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To the Graduate Council:

I am submitting herewith a thesis written by Whei-Ling Karin Pao entitled "Quality of soy fortified bread as affected by soy flour lipids and water dispersibility of soy protein." I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Food Science and Technology.

S.L. Melton, Major Professor

We have read this thesis and recommend its acceptance:

H.O. Jaynes, J.L. Collins

Accepted for the Council: Carolyn R. Hodges

Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

To the Graduate Council:

I am submitting herewith a thesis written by Whei-Ling Karin Pao entitled "Quality of Soy Fortified Bread as Affected by Soy Flour Lipids and Water Dispersibility of Soy Protein." I recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Food Technology and Science.

S. L. Melton, Major Professor

We have read this thesis and recommend its acceptance:

Velin

Accepted for the Council:

Vice Chancellor Graduate Studies and Research

Ag-VetMod



QUALITY OF SOY FORTIFIED BREAD AS AFFECTED BY SOY FLOUR LIPIDS AND WATER DISPERSIBILITY OF SOY PROTEIN

A Thesis

Presented for the

Master of Science

Degree

The University of Tennessee, Knoxville

Whei-Ling Karin Pao

December 1977

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Finally, the author wishes to express appreciation to Arthur Daniels Midland Company, Central Soya Company, and Wades Bakery which provided the materials of this research.

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ABSTRACT

The effects of defatted soy flours with different protein dispersibility index (PDI 10-25 and 65-75) alone and in combination with added lipids (polar and/or neutral at concentrations present in full fat soy flour) and a full fat soy flour (PDI 15-25) were investigated on quality of 12 and 24 percent (on full fat soy flour or its lipid free, dry material equivalent basis) fortified bread. The following characteristics were measured: specific loaf volume, crust and crumb color, total solids, and texture during staling (compressibility at 0, 2, 4, and 6 days). Bread was prepared according to the K-State Process, and the flavor and acceptability of 12 percent soy level were evaluated by a 25-member untrained sensory panel on a 6-point hedonic scale. Addition of neutral lipids to defatted soy flour caused greater loaf volume and more total solids than polar lipids. The addition of neutral and polar lipids to defatted soy flour gave bread the greatest loaf volume and highest level of total solids. PDI of defatted soy flour did not affect the specific loaf volume of bread significantly. Therefore, reported differences between loaf volume of bread fortified with full fat soy flour and with defatted soy flour and made by K-State Process (addition of sodium stearoy1-2 lactylate) would seem to be due to the lipid content of the full fat soy flour rather than protein quality, particularly the neutral lipids. The combined neutral and polar lipids also caused bread to have a lighter and less yellowish

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crumb color. Toasted defatted soy flour (TN, PDI 10-25) caused bread to have darker and more yellowish crumb color than a white defatted soy flour (BN, PDI 65-75). A consumer panel generally preferred the flavor of bread fortified with TN more than that of bread fortified with BN. The addition of neutral lipids to defatted soy flour caused bread to be softer than the addition of polar lipids and the combined lipids caused the softest texture of bread at 0, 2, 4, and 6 days storage, especially at the 24 percent soy level.

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CHAPTER I

INTRODUCTION

During the past decade, increased attention has been devoted to the use of numerous plant protein supplements as a potential source of additional protein in human food. Today's hungry world is in need of an economical source of high quality protein. Defatted soy flour, a byproduct of the oil extraction process, in particular, has received much attention because of the quantity produced, its low cost, and its high nutritive value. Although defatted soy flour possesses numerous desirable attributes, bread fortified with soy flour has some problems such as reduced loaf volume, course, open texture, off-white or yellowish color, and off-flavor described as bitter or beany.

Past research has indicated full fat soy flour is a superior additive to defatted soy flour in bread made with sodium stearoy1-2 lactylate. However, it is not known whether protein quality, fat, natural emulsifiers such as glycolipids, lecithin, or other substances are responsible for this superiority.

Numerous factors such as fermentation time, temperature, relative humidity, baking time and temperature, and all of the ingredients added affect quality characterisitcs of bread fortified with soy flour, especially in specific loaf volume. For a bread to be marketable, it must have a specific loaf volume of 6.00 cc/g. It would be desirable

to find conditions under which defatted soy flour could be added at high levels (above 12 percent) and still maintain that desirable specific loaf volume.

This study investigated factors (Protein Dispersibility Index and neutral and polar lipids of full fat soy flour, alone or in combination) which might be the cause of the difference in specific loaf volume between full fat soy flour and defatted soy flour fortified bread made by the K-State Process at 12 and 24 percent levels added soy flour. The effect of these factors on other characteristics of soy fortified bread, such as crust and crumb color, flavor, overall acceptability, staling (compressibility at 0, 2, 4, and 6 days storage), and protein and moisture content also was observed.

CHAPTER II

LITERATURE REVIEW

The food shortage brought on by World War II stimulated a strong interest in soy flour as a new source of food protein, especially for improving the nutritive value of bread and other bakery products. Currently, soy flour is used in bread manufacture for one or more of the following purposes (18, 33, 36, 45, 58):

- a. To extend or replace milk solids (non-fat dry milk) (16, 27, 67).
- b. To control moisture in the crumb (13, 41, 42).
- c. To whiten unbleached flour products.
- d. To increase protein content and to improve protein quality by increasing lysine content to get better amino acid balance (6, 7, 9, 11, 23, 25, 41): Soy flour contains 3.2 to 3.8 percent lysine compared with 0.38 percent in wheat flour. A mixture of wheat flour fortified with 12 percent defatted soy flour more than doubles the lysine content compared to wheat flour alone, and bread made from such a blended flour increases bread protein content by approximately 35 percent (61). Increasing the level above 12 percent would even increase protein content of soy fortified bread even more dramatically. Melton et al. (37) reported that protein

content of soy fortified bread increased from 15 to 21 percent as defatted soy flour level increased from 6 to 24 percent.

- e. To introduce variety (speckiness, flavor, etc.) (17, 44).
- f. Economics: Almost all of the major processors of soybean were geared in their oil extraction plants for the manufacture of soybean oil and meal, the residue primarily was used for animal feeds. The defatted soy flour is economical (20, 21, 31).

The acceptance of soy bread has been poor mainly because of functional disadvantages and nonuniformity of soy flour in early stages of its development (48, 49). The functional problems generally associated with the use of soy flours in bread dough in the past include: (a) alteration of water absorption, mixing and machining properties of dough; (b) adverse effects on loaf volume, grain, texture, color, and flavor; (c) changes in fermentation rates; and (d) effects on the gluten complex, including oxidation requirements (3, 5, 8, 10, 13, 14, 19, 54).

To alleviate these adverse effects, research workers generally selected wheat and different types of soy flours (45), used additives (12, 54), and modified the processing conditions (34, 61, 63).

I. SELECTION OF WHEAT AND SOY FLOURS

A number of workers showed that mixing properties and baking performance of bread containing fortified flour largely depended on the

qualities of wheat and soy flour (41, 42, 63). The enzyme system, the storage time, and temperature of storage for soy and wheat flours also affected the bread quality and dough properties (28, 29, 30, 34, 35). The functional characteristics of edible soy flours can be substantially affected by the method of processing the soya (65). Chemical-treated soy flours have more desirable functional characteristics for bread baking than the heat-treated flours (43). Heating soy flours reduced nitrogen dispersibility, increased water absorption of the soy flours in dough and darkened the flour color. Deleterious effects on the baking quality and the stability of dough of heated soy flours increased with severity of heat treatment (15, 48).

Particle size of soy flour also affected breadmaking and loaf quality (18). Finely powdered soy flours required more water and dough mixing and slightly more bromate than coarse soy flours (15). Adding coarse soy flour produced bread that had better crumb grain and color and larger loaf volume than did the less granular soy flour.

Studies with lean formula (without added shortening) showed the advantages of using lecithinated defatted soy flours or full fat soy flours in breadmaking (1, 64). Lecithinated or high fat soy flours had little or no advantage in bread baked by a formula which contained 3 percent vegetable shortening (15). By comparing 16 commercial soy protein products, the use of full-fat or high-fat soy flour was more suited for the production of good quality high protein bread (20 percent soy level) (52).

Melton et al. (37) found that defatted soy flour, nitrogen solubility index (NSI) of 77, had a significantly higher specific loaf volume, superior grain and higher overall acceptability at 6, 12, 18, and 24 percent soy flour addition than bread fortified with defatted soy flour, NSI of 20.

II. USE OF ADDITIVES

Pomeranz et al. (46, 47) found that adding natural and synthetic glycolipids or sucroesters to wheat flour permitted addition of up to 16 percent soy flour and other protein rich foodstuffs to bread formula without a significant loss in physical qualities. Sucroglycerides and sucrose monotallowate was essential for normal loaf volume of 12 percent soy fortified bread (24). Phosphatides were valuable for the emulsifying properties and the ability to reduce the interfacial tension of the constituents of the dough of soy bread, thereby producing a more uniform dispersion and more efficient action of the shortening (50). Tsen et al. (61, 62) reported that sodium stearoy1-2 lactylate (SSL), calcium stearoyl-2 lactylate (CSL) and ethoxylated monoglycerides formed a complex with gluten to stabilize the gluten network in dough and improved bread quality. SSL was more effective than CSL in sparing shortening when fortified flour was used for high protein bread. SSL and CSL have been approved by the Food and Drug Administration as safe food additives (62). Turro and Sipos (67) reported a new product, a specially processed soy flour with certain functional and nutritional

additives, which improved the buffering and mixing properties of the dough system. The food additives included calcium lactate, calcium sulfate, and ascorbic acid.

