl content of the NFDM was reduced on heating. The hydrogen peroxide oxidation further reduced the sulfhydryl content.

Undenatured whey protein nitrogen content was decreased by heat treatment. The heat treatment denatured 63.0 and 69.3% of serum protein for Milk No 1, Series A and B, (winter milk) respectively. When the milk was heated and treated with hydrogen peroxide, the whey protein nitrogen value was higher than observed for heat treated alone. This may be due to oxidation and subsequent fragmentation of casein which was not precipitated by saturated salt solution in the Harland and Ashworth procedure, or it may be due to the inability of B-lactoglobulin to form a complex with the Kappa casein moiety of Alpha casein.

Dough absorption was increased by incorporation of NFDM. This was due to the increase in absorbance sites resulting from the addition of milk proteins. The valorimeter values were decreased by incorporating the NFDM samples.

Statistically significant ($P < .05$) data for the bread produced by using winter milk showed that the best overall bread characteristics were obtained by the following combination of processing conditions and usage levels: 190 F/30 min heat treatment following oxidation of the milk with 0.05 percent hydrogen peroxide; incorporated the NFDM in the dough at the three percent level based on flour weight; mixing the dough in the developer head at a speed of 218 rpm. There was no statistical difference in the three levels of dough oxidant used. This was undoubtedly due to the interaction involving excessive water in the formula and the high percentage of total oxidizing power contributed by the residual hydrogen peroxide in the NFDM.

For studies involving the winter milk, the water absorptions used were arbitrarily fixed. This resulted in slack doughs due to more water being added than was indicated by the farinograms when NFDM was used in the formula. The excessive slackness of the doughs resulted in unrealistically low power requirements thus precluding the drawing of meaningful conclusions relative to these parameters.

The best overall characteristics of bread containing spring milk were produced when NFDM was used at the 3% level and preheated at 190 F/30 min following 0.05 percent hydrogen peroxide treatment using 90 ppm dough oxidant and mixing at 171 or 218 rpm. The smallest power requirement constant with a high total bread score and realistic rheological properties was when 50 ppm dough oxidant was used in conjunction with the incorporation of 3% NFDM preheated to 190 F/30 min following treatment with 0.05% H_2O_2 and mixed at 125 rpm.

The data obtained from this study indicate that loaf volume, bread cell structure and the power required to fully develop the dough can be altered favorably by the incorporation of hydrogen peroxide treated NFDM. Incorporation of NFDM at the 3% level gave better results than at the 6% level. However, it should be noted that the adverse effects observed as a result of the latter were attributed to over-oxidized doughs.

This study should be considered preliminary to a more exhaustive undertaking to determine the optimum conditions under which oxidized NFDM might be used to overcome some of the difficulties encountered when commercial NFDM is used in the continuous mix process.

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Special and sincere gratitude is extended to my parents whose encouragement and understanding made it all possible.

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APPENDIX

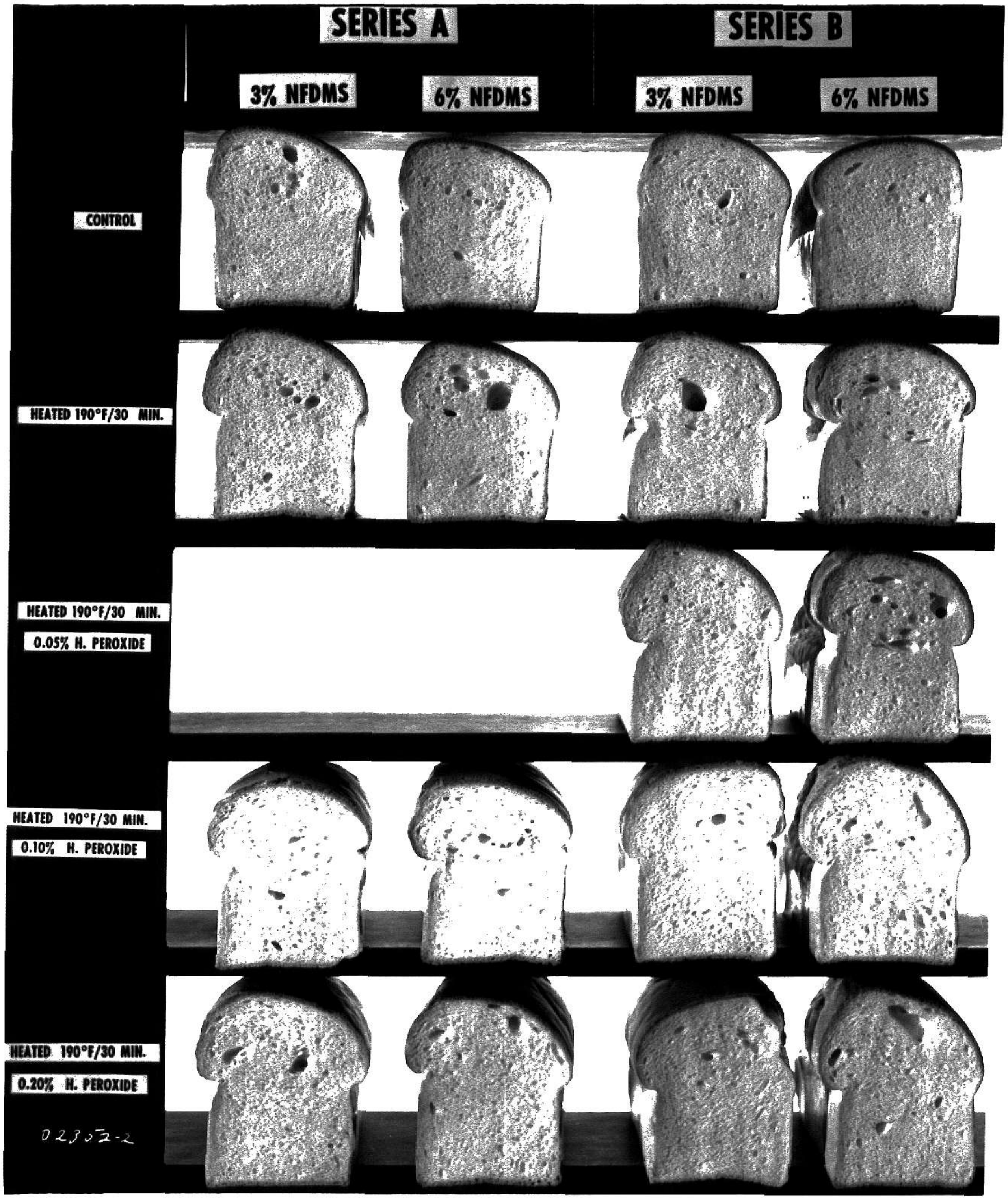

Figure 1. Cross section view of bread made by sponge dough process using NFDM series A and B.

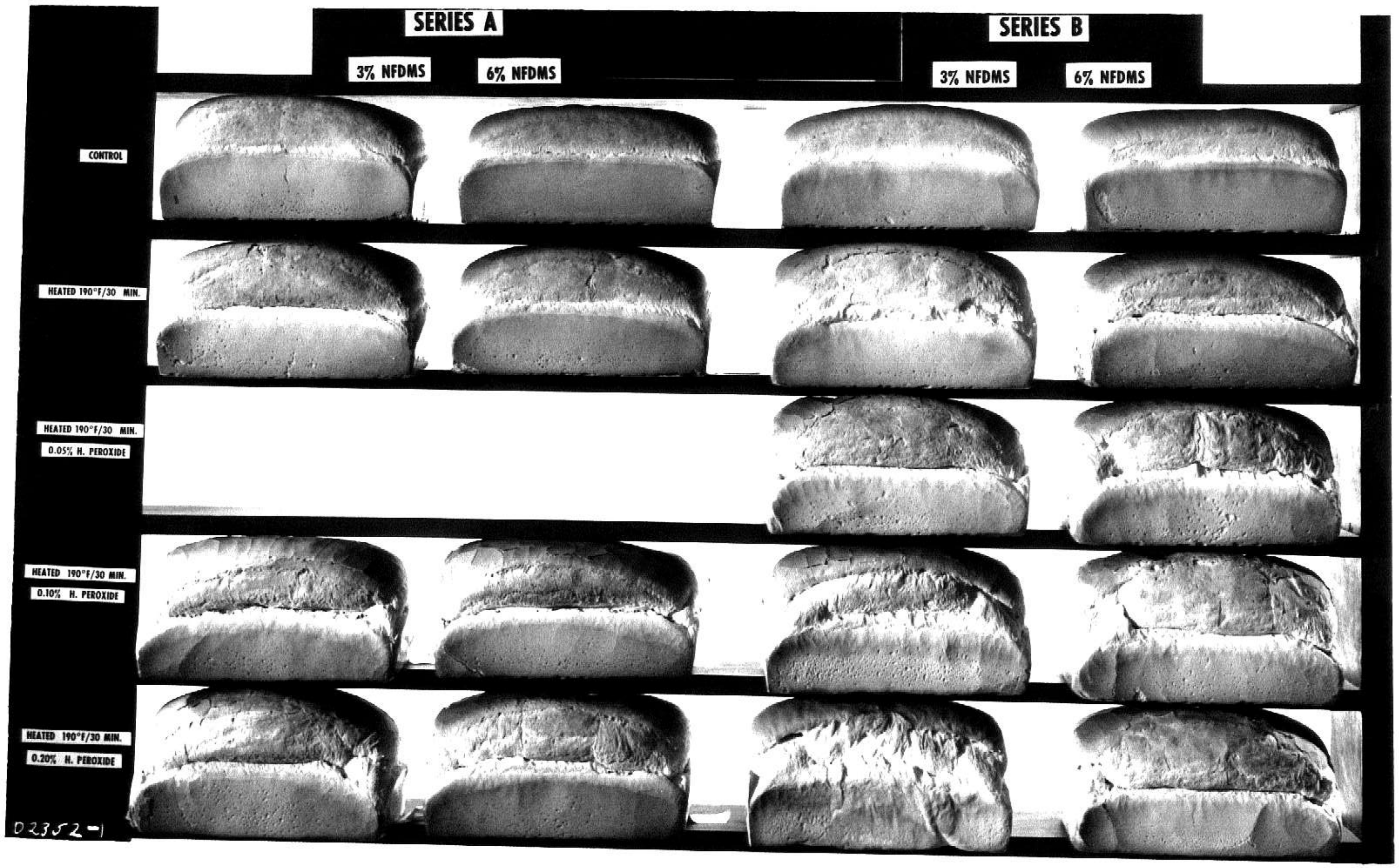

Figure 2. Longitudinal view of bread made by sponge dough process using NFDM series A and B.

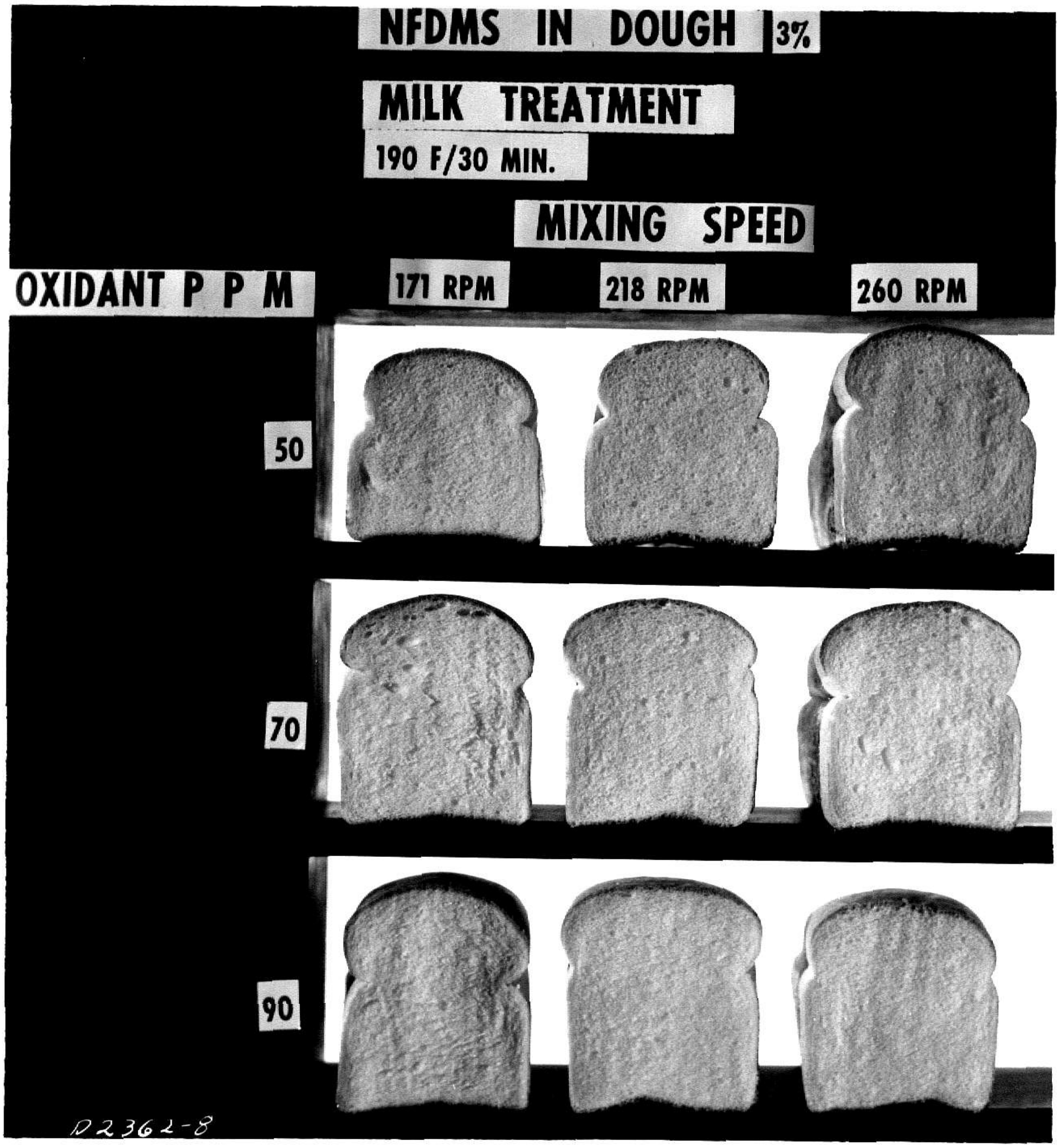

Figure 3. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min and used at 3% level.

Figure 4. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min and used at 3% level.

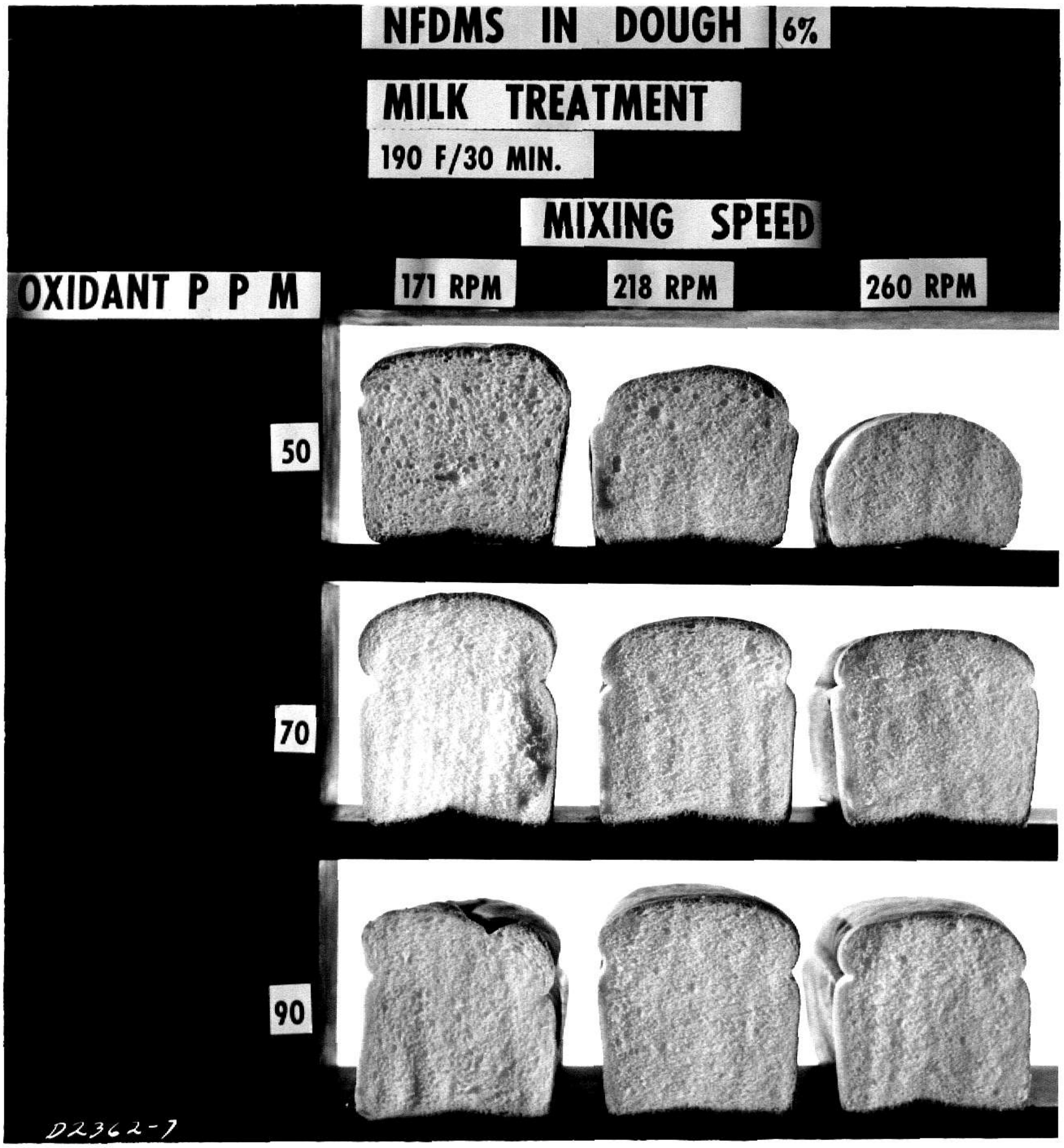

Figure 5. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min and used at 6% level.

