COMPARISION OF WHITE AND WHOLE WHEAT FLOUR BREWS IN THE LIQUID FERMENT PROCESS OF BREADMAKING

by

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B.S., KANSAS STATE UNIVERSITY, 1982

A MASTER'S THESIS

Submitted in partial fulfillment of the requirements for the degree

MASTER OF SCIENCE

Department of Grain Science and Industry

KANSAS STATE UNIVERSITY Manhattan, Kansas

1987

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INTRODUCTION

The most popular process for the production of white pan bread in the United States is the sponge-dough procedure. The sponge, which normally contains one-half to two-thirds of the flour, plus all of the yeast, yeast food and part of the water is mixed slowly to achieve a coherent mass with minimal gluten development (Hoseney 1986). After fermenting 3.5 to 5 hours, the remaining ingredients are combined with the sponge and mixed to a fully developed dough. Advantages of this procedure over the simpler straight dough process include greater scheduling flexibility, improved grain and texture, and a longer shelf life of the finished product (Pyler 1973).

Although fermented brews have been used for over a century in France and England, the large scale use of liquid ferment systems in the U.S. is only a few decades old (Pyler 1970). The pumpable brew, containing from 0% to 50% of the total flour, has many advantages over sponge-dough or straight dough methods. These include substantial space savings since sponge mixers, troughs, and fermentation rooms are eliminated; labor and processing time savings; and improved sanitation and easier equipment cleanup due to the use of stainless steel tanks, pumps and piping.

Brew ingredients typically include yeast, some sugar, yeast food, and part of the formula water. A buffer or buffering agent, such as nonfat dry milk, is necessary to

maintain an optimal pH range (Lee and Geddes 1959). Disadvantages of the process include less desirable grain and texture of the bread crumbs, a "yeasty" flavor, and day-to-day variations in ferment quality (Holder 1956). The liquid ferment process is used extensively in the production of soft rolls such as hamburger and hotdog buns and, to some extent, in the production of variety breads (Dubois 1984). Currently, almost 50% of plants in the United States are using the liquid ferment process (Kulp 1986).

attitudes (and sometimes consumer Changing misconceptions) have resulted in varied bread consumption patterns in recent years. In consumer surveys, wheat bread (bread in which some or all of the flour is whole wheat flour) rated more favorably in its nutrition than white bread. It is seen as being less filling, containing less starch, and having fewer calories (Wheat Industry Council 1983). Although white pan bread still maintains a majority of wholesale bakery output, per capita consumption has declined in recent years (Gaynor 1986). Variety bread consumption, on the other hand, has continued to exhibit Among variety breads, wheat breads steady growth. represent one of the fastest growing segments of the market. These breads typically contain 70-80% white flour and 20-30% whole wheat flour. The processing of wheat bread is the same as for most white breads with the exception being that whole wheat flour is generally added at the dough stage.

The studies described below compare the effects of

adding whole wheat flour at the dough stage to adding whole wheat flour in the brew stage of the liquid ferment process. Comparisons were based on wheat breads containing 30% whole wheat flour added at two different stages of production. Wheat breads containing 30% whole wheat flour added at two different stages were also made by the sponge-dough method. These breads represented the dominant industry procedure, allowing comparision both within and between the liquid ferment and sponge-dough processes.

The objectives of this study were:

- 1. To compare pH and total titratable acidity (TTA) values between fermenting whole wheat and white flour brews and between liquid ferment and sponge-dough breads containing 30% whole wheat flour.
- To compare micro-structural differences in doughs prepared from white and whole wheat flour brews.
- 3. To investigate possible differences in optimum proof time, specific volume, crumb color, and moisture content of breads containing 30% whole wheat flour added at various stages in the liquid ferment and sponge-dough processes.
- 4. To compare the firmness characteristics of liquid ferment and sponge-dough breads made with white and whole wheat flour brews and sponges.
- 5. To compare the residual sugar concentrations in crumb of

wheat breads made by liquid ferment and sponge-dough processes.

6. To investigate sensory evaluation characteristics of wheat breads made by the addition of whole wheat flour at various stages in the liquid ferment and sponge dough processes.

TITTERATURE REVIEW

FERMENTABLE SUGARS

Dough fermentation is an anaerobic process during which yeast ferments sugars to carbon dioxide and ethanol. Most of these fermentable sugars are produced by the enzymatic hydrolysis of formula sweetners and flour components (Tang et al. 1972) and include the monosaccarides glucose and fructose, and the disaccharides sucrose and maltose. Lactose, a dissaccharide present in formulas containing dairy ingredients, is not fermented by bakers yeast. Some polysaccharides such as starches and dextrins may also serve as sources of fermentable sugars if they are hydrolyzed by amylases (Ponte and Reed 1982).

Few studies have been undertaken to actually quantify the sugar levels in fermenting doughs. Early work by Rice (1938) indicated a higher content of fructose than glucose in bread made from a straight dough formula containing sucrose, suggesting the preferential metabolism of glucose. Several subsequent studies have confirmed these findings (Bohn 1954a, Koch et al. 1954, Tang et al. 1972). Similar findings have been reported using sponge-dough (Ponte et al. 1969) and liquid ferment processes (Piekarz 1963, Martinez-Anaya et al. 1984). Maltose levels in limited sugar systems increase until fructose and glucose levels reach critically low levels. At this point, the yeast is forced to ferment maltose which is associated with a temporary

decrease in gas production. When sucrose is added, the maltose levels increase throughout fermentation (Koch et al. 1954). Maltose fermentation appears to be more pH sensitive than does glucose fermentation (Cooper et al. 1968).

Bread-making processes differ markedly in their preferential use of fermentable carbohydrates. For example, bread made by the sponge-dough process contains less maltose than bread from straight doughs because the yeast utilizes the maltose formed in the sponge by enzymatic activity (Bohn 1954b). Koch (1954) traced fructose, glucose, and maltose in a straight dough process containing 3% yeast (based on flour wt.) and varying amounts of added sucrose. At the 5% sucrose level, glucose was fermented at a faster rate than was fructose. Maltose levels actually increased with fermentation time. Residual amounts of glucose, fructose, and maltose were 0.14%, 0.79%, and 1.5%, respectively.

BREAD STALING

The deleterious changes that take place in bread crumb and crust over time other than those resulting from spoilage organisms are referred to as staling (Bechtel and Meisner 1954). Staling reduces the value of an estimated 110 million pounds of bread annually (Maga 1975). Staling causes a loss of flavor, a leathery crust, a firmer crumb, and a decrease in soluble starch (Hoseney 1986). The transformations that occur in the crust apparently are simpler and less controversial than those of the crumb.

The primary reaction taking place in crust staling is the migration of water from the crumb to the hygroscopic crust (Pyler 1970). The end result is a crust with an undesireable taste and texture. Cathcart (1940) outlined various factors influencing crust staling rates including: amounts of yeast, salt and water in the formula; type and protein content of flour; type and amount of shortening added; fermentation temperature; and method and temperature of baking.