III. MODIFICATION OF THE PROCESSING CONDITIONS

Through various studies on modified processing conditions, it has been well documented that one of the following changes may improve baking performance of fortified flour:

- a. Changing the formulation (35).
- b. Decreasing the mixing time (8, 15, 33, 34, 61, 62): Soy fortified bread (12 percent) required 40 percent reduction in mixing time.
- c. Raising the content of water (13, 41, 42, 66): Soy fortified bread required a 5 percent increase in water over that expected for a dough composed of only wheat flour (33, 34).
- d. Increasing the oxidant treatment (1, 5, 8, 13, 41, 42, 61):
 Oxidizing agents of the bromate and iodate type improve the dough properties and soy fortified bread qualities.
- e. Reducing the fermentation period (5, 8).

The K-State Process (63), a no-time dough process with added SSL, has been the most successful to date in producing high protein bread. Fellers et al. (12) reported that a slightly heated defatted product (PDI 65-75 equivalent) was used in a 12 percent soy-wheat flour blend (K-State blend) for production of nutrition enriched bread in U.S.A. overseas aid programs. Soybean flour has been used in bread at a level of 6 percent or slightly more without serious impairment of flavor, texture or loaf volume (1, 14, 15, 41, 42) with only some alterations in the dough formula and baking procedure. The lower soy level (less than 6 percent) of fortified bread has been sold in supermarkets and used in school lunch programs. When higher amounts of soybean flour were used, however, the bread became decidedly less acceptable (14). With the K-State Process, a 12 percent soy fortified bread that is marketable may be produced. Melton et al. (37) showed that the flavor and overall acceptability score of bread containing SSL and fortified with 12 percent defatted soy flour was between "like moderately" and "like very much."

Tsen and Hoover (64) reported that addition of 0.5 percent SSL to soy fortified bread allowed the incorporation of up to 24 percent full fat soy flour (protein dispersibility index, PDI, 15-25) without severe suppression of specific loaf volume. Bread containing 28 percent full fat soy flour and 0.5 percent SSL had a specific loaf volume of 5.82 cc/g compared to 3.22 cc/g for bread containing 28 percent full fat soy flour and no SSL. In contrast to these findings, they reported that addition of 0.5 percent SSL to bread fortified with defatted soy flour (PDI 35-45) at levels above 12 percent did not cause dramatic improvement in specific loaf volume. Bread containing 28 percent defatted soy flour and 0.5 percent SSL had a specific loaf volume of 3.38 cc/g. Compared on an added equivalent soy flour level, full fat soy flour was a

superior additive to defatted soy flour in bread made with SSL. They further pointed out that it was not known whether protein quality, fat, natural emulsifiers such as glycolipids and lecithin, or other substances were responsible for this superiority.

CHAPTER III

MATERIALS AND METHODS

I. STATISTICAL METHODS

The statistical design of this experiment was a split plot (55, 59, 60). Within each replication, the order of completion for nine treatments was randomized for each soy flour level (12 and 24 percent full fat soy flour or its lipid free, dry matter equivalent, wheat flour basis) in bread. Two replications were run. The nine treatments were additions of different soy flours and/or lipids to bread and were as follows:

Treatment 1: full fat soy flour (PDI 15-25).

Treatment 2: defatted soy flour (PDI 10-25).

Treatment 3: defatted soy flour (PDI 65-75).

Treatment 4: defatted soy flour (PDI 10-25) + same quantity of neutral lipids present in full fat soy flour.

Treatment 5: defatted soy flour (PDI 10-25) + same quantity of polar lipids present in full fat soy flour.

- Treatment 6: defatted soy flour (PDI 10-25) + same quantities of polar and neutral lipids present in full fat soy flour.
- Treatment 7: defatted soy flour (PDI 65-75) + same quantity of neutral lipids present in full fat soy flour.

Treatment 8: defatted soy flour (PDI 65-75) + same quantity of polar lipids present in full fat soy flour.

Treatment 9: defatted soy flour (PDI 65-75) + same quantities of polar and neutral lipids present in full fat soy flour.

Efforts were made to assure the composition of the added polar and neutral lipids the same as those present in the full fat soy flour. Crude soybean oil, deslimed by the method of Rede et al. (53) which removed approximately 76 percent of the phospholipids, was added as the neutral lipids and crude soy lecithin was added for the polar lipids.

Analysis of variance was run for the dependent variables: specific loaf volume, crust and crumb color, and moisture content (Table 1, subtotal degrees of freedom is 35). Data for flavor and overall acceptability of bread containing 12 percent full fat soy flour or its lipid free, dry material equivalent were analyzed according to the analysis of variance in Table 2.

Compressibility of bread stored at 23°C for 0, 2, 4, and 6 days was determined, and these data were analyzed by complete analysis of variance shown in Table 1 (total degrees of freedom is 143). The complete analysis of variance showed effects of level, treatment and storage time on compressibility of soy fortified bread.

When significant treatment effects were found for a dependent variable, orthogonal comparisons (Table 3) (60) were used to find significant differences among treatments. When significant storage,

Source	Degrees of Freedom
Level (L)	1
Replication (R)	1
L x R (Error A)	1
Treatment (T)	8
Cl ^a	1
C2	1
C3	1
C4	1
C5	1
C6	1
C7	1
C8	1
LXT	8
L x Cl	1
L x C2	1
L x C3	1
L x C4	MARCENIC AND
L x C5	
L x C6	intervention 1 and the
L x C7	1
L x C8	1
T x R) Frror B	8) 16
L X T X R)	
	35 Subtotal
Storage Time (S)	3
S x L	3
SxT	24
SxLxT	24
SxR)	3)
SxLxR)	3) 54
S X T X R) Error C	24) 54
SxLxTxR)	24)
	108 Subtotal
Total	143

Table 1. Analysis of Variance for Dependent Variables Determined on 12 Percent and 24 Percent Soy Flour Fortified Bread.

 $a_{\rm C}$ = Comparison

Source	Degrees of Freedom
Treatment (T)	8
C1 -	
C4	1
C5	1
C6	1
C7	1
C8	1
Replication (R)	
T x R (Error)	ANESE ST
Total	17

Table 2.	Analysis of Variance for Flavor and Acceptability
	of 12 Percent Soy Flour Fortified Bread.

^aC = Comparison

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E.	1993
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	Full Fat Soy Floui				Defatted	Soy Flour			
Treatment	1	2	3	4	5	9	7	8	6
PDI ^a	15-25	10-25	65-75	10-25	10-25	10-25	65-75	65-75	65-75
Added Lipids	1	1	-	APP	Pp	NP+P	NP	P	NP+P
Comparison (C)									
1	-2	+1	+1	1+	+1	-2	+1	+1	-2
2	+2	0	0	0	0	-	0	0	1-
З	0	0	0	0	0	-1	0	0	+1
4	0	0	0	-1	-1	0	+1	+1	0
S	0	0	0	-1	+1	0	-1	+1	0
9	0	0	0	+1	۲.	0	-1	+1	0
- · · L	0	-1	+1	0	0	0	0	0	0
Ø	0	+2	+2	-	а Т - - - - - - - - - - - - - -	0	.	 1	0

^aMeans protein dispersibility index.

 $b_{P} = polar lipids$ and NP = neutral lipids which was added to defatted soy flour to the same concentration as in full fat soy flour.

treatment x storage, and level x storage effects were found for . compressibility, orthogonal polynomials were used first to estimate the effect of storage time on compressibility. However, after graphing the data it was seen that for each treatment-level combination with the exception of Treatment 9 at 12 percent soy flour level, the data best fit the model $Y = A + Be^{KT}$ where Y = compressibility, g/mm;T = storage time, days; and A, B, and K are constants. The constants were estimated from the data for each treatment-level combination with the exception of Treatment 9 at 12 percent full fat soy flour equivalent level by non-linear regression analysis. The effect of storage on compressibility of bread for Treatment 9 at 12 percent full fat soy equivalent level was estimated by orthogonal polynomials, and the model equation Y = a + bT where Y = compressibility, g/mm; T = storage time, days; and a and b are constants was determined. The equations and the actual compressibility means for each treatment-level combination were then graphed. Equations for the models of each treatment-level combination and means of compressibility are given in Appendix.

The values of crude protein of bread fortified with 12 and 24 percent full fat soy flour or its lipid free, dry material equivalent from defatted soy flour for Treatments 1, 2, and 3 were analyzed by analysis of variance (Table 4) and the means were separated by Student Newmans Keul's Test (60).

Source	Degrees of Freedom
Level (L)	1
Replication (R)	1
L x R (Error A)	•1
Treatment (T)	2
TxL	2
T x R)) Error B	2)
TxRxL)	2)
Total	11

Table 4. Analysis of Variance for Crude Protein in Soy Fortified Bread of Treatments 1, 2 and 3^a.