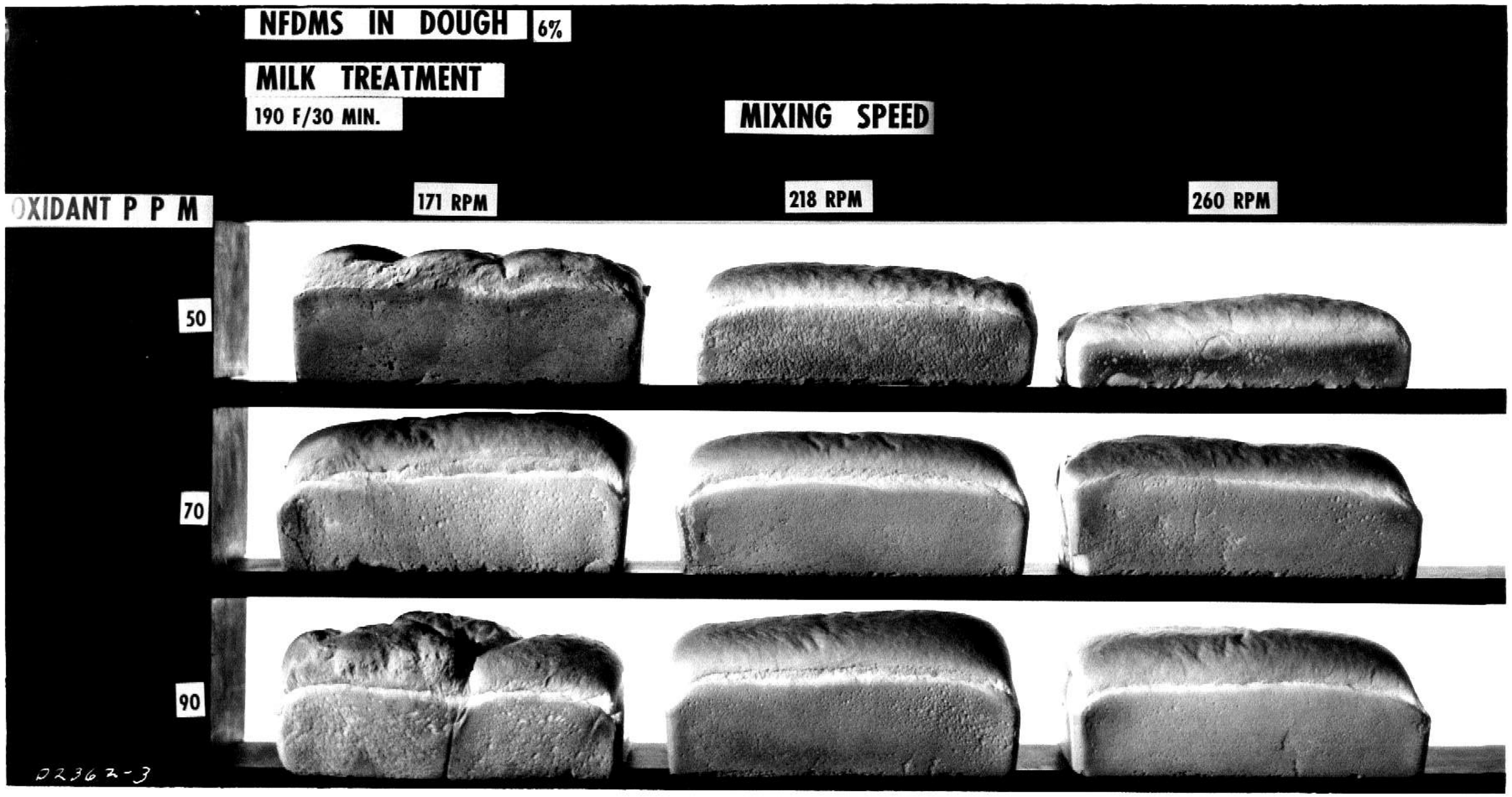

Figure 6. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min and used at 6% level.

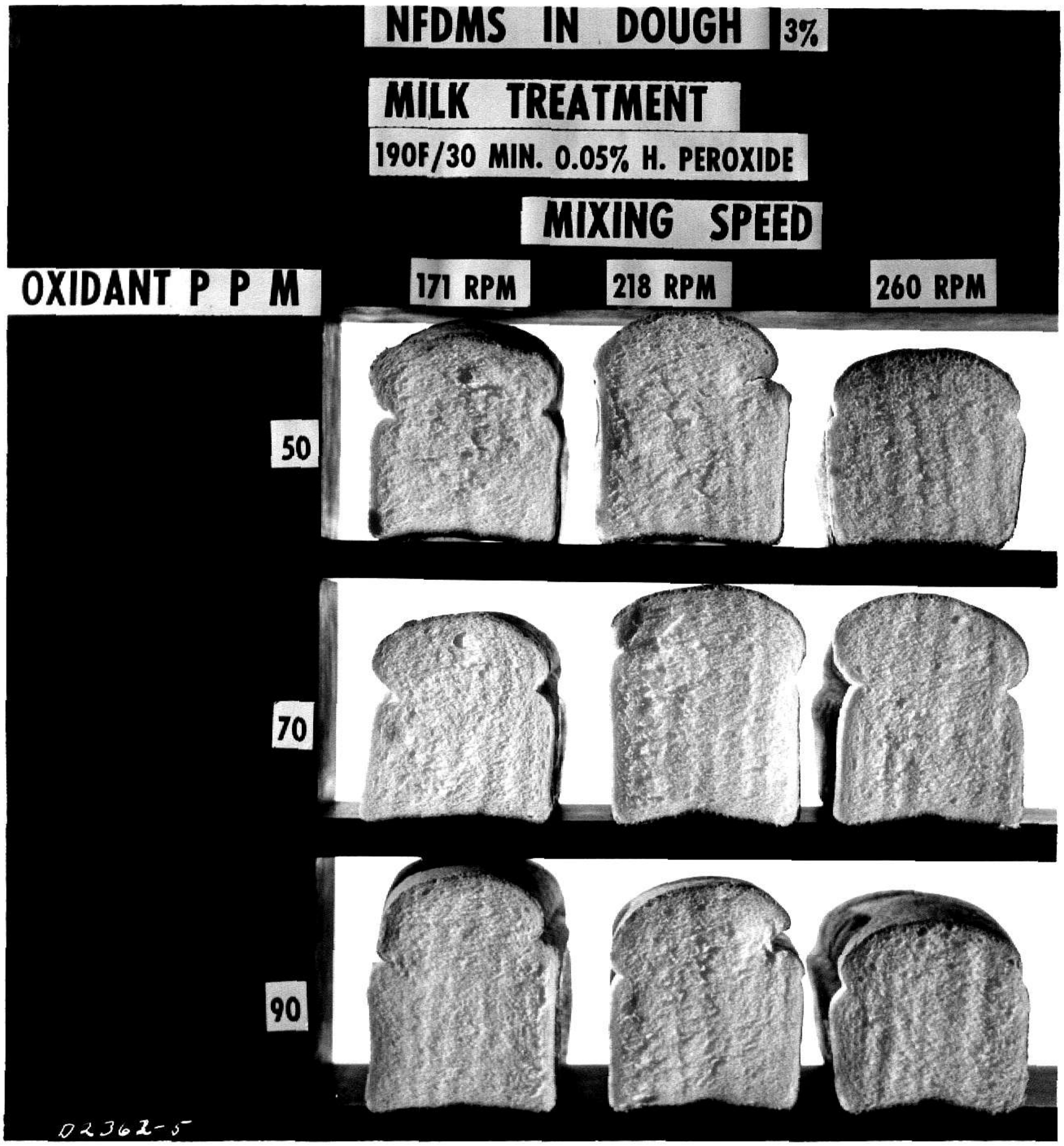

Figure 7. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.05% H_2O_2 and used at 3% level.

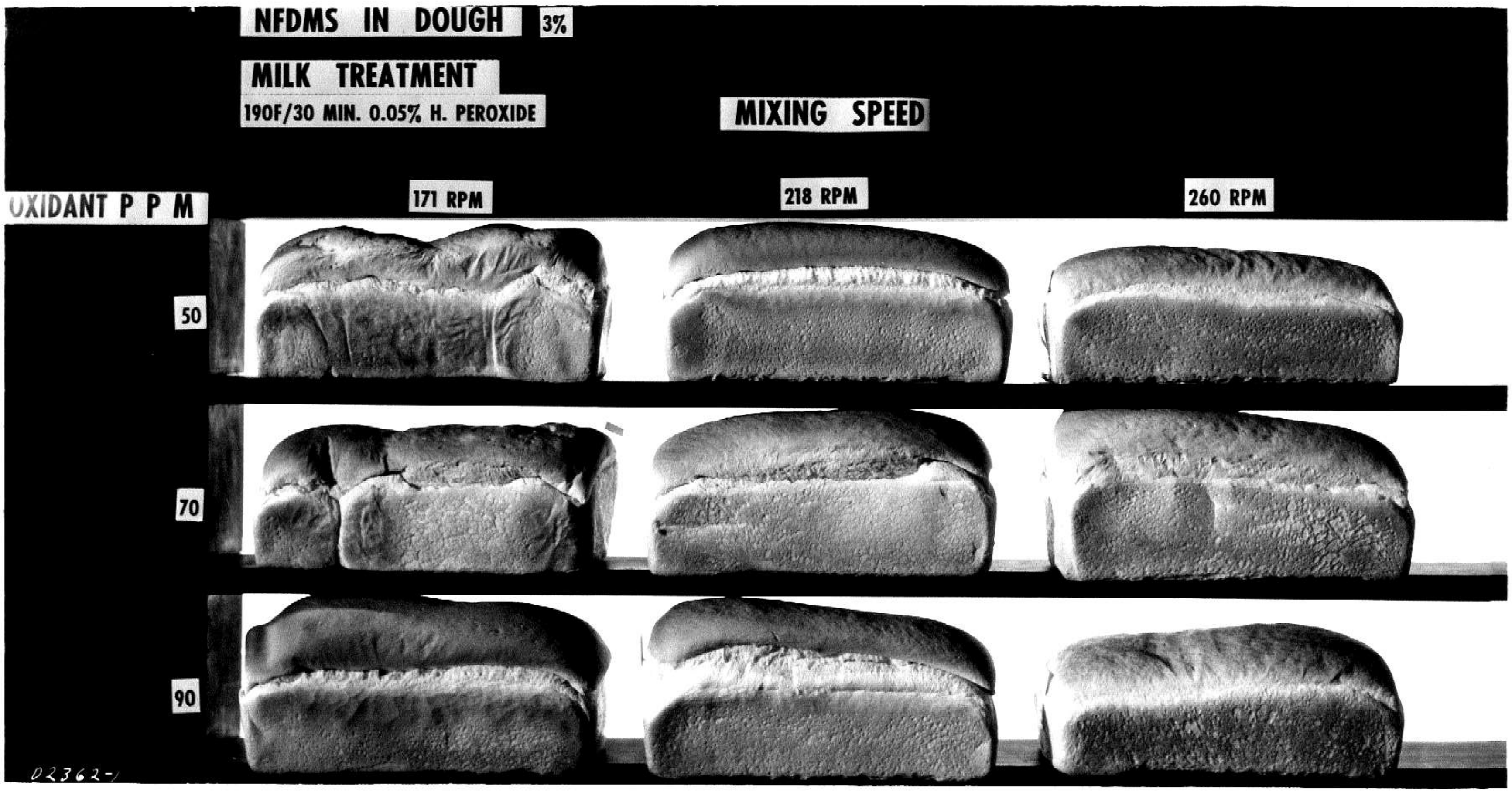

Figure 8. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.05% H_2O_2 and used at 3% level.

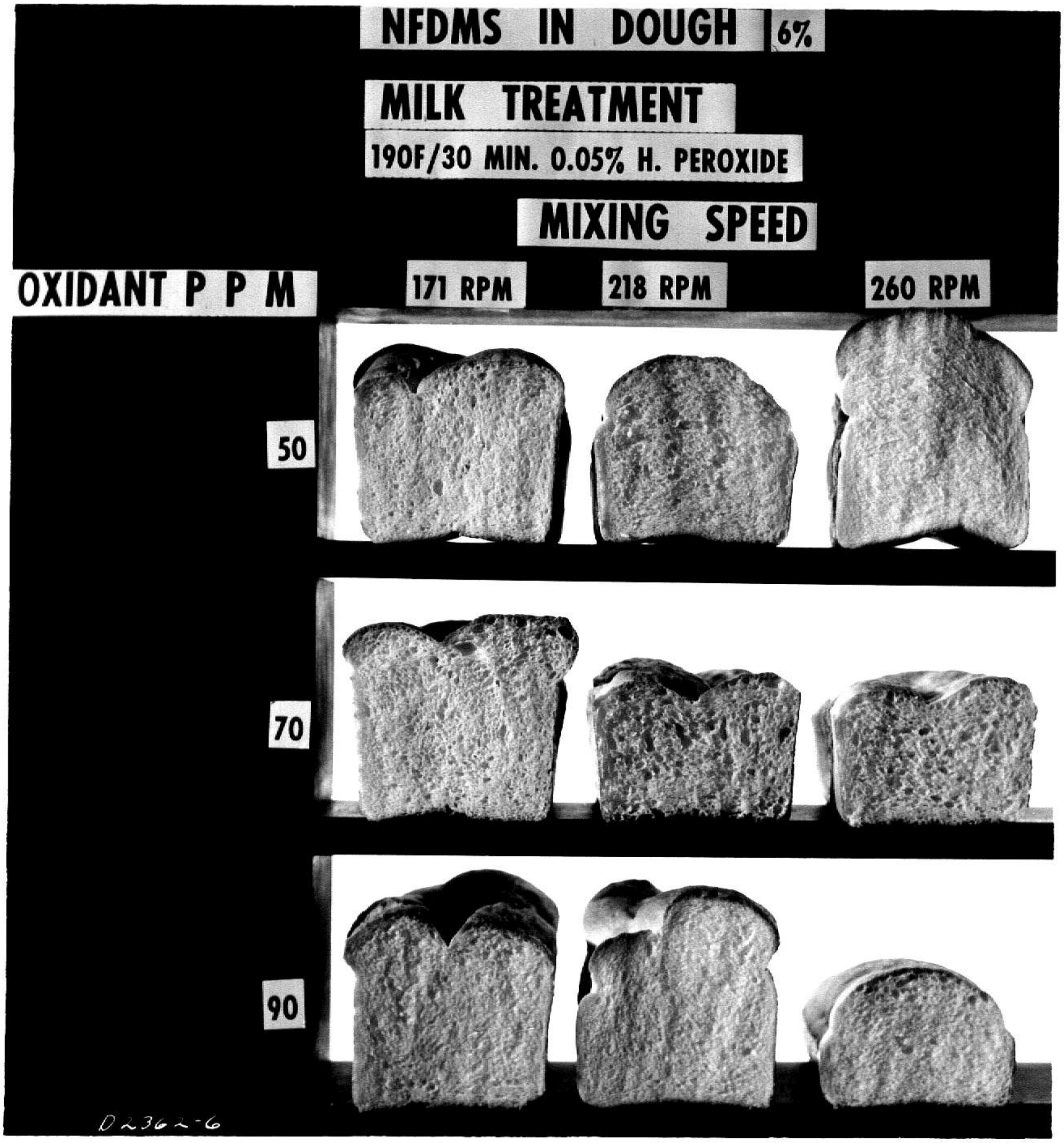

Figure 9. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.05% H_2O_2 and used at 6% level.