The bread crumb, on the other hand, undergoes a more complicated set of changes. Boussingault (1852) reported that crumb staling was not the result of moisture loss. Since the turn of the century, the focus of research on crumb staling has been on the recrystalization of starch components (Lindet 1902). Studies have shown that retrogradation of the amylopectin fraction of starch is primarily responsible for crumb firming during bread storage (Kulp 1979). However, the link between retrogradation and firming appears to be inconclusive. In fact, recent work (Dragsdorf et al. 1980) using x-ray diffraction techniques indicated that firming and starch recrystalization were not synonymous.

Over the years, a number of chemical, physical, and sensory methods have been developed to measure the rate and degree of staling. These include measurements of crumb-crust moisture level (Bradley and Thompson 1950), sedimentation rate (Banasik and Harris 1953), iodine

absorption (Pelshenke and Hampel 1962), and capacitanceconductance (Kay and Willhoft 1972). Unfortunately, most involve indirect measurements of staling and are difficult to correlate with actual consumer concepts of "stale" or consumer acceptance or rejection of a particular product.

Tests involving compressibility have been among the most commonly used by the baking industry to ascertain staleness in bread crumb. The Baker Compressimeter, described in an approved AACC Method (AACC Method 74-10), is simply a modernized version of an instrument developed almost half a century ago by Platt and Powers (1940). Many newer designs, including the GRL compression tester (Kilborn et al 1983) and the Voland-Stevens LFRA Texture Analyzer (Voland Corp., Hawthorne, NY), have expanded possible testing conditions. However, these instruments all involve application of uniaxial compression to the bread crumb surface. Newer rheological methods involving dynamic stress-strain measurements may eventually provide a better means of understanding changes in bread texture as a function of bread staling (Ponte and Faubion 1985).

COMPARISON OF WHITE AND WHEAT BREADS

Although wheat breads obviously contain a greater amount of fibrous materials such as bran, shorts and germ then do white breads, the differences in chemical composition of these breads may not be so apparent. The enrichment vitamins (niacin, thiamine, riboflavin) and trace

mineral (iron) added to white bread are typically as high or even higher in concentration than in wheat bread (Miller 1981).

Other nutrients including vitamins B_6 , folacin, and pantothenic acid as well as most trace minerals are often considerably higher in breads containing whole wheat flour (Miller 1981). This may be misleading, however, since the high phytate in bran may interfere with vitamin and mineral bioavalibility (Davies 1979, Daniels et al. 1981, Tangkongchitr 1981).

The most publicized virtue of whole grain breads is their high dietary fiber content. A large number of colonic diseases in western man have been associated with the decreased amount of fiber in the diet including: cancer of the colon, diverticular disease, hemorrhoids, and constipation (Painter et. al. 1972, Piepmeyer 1974). Therefore, a diet containing adequate quantities of dietary fiber, such as one including whole grain products, may substantially reduce the incidence of these diseases.

The baked goods classed as wheat breads consists of a number of products, including whole wheat breads. The Food and Drug Administration (FDA) standards of identity require packages labeled as whole wheat bread to contain only whole wheat flour or bromated whole wheat flour. No white flour may be used (Code of Federal Regulations 1986). No standards of identity have been established for wheat breads containing any lesser proportions of whole wheat

flour. Other wheat-derived ingredients often added to wheat breads in varying amounts to provide texture and flavor include cracked wheat, rolled wheat, crushed wheat and bran.

Whole wheat breads are typically darker in color and denser than wheat breads containing some white flour. The granulation of whole wheat flour in the formula is finer than that used in wheat breads containing some white flour. This is done to give whole wheat breads maximum loaf volume potential. Wheat breads made with a strong white bread flour have good volume and crumb characteristics similar to those of white pan bread (Ponte 1981). Bakers often add small amounts of vital wheat gluten to "carry" the whole wheat flour in these breads.

MATERIALS AND METHODS

FLOUR

The white flour used in this study was milled from a hard red winter wheat supplied by Ross Milling Company, Wichita, KS. The Whole wheat flour was supplied by General Mills Inc., Minneapolis, MN. The 50 lb. bags were labeled Stone Ground Whole Wheat, Hi-Protein (Fine Ground). Malted barley flour and oxidants were added at the mill for both flours. Moisture, Kjeldahl protein (N X 5.7), and ash values were determined according to AACC standard methods 44-15A (AACC 1967), 46-10 (AACC 1976), and 08-01 (AACC 1961), respectively. Results are shown in Table 1.

Granulation of the whole wheat flour was determined by a Ro-tap Sifter (W. S. Tyler Co., Cleveland, OH). A 100 gram sample was sifted for 2 minutes on a stack of Tyler Test Sieves of various screen sizes. The percentage of the stock over and thru each of the sieves was measured. Two replications were performed and an average determined (Table 2).

PHYSICAL DOUGH TESTS

<u>Farinograph</u>: The Brabender Farinograph equipped with the small (50 g.) bowl was used according to AACC method 54-21 (AACC 1961). Water absorption, arrival time, peak time, stability, departure time, Valorimeter, and mixing tolerance index (MTI) readings were recorded. Results are summarized

Table 1. FLOUR QUALITY DATA

Flour	Protein ^{a,b} (%)	Moisture (%)	Ash ^a (%)
White	11.75	13.9	0.48
Whole Wheat	14.70	11.2	1.70

a_{14%} moisture basis

b_{N x 5.7}

Table 2. WHOLE WHEAT FLOUR GRANULATION DATA

		OPE	NING	,
	WIREa	MICRONS	INCHES	PERCENT
OVER OVER OVER OVER OVER THRU	48 W 60 W 65 W 80 W 100 W	297 250 212 178 150	0.0117 0.0098 0.0083 0.0070 0.0059	18 % 9 % 15 % 40 % 8 %

^aTyler Test Sieve

bBased on two replications

in Table 3. Farinograms are shown in Figure 1.

Amylograph and Falling Number: Amylograms and Falling Number data were obtained according to AACC methods 22-10 (AACC 1960) and 56-81B (AACC 1972), respectively (Table 4).

SPONGE-DOUGH AND LIQUID FERMENT PROCEDURES

Wheat breads were made by the addition of whole wheat flour (30% of the total formula flour) added at various stages in the liquid ferment and sponge-dough processes. In the liquid ferment process, whole wheat flour brews contained all of the whole wheat flour (30% of the total flour). Breads made from white flour brews had the whole wheat flour plus the remaining white flour added at the dough stage of production (Figure 2). The sponge-dough breads made from whole wheat flour sponges had all of the whole wheat and part of the white flour added at the sponge stage. Breads made from white flour sponges had only whole wheat flour added at the dough stage (figure 3).

The sponge-dough and liquid ferment formulations are given in Tables 5 and 6, respectively. These formulations are similar to those typically used in the U.S. wholesale baking industry with some modifications to facilitate processing and comparision of breads produced by the two methods. These modifications include elimination of dough strengtheners, surfactants, and preservatives.