^aTreatments are defined on pages 10-11.

II. MATERIALS AND THEIR ANALYSES

Three soy flours (Nutrisoy 220, a full fat soy flour with PDI 15-25; Toasted Nutrisoy flour, a more yellow defatted soy flour with PDI 10-25; and Bakers Nutrisoy, a less yellow defatted soy flour with PDI 65-75) were obtained from Arthur Daniels Midland Company, Decatur, Illinois (Table 5). These three flours represented varying degrees of protein dispersibility.

These soy flours also were analyzed for moisture, total lipids, polar lipids (glycolipids and phospholipids), neutral lipids (mono-, di- and triglycides, sterol and sterol ester), phospholipids in total lipids, and lecithin and cephalin in polar lipids.

A high gluten hard wheat flour (with 3 ppm bromate) utilized for breadmaking and fresh granular compressed yeast (Federal Yeast Corporation, Baltimore, Maryland), were provided by Wades Bakery, Knoxville, Tennessee. Sodium stearoyl-2 lactylate (SSL) was obtained from the C. J. Patterson Company, Kansas City, Missouri. Yeast food was provided by Kern's Bakery, Knoxville, Tennessee.

Crude soybean oil and crude lecithin were obtained from Central Soy Company, Chicago, Illinois. After desliming the crude soybean oil, phospholipid content of oil was determined. Polar lipids, neutral lipids, phospholipids, and lecithin and cephalin content in crude lecithin were also determined.

the second s	the second se	the second se
FF ^a	TN ^b	BNC
7.25	8.23	7.25
43.0	52.0	52.0
22-23	1.0	1.0
2.5	3.5	3.5
2.40	2.20	2.20
15-25	8-20	70-79
4.5	5.7	5.7
27.0	34.5	34.5
465	360	360
	FF ^a 7.25 43.0 22-23 2.5 2.40 15-25 4.5 27.0 465	FF ^a TN ^b 7.25 8.23 43.0 52.0 22-23 1.0 2.5 3.5 2.40 2.20 15-25 8-20 4.5 5.7 27.0 34.5 465 360

Table 5. Proximate Composition of Three Types of Soy Flours.

^aNutrisoy 220 (full fat soy flour, PDI 15-25).

^bToasted Nutrisoy (defatted soy flour, PDI 10-25). ^cBakers Nutrisoy (defatted soy flour, PDI 65-75). ^dDetermined by A.O.A.C. method (2), percent. ^eAnalyzed by Arthur Daniels Midland Company, percent. ^fAnalyzed by Arthur Daniels Midland Company, calories.

III. EXPERIMENTAL PROCEDURES

- A. Analyses of Soy Flour Lipids.
 - Lipids of full fat soy flour and the two defatted soy flours were extracted and quantitated by the method of Moyer et al. (39). Duplicate samples were extracted for each flour.
 - 2. The quantities of neutral lipids and polar lipids in total lipids extracted from each flour were estimated by elution from silicic acid column by chloroform : methanol, 20 : 1, v/v (neutral lipids) and by chloroform : methanol, 1 : 1, v/v and methanol (polar lipids) (22).
 - a. The type of lipids in each fraction was identified by thin layer chromatographic techniques, molybdenum blue test for phospholipids, and diphenylamine test for glycolipids (37, 57).
 - b. The quantity of lecithin and cephalin in polar lipid fraction was estimated by Infrared Spectrophotometry (40).
 The percent transmittance was read at 5.8 μ wavelength on Pye Unicam SP 1100 Infrared Spectrophotometer. Two standard curves, Y = 28.79 X -0.89 and Y = 23.78 X -0.94 where Y = mg pure lecithin in carbon disulfide and X = percent transmittance, were determined. The correlation coefficients for those curves were 0.9931 and 0.9983, respectively. Phospholipid quantity in total lipids was determined by method of Bartlett (4). The absorbance was read at 800 nm

on Perkin-Elmer (Coleman 124) Double Beam Spectrophotometer. The following standard curve was determined: Y = 19.231 X-0.173 where Y = mg of phosphorus and X = absorbence. The equation had a correlation coefficient of 0.9969.

- B. Lipid Analyses of Crude Deslimed Soybean Oil and Lecithin.
 - The type of lipids in crude lechithin was identified by thin layer chromatographic method described previously for soy flour lipid analysis.
 - Polar lipids, neutral lipids, and lecithin and cephalin in crude lecithin were determined by the same methods which were used to measure each respective component in soy flour lipids.
 - Phospholipid content in crude lecithin and deslimed soybean oil were determined as previously described.
- C. Moisture content of soy flours. This was determined by A.O.A.C. method (2) (Table 5).
- D. Bread Processing.

The bread formula for full fat soy flour fortified bread is shown in Table 6. For other experimental treatments, the full fat soy flour was replaced by defatted soy flour (Footnote C, Table 6), and dependent upon treatment, polar and neutral lipids were added alone, in combination or not at all. Actual quantities of soy flours and lipids added in experimental treatments are shown in Table 7. The quantity of neutral lipids (deslimed soybean oil) added in Treatments 4, 6, 7, and 9 was the

	Level of Full Fa Percent Wheat F	t Soy Flour lour Basis
Ingredients ^a	12 percent	24 percent
Wheat Flour	700	700
Water ^b	500	540 - 550
Soy Flour, Full Fat ^C	84	168
	and the second se	

Table 6. Bread Formula for Full Fat Soy Flour Fortified Bread.

^aOther ingredients were 21.0 g yeast, 14.0 g salt, 35.0 g sugar, 1.75 g yeast food, 3.5 g SSL (sodium stearoy1-2 lactylate) and 67 ppm bromate (added by addition of KBrO₂).

^bWater temperature at time of mixing was 32[°]C and quantity was adjusted to obtain optimum dough consistency.

^CFor other treatments full fat soy flour was replaced by defatted soy flour; the quantity of which was adjusted to the same quantity of lipid free, dry material added by full fat soy flour, and lipids added were dependent upon treatment.

Treatment ^b	Soy Flour	Lipids	
		Soybean Oil (Neutral)	Crude Lecithin (Polar)
		g	
1 FF	84.00		
2 TN	66.84		·
3 BN	66.12		
4 TN + NP	66.84	16.27	
5 TN + P	66.84		0.42
6 TN + NP + P	66.84	16.27	0.42
7 BN + NP	66.12	16.33	
8 BN + P	66.12		0.36
9 BN + NP + P	66.12	16,33	0.36

Table 7. Actual Quantities of Soy Flour and Lipids Added in Experimental Treatments for 12 Percent Soy Fortified Bread^a.

^aFor 24 percent soy fortified bread, the amount of soy flour and lipids in each treatment was doubled.

^bFF = full fat soy flour (PDI 15-25); TN = toasted defatted soy flour (PDI 10-25); BN = Bakers defatted soy flour (PDI 65-75); NP = neutral lipids (deslimed soybean oil); and P = polar lipids (crude lecithin).

difference in neutral lipid content between the defatted soy flours and full fat soy flour.

The baking procedure of soy fortified bread was followed:

- 1. Add all ingredients at room temperature (23°C).
- Mix dough in Hobart A-200 mixer with a bowl and dough hook at first speed for 1 minute and then at second speed to develop dough for 5 minutes.
- 3. Scale dough (dough temperature is about 26.7°C) to 500 g and round by hand. (Two 500 g loaves and a smaller loaf resulted from previous formula.)
- Relax for 40 minutes at 30°C and 85 percent relative humidity in proof cabinet.
- Roll by hand and put in greased pans of appropriate size (21.6 x 11.4 x 6.3 cm³).
- 6. Proof at 35.5°C and 92 percent relative humidity for 50 minutes in different proof cabinet. (This amount of time is sufficient to allow center of loaf to rise 3.8-5.0 cm above the center of the pan.)
- 7. Bake at 210°C oven for 25 minutes in Rotary Despatch Oven.

E. Evaluation of Soy Fortified Bread.

The bread formula (Table 6) produced two 500 g loaves (dough stage) which were used for determining the specific loaf volume of bread. Within 10 minutes after baking, the loaf volume of each loaf was determined by rape seed displacement in a loaf volume meter. The weight of each loaf was determined to the nearest 0.1 g. The mean specific loaf volume (cc/g) of the two loaves was determined for each treatment-level combination in each replication.

Two 500 g loaves (dough stage) of bread was also evaluated for crust and crumb color. The means of lightness index and dominant wavelength (nm) which can be used to represent the crust and crumb color of bread were determined for each treatment-level combination. One loaf of bread was utilized for the staling experiment (compressibility of bread slices stored at 23°C for 0, 2, 4, and 6 days). The other loaf of bread and a smaller loaf were sealed in plastic bags and were frozen for future sensory evaluation and protein and moisture analyses.

The crust color was determined at three predetermined places on top, bottom, and each long side of the loaf and at one spot centrally located on each short side of the loaf by the Hunter Color Difference Meter which had been standardized against a brown tile (L = 36.1, a =12.0 and b = 13.9). The measurements (L, a, or b) were averaged for top crust and for bottom and side crust (BSC) to obtain a single value of L, a, and b for top crust and BSC. The lightness index (Hunter L value) and dominant wavelength of top crust and BSC color were calculated (32).