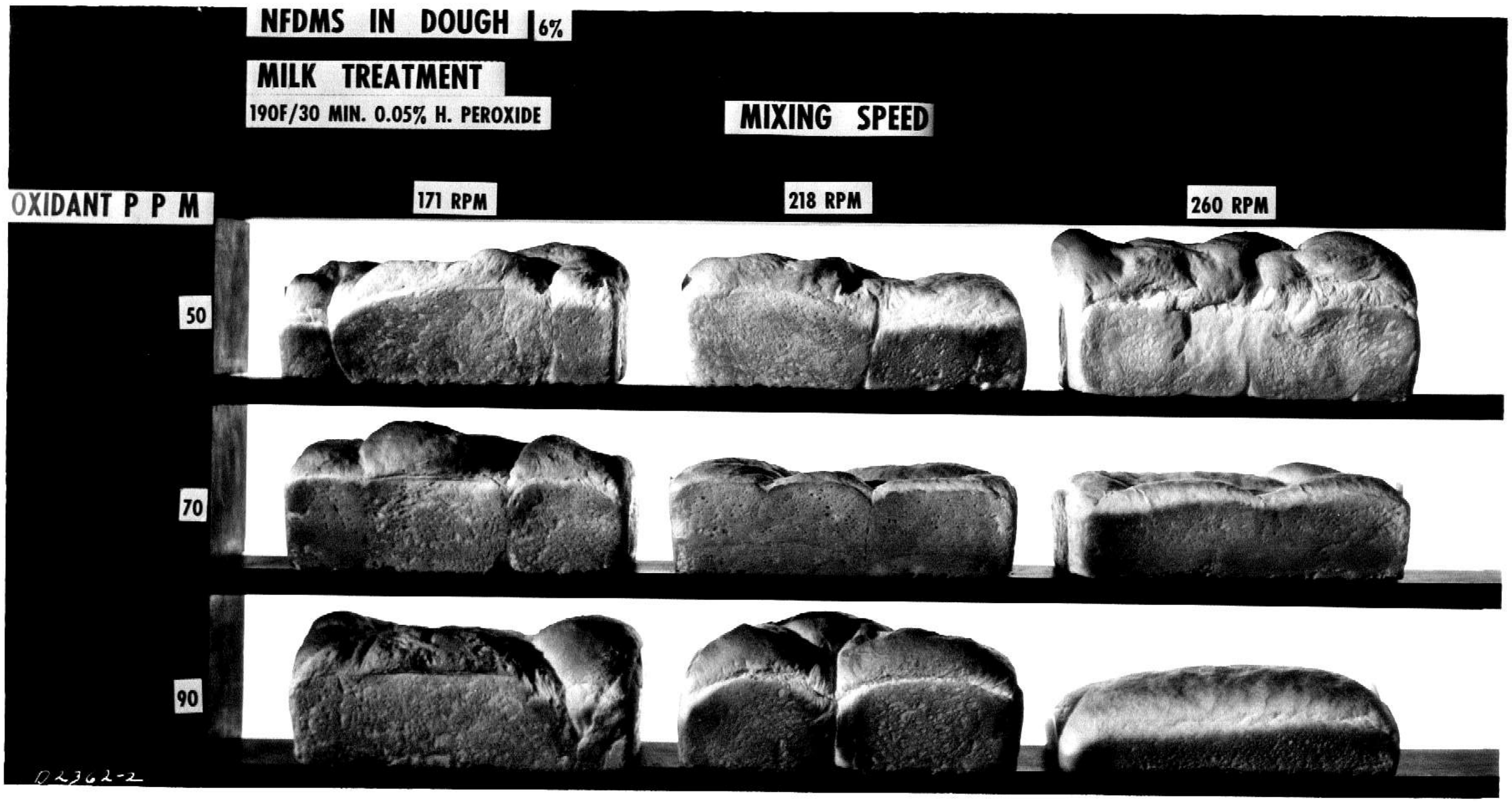

Figure 10. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.05% H_2O_2 and used at 6% level.

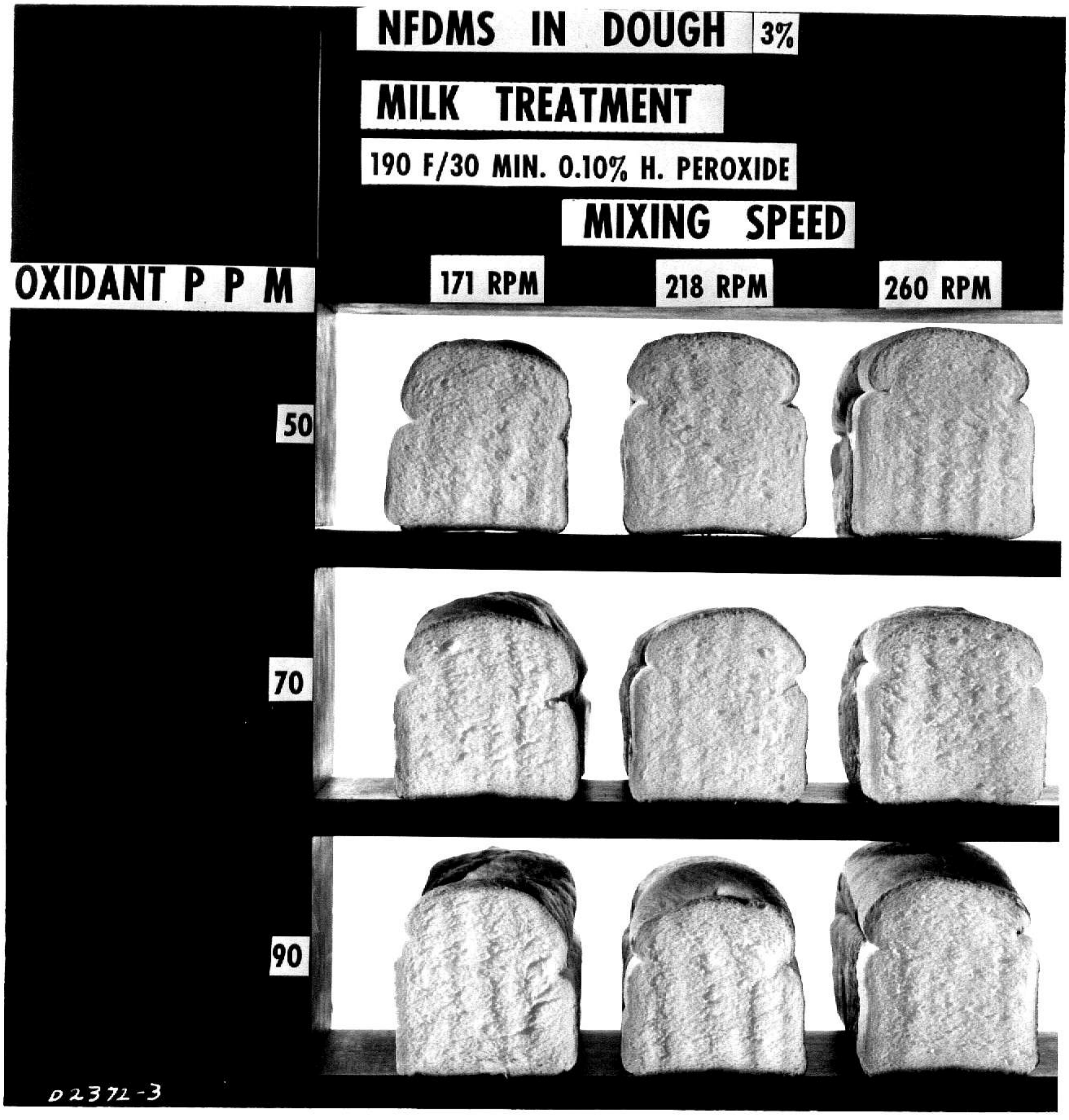

Figure 11. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.10% H_2O_2 and used at 3% level.

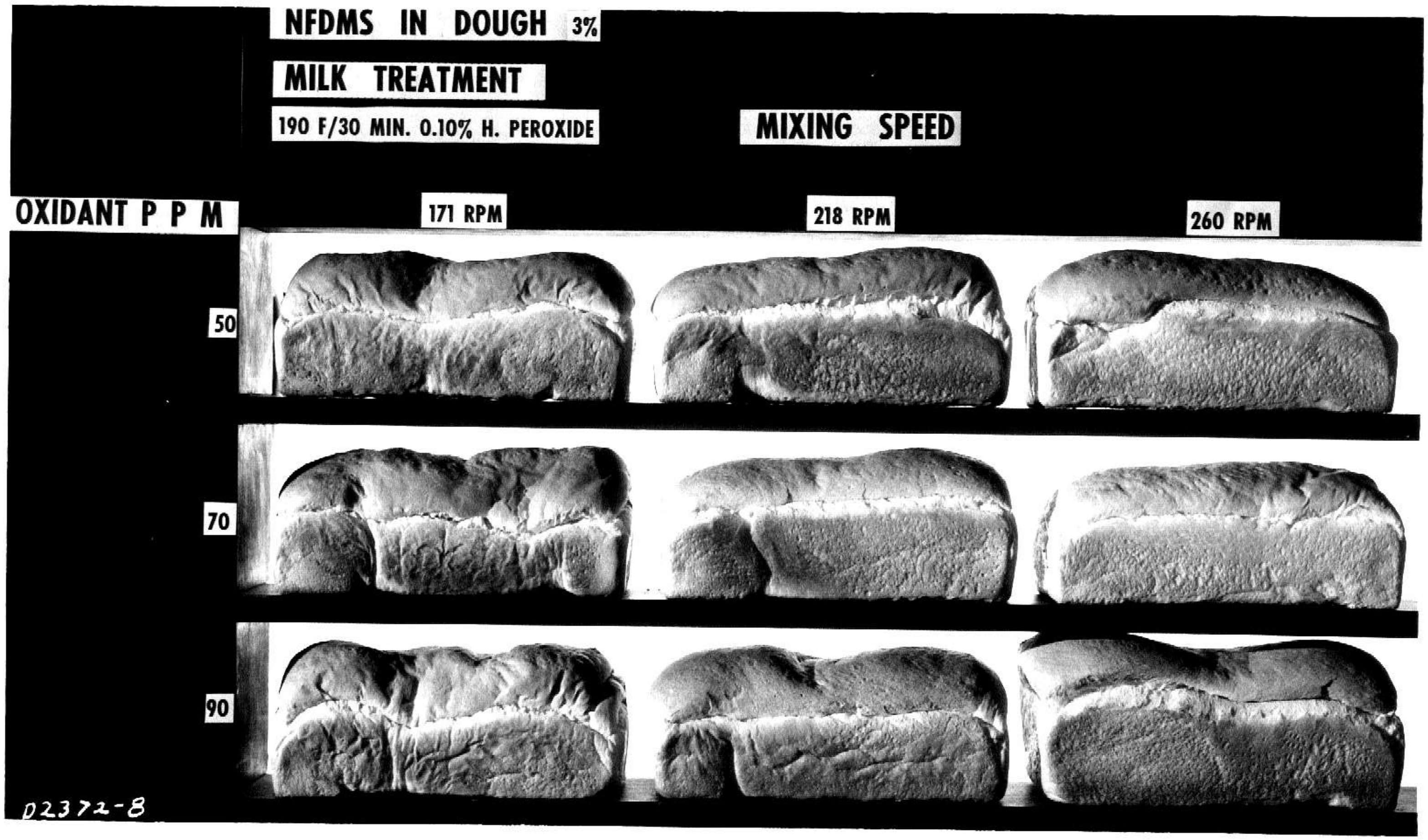

Figure 12. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.10% H_2O_2 and used at 3% level.

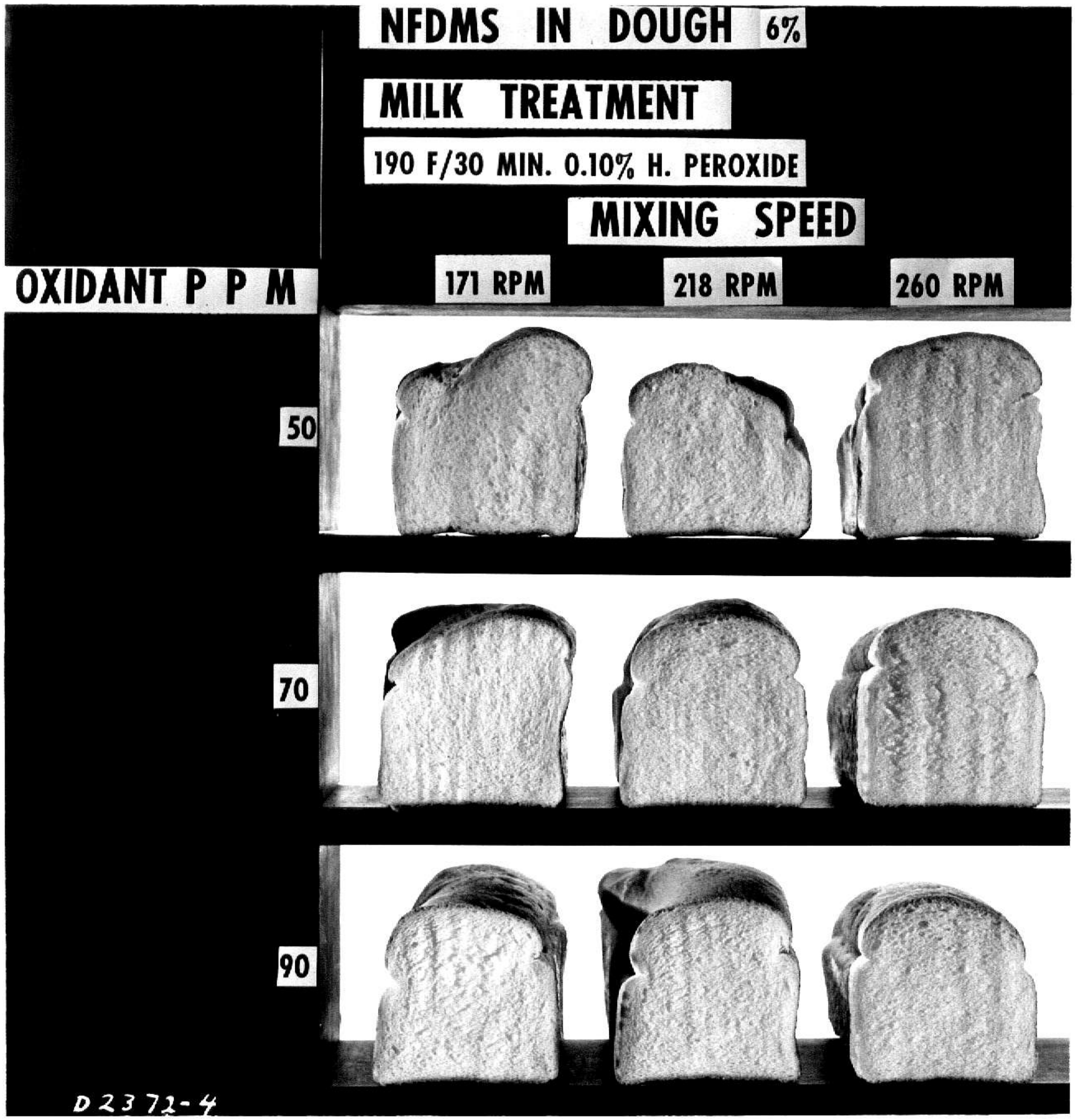

Figure 13. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.10% H_2O_2 and used at 6% level.

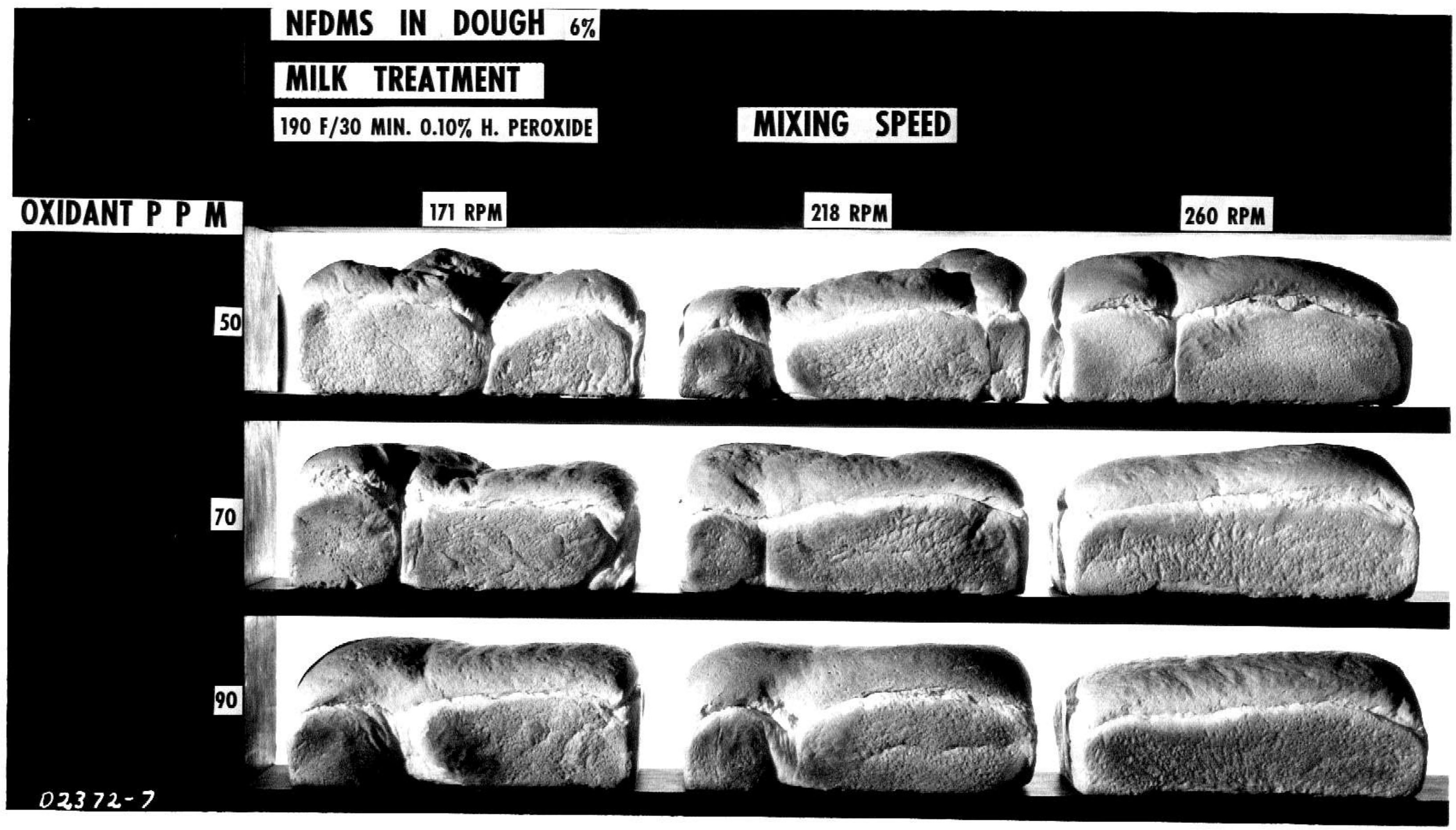

Figure 14. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.10% H_2O_2 and used at 6% level.