Flow diagrams of the liquid ferment and sponge-dough

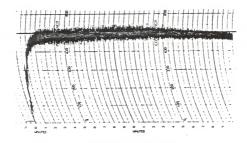
FARINOGRAPH DATA

Flour	Water Abs.	Arrival Time (min)	Time (min)	(min)	Time (I	(BU)	(BU)
White	58.8	2.0	18	19	21	1.0	8.8
Whole Wheat	73.0	3.5	6.5	10.5	14	35	99

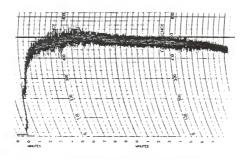
15

Average of 2 replications.

Figure 1. Farinograms of Experimental Flours



WHITE FLOUR



WHOLE WHEAT FLOUR

17

Table 4. FALLING NUMBER AND AMYLOGRAPH DATA

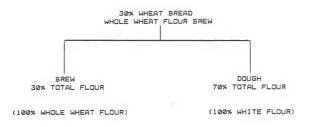
	Falling Number Value	Amylograph Value (BU)
White Flour	292	520
Whole Wheat Flour	319	550

^aAn average of three replications.

bAn average of two replications.

Figure 2. Stages of Whole Wheat Flour Addition in Wheat Breads Made by the Liquid Ferment Process

LIQUID FERMENT PROCESS



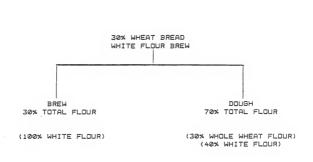
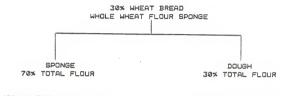


Figure 3. Stages of Whole Wheat Flour Addition in Wheat Breads Made by the Sponge-Dough Process

SPONGE-DOUGH PROCESS



(30% WHOLE WHEAT FLOUR) (40% WHITE FLOUR)

(100% WHITE FLOUR)

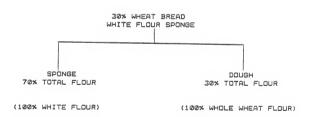


Table 5. SPONGE-DOUGH FORMULATION

Ingredients ^a	Sponge	Dough	Total
Flour	70	30	100
	43	23	66
Water Yeast	1.0		1.0
Veset Food	0.25		0.25
Potassium Bromate ^d Ascorbic Acid	Optimum		Optimum
Ascorbic Acide	Optimum		Optimum
Salt		2.0	2.0
Sugar		6.0	6.0
Shorteningf		3.0	3.0
Non Fat Dry Milk		3.0	3.0

aIngredients, % based on total flour as 100%.

bInstant Active Dry Yeast (Fermipan; Gist-brocades; Delft, Holland)

CArkady, Archer Daniels Midland, Co., Decatur, IL.

d,eAdded in parts per million of flour.

fNon-emulsified commercial shortening.

Table 6. LIQUID FERMENT FORMULATION

Ingredient ^a	Brew 1st Stage	Brew 2nd Stage	Brew 3rd Stage	Dough Stage	Total
Flour	30.0			70.0	100.0
Water	43.0	5.0		8.0	66.0
Yeast ^D	1.2				1.2
Yeast Food ^C	0.25				0.25
Oxidants	Variable				Variable
Salt		2.0			2.0
Sugar	1.0			5.0	6.0
Shorteninge				3.0	3.0
NFDM				3.0	3.0
Ice ^I			Variable		Variable

^aIngredients, % based on total flour as 100%.

^bInstant Active Dry Yeast (Fermipan; Gist-brocades; Delft, Holland).

CArkady, Archer Daniels Midland, Co., Decatur, IL.

 $[\]ensuremath{^{\text{d}}}\xspace\textsc{Potassium}$ bromate and ascorbic acid added in parts per million of flour

eNon-emulsified commercial shortening.

fAdded to cool brew to 45° F.

processes are shown in figures 4 and 5. The steps involved from the moulding through the oven stages are virtually the same for both methods.

Sponge-Dough Procedure

A Hobart Mixer, Model A-200 (Hobart Manufacturing Co., Troy, Ohio) equipped with a Mcduffee bowl and mixing fork was used to mix both sponges and doughs. The following is an outline of the procedure:

- The sponge and dough ingredients are weighed separately and put into sealed cans to prevent moisture loss.
- 2. Sponge ingredients are added (except water) to a mixing bowl and mixed at speed 1 (120 RPM) for 30 seconds to homogenize the dry ingredients. The water is then added and the sponge mixed for 2 minutes. Sponge temperature should be 76 \pm 10 F.
- 3. Place the sponge in a greased fermentation bowl and set the bowl into a fermentation cabinet for 4 hours. Cabinet temperatures are maintained at 85° F and 85% relative humidity (R.H.).
- 4. After fermentation, dough ingredients are added to a mixing bowl and mixed for 30 sec. at speed 1.
- 5. Fermented sponge is added and mixed 1 1/4 min. on speed
- 1, then on speed 2 until the dough is mixed to optimum.

Figure 4. Sponge-Dough Process Flow Diagram

SPONGE-DOUGH FLOW DIAGRAM

MIX SPONGE

4

SPONGE FERMENTATION
(4 HR. 85° F. 85% R.H.)

MIX DOUGH

DOUGH FERMENTATION
(30 MIN. 85° F 85% R.H.)

٠.

DIVIDE & ROUND

+

FLOOR TIME (20 MIN.)

•

MOULD & PAN

4

PROOF TO HEIGHT (1.5 CM. ABOVE PAN) (110° F 90% R.H.)

+

BAKE (425° F. 20 MIN.) Figure 5. Liquid Ferment Process Flow Diagram

LIQUID FERMENT FLOW DIAGRAM

MIX BREW

+

BREW FERMENTATION
(1 1/2 HR. 30° C. WATER BATH)

+

2ND STAGE BREW

(ADD SALT = 1/2 HR FERMENTATION)

+

3RD STAGE BREW (ADD ICE - COOL BREW TO APPROX. 45° F.)

÷

MIX DOUGH

4

DIVIDE & ROUND

+

FLOOR TIME

+

MOULD & PAN

+

PROOF TO HEIGHT

+

BAKE

Dough temperature after mixing should be 82° F.

- 6. Return dough to cabinet for an additional 30 minute fermentation.
- 7. Scale dough into two pieces (539 g. each).
- 8. Round the dough pieces and rest 20 min. covered to minimize outer skin moisture loss.
- 9. Mould dough pieces using a Moline Model 100 Moulder (Moline Co., Duluth, MN) adjusted as follows: top roller,
- 1.5; bottom roller, 1.0; pressure plate (front), 1 1/4 in.; pressure plate (back), 1.0 in.
- 10. Pan and proof to height (1.5 cm. above loaf pan) at 110° F. and 90% R.H.
- 11. Bake at 425° F. for 25 minutes in a Reed Reel Oven (Bakers Engineering and Equipment Co., Kansas City, KS).
- Measure volume (rapeseed displacement) and weigh immediately after the oven.
- 13. Cool bread on rack for one hour before bagging.