Each end of the loaf was sliced with an electric knife in a slicing box. The crumb color was determined by measuring at three predetermined spots on each of these slices with the Hunter Color Difference

Meter which had been standardized against a white standard tile (standard number C2-136; L = 93.4, a = -1.10 and b = 1.90). The measurements (L, a or b) were averaged for crumb color to obtain a single value for L, a or b of crumb color for each treatment-level combination. The lightness index and dominant wavelength of crumb color were calculated (32). Total solids content of soy fortified bread (used small loaf) was determined by A.O.A.C. method (2).

For the staling experiment, a 1.25 cm slice of bread was sliced off with an electric knife in a slicing box, as quickly as the bread had cooled, and the compressibility was determined. This was the compressibility of bread at 0 day. The bread was sealed inside a plastic bag and stored at 23°C. At 2, 4, and 6 days, a 1.25 cm slice of bread was removed from the end of the loaf and discarded, a second 1.25 cm slice of bread was removed from the same end, and the compressibility of that slice was determined.

Compressibility of the bread slice was determined by compressing the top, center and bottom of the slice in a compression cell (Model AR 1859-1, Serial No. 2) of the Instron Universal Testing Instrument, Model 1132, equipped with a 5,000 g load cell. The bread slice was compressed at a speed of 10 cm/min from 12.5 to 7.5 mm by a plunger with a flat surface of 25.75 cm² area. The maximum force (g) required to compress the bread in each of three parts was divided by distance (mm) compressed and averaged to obtain a single value for compressibility in g/mm (texture).
For sensory evaluation, a 25-member untrained panel (male and female students and staff members from food-oriented departments) was randomly selected. Each panelist evaluated the samples under white fluorescent light in a separate booth in the sensory laboratory. The panel evaluated at one sitting samples representing all nine treatments of bread fortified with 12 percent full fat soy flour or its lipid free, dry matter equivalent for overall acceptability and flavor. Bread made with 24 percent full fat soy level had a specific loaf volume of approximately 4 cc/g which is not acceptable as a marketable product. Therefore, the loaf volume of 24 percent soy fortified bread should be increased before the flavor and acceptance were evaluated. A 6-point hedonic scale ranging from 1 = dislike very much to 6 = like very much was used for scoring the samples (26). The score sheet was shown in Table 8 (51).

The samples were presented by cutting each slice of 12 percent soy fortified bread for a given treatment into 3.18 cm circular pieces with a thickness of 1.25 cm leaving part of the crust attached. These pieces were kept in plastic bags until evaluated. The order of presentation of the samples in serving dishes to the panelists was randomized each time. An average score for overall acceptability and flavor of two replications of 12 percent soy fortified bread was calculated.

Crude protein content of bread fortified with 12 and 24 percent full fat soy flour or its lipid free, dry material equivalent from defatted soy flour (Treatments 1, 2, and 3) was determined in the

Table 8. Score Sheet for Sensory Evaluation.

Date	Taster .			Prod	uct <u>12</u>	% Soy	Fortif	ied Br	<u>ead</u>
Taste test sample by marking sample. Then eval Please give a reas	each sam the poin luate the son for y	ple. t that sampl our ev	Indica best e for aluati	te you descri flavor on.	r over bes yo and m	all ac ur fee ark th	c eptan ling a e scal	ce of bout t e belo	each he w.
ACCEPTABILITY	CODE	CODE	CODE	CODE	CODE	CODE	CODE	CODE	CODE
Like Very Much Like Moderately Like Slightly Dislike Slightly Dislike Moderately Dislike Very Much	y								
FLAVOR									
Like Very Much	<u> </u>								<u>.</u>
Like Moderately							<u> </u>		
Like Slightly			1. <u>1. 1. 1.</u>				· <u>· · · · ·</u> ·	<u> </u>	
Dislike Slightly	1 1 <u>1 1</u>	-77	<u></u>					<u></u>	
Dislike Moderatel	у	<u> </u>	(LAI)	(<u>cc</u>)	4 <u>44</u>		(n) tr	Critica Contraction	
Dislike Very Much			<u>.</u>		1	<u>22 -</u>			
			Reas	son for	Each	Sample	e Evalu	ation	

following manner: Nitrogen content of the bread was determined by the semimicro modification of nesslerization method (38). The absorbence was read at 420 nm in Perkin-Elmer (Coleman 124) Double Beam Spectrophotometer. Two standard curves, $Y = 40.000 \times -0.464$ and Y =40.650 \times -0.337 where Y = mg of nitrogen and X = absorbence, were determined. The equations had correlation coefficients of 0.9994 and 0.9997, respectively. Crude protein content was calculated by multiplying percent nitrogen of the bread sample by the conversation factor 5.7.

CHAPTER IV

RESULTS AND DISCUSSION

I. LIPID ANALYSES AND COMPOSITION IN TREATMENTS

The lipid quantity and composition of full fat soy flour (Nutrisoy 220, PDI 15-25), defatted soy flours (Toasted Nutrisoy, PDI 10-25, and Bakers Nutrisoy, PDI 65-75, lipid composition of crude lecithin (source of polar lipids) and phospholipid content of deslimed soybean oil (source of neutral lipids) are shown in Table 9. On as is basis, Full fat soy flour contained 21.85 percent total lipids which were composed of 91.97 percent neutral lipids and 8.03 polar lipids. Toasted Nutrisoy flour contained 2.90 percent total lipids which was composed of 36.17 percent neutral lipids and 63.83 percent polar lipids. Bakers Nutrisoy contained 2.89 percent total lipids which was composed of 35.36 percent neutral and 64.64 percent polar lipids. It could be calculated that full fat soy flour contained 20.08 percent neutral lipids and 1.77 percent polarlipids and defatted soy flours averaged together had 1.04 percent neutral and 1.86 percent polar lipids. This indicates that the main difference of lipid content between full fat soy flour and defatted soy flour was neutral lipids.

Figure 1 shows the thin layer chromatogram of polar soy flour lipids and crude lecithin which was developed by the chloroformmethanol-water 65:35:4 v/v/v solvent system. Lipids A, B, and C

Table 9. Analysis of Soy Flours, Crude Lecithin, and Soybean Oil.

Source	Total Lipids	Neutral Lipids	Polar Lipids	Phospholipids	Lecithin and Cephalin
	Percent		Percent	of Total Lipids	
rn ^b	2.90	36.17	63.83	50.26	43.93
BN ^c	2.89	35.36	64.64	52.67	39.31
Pad	21.85	91.97	8.03	6.49	4.80
Crude Lecithin	1	44.08	55.92	51.34	44.92
Soybean Oil		:		0.42	

^aAs is basis.

broasted Nutrisoy, defatted soy flour with PDI 10-25.

^CBakers Nutrisoy, defatted soy flour with PDI 65-75.

dNutrisoy 220, full fat soy flour with PDI 15-25.



Figure 1. Thin Layer Chromatogram of Polar Soy Flour Lipids and Crude Lecithin.

- BN. Bakers Nutrisoy, PDI 65-75 defatted soy flour.
- TN. Toasted Nutrisoy, PDI 10-25 defatted soy flour.
- CL. Crude Lecithin
- FF. Nutrisoy 220, PDI 15-25 full fat soy flour.
- A. Cephalin.
- B. Phosphatidial inositol.
- C. Glycolipids.

reacted positively to the molybdenum blue test which indicated they contained phosphorus, and A, B, and C had the same r_f values as cephalin, lecithin, and phosphatidyl inositol, respectively. All lipids indicated by D reacted positively to the diphenyl amine test which indicated they were glycolipids.

Crude lecithin (Column L, Figure 1) had less glycolipid quantity than polar lipids extracted from the soy flours. Data in Table 9 allow an estimation of amount of glycolipids present in each lipid analyzed. Crude lecithin contained 55.92 percent polar lipids. Phospholipids (51.34 percent of total lipids) composed 91.81 percent of the polar lipids in the crude lecithin and glycolipids, 8.19 percent. Lipids extracted from defatted soy flour PDI 10-25 (Toasted Nutrisoy, TN), defatted soy flour PDI 65-75 (Bakers Nutrisoy, BN), and full fat soy flour PDI 15-25 (Nutrisoy 220, FF) contained 63.83, 64.64, and 8.03 percent polar lipids, respectively (Table 9). The polar lipids from soy flours: TN, BN, and FF were composed of 78.74, 81.48, and 80.82 percent phospholipids, and 21.26, 18.52, and 19.18 percent glycolipids, respectively. Therefore, the polar lipids extracted from any soy flour contained approximately 2.4 times the quantity of glycolipids present in the crude lecithin.

An estimation of the quantity of phosphatidyl inositol present in lipids may also be made since the quantity of lecithin and cephalin was analyzed (Table 9), and the phospholipids present were chiefly lecithin, cephalin, and phosphatidyl inositol (Figure 1). The amount of lecithin and cephalin extracted from TN, BN, FF, and crude lecithin

was 87.41, 74.62, 73.96, and 87.50 percent of the phospholipids, respectively. The amount of phosphatidyl inositol in each case would be the difference between 100 percent and the percentage of phospholipids that is lecithin and cephalin.