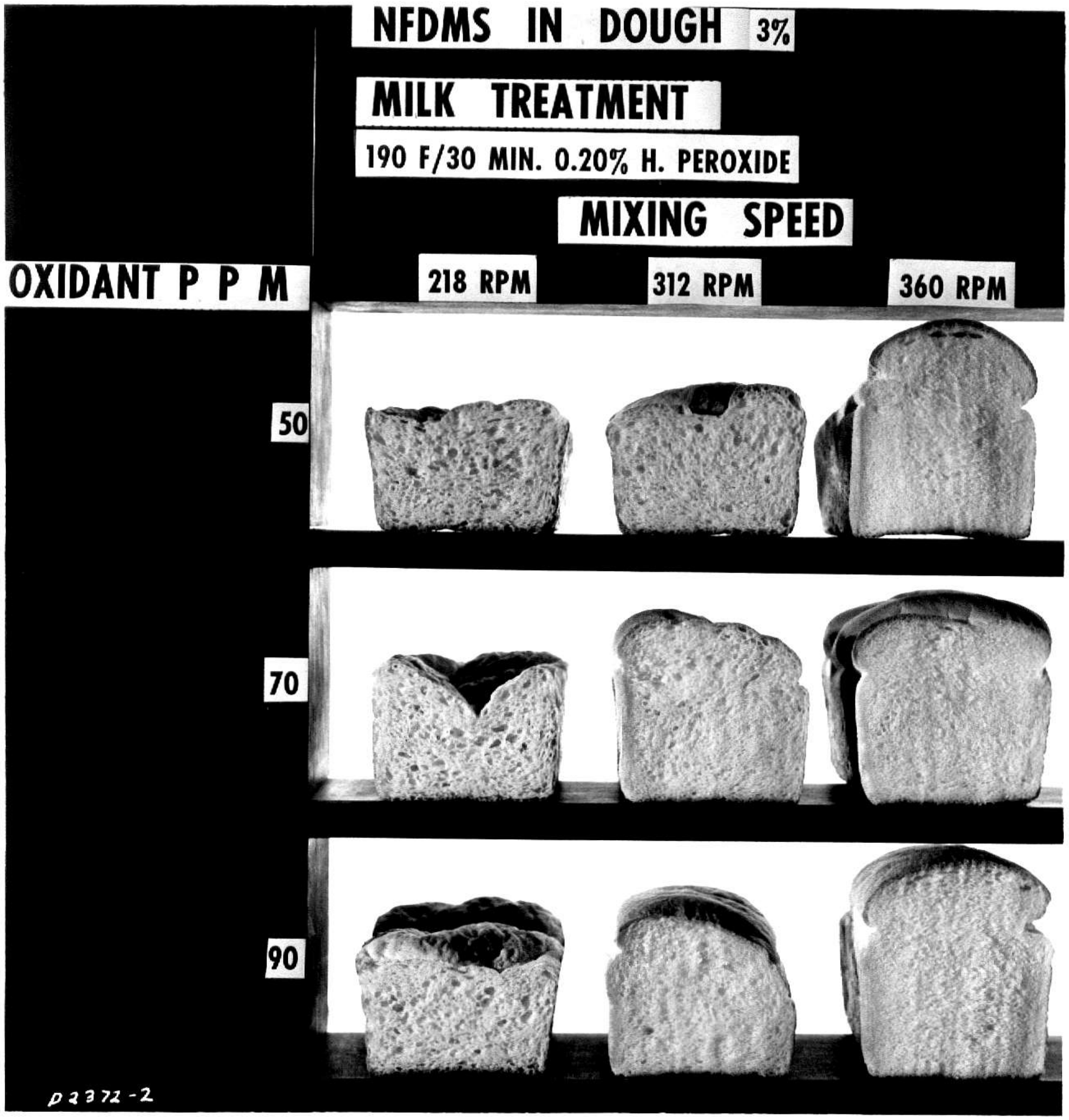

Figure 15. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.20% H_2O_2 and used at 3% level.

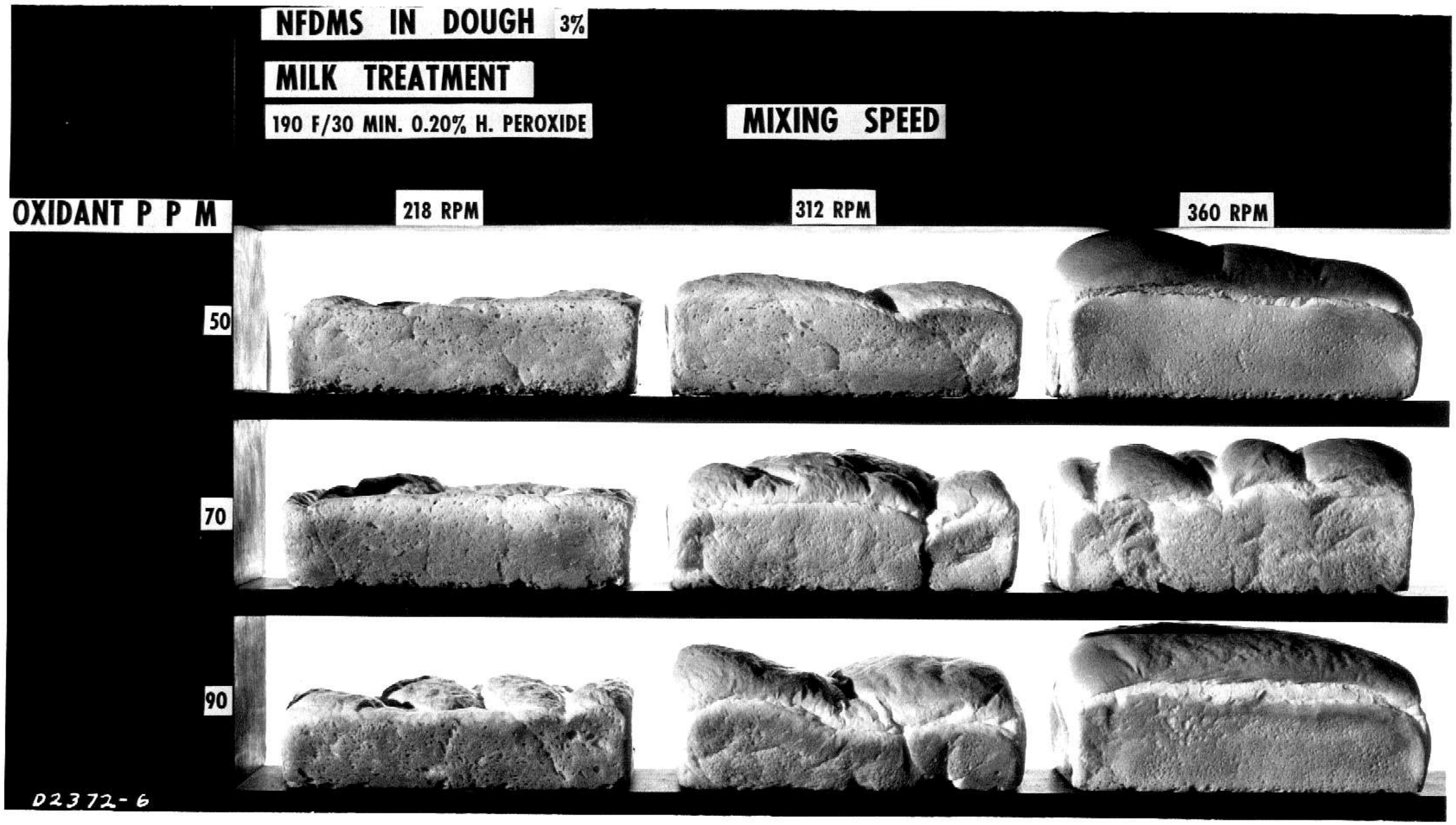

Figure 16. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.20% H_2O_2 and used at 3% level.

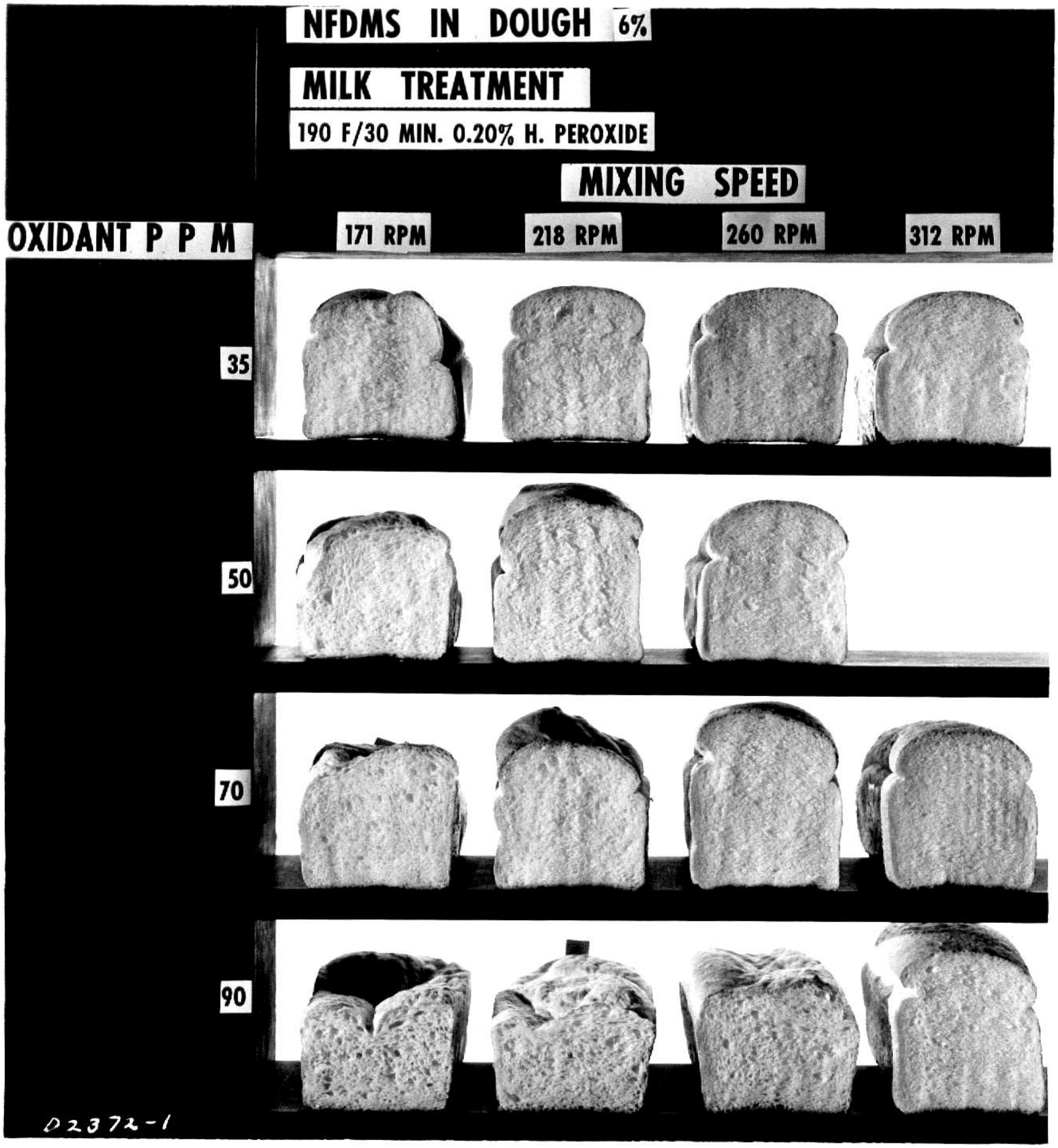

Figure 17. Cross section view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.20% H_2O_2 and used at 6% level.

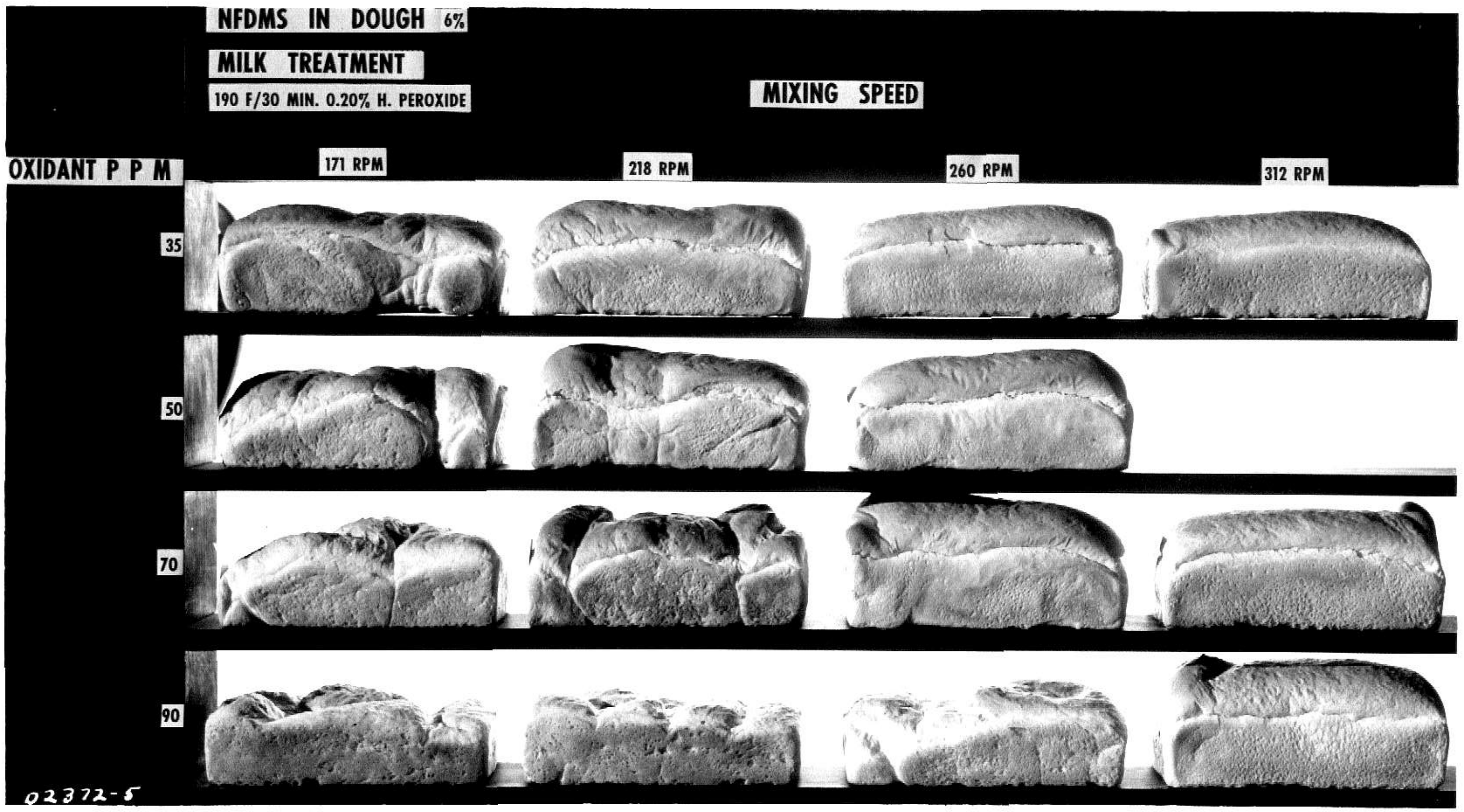

Figure 18. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 1, series B treated 190 F/30 min, 0.20% H_2O_2 and used at 6% level.