Liquid Ferment Procedure

- weigh-up first, second, and dough stage ingredients (except water, yeast, and shortening) separately.
- 2. Pour first stage water at 85° F into a container and

dissolve the yeast.

- 3. Add remaining first stage ingredients and mix until smooth. Allow to ferment for 1 1/2 hours in a 30° C water bath.
- 4. After 1 1/2 hours, add second stage salt to brew. Allow to ferment for 1/2 in water bath.
- 5. At the two hour brew fermentation mark, add ice as indicated in the formulation. The ice should cool the brew to approximately 45° F.
- Pour brew into mixing bowl containing dough ingredients. Mix at speed 1 for 30 seconds.
- 7. Mix dough to optimum using speed 2. Dough temperature out of mixing bowl should be 81 +/- 1 $^{\circ}$ F.
- Scale dough into two pieces (539 g. per piece). Round the dough pieces and rest in drawer for 20 min.
- 9. 12. Same as sponge dough process.

BREW pH and TTA

<u>pH Measurement:</u> The hydrogen ion activity (pH) of white and whole wheat brews were measured at twenty minute intervals according to AACC method 02-52 (AACC 1961). Yeast activity was terminated at each interval with 1 ml. of formaldehyde solution (37% w/w). Certified buffer solutions of pH 4.00, pH 7.00, and pH 10.00 from Fisher Scientific Co. (Fairlawn, NJ) were used as standards. Three replications at each time period were performed.

TTA Measurement: The total titratable acidity (TTA) was determined by titrating 20 grams of first stage liquid brew with 0.1 normal (N) sodium hydroxide (NaOH) to pH 6.6. This was done immediately following pH determination. Three replications at each 20 min. time interval were measured.

BREAD PH AND TTA

pH Measurement: The hydrogen ion activity (pH) of whole wheat and white flour brew breads produced by the liquid ferment and sponge dough methods were measured according to AACC method 02-52 (AACC 1961). Certified buffer solutions of pH 4.00, pH 7.00, and pH 10.00 from Fisher Scientific Co. (Fairlawn, NJ) were used as standards. Three replicants were performed.

TTA Measurement: The total titratable acidity (TTA) was determined by titrating 20 grams of bread crumb with 0.1 Normal (N) sodium hydroxide (NaOH) to pH 6.6 after pH measurements were recorded. Three replications were measured.

SCANNING ELECTRON MICROSCOPE ANALYSIS

Scanning electron micrographs were taken of liquid

ferment doughs at the whole wheat flour/brew stage and the whole wheat flour/dough stage. The SEM used was a ETEC Autoscan Model U-1 (Electron Beam Technology, Inc., Hayward, CA). Samples were frozen in freon and subsequently freezedried at -60° C in an Edwards Tissue Dryer (Edwards High Vacuum, Inc., Grand Island, NY) to minimize ice crystal formation and resulting artifacts. Samples were then mounted for subsequent veiwing.

BREAD MOISTURE

Moisture was determined in bread samples according to AACC method 44-15A (American Association of Cereal Chemists 1981) after preparation of bread sample by AACC method 62-05.

BREAD CRUMB FIRMING MEASUREMENTS

Firmness measurements were taken at Day 0 (2 hours after baking) and at day 5 (120 hours later) using the Voland-Stevens LFRA Analyzer (Voland Corporation, Hawthorne, NY). The instrument was adjusted to the following settings: plunger speed, 2.0 mm/sec; plunger penetration, 4.0 mm; and cycle selection, normal. A plunger with a diameter of 3.6 cm was used, resulting in a plunger surface area of 10.18 cm². The force required to deform the bread crumb is a measurement of load and is indicated in gram units.

The bread was sliced with an electric knife in a miter-

box type apparatus, yielding seven 1-inch slices plus the 2 heels. The heels and center slice were discarded. The measurements were made on the center of each slice with the force being applied from the ends of the loaf towards the center.

A bread slice was placed on the texture analyzer platform and the LED readout was adjusted to 0 grams force. The start button was then pressed, starting the plunger in motion. As the the plunger made contact with the bread surface, the force required to deform the slice was recorded continuously. At the maximum penetration depth (4 mm) the load also reached a maximum and the plunger returned to its ready position.

Two loaves from each dough were measured and the results were averaged to represent the crumb firmness of the overall loaf. Each bread was measured only once. New loaves were used for subsequent trials.

BREAD CRUMB COLOR MEASUREMENT

To analyze possible bread crumb color differences between experimental conditions, an Agtron reflectance spectrophotometer model M-500-A (Magnuson Engineer Inc., San Jose CA) was used. Although no AACC Approved Method exists for bread crumb, AACC Method 14-30 (American Association of Cereal Chemists 1974) for flour was followed with some modification. The standard green mode (wavelength 546 nm) was used.

The instrument was set to zero reflectance using a number 00 disk and to 100% reflectance using a number 85 disk. This was checked and adjusted as necessary after each sample. The center portion of the bread slice was cut to the same inside diameter as the sample cup. A number 85 disk was placed on top of the sample to reduce errors resulting from diffused light. Both sides of the sample were measured. Two slices from each loaf were selected at random.

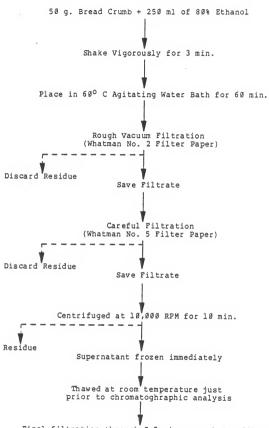
BREAD CRUMB RESIDUAL SUGAR EXTRACTION

High performance liquid chromatography techniques were used to determine residual sugar percentages in breads made from both liquid ferment and sponge-dough procedures containing 30% whole wheat added at either brew/sponge or dough stages. The extraction procedure was similar to that used by Kai (1985) and is shown in Figure 6.

HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

Equipment: The high performance liquid chromatograph used in the experiment was a Varian (Palo Alto, CA) Model 5000 equipped with a 20 ul injection loop. A Varian Aerograph Refractive Index Detector was also used (cell volume, 6 ul; minimum detectability, +/- 1 X 10⁻⁷ refractive index units for aqueous solvents). A Hewlett Packard Model 3392A Integrator plotted the chromatograms, integrated the

Figure 6. Bread Crumb Residual Sugar Extraction Procedure



Final filtration through 0.5 micron membrane filter

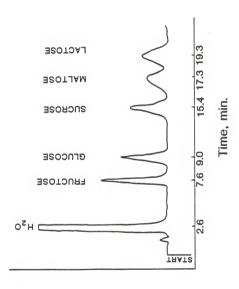
peaks, and printed results of peak area and retention times. The analytical and guard columns were obtained from Alltech Associates (Deerfield, IL). The 600CH Carbohydrate Analysis (amino phase) column is 300 mm X 4.1 mm with particle size of 10 u. The guard column was an Alltech 601CH with dimensions of 50 mm X 4.1 mm.