Some differences between composition of polar lipids present in full fat soy flour and crude lecithin did exist. However, it was found that only small quantities of crude lecithin were required in Treatments 5, 6, 8, and 9 (Table 7, p. 22) for the level of phospholipids to be equal to phospholipid content of Treatment 1. It was felt that to add crude lecithin on the basis of phospholipid content to Treatments 5 and 8 gave as good a match on polar lipid composition of defatted soy flour with polar lipids in full fat soy flour as could be achieved. Table 10 shows the components of polar lipids in Treatments 5 and 8 compared with Treatment 1 for the 12 percent full fat soy flour fortification or its lipid free, dry material equivalent in bread.

In Treatments 4, 6, 7, and 9, deslimed soybean oil was added to defatted soy flours to bring the level of neutral lipids to the level of neutral lipids in Treatment 1. This crude oil contained 0.42 percent phospholipids (Table 9). This phospholipid content was considered negligible since only 0.068 g of phospholipids were added to Treatments 4, 6, 7, and 9 at the 12 percent full fat soy flour equivalent.

II. QUALITY CHARACTERISTICS OF SOY FORTIFIED BREAD

Table 11 shows the sum of squares from analysis of variance for both levels of soy fortified bread (12 and 24 percent full fat soy

Quantities of Polar Lipid Components in Different Treatments at 12 Percent Full Fat Soy Flour Level in Bread. Table 10.

				Lecithin and	Phosphatidial
Treatment	Polar Lipids	Glycolipids	Phospholipids 8	Cephalin	Inositol
18	1.4738	0.2827	1.1912	0.8810	0.3102
2p	1.4721	0.2823	1.1898	1.0401	0.1496
8c	1.4365	0.2452	1.1913	0.9129	0.2785

^aFull fat soy flour, PDI 15-25.

^bDefatted soy flour, PDI 10-25, and added by crude lecithin.

^cDefatted soy flour, PDI 65-75, and added by crude lecithin.

Sums of Squares for Quality Characteristics of Soy Fortified Bread. Table 11.

					Sums of	Squares	8		y my
Source	Degrees of Freedom	Specific Loaf Volume	Total Solids Content	Top Crust	Shtness Ind Bottom and Side Crust	Crumb	Domi Top Crust	nant Waveler Bottom and Side Crust	lgth Crumb
Level (L)	1	14.5542	41.8825	22.8803	607, 6225	89.3025	2.5069	48.0711	3.0625
Replication (R)	1	0.0793	0.0568	6.8469	19.2136	2.8336	64.8025	7.4711	0.0025
Error for L and R	1	0.1100	0.6861	8.7025	11.6736	1.6469	70.2803	6.9344	0.2025
Treatment (T)	œ	2.0065 ^a	15.5103 ^a	19.3956	19.4106	19.5800 ^b	84.4789	13.4500	6.1550 ^b
60000000000000000000000000000000000000		0.7896 ^a 0.2838 ^b 0.0000 0.0798 0.5148 ^a 0.1208 ^c 0.1208 ^c 0.2174 ^c	4.2973 ^a 0.8740 ^b 0.5513 ^c 0.7268 ^c 5.8685 ^a 0.0018 0.1152 3.0755 ^a	0.2335 1.8150 8.8200 3.0625 2.5600 2.8900 0.0013 0.0133	0.3335 6.3038 6.3038 6.3038 0.4225 0.4225 0.025 0.025 0.2113 1.7633	1.8050 ^b 4.2504 ^b 7.8013 ^a 3.3306 0.1806 0.1806 2.1756 0.0013 0.0352	29.7735 8.1667 0.8450 0.1406 7.9806 2.9756 2.9756 2.4200 32.1769	2.1013 0.0338 0.6613 0.0756 2.9756 1.2656 0.2450 6.0919 ^c	0.9113 ^c 0.1667 2.4200 ^a 0.2756 ^c 0.8556 ^c 0.0056 ^b 1.4450 ^b 0.0752
T*T	80	0.2300	0.4590	3.8422	40.3350 ^b	5.5400	119.1456	7.6689	0.8050
Error for T and T*L	16	0.4261	1.4626	21.4956	18.0478	7.4644	117.3822	16.7544	1.9800

^aMeans significant difference at the P < 0.001 level.

Table 11 (continued)

 b Means significant difference at the P $\angle 0.01$ level.

^cMeans significant difference at the $P \prec 0.05$ level.

 d_{C} = comparison; these comparisons are defined in Table 3.

flour, FF, or its lipid free, dry material equivalent) and for the following variables: specific loaf volume, total solids, and crust and crumb color. Table 12 shows the sums of squares from analysis of variance for sensory evaluation of flavor and overall acceptability of bread fortified with 12 percent FF level or its lipid free, dry material equivalent. The only significant difference found between replications was for overall acceptability. Tables 13 and 14 show the treatment means for the dependent variables listed above.

A. Specific Loaf Volume

A difference was found for specific loaf volume between bread fortified with 12 and 24 percent FF or its lipid free, dry material equivalent that approached significance (P = 0.0547) at the P<0.05 level (Table 11, page 35, and Figure 2, page 41). The experimental design for this experiment did not give a sensitive test for soy flour level since the error mean square for testing level had only one degree of freedom (Table 11). The means of specific loaf volume (Table 13, page 39) show that 12 percent FF or its lipid free, dry material equivalent soy fortified bread had greater loaf volume than 24 percent FF or its lipid free, dry material equivalent soy fortified bread (Figure 2). There are obviously significant differences among the nine different treatments (Table 11 and Figure 2).

The addition of either polar or neutral lipids to defatted soy flour increased the specific loaf volume of bread significantly when compared to bread made with defatted soy flour alone (C8, Table 11, and Table 3, page 14; specific loaf volume means T4, T5, T7, and T8

Source	Degrees of Freedom	Flavor	Overal1 Acceptability
Replication (R)	1	0.1369	0.3147 ^c
Treatment (T)	8	1.5559 [°]	1.2959 ^c
Cld	1	0.1820	0.1965
C2 ^d	1	0.0954	0.0432
C3 ^d	1	0.1406	0.1936
C4 ^d	1	0.3362 ^c	0.4325 ^c
C5 ^d	1	0.0338	0.2205
C6 ^d	1	0.4418 ^c	0.1301
C7 ^d	1	0.1849	0.2500 ^c
C8 ^d	1	0.1411	0.0280
Error for T	8	0.3633	0.3311

Table 12. Sums of Squares for Flavor and Overall Acceptability of Bread Fortified with 12 Percent Soy Flour.

^cMeans significant difference at the P ≤ 0.05 level.

 d C = comparison; these comparisons are defined in Table 3.

Table 13. Means of Quality Characteristics of Soy Fortified Bread

576.00 577.20 576.05 576.40 576.85 576.80 575.95 576.38 576.55 575.65 24% Crumb 576.10 575.40 575.90 576.20 575.80 575.85 576.40 575.85 575.15 575.35 127 Dominant Wavelength (nm) Bottom and 585.70 586.00 584.95 585.35 587.05 586.05 585.10 586.05 587.20 585.94 Side Crust 12% 24% 582.15 582.50 583.30 583.63 584.40 584.40 584.05 583.70 584.45 583.70 583.40 588.90 588.00 588.35 589.55 584.65 587.35 593.05 588.14 584.80 587.35 588.65 Top Crust 12% 24% 585.65 585.30 589.30 589.50 589.85 591.10 587.10 587.62 72.70 71.40 71.00 73.45 70.45 70.90 70.45 70.15 71.20 71.30 24% Crumb 74.40 74.45 74.25 74.65 75.75 74.90 73.90 73.90 73.85 74.47 127 Lightness Index 41.30 42.70 40.50 42.40 42.20 41.35 42.05 50.14 41.93 42.55 42.30 Side Crust 12% 24% **BOLLOM and** 50.75 52.15 49.05 49.25 53.05 50.05 50.75 49.20 47.05 30.30 31.75 30.70 30.05 30.40 29.15 32.00 31.60 30.50 30.65 30.68 Top Crust 12% 24% 30.70 33.35 31.90 31.65 32.25 32.50 33.65 31.45 31.16 62.05 60.03 60.37 61.67 62.05 60.40 63.26 61.13 Total Solids 61.25 61.54 63.03 60.59 12% 24% Content 3 63.34 62.60 63.95 64.21 62.36 63.53 62.71 63.65 4.05 4.12 3.88 3.70 4.08 3.92 4.26 4.44 3.77 4.28 Loaf Volume 24% Specific (cc/g) 5.02 5.26 5.73 5.32 5.24 5.18 5.05 5.75 5.53 5.13 12% Soy Level Treatment Means Overal1 -00 0 N

Treatment (T)	Flavor Score ^a	Acceptability Score ^a
1	4.70	4.74
2	4.37	4.58
3	3.94	4.08
4	4.76	4.74
5	4.42	4.59
6	4.62	4.78
7	3,88	4.02
8	4.48	4.38
9	4.25	4.34
Overall means	4.38	4.47

Table	14.	Means of Flavor and Overall Acceptability
		of Bread Fortified with 12 Percent Soy
		Flour.