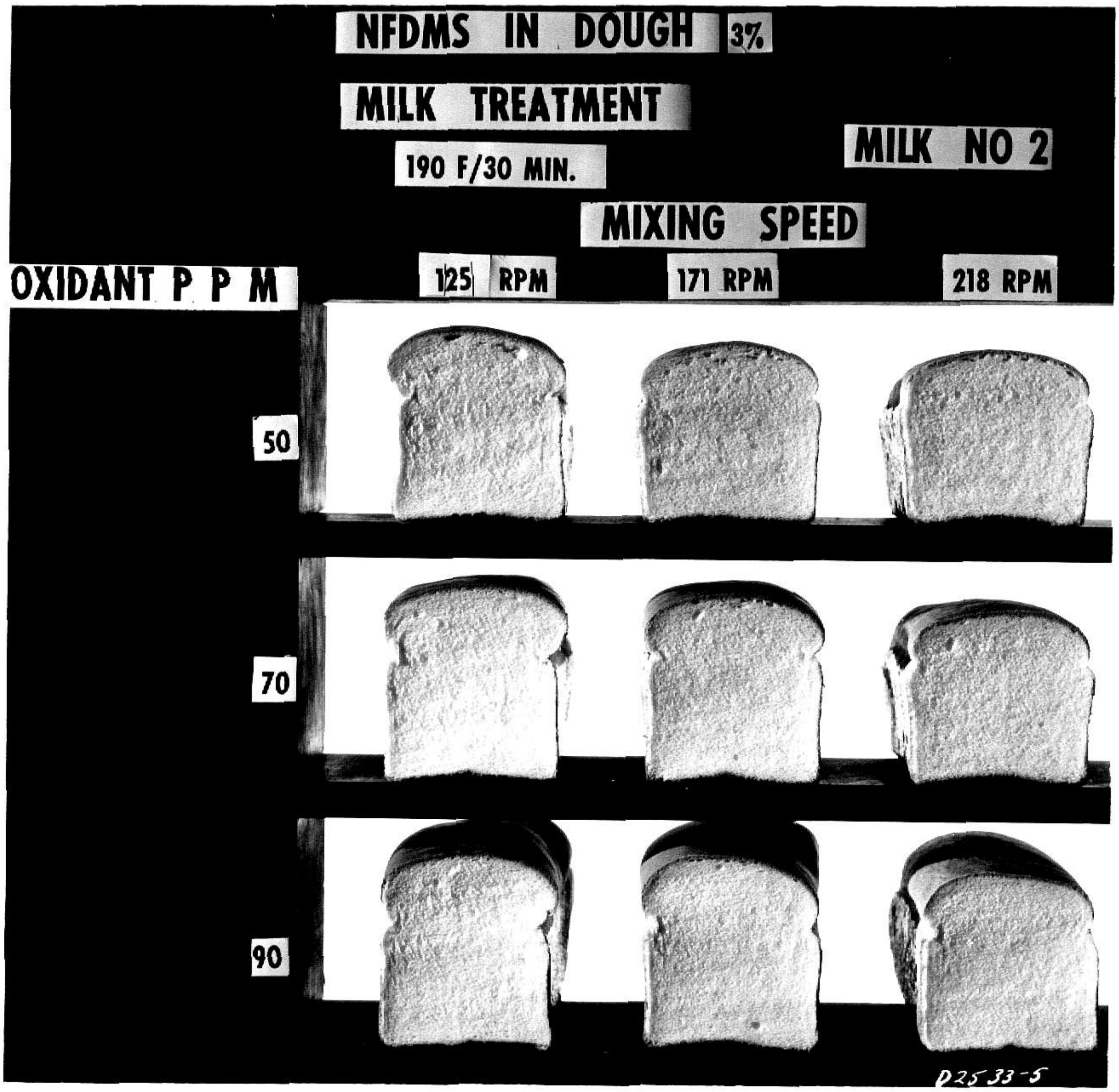

Figure 19. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2 treated 190 F/30 min, and used at 3% level.

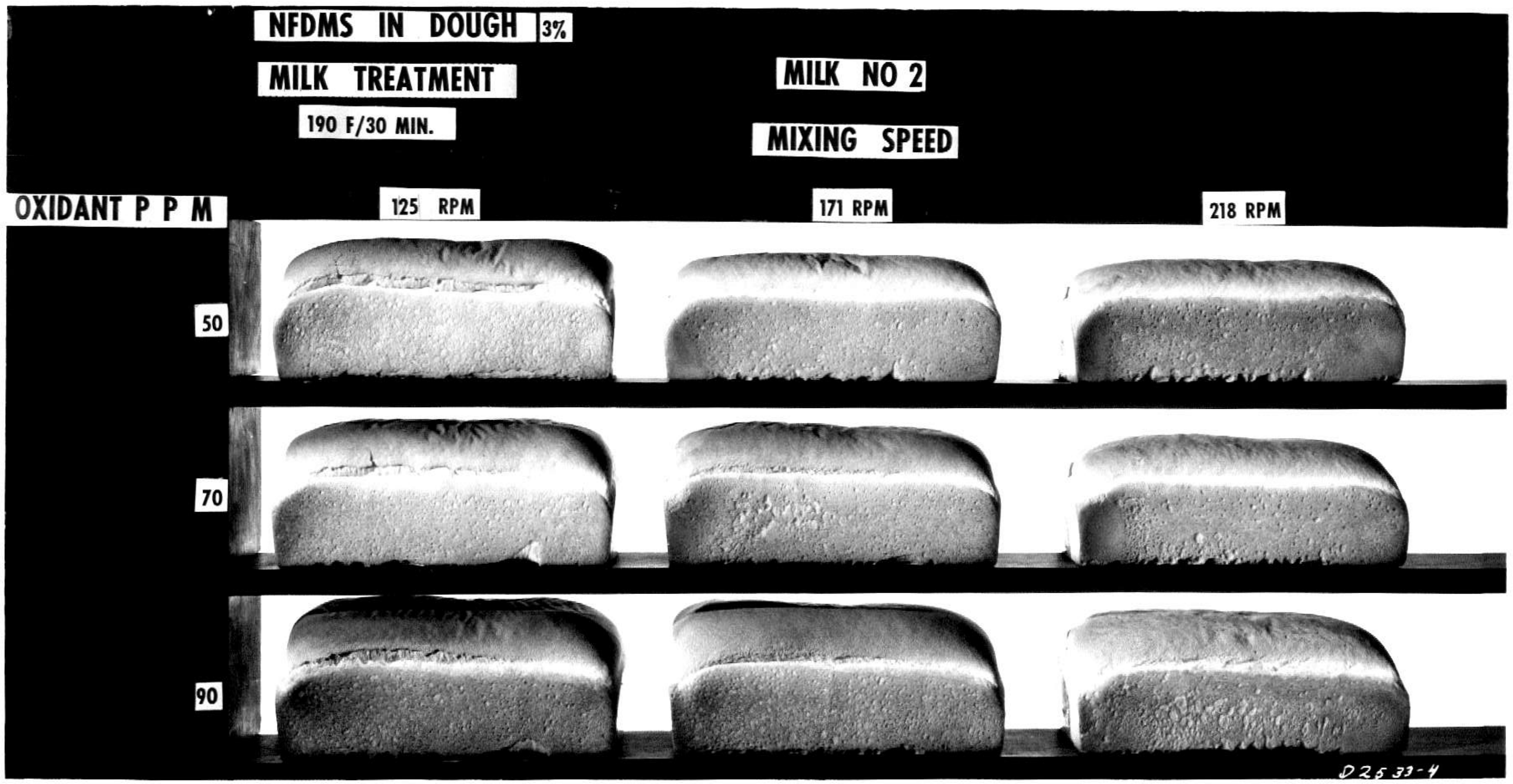

Figure 20. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2 treated 190 F/30 min and used at 3% level.

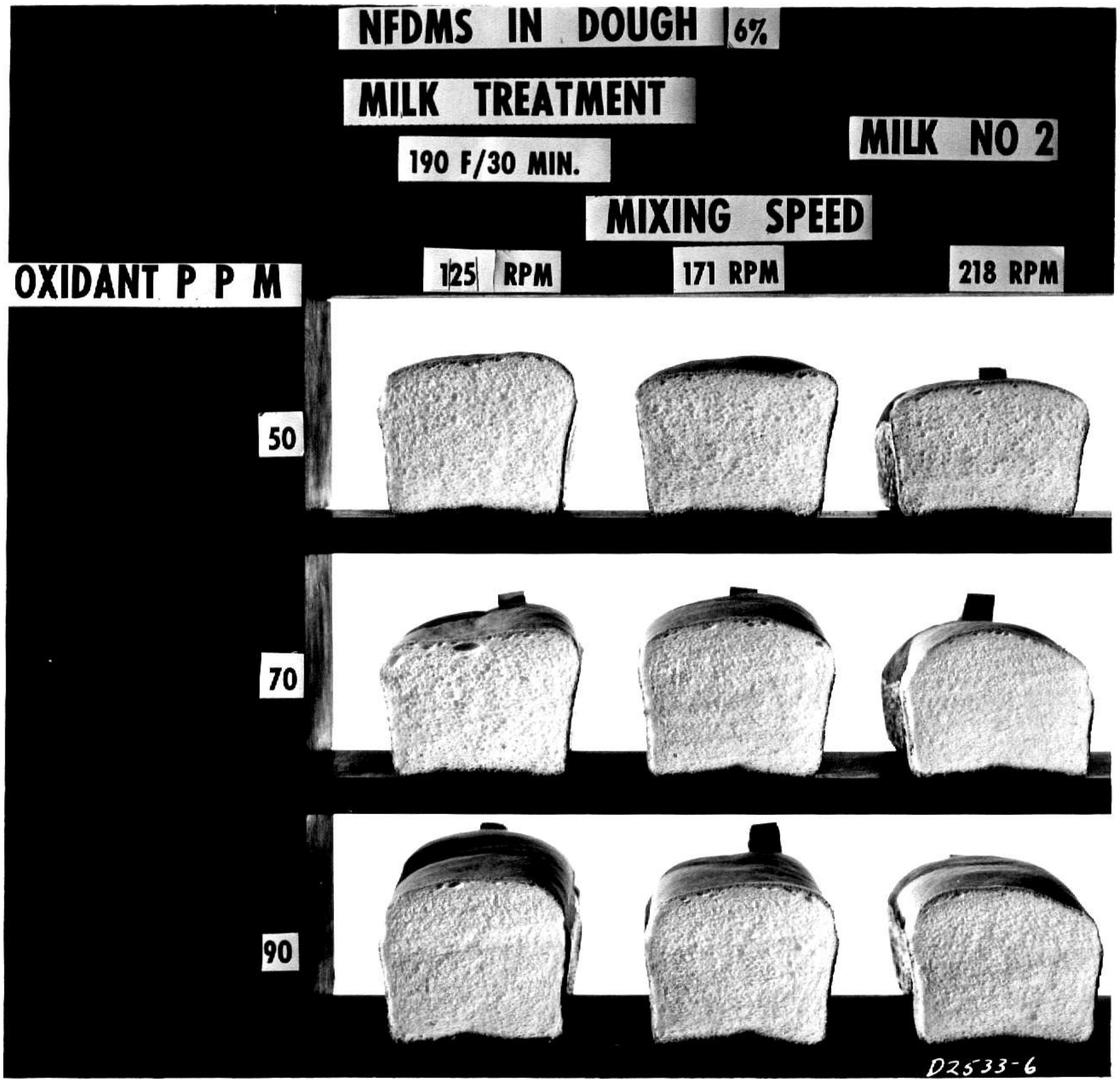

Figure 21. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2 treated 190 F/30 min and used at 6% level.

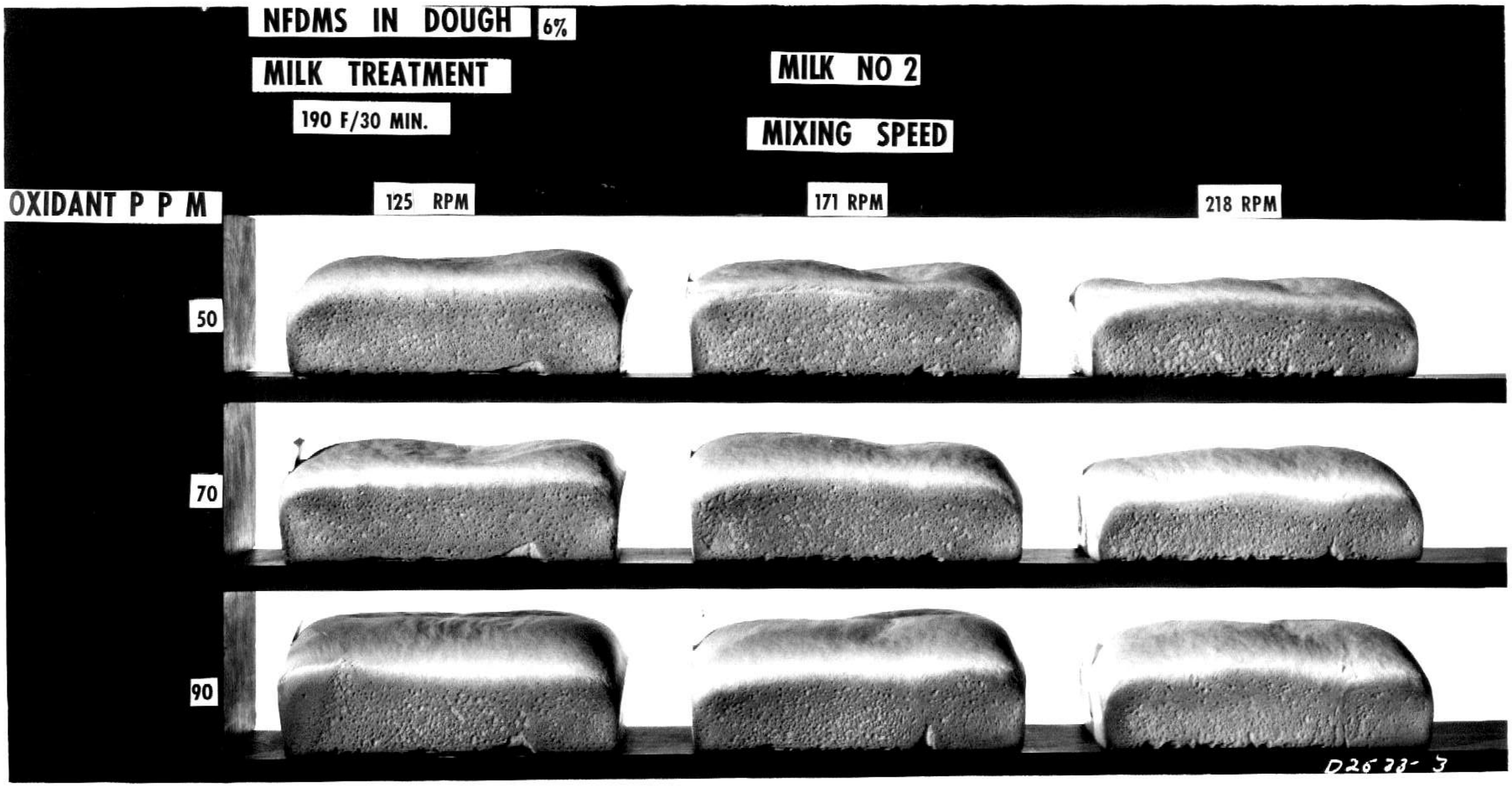

Figure 22. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min and used at 6% level.

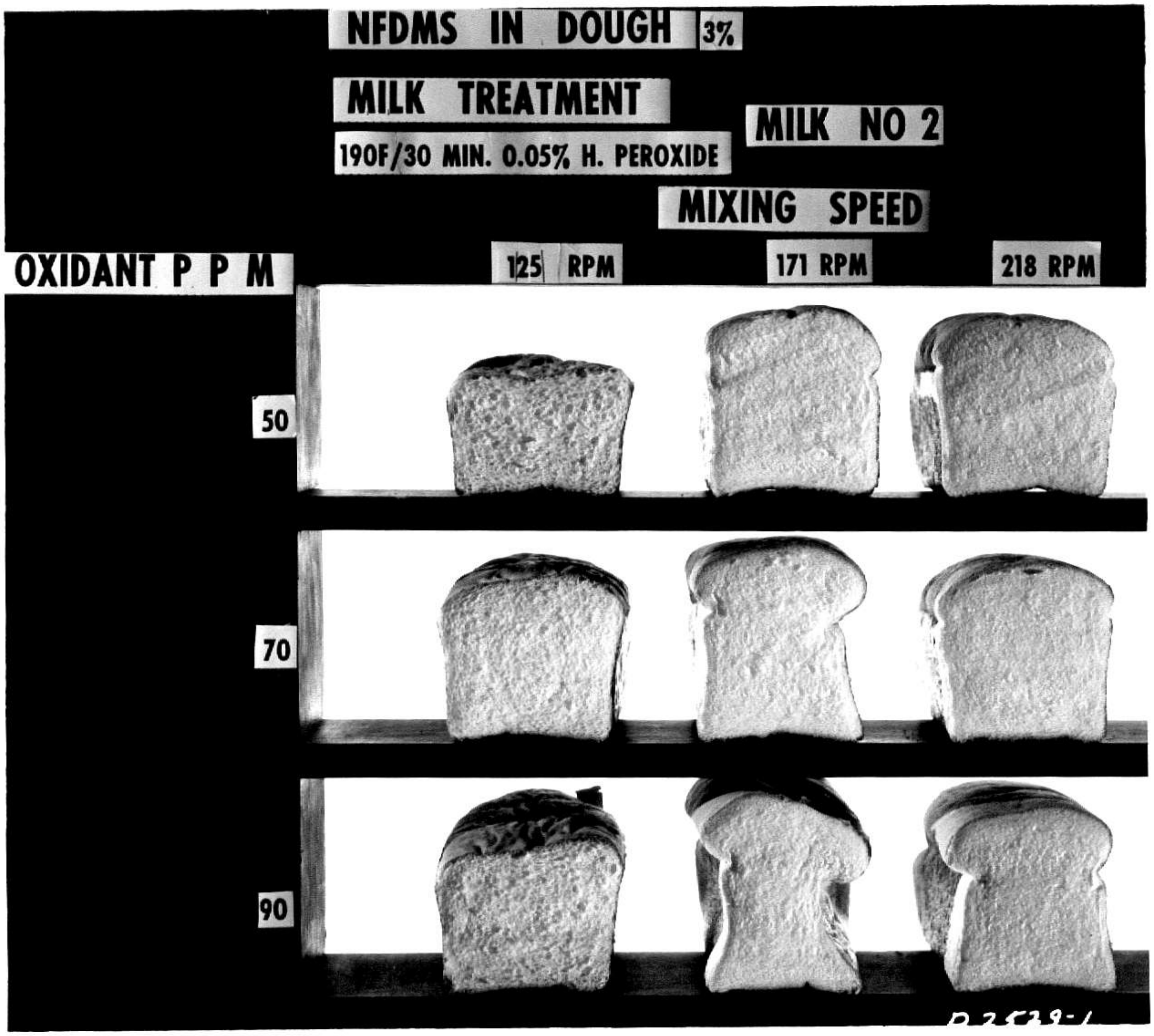

Figure 23. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.05% H_2O_2 and used at 3% level.