Experimental Conditions: A considerable amount of preexperimental work was required to determine the most
suitable conditions for HFLC analysis. The mobile phase was
an 80:20 mixture of HPLC grade acetonitrile and water,
respectively. This ratio gave adequate resolution while
minimizing retention times of dissacharides (Figure 7). An
eluant flow rate of 1.5 ml/min was used. Column temperature
was maintained at 25 degrees Centigrade. Integrator chart
speed was 0.5 cm/min. A summary of HPLC equipment and
conditions is given in Figure 8.

Standard Curves: Standard solutions of fructose, glucose, sucrose, maltose, and lactose were prepared from stock standards supplied by Fisher Scientific Co. (Fair Lawn, NJ). Solutions ranging in concentration from 0.1% to 2.0% were prepared using de-ionized water immediately prior to injection. Each standard solution was injected in triplicate and an average peak area was obtained. No impurities were detected in any of the standards during chromatographic analysis.

Standard curves relating peak area to saccharide

Figure 7. Chromatogram of Standard Sugar Solutions



Resbouse

Figure 8. Summary of HPLC Experimental Conditions

HIGH PERFORMANCE LIQUID CHROMATOGRAPHY SUMMARY

HPLC : Varian Model 5000

GUARD COLUMN : Alltech Model 601CH

COLUMN : Alltech Carbohydrate Analysis Column

Model 600CH

PACKING : Alltech Amino Phase (10 μ)

ELUANT : 80:20 Mixture CH₃CN/H₂O

FLOW RATE : 1.5 ml/min

INJECTION : 20 µl

TEMPERATURE : 25°C

DETECTOR : Varian Aerograph Refractive Index Detector

INTEGRATOR : Hewlett Packard Model 3392A

concentration were obtained using linear regression analysis (Ott 1984). The curves and coefficients of correlation are shown in Figures 9 - 13.

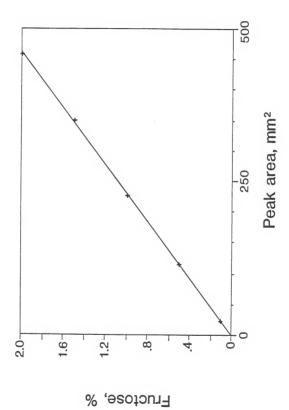
SENSORY EVALUATION

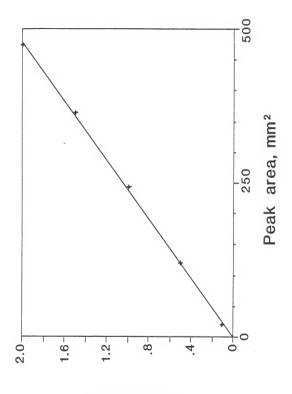
Sensory evaluations of bread were made by a taste panel consisting of 25 untrained participants using the triangle test (Larmond 1977). In this test, the panelist received three coded samples (two of which were identical) and was asked to identify the odd sample. Panelists were also asked to indicate degree of acceptability and difference between odd and duplicate samples, however only responses of correct panelists were tabulated. A sample questionaire is shown in Figure 14.

STATISTICAL METHOD OF ANALYSIS

Analysis of variance (ANOVA) using the least significant difference (LSD) multiple-comparision procedure (Snedecor and Cochran 1980) was followed in determining differences among means in the experiment. Statistical assistance was provided by Dr. Dallas Johnson, Department of Statistics, Kansas State University.

Figure 9. Standard Curve for Fructose Determination Coefficient of Correlation (r) = 0.9999





Glucose, %

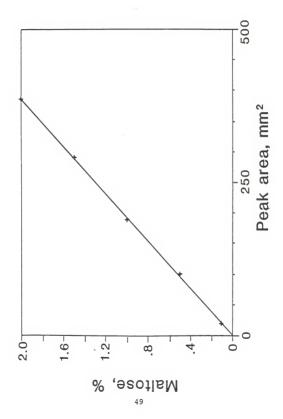
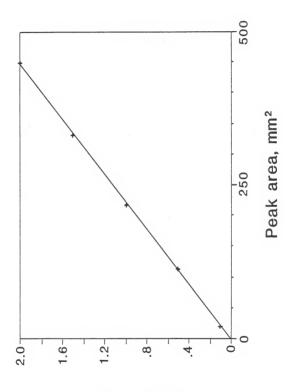


Figure 12. Standard Curve for Sucrose Determination

Coefficient of Correlation (r) = 0.9999



Sucrose, %

Figure 13. Standard Curve for Lactose Determination

Coefficient of Correlation (r) = 0.9998

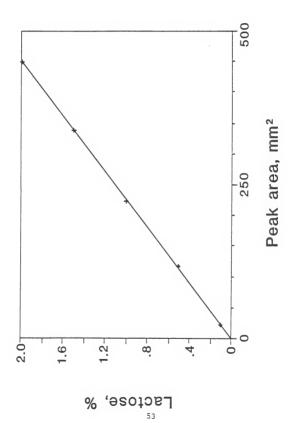


Figure 14. Sensory Evaluation Sample Questionaire

QUESTIONNAIRE FOR TRIANGLE TEST

MAK	EDATE_				
PRO	DUCT				
Two	of these three samples are identical, the third is different.				
1.	Taste the samples in the order indicated and identify the odd sample.				
	Code Check odd sample				
2.	 Indicate the degree of difference between the duplicate samples and the odd sample. 				
	Slight				
	Moderate				
	Much				
	Extreme				
3.	Acceptability:				
Odd sample more acceptable					
Duplicates more acceptable					
4.	Comments:				

RESULTS AND DISCUSSION

BREW pH and TTA

Brew pH measurements of white flour and whole wheat brews taken at 20 minute intervals are shown in Figure 15. The white flour brew pH reading at time zero was approximately 5.1. It continued to drop steadily, reaching less than 4.6 by 2 hours of fermentation. The whole wheat flour brew had an initial pH of greater than 5.8 which dropped slowly to slightly above 5.5 over 2 hours. The whole wheat flour seemed to have a greater buffering effect on the system compared to white flour, resulting in an initially high pH with only a slight decrease in pH over the remainder of the fermentation time. This may have been due, in part, to the higher protein content of the whole wheat flour.

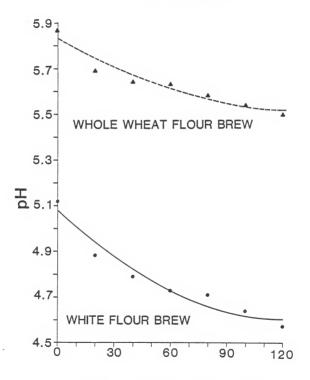
The whole wheat flour brew TTA increased more rapidly than the white flour brew during the first 60 minutes of fermentation (Figure 16). After that time, little increase in whole wheat brew TTA values were noted, however, white flour TTA values continued to increase rapidly after a plateau in the 60-80 minute range.