^aThe six point hedonic scale ranged from 1 = dislike very much, 2 = dislike moderately, 3 = dislike slightly, 4 = like slightly, 5 = like moderately, and 6 = like very much.



- Fortified Bread (12 and 24 Percent Full Fat Soy Flour or Its Lipid Free, Dry Matter, Equivalent) Made with Soy Flour of Varying Protein Dispersibility Indices and With or Without Added Lipids. Figure 2.
- Toasted Nutrisoy (defatted soy flour, PDI 10-25). IN.
 - Bakers Nutrisoy (defatted soy flour, PDI 65-75).
 - Nutrisoy 220 (full fat soy flour, PDI 15-25).
 - Polar lipids (crude lecithin). FF. P.
- Neutral lipids (deslimed soybean oil).

versus T2 and T3, Table 13). However, the addition of neutral lipids to defatted soy flour increased the specific loaf volume of bread more than the addition of polar lipids (C5, Table 11 and Table 3; specific loaf volume means T4 and T7 versus T5 and T8, Table 13). This means that the addition of neutral lipids to defatted soy flour was the chief cause of the difference between defatted soy flour alone and defatted soy flour with the addition of neutral or polar lipids. The addition of a combination of neutral and polar lipids to defatted soy flour and addition full fat soy flour alone gave bread with a significantly greater loaf volume than defatted soy flour alone or with the addition of neutral or polar lipids (C1, Table 11 and Table 3; specific loaf volume means T1, T6, and T9 versus T2, T3, T4, T5, T7, and T8, Table 13). The addition of the combined neutral and polar lipids to defatted soy flour increased the specific loaf volume of bread significantly more than bread made with full fat soy flour (C2, Table 11 and Table 3; specific loaf volume means T6 and T9 versus T1, Table 13). This means that the addition of a combination of neutral and polar lipids to defatted soy flour gave bread with the highest specific loaf volume of all treatments in this investigation.

As has been shown previously (1) inclusion of lipids in a lean bread formula gave greater loaf volume to soy fortified bread. Tsen and Hoover (62) reported that SSL had a shortening sparing effect. However, the results of this investigation show that neutral lipids in combination with SSL gave significantly greater volume to soy fortified bread. The addition of lecithin, another surface active agent, to

SSL and neutral lipids gave an even greater loaf volume in soy fortified bread. The interactions of SSL and lecithin with the different protein in soy fortified bread and neutral lipids have not been investigated. It is, therefore, impossible to even project what might be causing these significant differences.

The PDI of defatted soy flour did not have any significant effect on specific loaf volume of bread fortified at 12 and 24 percent FF or its lipid free, dry material equivalent level in this investigation. No significant differences were found between defatted soy flours with PDI 10-25 and PDI 65-75 when added to bread alone, with a single source of lipids (neutral or polar) or with a combination of neutral and polar lipids (C7, C4, and C3, respectively, Table 11 and Table 3).

A significant interaction between PDI of defatted soy flour and the addition of a single source of lipids to defatted soy flour was found for specific loaf volume of bread (C6, Table 11 and Table 3). From the specific loaf volume means (Table 13), the addition of neutral lipids to defatted soy flour with PDI 65-75 (T7) caused bread with the highest specific loaf volume when compared to T4, T5, or T8 (Figure 2).

B. Total Solids Content (Moisture Content)

The means of total solids content (Table 13, page 39) show that 12 percent FF or its lipid free, dry matter equivalent soy fortified bread had higher total solids (less moisture) than 24 percent FF or its lipid free, dry matter equivalent soy fortified bread. There were

significant differences in total solids content among nine treatments at the $P \le 0.001$ level (Table 11, page 35).

The addition of either neutral or polar lipids to defatted soy flour increased total solids content of bread when compared to bread made with defatted soy flour alone (C8, Table 11 and Table 3, page 14; total solids means T4, T5, T7, and T8 versus T2 and T3, Table 13). The addition of neutral lipids to defatted soy flour increased the total solids content of bread more than the addition of polar lipids (C5, Table 11 and Table 3; total solids means T4 and T7 versus T5 and T8, Table 13). This indicates that the addition of neutral lipids to bread fortified with defatted soy flour was the main reason it contained more solids than bread made with defatted soy flour alone or with added polar lipids. The addition to bread of a combination of neutral and polar lipids and defatted soy flour or the full fat soy flour caused bread to have significantly greater total solids than bread made with defatted soy flour alone and with an added single source of lipids (neutral or polar) (C1, Table 11 and Table 3; total solids means T1, T6, and T9 versus T2, T3, T4, T5, T7, and T8, Table 13). The addition of the combined neutral and polar lipids to defatted soy flour fortified bread made the total solids content of the bread significantly higher than bread made with full fat soy flour (C2, Table 11 and Table 3; total solids means T6 and T9 versus T1, Table 13). The bread in the treatment where combined neutral and polar lipids were added to defatted soy flour had the least moisture of any bread in this experiment.

Bread containing PDI 65-75 defatted soy flour and a single lipid source (neutral or polar) contained significantly less moisture (more total solids) than bread containing PDI 10-25 defatted soy flour and the same lipids (C4, Table 11 and total solids means T4 and T5 versus T7 and T8, Table 13). Bread with PDI 65-75 defatted soy flour and combined lipid sources also had significantly less moisture than bread with PDI 10-25 defatted soy flour and combined lipid sources (C3, Table 11 and total solids means T6 versus T9, Table 13). The PDI of soy flour did not significantly affect total solids of bread made with defatted soy flour alone (C7, Table 11). These findings indicate some type of interaction occurred between added lipids and the water soluble proteins causing less moisture in the resultant bread. No significant interaction was found for total solids content of bread between added lipids and the PDI of defatted soy flour (C6, Table 11).

C. Lightness Index of Bread Color

1. Top crust of bread

The means of lightness index of top crust color (Table 13, page 39) show that 12 percent FF or its lipid free, dry material equivalent soy fortified bread had higher lightness index (lighter) than 24 percent FF or its lipid free, dry material equivalent soy fortified bread. Only one significant comparison for differences among treatments was found at the $P \leq 0.05$ level (C3, Table 11, page 35, and Table 3, page 14). Bread made with PDI 10-25 defatted soy flour (Toasted Nutrisoy,

TN) and combined neutral and polar lipids had a higher lightness index (lighter) for top crust than bread made with PDI 65-75 defatted soy flour (Bakers Nutrisoy, BN) and the combined lipids (lightness index means, T9 versus T6, Table 13).

2. Bottom and side crust color of bread

The means of lightness index of bottom and side crust (BSC) color (Table 13, page 39) show that 12 percent FF or its lipid free, dry material equivalent soy fortified bread had much higher lightness index (lighter) than 24 percent FF or its lipid free, dry material equivalent soy fortified bread. Two significant treatment comparisons (C3 and C4, Table 11, page 35, and Table 3, page 14) at the P<0.05 level and a significant interaction (T x L) for the lightness index of BSC were found (Table 11).

The addition of combined neutral and polar lipids to defatted soy flour in bread caused a significantly higher lightness index (lighter) in BSC than the addition of full fat soy flour (C2, Table 11 and lightness index means T6 and T9 versus T1, Table 13). PDI 10-25 defatted soy flour (TN) with combined lipids also caused bread to have a significantly higher lightness index (lighter) of BSC than PDI 65-75 defatted soy flour (BN) and combined lipids (C3, Table 11 and lightness index means T6 versus T9, Table 13). No other comparisons showed significant differences for the lightness index of BSC (Table 11).

3. Crumb color of bread

The means of lightness index of crumb color (Table 13, page 39) show that 12 percent FF or its lipid free, dry material equivalent soy fortified bread had higher lightness index (lighter) than 24 percent FF or its lipid free, dry material equivalent soy fortified bread. A significant effect was found for the lightness index of bread crumbs among treatments at the P<0.01 level (Table 11, page 35).

The addition of a combination of neutral and polar lipids to defatted soy flour or full fat soy flour alone caused bread to have a significantly higher crumb lightness index (lighter) than defatted soy flour alone or in combination with a single source of lipids (neutral or polar) (C1, Table 11 and Table 3, page 14; and lightness index means T1, T6 and T9 versus T2, T3, T4, T5, T7, and T8, Table 13). The addition of the combination of neutral and polar lipids to defatted soy flour gave bread with higher lightness index (lighter) than full fat soy flour (C2, Table 11 and lightness index means T6 and T9 versus T1, Table 13). On the other hand, PDI 65-75 defatted soy flour (BN) caused lighter crumb color in bread (higher lightness index) than PDI 10-25 defatted soy flour (TN) when added with a combination or a single source of neutral or polar lipids (C3 and C4, Table 11 and lightness index means T9 versus T6; and T7 and T8 versus T4 and T5, Table 13). This indicates that the addition of a combination of neutral and polar lipids to PDI 65-75

defatted soy flour (T9, Table 13) caused bread to have the lightest crumb color of any treatment in this investigation.