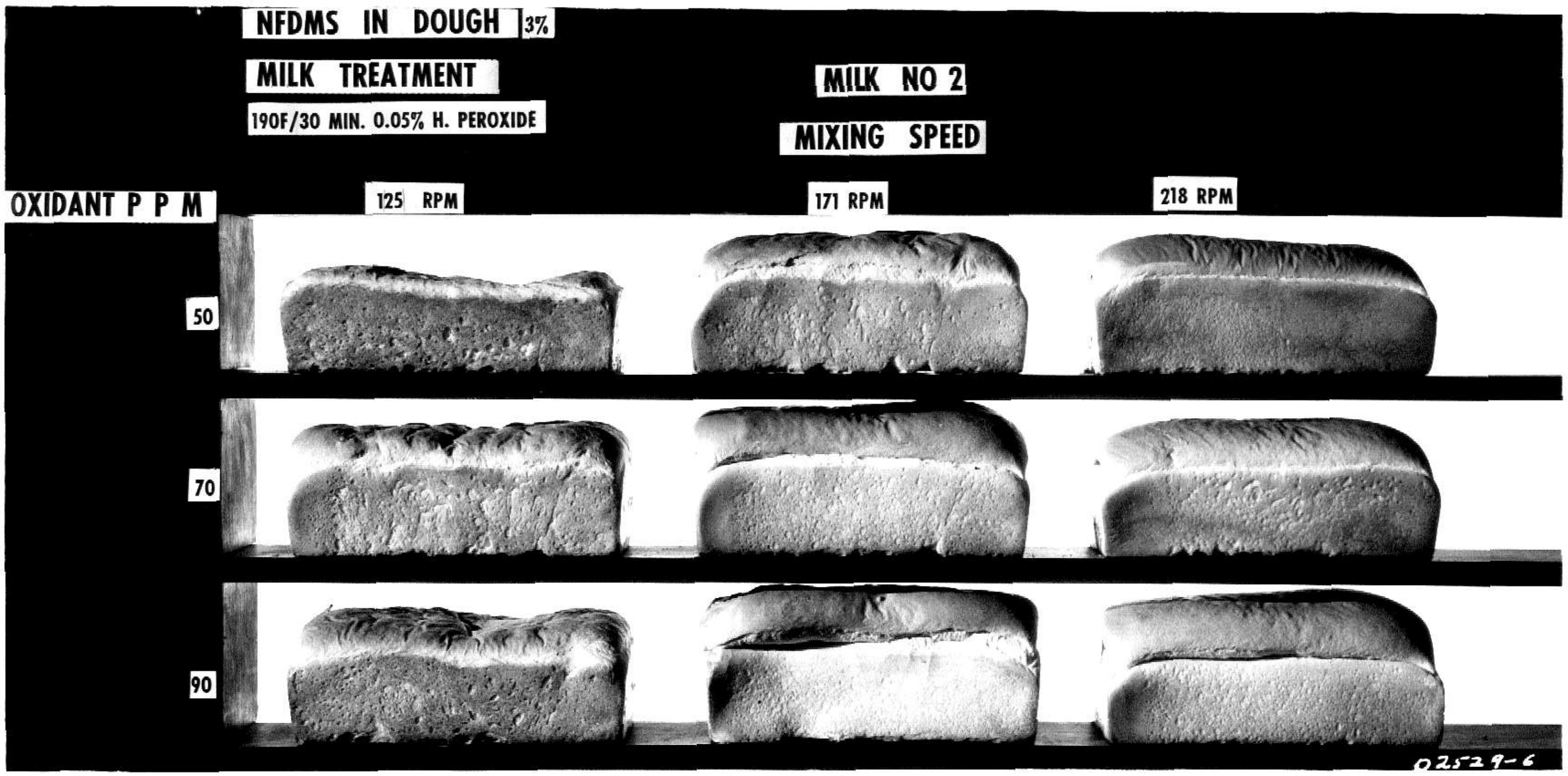

Figure 24. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.05% H_2O_2 and used at 3% level.

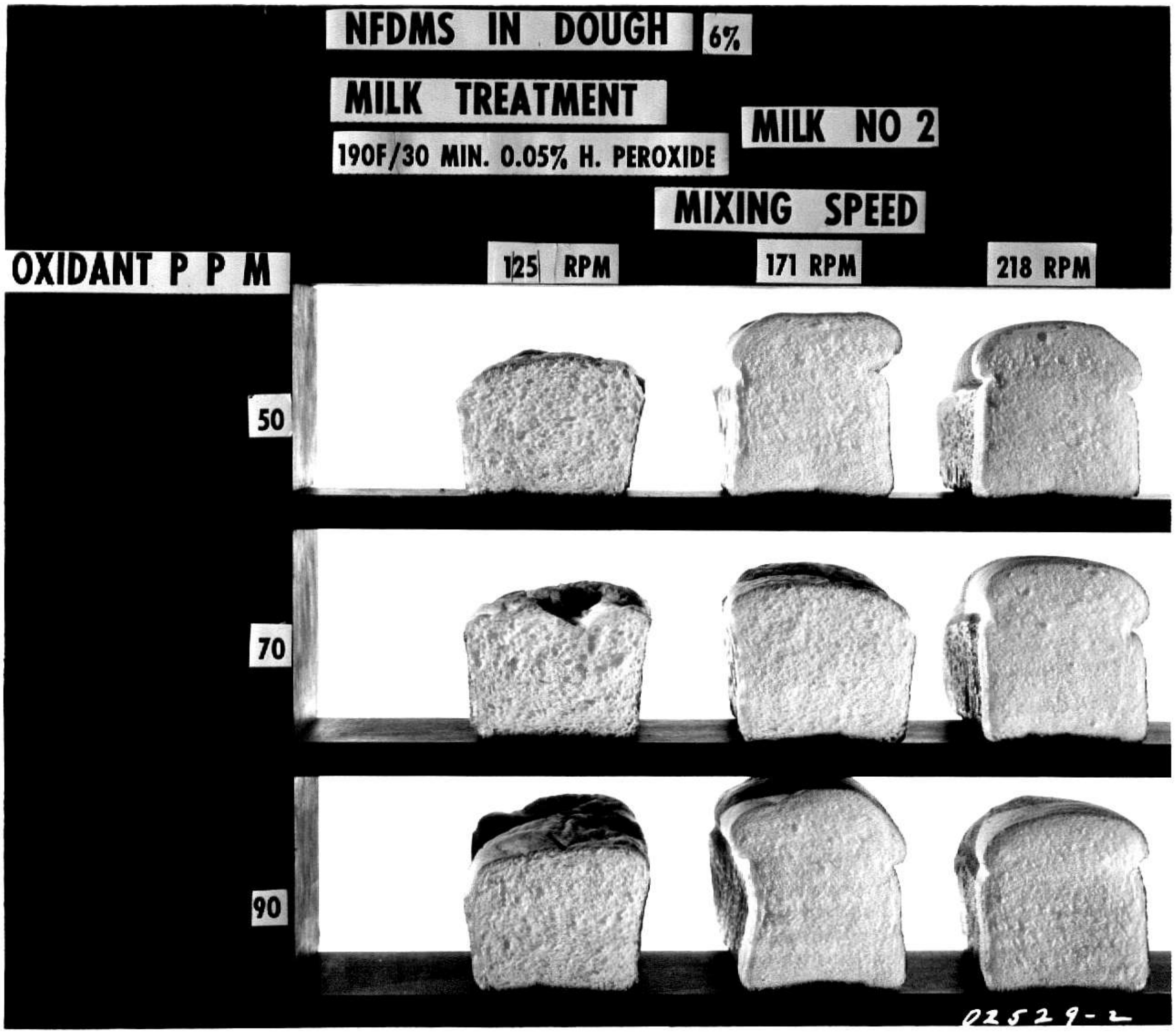

Figure 25. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.05% H_2O_2 and used at 6% level.

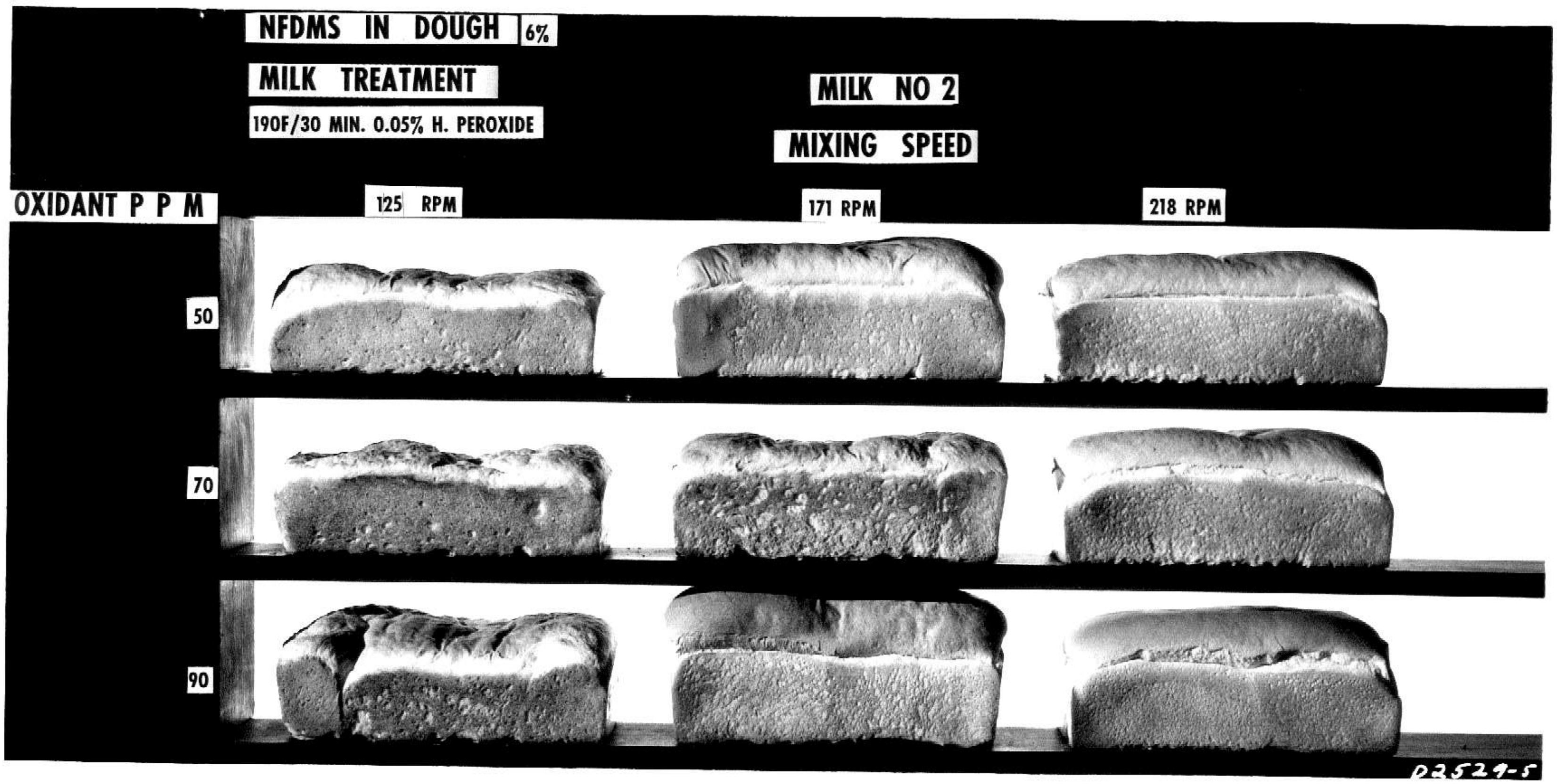

Figure 26. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.05% H_2O_2 and used at 6% level.

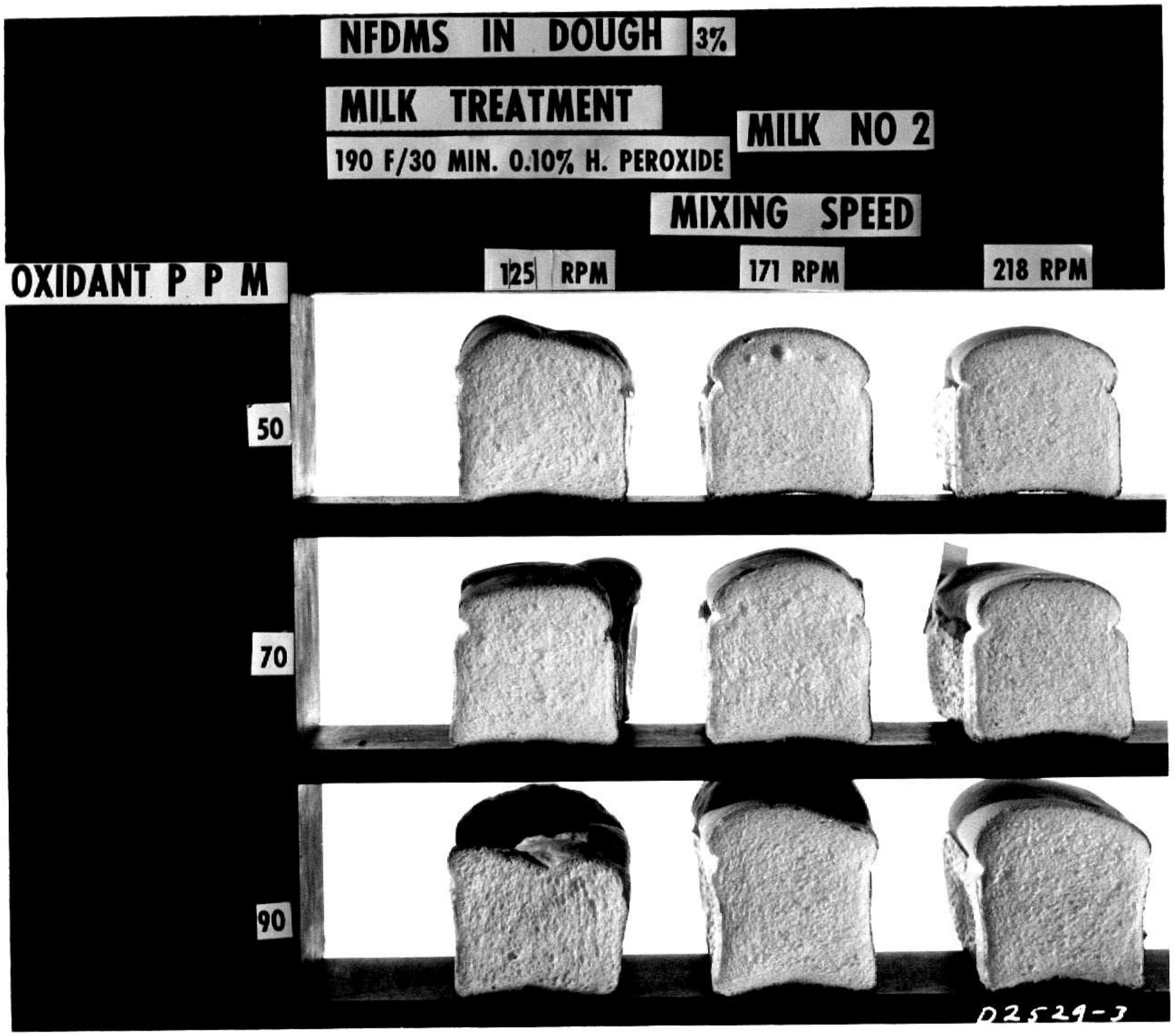

Figure 27. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.10% H_2O_2 and used at 3% level.

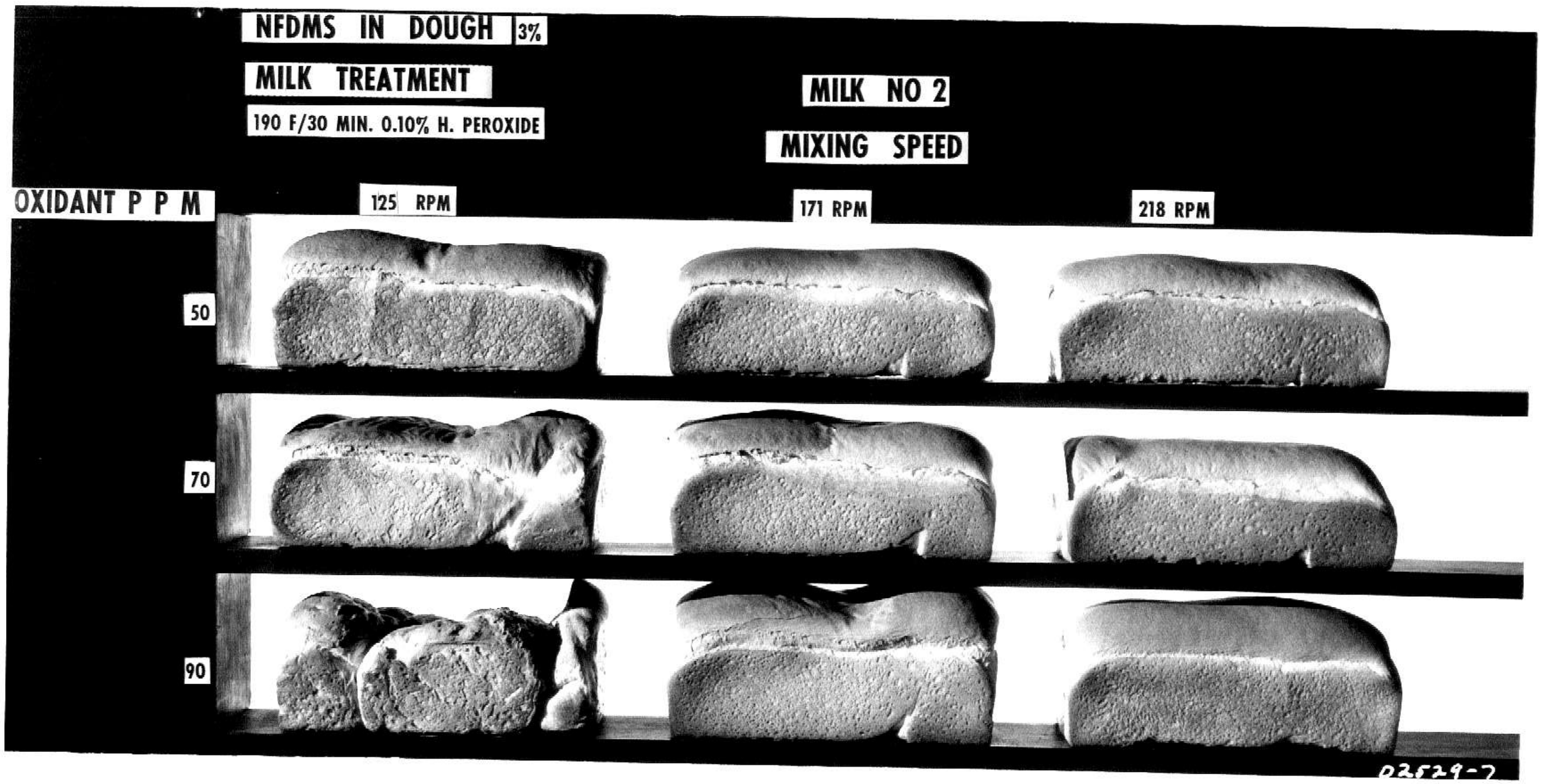

Figure 28. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.10% H_2O_2 and used at 3% level.