BREAD pH and TTA

No significant differences in pH or TTA were detected among the breads tested (Table 7). The stage at which whole

Figure 15. Brew pH During 2 Hour Fermentation

BREW pH



Fermentation time, min

Figure 16. Brew TTA During 2 hour Fermentation

BREW TTA

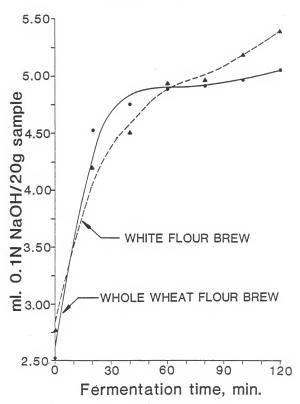


Table 7. STATISTICAL ANALYSIS OF pH and TTA OF WHEAT BREAD MADE BY THE ADDITION OF 30% WHOLE WHEAT FLOUR AT VARIOUS STAGES IN THE LIQUID FERMENT AND SPONGE-DOUGH PROCESSES

		BAKING	PROCESS	
	LIQUID FERMENT		SPONGE-DOUGH	
	WHITE BREW	WHOLE WHEAT BREW	WHITE SPONGE	WHOLE WHEAT SPONGE
	MEAN	MEAN	MEAN	MEAN
pH	5.75A	5.78A	5.76A	5.78A
TTA*	2.5A	2.3A	2.4A	2.5A

^{*}Milliliters of 0.1 normal NaOH required to raise sample to pH 6.6.

Means with the same letter compared horizontally are not significantly different (Alpha = 0.05).

wheat flour was added in either process did not affect measured values.

SCANNING ELECTRON MICROGRAPHS

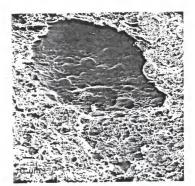
Figures 17 and 18 are scanning electron micrographs of doughs containing white and whole wheat flour added at various stages in the liquid ferment and sponge-dough processes. It had been theorized that addition of whole wheat flour at the dough stage may cause a weakening or tearing of the protein matrix due to bran particle size or sharpness. If this was the case, it was not evident by scanning electron microscopy.

CRUMB FIRMNESS

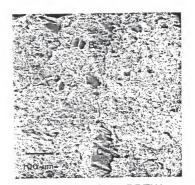
Table 8 summarizes the crumb firmness data obtained using 30% wheat bread made by white flour and whole wheat flour brews and sponges. Sponge-dough bread had significantly firmer crumb than did bread produced using the liquid ferment process (Figure 19). No differences existed between white flour brews and whole wheat brews or white flour sponges and whole wheat sponges on either day 0 or day 5.

The difference in firmness bewteen sponge-dough and liquid ferment bread can be partially explained by differences in specific volume of the loaves (Table 9). Axford et al. (1968) showed that specific volume was a major factor affecting both the absolute extent and rate of

- Figure 17. Scanning Electron Micrographs of Doughs Made From White Flour Brews (Whole Wheat Flour Added at Dough Stage)
 - A. Air Cell Showing Starch Granules and Smooth Protein Matrix
 - B. Aleurone Cells
 - c. Bran Particle



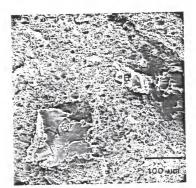
WHITE FLOUR BREW



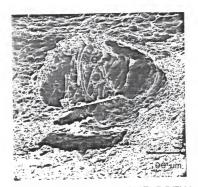
WHITE FLOUR BREW

Figure 18. Scanning Electron Micrographs of Doughs Made From Whole Wheat Flour Brews (White Flour Added at Dough Stage)

- A. Bran Particle
- B. Aleurone Cells
- C. Wheat Kernel Brush Hairs



WHOLE WHEAT FLOUR BREW



WHOLE WHEAT FLOUR BREW

Table 8. STATISTICAL ANALYSIS OF DAY 0 AND DAY 5 FIRMNESS MEASUREMENTS OF WHEAT BREAD MADE BY THE ADDITION OF 30% WHOLE WHEAT FLOUR AT VARIOUS STACES IN THE LIQUID FERMENT AND SPONGE-DOUGH PROCESSES

	LIQUID	BAKING FERMENT	PROCESS SPONGE	-DOUGH
	WHITE BREW	WHOLE WHEAT BREW	WHITE SPONGE	WHOLE WHEAT SPONGE
	*	*	*	*
DAYS STORED	MEAN	MEAN 	MEAN 	MEAN
Day 0	110.7A	104.2A	82.9B	82.1B
Day 5	488.0A	476.1A	399.5B	400.8B

^{*}Grams of force measured by Voland-Stevens-LFRA Texture Analyzer

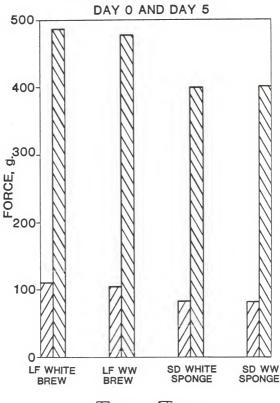
Average of 4 replications.

Means with the same letter compared horizontally are not significantly different (Alpha = 0.05).

Day 0 was measured at 2 hr. after baking.

Figure 19. Day 0 and Day 5 Crumb Firmness

CRUMB FIRMNESS



DAY 0 DAY 5 69

firming. As expected, breads of higher loaf specific volumes (those made by the sponge-dough process) had lower crumb firmness values than those of lower specific volumes (liquid ferment breads). The remaining differences were presumably caused by changes associated with variations in fermentation time.

CRUMB COLOR

Differences in crumb color were not significant between the liquid ferment and sponge-dough processes or between stages of whole wheat flour addition within each process (Table 9 and Figure 20). It was possible that "mellowing" of the crumb color by the early addition of whole wheat flour in the process or by the longer sponge-dough fermentation time might result in lighter crumbs. This, however, was not the case.

PROOF TIME

Table 10 lists in minutes the times required to proof doughs of white and whole wheat flour brews and sponges to a constant height. A shorter proof-to-height time was required for the sponge-dough process than for the liquid ferment procedure (Figure 21). Although studies have suggested a possible relationship between proof time and average firming rate, evidence appears inconclusive (Ponte et al. 1962).

Table 9. STATISTICAL ANALYSIS OF CRUMB COLOR OF WHEAT BREAD MADE BY THE ADDITION OF 30% WHOLE WHEAT FLOUR AT VARIOUS STAGES IN THE LIQUID FERMENT AND SPONGE-DOUGH PROCESSES

		BAKING	PROCESS	
	LIQUID FERMENT		SPONGE-DOUGH	
	WHITE BREW	WHOLE WHEAT BREW	WHITE SPONGE	WHOLE WHEAT SPONGE
	MEAN	MEAN	MEAN	MEAN
CRUMB* COLOR	53.7A	53.9A	54.1A	53.3A

 $^{^{\}star} \text{Agtron}$ values based on % Relative Reflectance at wavelength = 546nm.