A significant interaction was found between the PDI of defatted soy flour and defatted soy flour with the addition of a single source of lipids (neutral or polar) (C6, Table 11). No significant effects on the lightness index of bread crumb were found for C5, C7, and C8 at the P<0.05 level (Table 11).

D. Dominant Wavelength of Bread Color

1. Top crust color of bread

2.

No significant effects on the dominant wavelength of top crust were found either among treatments or between the two levels (12 and 24 percent FF soy lipid free, dry material equivalent) of soy fortified bread at the P<0.05 level (Table 11). Bottom and side crust (BSC) color of bread

The means of dominant wavelength of BSC color (Table 13, page 39) show that 12 percent FF or its lipid free, dry material equivalent soy fortified bread had lower dominant wavelength (less brown) than 24 percent FF or its lipid free, dry material equivalent soy fortified bread. Only one treatment comparison was found to be significant for dominant wavelength of BSC at the P \angle 0.05 level (Table 11, page 35). Bread made with the addition of polar or neutral lipids to defatted soy flour had higher dominant wavelength of BSC than bread made with defatted soy flour alone (C8, Table 11 and Table 3, page 14; and dominant wavelength means T4, T5, T7, and T8 versus T2 and T3, Table 13).

3. Crumb color of bread

The means of dominant wavelength of crumb color (Table 13, page 39) show that 12 percent FF or its lipid free, dry material equivalent soy fortified bread had lower dominant wavelength (less yellow) than 24 percent FF or its lipid free, dry material equivalent soy fortified bread. A significant difference for the dominant wavelength of crumb was found among the treatments at the P<0.01 level (Table 11, page 35).

The addition of combined neutral and polar lipids to defatted soy flour or full fat soy flour alone in bread gave it a lower dominant wavelength for crumb color than defatted soy flour alone or in combination with a single source of lipids (neutral or polar) (C1, Table 11, and Table 3, page 14; and crumb dominant wavelength means T1, T6, and T9 versus T2, T3, T4, T5, T7, and T8, Table 13). On the other hand, bread made with PDI 65-75 defatted soy flour (BN) and combined neutral and polar lipids had lower dominant wavelength for crumb color than bread made with PDI 10-25 defatted soy flour (TN) and the combined lipids (C3, Table 11 and crumb dominant wavelength means T9 versus T6, Table 13). The addition of the combined lipids to PDI 65-75 defatted soy flour gave bread the least yellowish crumb color (the lowest dominant wavelength) of all nine treatments at 12 and 24 percent soy level in this investigation. The addition of neutral lipids to defatted soy flour gave bread with lower crumb dominant wavelength (less yellow) than the addition of polar lipids to defatted soy flour (C5, Table 11 and crumb dominant wavelength means T4 and T7 versus T5 and T8, Table 13). For defatted soy flour alone without any lipid addition, PDI 65-75 defatted soy flour gave bread with lower crumb dominant wavelength (less yellow) than PDI 10-25 defatted soy flour (C7, Table 11 and crumb dominant wavelength means T3 versus T2, Table 13).

E. Flavor of 12 Percent Soy Fortified Bread

No significant difference on the flavor of 12 percent soy fortified bread was found between replications. Significant effects on the flavor of 12 percent soy fortified bread (FF or its lipid free, dry material equivalent) were found among nine treatments at the P < 0.05level (Table 12, page 38).

Flavor of bread containing toasted defatted soy flour PDI 10-25 (TN) in combination with a single lipid source (polar or neutral) was liked significantly more than that of bread containing defatted soy flour PDI 65-75 (BN) (C4, Table 12 and Table 3, page 14; and flavor score means T4 and T5 versus T7 and T8, Table 14, page 40). The flavor score (4.37) of bread containing PDI 10-25 defatted soy flour alone approached a significant difference (P = 0.0783; C7, Table 12) over the flavor score (3.94) of bread containing only PDI 65-75 defatted soy flour.

A significant interaction for flavor score between PDI and type of single lipid source added was also found (C6, Table 12). Addition of neutral lipids to PDI 10-25 (TN) defatted soy flour gave bread a higher flavor score than addition of polar lipids (T4 and T5, Table 14). However, addition of neutral lipids to PDI 65-75 defatted soy flour (BN) gave bread with a much lower flavor score than addition of polar lipids (T7 and T8, Table 14). No significant differences in flavor score were found between T6 (TN, combined lipid sources) and T9 (BN, combined lipid sources) at the P<0.05 level (C3, Table 12). Comparison flavor scores of bread containing defatted soy flour (TN and BN) alone or combined with a single lipid source with bread containing defatted soy flour and combined lipid sources or full fat soy flour was significant at the P = 0.08 level (C1, Table 12).

Except for T3 and T7, all treatments had acceptable bread flavor. The flavor score means for these treatments were between "like slightly" and "like moderately" (Table 8, page 27, and Table 14, page 40).

F. Overall Acceptability of 12 Percent Soy Fortified Bread

The significant differences for overall acceptability of 12 percent soy fortified bread were found between replications and among nine treatments at the P<0.05 level (Table 12, page 38).

Bread made with PDI 10-25 defatted soy flour (TN) alone had a significantly higher acceptability score than bread made with PDI 65-75 defatted soy flour (BN) alone (C7, Table 12 and Table 3, page 14; and acceptability score means T2 versus T3, Table 14, page 40). Also bread

made with TN flour and a single lipid source (neutral or polar) was more acceptable than bread made with BN flour and a single lipid source (C4, Table 12 and acceptability score means T4 and T5 versus T7 and T8, Table 14). Acceptability of bread made with TN flour and combined lipids was (significant at the P< 0.08 level) greater than that of bread made with BN flour and combined lipids. Also C1 (Table 12) was at the P = 0.061 significant difference level indicating bread with FF soy flour alone or with BN and TN in combination with both lipid sources was more acceptable than bread made in other treatments.

All of the breads fortified with 12 percent soy flour (FF or its lipid free, dry material equivalent) had overall acceptability between "like slightly" and "like moderately" (Table 8, page 27, and Table 14, page 40).

G. Compressibility of Soy Fortified Bread During Storage

Compressibility increased at a decreasing rate with increasing storage time for each treatment of 12 and 24 percent soy fortified bread (or its full fat soy flour lipid free, dry matter equivalent). This means that bread became firmer with increasing storage time from 0 to 6 days at 23°C. Twenty-four percent soy fortified bread was firmer and showed more differences in compressibility as a function of time among the nine treatments than 12 percent soy fortified bread (Figures 3, 4, 5, and 6). Significant effects were found for soy levels (12 and 24 percent FF or its lipid free, dry material equivalent), treatment storage time (0, 2, 4, and 6 days), treatment x level interaction,

storage time x level interaction, and storage time x treatment interaction (Table 15). No significant difference was found between replications (Table 15). The compressibility means are shown in the Appendix for each soy level, treatment and storage time combination, and the equations of compressibility as function of storage time for each treatment and soy flour level.

Figures 3 and 4 show the compressibility as a function of storage time at 23°C for all treatments of 12 percent soy fortified bread. Bread made with the addition of a combination of neutral and polar lipids to (TN) PDI 10-25 defatted soy flour (Treatment 6) was the softest (had lowest compressibility) of all treatments throughout storage at 23°C. The addition of a combination of lipids to (BN) PDI 65-75 defatted soy flour (Treatment 9) or the addition of neutral lipids to PDI 65-75 defatted soy flour (Treatment 7) made bread softer than either defatted soy flour alone (PDI 10-25 or PDI 65-75, Treatments 2 and 3), the addition of polar lipids to either defatted soy flour (PDI 10-25 or 65-75, Treatments 5 and 8), the addition of neutral lipids to PDI 10-25 defatted soy flour (Treatment 4), or full fat soy flour (Treatment 1) at 2, 4, and 6 days storage. This indicates that the addition of a combination of lipids to defatted soy flour (either PDI 10-25 or PDI 65-75) caused 12 percent soy fortified bread to be softer than the others when the bread was stored at 23°C up to six days.

For all treatments of 24 percent soy fortified bread, Figures 5 and 6, show the compressibility as a function of storage time at 23°C. Bread made with the addition of a combination of neutral and polar

Source	Degrees of Freedom	Sums of Squares	Mean Squares
Replication (R)	1	6222.84	6222.84
Level (L)	i	1113401.32	1113401.32 ^c
Error for R and L	1	698.02	698.02
Treatment (T)	8	419762.07	52470.26 ^b
T * L	8	237133.79	29641.72 ^d
Error for T and T*L	16	193621.85	12101.37
Storage Time (S)	3	1103719.42	367906.47 ^a
S * L	3	30500.53	10166.84 ^a
S * T	24	70407.35	2933.64 ^b
S * L * T	24	30226.29	1259.43
Error for S S*L S*T and S*L*T	54	58453.56	1082.47

Table 15. Analysis of Variance for Compressibility of Soy Fortified Bread during Storage.

^aMeans significant difference at the P \leq 0.001 level. ^bMeans significant difference at the P<0.01 level. ^cMeans significant difference at the P<0.05 level. ^dMeans significant difference at the P<0.07 level.