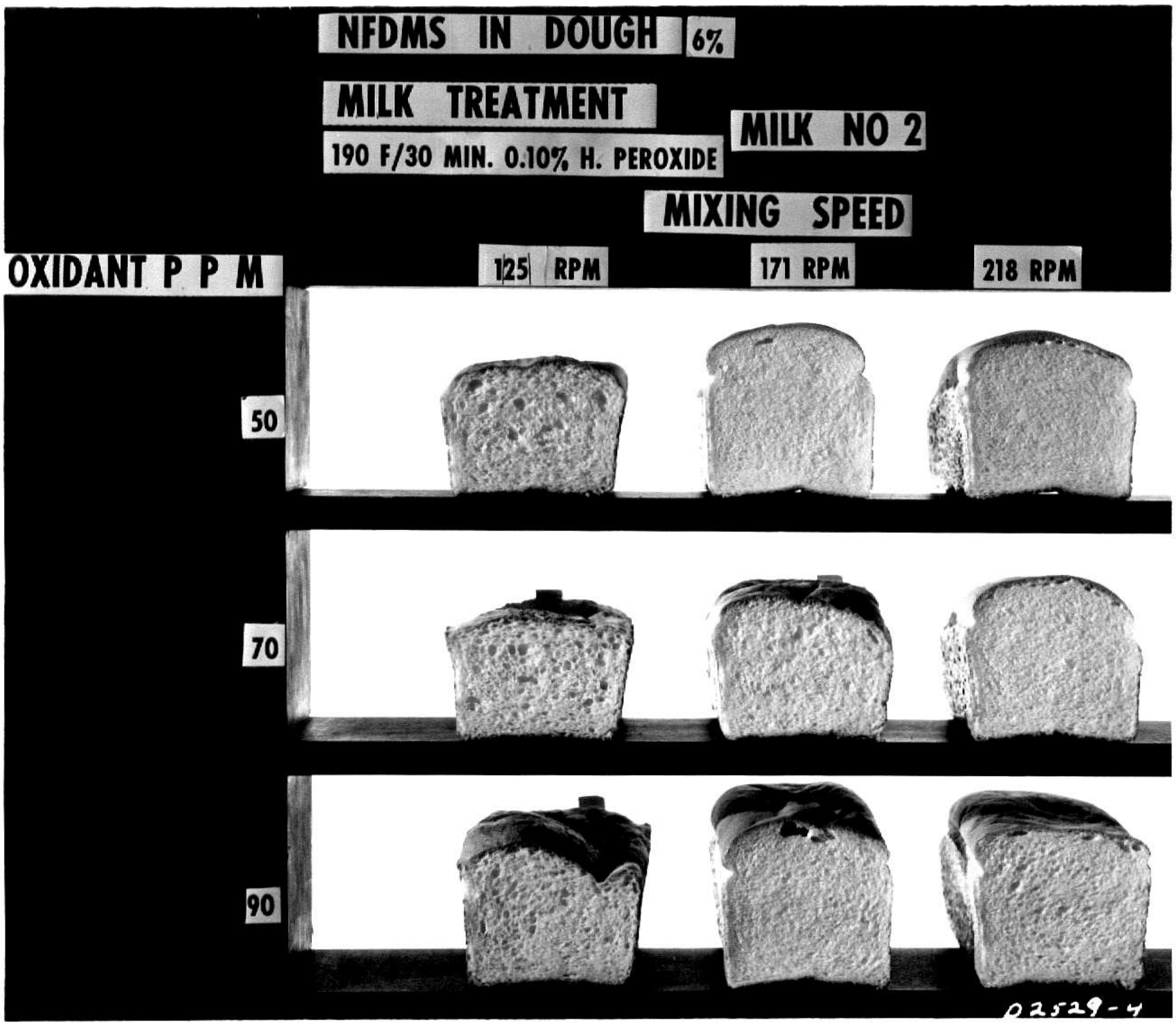

Figure 29. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.10% H_2O_2 and used at 6% level.

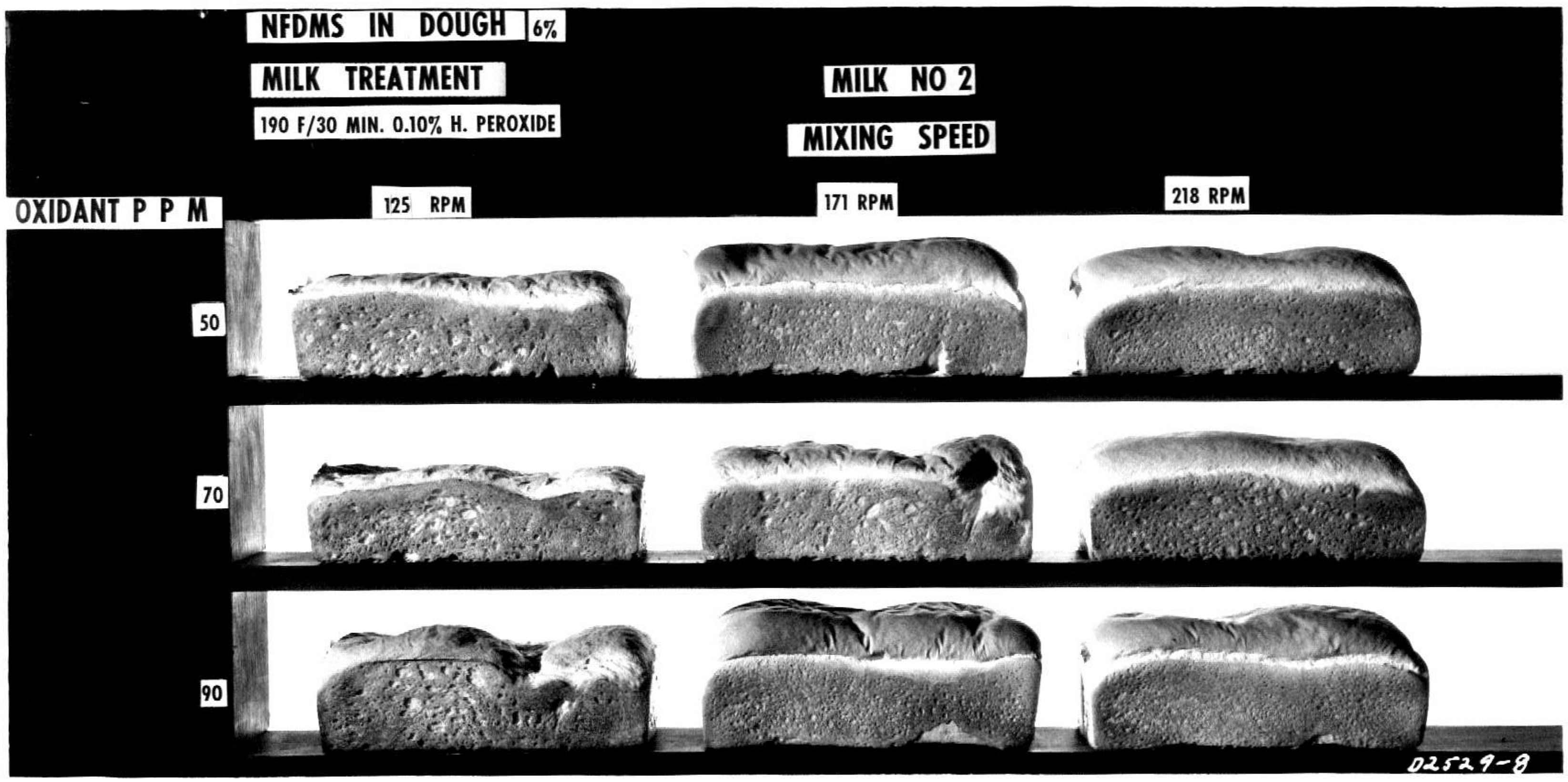

Figure 30. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.10% H_2O_2 and used at 6% level.

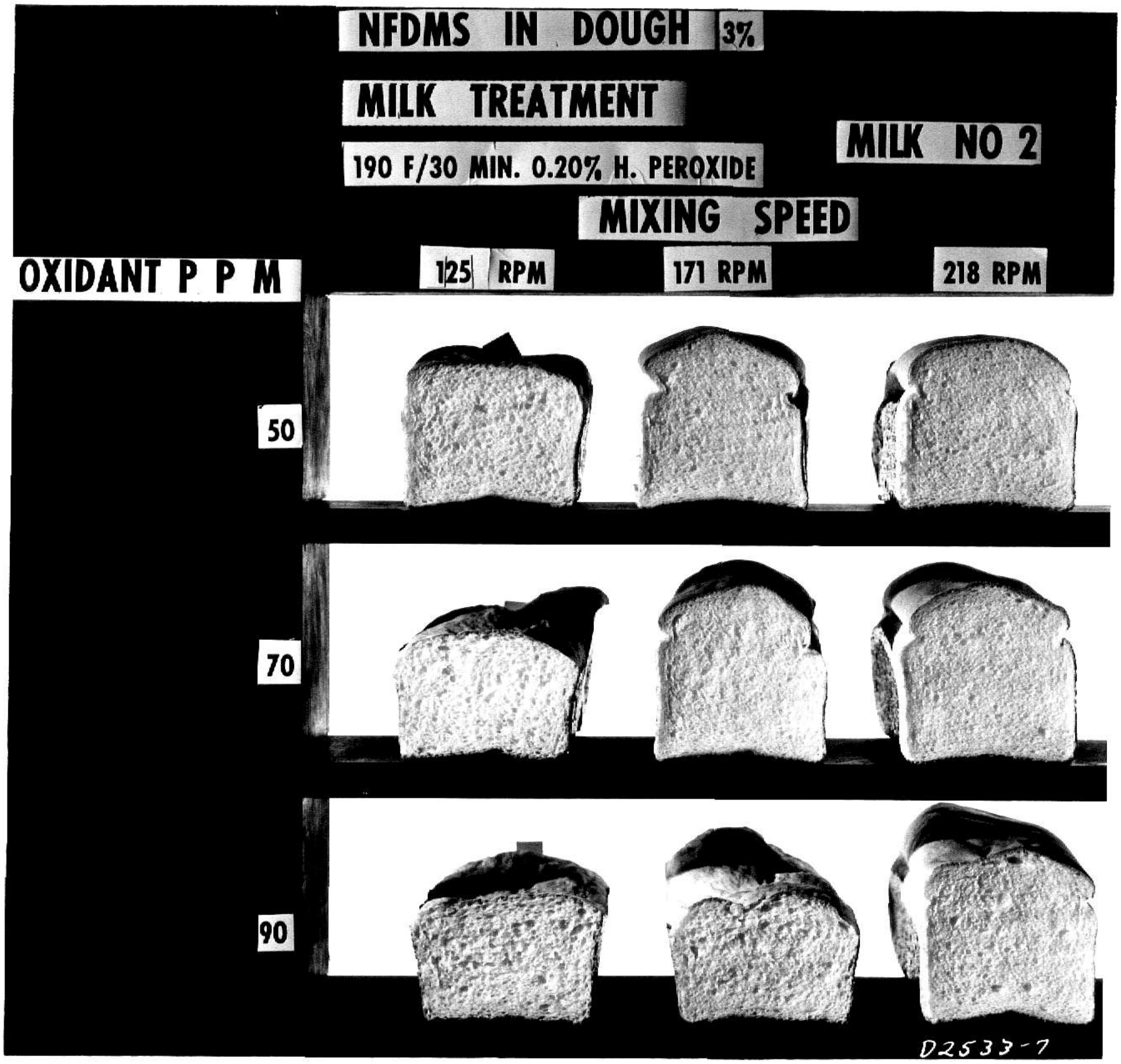

Figure 31. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.20% H_2O_2 and used at 3% level.

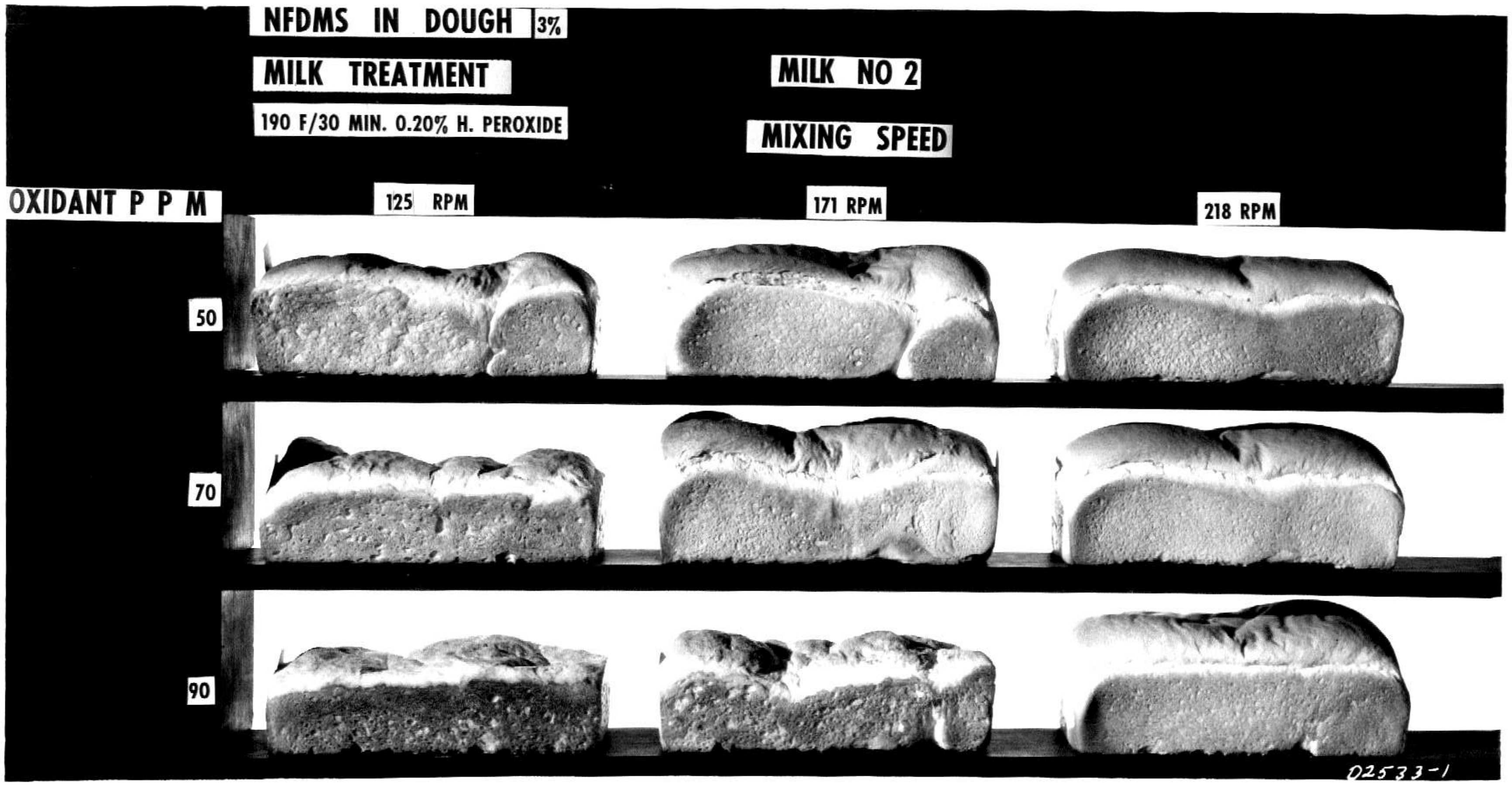

Figure 32. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.20% H_2O_2 and used at 3% level.