Average of 4 replications.

Means with the same letter are not significantly different (Alpha = 0.05).

Figure 20. Crumb Color

CRUMB COLOR AGTRON

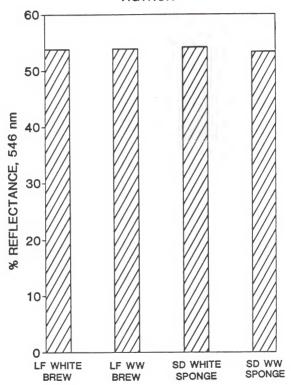


Table 10. STATISTICAL ANALYSIS OF SPECIFIC VOLUME AND PROOF TIME OF WHEAT BREAD MADE BY THE ADDITION OF 30% WHOLE WHEAT FLOUR AT VARIOUS STAGES IN THE LIQUID FERMENT AND SPONGE-DOUGH PROCESSES

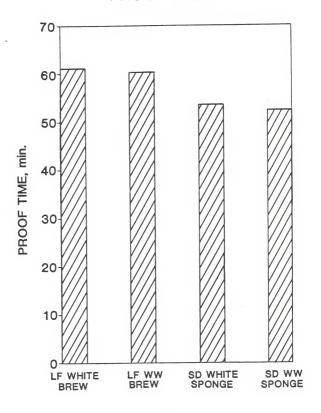
		BAKING	PROCESS	
	LIQUID FERMENT		SPONGE-DOUGH	
	WHITE BREW	WHOLE WHEAT BREW	WHITE SPONGE	WHOLE WHEAT SPONGE
	MEAN	MEAN	MEAN	MEAN
VOLUME	2635.9A	2700.0B	2942.2C	2937.5C
WEIGHT (g)	494.2A	494.4A	492.0AB	489.8B
SPECIFIC VOLUME (cc/g)	5.3A	5.5B	6.0C	6.0C
PROOF TIME (min)	61.2A	60.4A	53.6B	52.5B

Average of 4 replications.

Means with the same letter compared horizontally are not significantly different (Alpha = 0.05).

Figure 21. Proof Time

PROOF TIME



SPECIFIC VOLUME

Significant differences in loaf specific volume existed both within the liquid ferment and between liquid ferment and sponge-dough procedures (Table 10). No significant differences were found in loaf specific volume between white and whole wheat flour sponges.

Wheat breads produced from whole wheat flour brews had a higher loaf specific volume than those of white flour brews. However, this value was less than that of breads made by sponge-dough methods, regardless of the sponge type (Figure 22).

BREAD MOISTURE

Table 11 summarizes the results of bread moisture determinations performed on wheat breads made by liquid ferment and sponge-dough processes containing 30% whole wheat flour (added at various stages). No significant differences in bread moistures were noted. Thus, further comparisions can be made between breads on a dry solids basis.

RESIDUAL SUGARS

Table 12 shows the amounts of residual sugars detected in bread crumb samples using high performance liquid chromatography analysis. Fructose and glucose levels were significantly higher in breads made by the sponge-dough

Figure 22. Specific Volume

SPECIFIC VOLUME

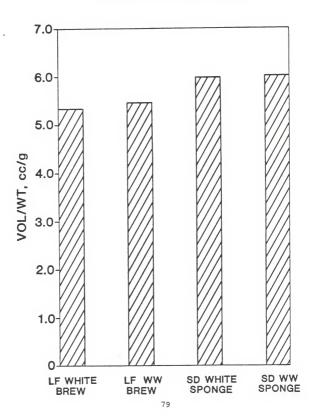


Table 11. STATISTICAL ANALYSIS OF BREAD MOISTURES OF WHEAT BREAD MADE BY THE ADDITION OF 30% WHOLE WHEAT FLOUR AT VARIOUS STAGES IN THE LIQUID FERMENT AND SPONGE-DOUGH PROCESSES

		BAKING	PROCESS		
	LIQUID	LIQUID FERMENT		SPONGE-DOUGH	
	WHITE BREW	WHOLE WHEAT BREW	WHITE SPONGE	WHOLE WHEAT SPONGE	
	MEAN	MEAN	MEAN	MEAN	
BREAD MOISTURE	36.2A	36.6A	36.7A	36.9A	

Means with the same letter are not significantly different (Alpha = 0.05).

Table 12. STATISTICAL ANALYSIS OF BREAD CRUMB RESIDUAL SUGARS OF WHEAT BREAD MADE BY ADDITION OF 30% WHOLE WHEAT FLOUR AT VARIOUS STAGES IN THE LIQUID FERMENT AND SPONGE-DOUGH PROCESSES

	BAKING PROCESS			
	LIQUID FERMENT		SPONGE-DOUGH	
	WHITE BREW	WHOLE WHEAT BREW	WHITE SPONGE	WHOLE WHEAT SPONGE
RESIDUAL SUGAR	MEAN ^a (%)	MEAN ^a	MEAN ^a (%)	MEAN ^a (%)
FRUCTOSE	1.34A	1.42A	1.47AB	1.58B
GLUCOSE	0.89A	0.88A	1.08B	1.04B
SUCROSE	ND_p	ND	ND	ND
MALTOSE	0.76A	0.83A	0.26B	0.36B
LACTOSE	0.55A	0.54A	0.49A	0.52A

^aDry solids basis

Average of 4 replications.

Means with the same letter compared horizontally are not significantly different (Alpha = 0.05).

b_{Non-detectable}

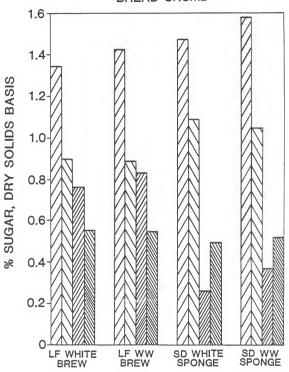
process than in those made by the liquid ferment procedure. No significant differences in fructose or glucose contents were found between white and whole wheat flour brews or sponges. More fructose than glucose was present in the crumb of all breads (Figure 23), consistant with previous work (Rice 1938 and Tang et al. 1972). It is due to the preferential utilization of glucose over fructose as a yeast carbohyrate source.

Maltose levels, on the other hand, were substantially higher in liquid ferment breads than in sponge-dough breads (Table 12). During fermentation, maltose becomes available due to amylytic hydrolysis of starch mechanically damaged during the milling process. Maltose levels continued to increase in the dough until the monosaccharides such as fructose and glucose are no longer present in sufficient quantities to support yeast activity. At that point, the yeast adapts to the utilization of maltose and the maltose level begins to drop. Thus, the longer fermentation time required in the sponge-dough process necessitates the use of greater quantities of maltose compared to that of the shorter liquid ferment process. This was reflected by the lower levels of maltose in the sponge-dough bread crumb.