MEAN COMPRESSIBILITY, (g/mm)









lipids to defatted soy flour (Treatments 6 and 9) or bread made with the addition of neutral lipids to defatted soy flour (Treatments 4 and 7) was softer than the other treatments at 0, 2, 4, and 6 days storage time. The addition of polar lipids to defatted soy flour (Treatments 5 and 8) caused bread to be softer than bread with defatted soy flour alone (Treatments 2 and 3). Bread made with full fat soy flour (Treatment 1) was softer than bread of Treatments 2, 3, 5, or 8 at four and six days storage time. The compressibility means (Appendix, 24 percent full fat soy flour or its lipid free, dry matter equivalent) show that bread made with PDI 10-25 defatted soy flour (TN) was softer than bread made with PDI 65-75 defatted soy flour (BN) (i.e., Bread of treatments 2, 4, 5, and 6 was softer than bread of treatments 3, 7, 8, and 9, respectively) at 2, 4, and 6 days storage.

Mold grew on 24 percent soy fortified bread at 23^oC during the storage of four and six days. Therefore, further work should be conducted by addition of mold inhibitor to soy fortified bread in order that bread can be stored longer and still keep good qualities for commercial purposes.

H. Crude Protein Content

No significant difference in crude protein content of bread fortified with the same level (12 or 24 percent full fat soy flour or its lipid free, dry matter equivalent) of soy flour was found among Treatments 1, 2, or 3 at the P<0.01 level (Table 16). Crude protein content of bread for treatments 4, 5, 6, 7, 8, and 9 were not determined

	Level of Full Fat Soy Flour ^a			
Treatment	12%	24%		
	%, Dry Matter Basis			
1	17.64 ^b	20.76 [°]		
2	17.69 ^b	21.38 ^c		
3	18.34 ^b	21.34 ^c		

Table 16. Crude Protein Content of Bread Fortified with Soy Flour.

^aIn treatments 2 and 3, defatted soy flour replaced full fat soy flour. The quantity of defatted soy flour was adjusted to the equivalent amount of soy protein which was added by treatment 1 (full fat soy flour).

^{bc}Means in row or column bearing like superscript letters are not significantly different at the P<0.01 level. because the same quantity of toasted defatted soy flour (Treatment 2) was added in treatments 4, 5, and 6, and the same quantity of bakers defatted soy flour (Treatment 3) was added in treatments 7, 8, and 9 (Table 7, p. 22). This means all nine treatments at one level of soy fortified bread should have contained the same amount of protein. On the other hand, a significant difference was found between 12 and 24 percent full fat soy levels of bread (Table 16).

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CHAPTER V

CONCLUSION

Added lipids (neutral and/or polar) influenced loaf volume of soy supplemented bread which was made by the K-State Process. When lipids were added to bread fortified with defatted soy flour (12 or 24 percent full fat soy flour or its lipid free, dry material equivalent), neutral lipids caused greater loaf volume than polar lipids. The addition of combined polar and neutral lipids to toasted (TN) or bakers (BN) defatted soy flour gave bread with the significantly greatest loaf volume of all treatments. Protein dispersibility index (PDI) of soy flour, TN (PDI 10-25) or BN (PDI 65-75), did not affect the specific loaf volume significantly. Therefore, the difference between loaf volumes of bread fortified with full fat and with defatted soy flour reported by Tsen and Hoover (64), was probably due to the lipid content rather than protein quality, particularly because of the large quantity of neutral lipids present in the full fat soy flour. The previous observation (37) mentioned that nitrogen dispersibility index (NSI) of defatted soy flour had an influence on loaf volume of soy fortified bread made with sodium stearoy1-2 lactylate. However, the defatted soy flours used, NSI 77 and 20, were obtained from two sources, Swifts Edible Oil Company and Central Soya Company, respectively. This indicates the defatted soy flours from different

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suppliers probably were the cause of the difference rather than NSI of defatted soy flour.

Bread made with added neutral lipids to defatted soy flour contained more solids (less moisture) than bread made with added polar or no lipids to defatted soy flour. The addition of combined neutral and polar lipids to defatted soy flour also gave bread with the significantly higher level of solids (least moisture) of all treatments. Bread made with a single source (neutral or polar lipids) or combined lipids and PDI 65-75 defatted soy flour had significantly more solids than bread made with PDI 10-25 defatted soy flour and a single source (neutral or polar lipids) or combined lipids.

In treatments where combined lipids were added, PDI 65-75 defatted soy flour gave bread a lighter crust color than PDI 10-25 defatted soy flour. PDI 65-75 defatted soy flour also gave bread lighter and less yellowish crumb color than PDI 10-25 defatted soy flour in treatments where combined lipids were added. These results agree with the report of Fellers et al. (12) that TN caused bread to be darker than BN. Neutral lipids caused the crumb color of bread to be less yellowish than polar lipids, and combined lipids caused bread crumb color to be lighter and less yellowish than either neutral, polar, or no added lipids.

Fellers et al. (12) reported that a lightly heated product (PDI 65-75 equivalent) was used in a 12 percent defatted soy-wheat flour blend (K-State blend) for production of soy fortified bread in U.S.A. overseas aid programs. Sensory evaluation of 12 percent soy fortified bread in our investigation showed that our sensory panel which was composed of United States citizens liked the flavor and accepted better the bread fortified with TN than bread fortified with BN. These results indicate that more research should be done with defatted soy flours from other sources before a recommendation that BN be the defatted soy flour used for fortification of white bread in the United States. Combined lipids caused 12 percent soy fortified bread to be more acceptable and have a flavor which was liked better than other lipid treatments in this investigation, however, the differences among scores were not large. The flavor and acceptability score for all treatments was between "like slightly" and "like moderately."

Soy fortified bread became firmer with increasing storage time at 23°C from 0 to 6 days. Twenty-four percent soy fortified bread was firmer and showed greater differences in compressibility as a function of time than 12 percent soy fortified bread. Neutral lipids made bread softer than polar lipids. Bread made with the addition of combined lipids to defatted soy flour was the softest of all treatments. At the same lipid content treatments, PDI 10-25 defatted soy flour (TN) caused 24 percent soy fortified bread to be softer than PDI 65-75 defatted soy flour (BN) at 2, 4, and 6 days storage.

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APPENDIX

Treatment	Soy Level (%)	Storage Time (days)	Compressibility Means (g/mm)	Equation	
		0	110 160	V-370 4038+251 021300.3080T	
1	12	0	110.100	1-370.4930+231.0213e	
		2	241.085		
		4	287.905		
		6	335.440	W. 260 1/21/228 5061 0.2857T	
2	12	0	129.365	Y=369.1421+238.3961e	
		2	239.760		
		4	285.160		
		6	329.870	0.1065T	
3	12	0	111.755	Y=642.6022+530.4543e	
		2	215.265		
		4	294.560		
		6	363.195	0,2088T	
4	12	0	128.085	Y=422.0404+296.1150e	
	11000	2	218.305		
		4	305.140		
		6	332.475	0 2713T	
5	12	0	150.205	Y=384.9291+232.7145e	
		2	258.595		
		4	293.450		
		6	345.185	0.39007	
6	12	0	103.550	Y=232.9516+127.9458e	
		2	180,915		
		Ā	192.150		
		6	226.615	0.05070	
7	12	Ő	120,995	Y=341.5550+218.2396e ^{0.25271}	
and the second of the		2	219,905		
		4	248.055		
		6	300.010	0./2095	
Q	12	0	121,430	¥=355.0727+235.6145e	
0	12	2	245.655		
		1	335,240		
		6	326 730		
0	12	0	117 680	Y=115,7095+33,0748T	
У	12	2	187 215		
		2	231 385		
		4	202.505		
		O	JLJ,4JJ		

Table 17.	Equations and Means of	Compressibility	of	Soy	Fortified
	Bread during Storage				

Equation	Compressibility Means (g/mm)	Storage Time (days)	Soy Level (%)	Treatment
	000 700			
I=434.7233+210.9096e	223.790	0	24	1
	416.305	2		
	429.560	4		
0.4567	437.500	6	1.590.	1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1. 1
Y=566./188+305.1666e	263.760	0	24	2
	431.085	2		
	542.335	4		
0.3145	533.300	6		
Y=829.9551+448.3866e	381.460	0	24	3
	591.425	2		
	701.725	4		
0.1982	762.395	6		
Y=508.7785+307.7781e	202.465	0	24	4
	295.895	2		
	377.060	4		
0 5795	411.820	6		
Y=515.6999+236.7364e	277.880	0	24	5
	449.405	2		
	474.480	4		
¥=422,2200+198.6586e ^{0.2709}	519.390	6		
	224.150	0	24	6
	304.055	2		
	358.770	4		
0 2111	381.385	6		
¥=471.5257+281.3090e	190.515	0	24	7
	319.130	2		States and the
	392,635	4		
0 1000	426,995	6		
Y=880.8138+603.4806e ^{0.1882}	271,930	0	24	8
	487.795	2		· · · ·
	569.355	4		
	697.210	6		
Y=542.1719+335.2981e ^{0.2623}	201,700	0	24	9
	366.410	2		States .
	392.485	4		
	487.460	6		

Table 17 (continued)

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VITA