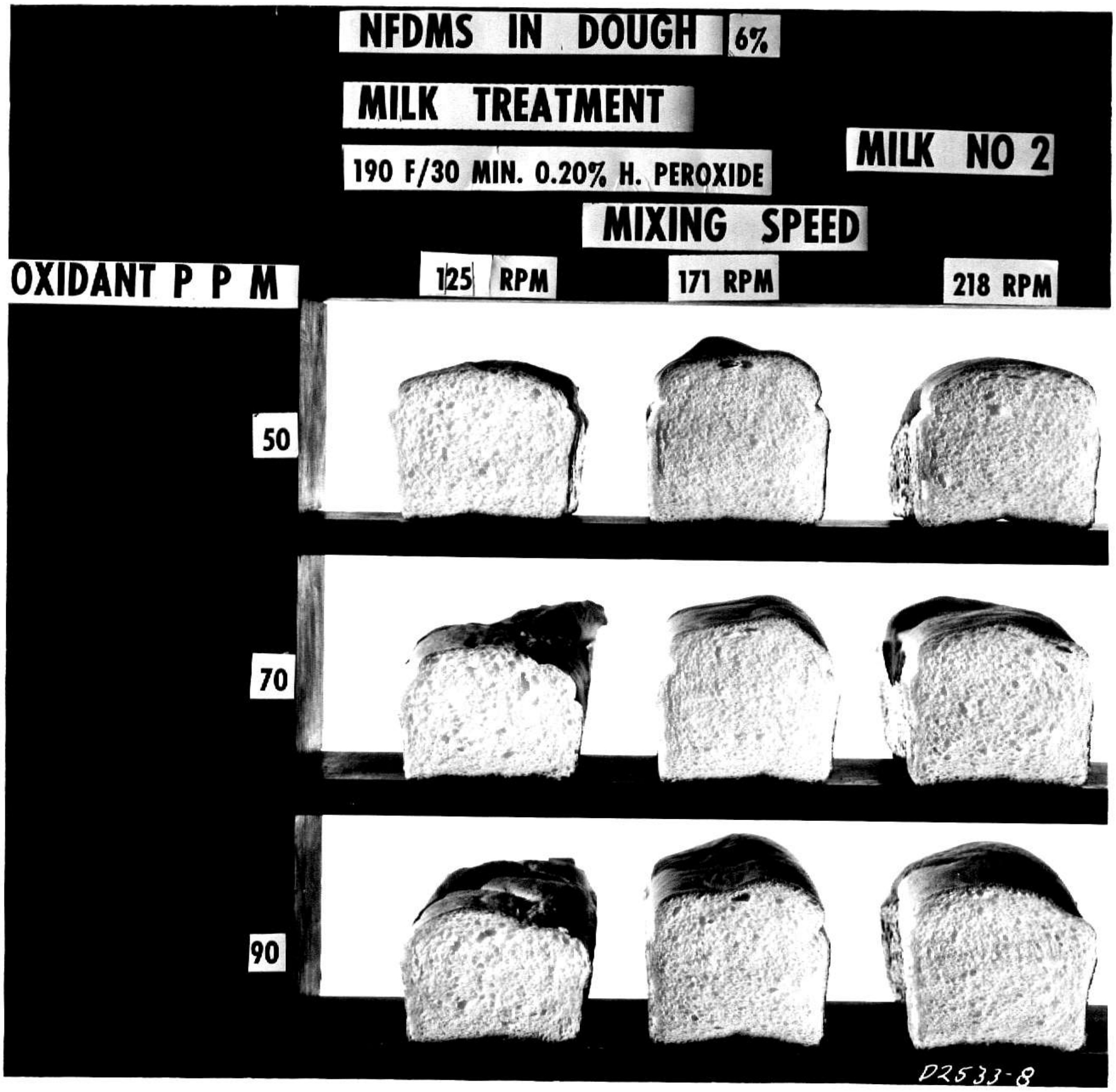

Figure 33. Cross section view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.20% H_2O_2 and used at 6% level.

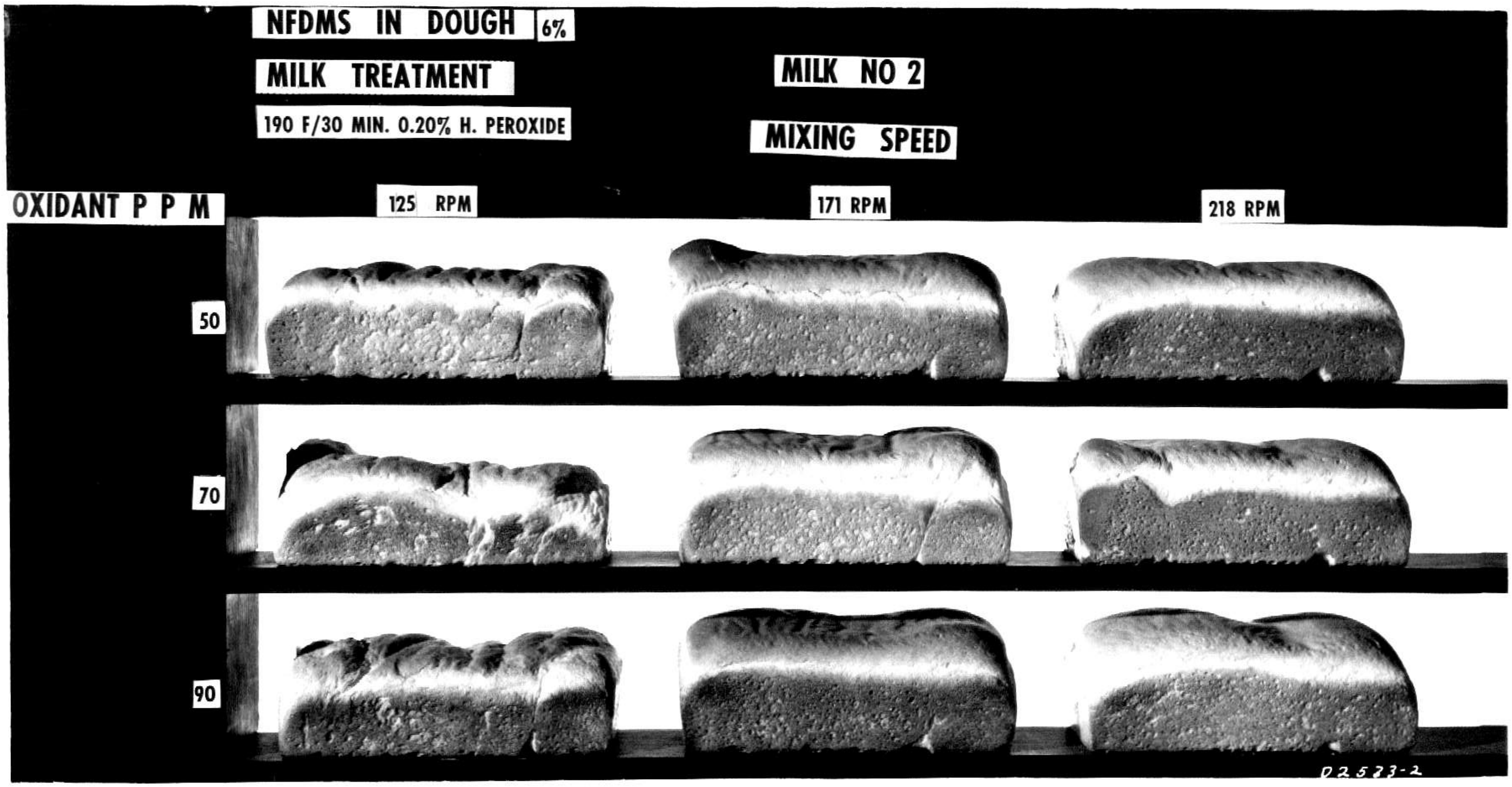

Figure 34. Longitudinal view of bread made by continuous dough mixing process containing NFDM no. 2, treated 190 F/30 min, 0.20% H_2O_2 and used at 6% level.

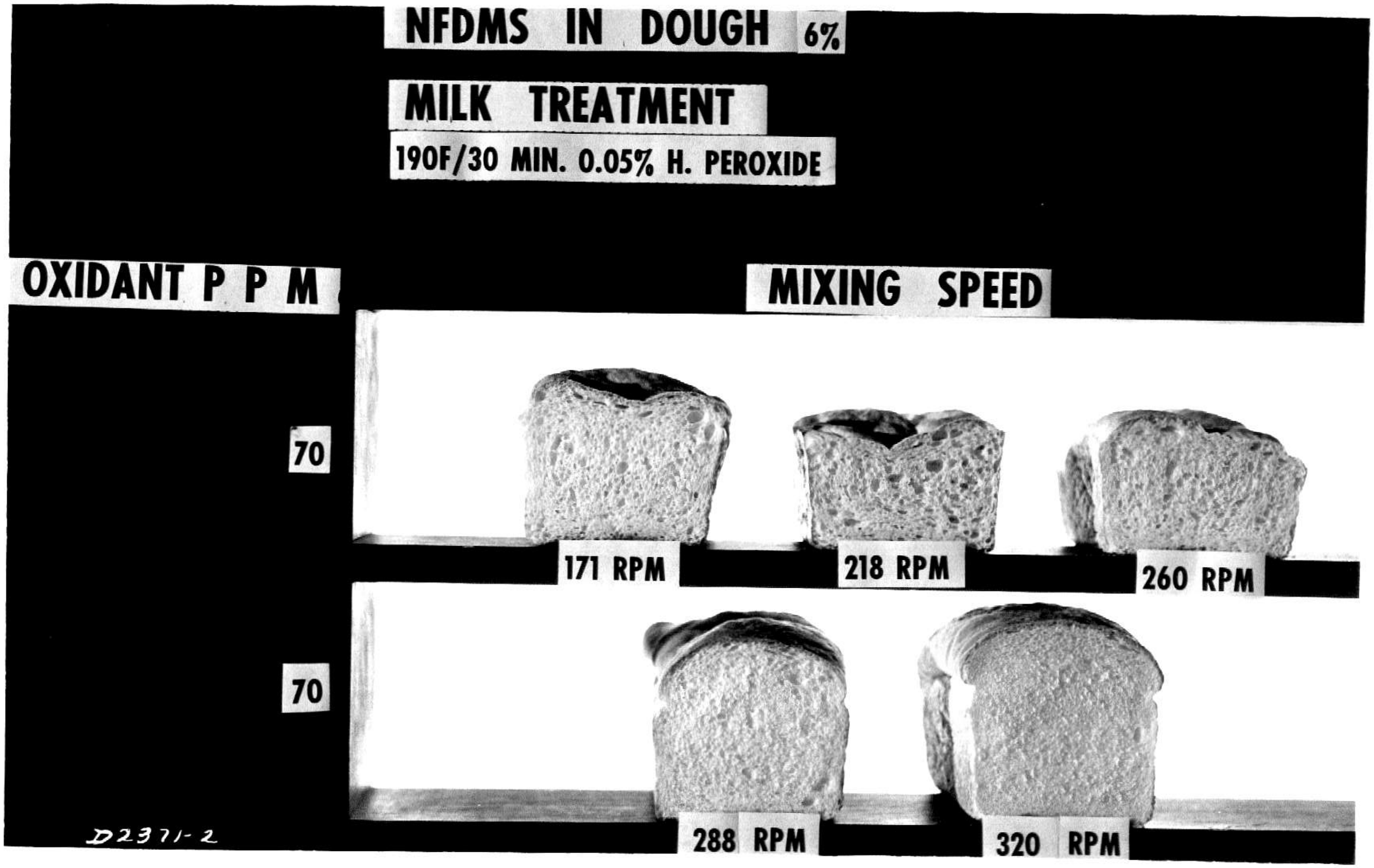

Figure 35. Cross section view of continuous mix bread containing 6% NFDM treated 190 F/30 min, 0.05% H_2O_2 with wide range of variation in mixing speed.

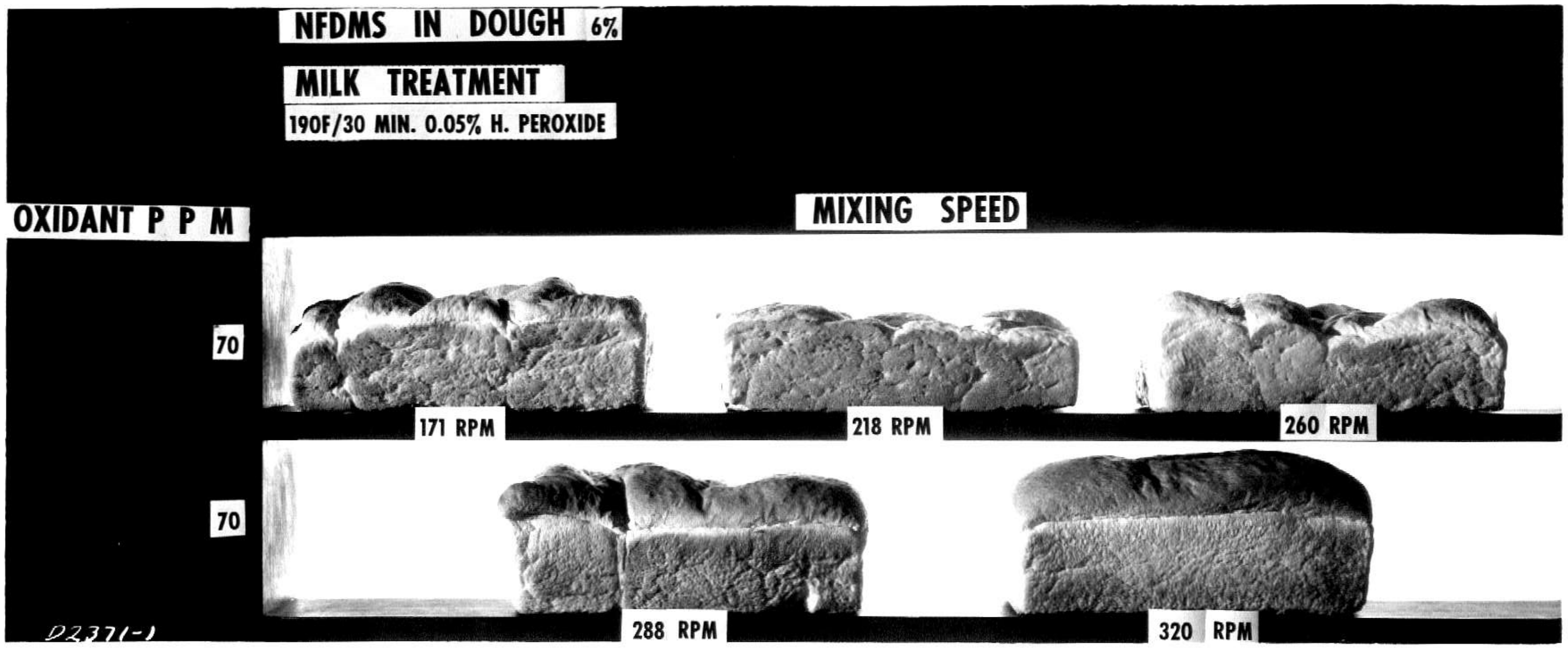

Figure 36. Longitudinal view of continuous mix bread containing 6% NFDM treated 190 F/30 min, 0.05% H_2O_2 with wide range of variation in mixing speed.

USE OF HYDROGEN PEROXIDE TREATED NONFAT DRY MILK IN THE CONTINUOUS DOUGH MIXING PROCESS

by

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The inclusion of commercial nonfat dry milk (NFDM) in the continuous bread dough mixing formula causes many quality problems such as loss of volume, loss of grain structure, dull crumb color and harsh texture. The power required for development of dough containing NFDM also is high. These difficulties are responsible for a decrease in NFDM usage in commercial bread.

It has been known that the sulfhydryl-disulfide interchange influences the dough structure and it is believed that the sulfhydryl groups of milk might affect this interchange. The total fermentation time given to the continuously mixed dough is short as compared to sponge dough. The sulfhydryl groups of milk might not be oxidized when incorporated into continuously mixed dough as they may be in the case of the sponge dough.

The purpose of this investigation was to study the effects of hydrogen peroxide oxidized NFDM on bread quality when made by the continuous dough mixing process. NFDM samples were prepared by subjecting skim milk to varying concentrations of hydrogen peroxide prior to pre-heating and drying. The NFDM samples were examined for sulfhydryl content, undenatured whey protein nitrogen content and for residual hydrogen peroxide. The rheological properties of doughs containing 3 and 6% of the oxidized NFDM samples were studied. Doughs containing 3 and 6% oxidized NFDM samples were mixed on an AMFLO continuous dough mixer at three mixing speeds, using three levels of dough oxidant. The power required to mix each combination of variables was determined. Following baking of the bread, the data were statistically analyzed using loaf volume, grain, texture and total scores as parameters.

The sulfhydryl content of the milk was decreased by heating and was further reduced by treating with hydrogen peroxide. The undenatured whey protein nitrogen content was increased by the hydrogen peroxide treatment.

The residual hydrogen peroxide in the NFDM after forewarming and drying ranged from 38.28 to 91.76% of the total oxidizing power present in the dough formula. The addition of oxidized NFDM did not materially alter the water absorption properties of the dough, however, all doughs containing NFDM showed greater water absorption than doughs not containing NFDM.

Statistical analysis of the parameters studied showed that the best overall breads were produced by the following conditions: (a) for NFDM made from winter milk, 190 F/30 min heat treatment following oxidation by 0.05% hydrogen peroxide, incorporated in the dough in amounts equal to 3% of the flour weight, at a mixing speed of 218 rpm, and using 50, 70 or 90 ppm of dough oxidant; (b) for NFDM made from spring milk, 190 F/30 min heat treatment following oxidation by 0.05% hydrogen peroxide, incorporated in the dough at 171 or 218 rpm, using 90 ppm dough oxidant. The high residual hydrogen peroxide content of some NFDM samples caused the over oxidation of certain doughs.

Optimum power requirements consistent with commercial rheological dough properties were observed when the NFDM was oxidized with 0.05% hydrogen peroxide and incorporated in the dough in amounts equal to 3% of the flour weight, at 125 rpm mixing speed, using 50 ppm dough oxidant.