No significant differences in lactose were detected among the breads tested. This was expected due to the inability of baker's yeast to utilize lactose as a carbohydrate source. No sucrose was detected in any of the bread crumb samples. This is the result of almost immediate

Figure 23. Bread Crumb Residual Sugars

RESIDUAL SUGARS BREAD CRUMB



FRUCTOSE

MALTOSE

GLUCOSE 84

M LACTOSE

hydrolysis of the dissacharide to its glucose and fructose components by yeast invertase.

SENSORY EVALUATION

Results of the taste panel triangle test are shown in Figure 24. Of the 25 panelists involved in the test, 13 were able to correctly identify the odd sample. Of those 13. seven indicated a slight difference beween duplicates and odd sample, four suggested a moderate difference and two said there was much difference. Seven "correct" panelists preferred the white flour brew-30% wheat bread, four preferred the whole wheat flour brew-30% wheat bread and two indicated no preference. The "correct" panelists chose the white flour brew as being the best tasting bread due to its milder flavor as compared with the whole wheat brew bread. These panelists used words such as "sharp", "acidic", and "strong after taste" in referring to the whole wheat brew bread. The four panelists preferring the whole wheat brew bread indicated it had a "nutty" or "branny" flavor while the white flour brew bread was "bland".

According to statistical analysis supplied by Larmond (1977), 13 out of 25 panelists must correctly identify the odd sample in order to identify statistically significance differences among samples at the alpha = 0.05 level. Therefore, the results of this study indicate a significant flavor difference between white flour and whole wheat flour brew wheat breads.

Figure 24. Sensory Evaluation Summary

TRIANGLE TASTE TEST RESULTS OF 25 PARTICIPANTS

Number of Correct Respo	onses		13
Degree of Difference I Between odd and even s		Slight Moderate Much	 7 4 2
Sample preferred	White Flour Whole Wheat No Preference	Flour Brew	 7 4 2

13 OUT OF 25 CORRECT RESPONSES INDICATES A STATISTICALLY SIGNIFICANT DIFFERENCE AMONG SAMPLES AT ALPHA=0.05 LEVEL.

CONCLUSIONS

Brews containing whole wheat flour had higher pH values than white flour brews during the two hour fermentation. A buffering effect was observed after 1 hour of fermentation according to pH and TTA readings. This was at least partially attributed to the higher protein content of the whole wheat flour. Breads containing 30% whole wheat flour added at either brew/sponge or dough stages showed no significant differences in pH or TTA.

Scanning electron micrographs of doughs containg 30% whole wheat flour added at the dough stage showed no apparent break-down or weakening of the protein matrix when contrasted with whole wheat flour brews. Thus, the late addition of whole wheat particles did not appear to cause structural changes in the dough.

Crumb firmness values were not significantly different between early and late addition of whole wheat flour in either the liquid ferment or sponge-dough processes. Therefore, the stage of whole wheat flour addition does not appear to affect crumb firmness. Significant differences do exist between liquid ferment and sponge-dough processes, however, with the liquid ferment process having a firmer crumb.

No crumb color differences existed among breads regardless of the stage in which whole wheat flour was added. This was true in both the liquid ferment and spongedough processes.

A shorter proof time was required for the sponge-dough process than for the liquid ferment process. No significant differences occured within each process.

Wheat breads produced from a whole wheat flour brew had a significantly higher loaf specific volume than breads of white flour brews. No differences existed between spongedough breads. Sponge-dough bread specific volumes were greater than those of liquid ferment breads.

Analysis of bread crumb residual sugars indicated no significant differences in fructose, glucose, maltose, or lactose levels among white or whole wheat flour brews with in the two processes. Sucrose was not detected in the crumb of the breads tested. This was expected due to rapid hydrolysis by yeast invertase to its fructose and glucose monosaccharide units. Significantly lesser amounts of maltose were found in breads made by the sponge-dough process than in those made by the liquid ferment procedure. This was due to the required use of maltose by yeast as fructose and glucose levels reached critically low levels during the four hour sponge fermentation.

Sensory evaluation involving a triangle test taste panel indicated a slight difference between wheat breads in which the whole wheat flour was added at either the brew or dough stage of production. Wheat breads made from white flour brews were perceived as being milder in flavor than those from whole wheat flour brews.

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ACKNOWLEDGEMENTS

The author would like to express his sincere appreciation to Professor J. G. Ponte, Jr. for his guidance and leadership throughout this research. The author would like to thank advisory committee members Dr. Jon Faubion and Dr. Eli Posner and also American Institute of Baking consultant Mr. Karel Kulp for their technical assistance and sound advice. Finally, a special thanks goes to the author's family for their unrelenting support and encouragement throughout his academic endeavors.

COMPARISION OF WHITE AND WHOLE WHEAT FLOUR BREWS IN THE LIQUID FERMENT PROCESS OF BREADMAKING

by

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B.S., KANSAS STATE UNIVERSITY, 1982

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Grain Science and Industry

KANSAS STATE UNIVERSITY Manhattan, Kansas

This study compared the effects of adding whole wheat flour at two different stages of the liquid ferment process. Comparisions were based on a formulation containing 30% whole wheat flour added at either the brew or dough stages. Wheat breads were also produced by the sponge-dough process with similar variations in whole wheat flour addition. These breads represented the dominant industry procedure, allowing comparisions both within and between liquid ferment and sponge-dough processes.

Brew stage comparisions were made using pH and total titratable acidity (TTA) analysis. Whole wheat flour in the brew showed a buffering effect after one hour of fermentation. Bread ph and TTA values were not significantly different.

Scanning electron micrographs of doughs showed similar protein matrix and starch granule characteristics. Thus, stage of whole wheat flour addition does not appear to affect dough structure.

Bread crumb firmness was measured on day 0 and day 5. Crumb firmness values were not significantly different between early and late addition of whole wheat flour within either breadmaking process, however differences did exist between processes. Higher firmness values in liquid ferment breads compared to sponge-dough breads were thought to be related to lower loaf specific volumes.

High performance liquid chromatography was used to

estimate residual sugars contained in the bread crumb. There were no significant differences in fructose, glucose, maltose, or lactose levels between early and late whole wheat flour addition within each process. Residual maltose was lower in the crumb of breads made by the sponge-dough process. This was due to the required use of maltose by yeast as other substrates reached critically low levels in the four hour sponge fermentation. Residual fructose and glucose levels were higher in the sponge-dough process, while lactose was not significantly different.

Crumb color and bread moisture values were not significantly different between breads. No differences in proof time occured within sponge-dough or liquid ferment processes. Wheat breads produced from a whole wheat flour brew had a higher loaf specific volume than breads of white flour brews.

Sensory evaluation indicated a slight difference between white and whole wheat flour brews in 30% wheat bread. A triangle test taste panel preferred breads in which the whole wheat flour was added at the dough stage of production.