# EFFECTS OF VARYING GRIND AND LEVEL OF GLANDLESS COTTONSEED FLOUR ON QUALITY AND AMINO ACID CONTENT OF PLAIN SUGAR COOKIES

by 650 p

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#### INTRODUCTION

Presently, cereal grains are among the most important feeds in the world because they supply the most calories per acre, can be stored for comparatively long times, and can be processed into a large number of familiar, widely acceptable foods. However, cereal grains, the major source of both calories and protein in the world diet, do not provide adequate protein, as they are deficient in certain essential amino acids (Bressani, 1965).

According to Milner (1966), one of the main solutions to the problem of malnutrition is development of marketable supplemented food products native to the population. The development of suitable vegetable protein concentrates is a widely accepted, important, and practical approach to supplying needed dietary protein supplements (Bressani, 1965). The potential contribution of oilseeds, in the form of protein concentrates, is great. The amount of protein potentially available is about equal to that of all protein of animal origin being produced in the world today (Milner, 1966).

Cottonseed is a good potential source of high quality protein and is one of the principle oilseeds available. Cottonseed protein became recognized as food for human consumption in 1959, when commercial production of protein-rich food mixtures that contained cottonseed flour began. Up to 38% cottonseed flour was used to enrich cereal flours, noodles, flaked breakfast foods, sausages, and other foods. The nutritional value, stability, tolerance, acceptability, and shelf life of those foods was good (Bressani et al., 1966).

Protein deficiency, a widely recognized problem in persons of all

ages throughout the world, is not as acute in the United States as in the rest of the world, but the problem does exist. In areas of the world where animal products are relatively plentiful, the population derives a large part of its total protein supply from cereal grains and their derivatives. Dependence on cereals for protein is expected to continue (West, 1969). In North America, the condition of adequate food for everyone is theoretically possible. However, a significant percentage of the population has poor or inadequate diets, which Senti (1969) attributed to non-uniform food distribution among other things.

The classical solution to problems of protein malnutrition has been the addition of more animal protein from meat, milk, and eggs to the diet. Animal protein is expensive, and Altschul (1968) and Mottern (1968) predicted a gradual shift from costly, high-quality, traditional animal proteins to consumption of cheaper vegetable proteins of high quality.

Matthews et al. (1963) found that baked products were a prominent part of the diet of most Americans. Eagles (1969) indicated that, in general, the low-income families in the United States used more of the foods in the bread-cereal group, but less of other foods, than families living on higher incomes. Consumption of convenience-type cereal products has shifted from use of mixes to consumption of fully prepared bakery products (Bivens, 1967). Thus, it appears that supplementation of cereal foods or baked products with protein concentrates is of importance. A low-cost vegetable protein additive such as glandless cottonseed flour to supplement cereal products should improve the quality of diets in the United States.

Such supplementation is possible, can have immediate effect, and can improve both the protein quantity and quality of the diet. However, use

of protein concentrates is feasible only where the type of foods commonly consumed allow their addition without significant alteration of the flavor, texture, and appearance of the original product (West, 1969).

No research was found in the literature comparing effects of a fine grind of glandless cottonseed flour with those of a coarse grind substituted at varying levels for wheat flour in a plain sugar cookie. Therefore, a study was proposed to investigate acceptability, physical properties, and amino acid content of cookies made with fine and coarse glandless cottonseed flours substituted at two levels for wheat flour.

#### REVIEW OF LITERATURE

## Malnutrition in the United States

In a survey by the United States Department of Agriculture, it was found that 2 to 7% of the low-income families studied received less than two-thirds of the National Research Council recommended allowance for protein (Eagles, 1969). Lamb (1969) noted that poor diets occurred at each income level, with four times as many occurring at the lower income levels. Preliminary results of a recent survey of inhabitants of ten representative states have shown cases of protein-calorie malnutrition, low serum-protein levels, marasmus, and kwashiorkor. These problems were definitely associated with other health, educational, social and economic problems (Shaefer, 1969).

## Cottonseed Protein as Human Food

In the United States alone, the annual production of cottonseed could yield over 8 tons of cottonseed flour with 60% high-quality protein

(Vix, 1968). Aside from serving as a major protein constituent in protein-rich foods, Bressani et al. (1966) indicated that cottonseed flour could be used as a supplement to the diet and cereal grain flours. Several workers (Wilcke et al., 1965; Smith et al., 1968) stated that cottonseed flour had a great potential as a source of protein in human feeding programs, in nutritional improvement of staple food items, and in domestic uses of flour.

Braham et al. (1965a), Bressani et al. (1965), Bressani et al. (1966), and Scrimshaw et al. (1961) conducted extensive research with human consumption of vegetable protein mixtures that contained cottonseed flour as the main source of protein. This flour was found safe, nutritious, palatable, and of high protein quality.

In early use of cottonseed flour for human food, several interrelated difficulties were encountered: gossypol, lysine content, and color. The dual role of gossypol as a toxic constituent and as a detriment to cotton-seed protein quality has been documented (Altschul et al., 1958). Gossypol is a naturally occurring, polyphenolic pigment located in pigment glands in cottonseed. The inactivation of that pigment during processing resulted in a decrease of the essential amino acid lysine through binding of lysine with gossypol (Braham et al., 1965b; Bressani et al., 1966; Lyman, 1967; Gastrock, 1968; Smith et al., 1968). The "bound" gossypol that remained in cottonseed flour imparted an undesirable, intense yellow-green or chrome yellow color to vegetable protein concentrates and to food products that contained the oilseed flour (Bressani, 1965; Harper, 1965; Bressani et al., 1966; Harper, 1968; Vix et al., 1968). The consensus of opinion was that this color had to be improved before cottonseed flour would be accepted

generally by the food industry and the consumer (Verdery, 1968).

## The Advent of Glandless Cottonseed Flour

The presence of a naturally occurring nutritional toxin gossypol decreased the nutritive value of cottonseed flour as a protein source, limited the unrestricted usefulness of cottonseed protein, and thus, prompted research in development of a glandless variety of cottonseed. Hopi cotton was reported to have a variable number of pigment glands in the boll (Lewton, 1912; Fulton, 1938). In studies of the possibilities that Hopi cotton might be used as a commercial source of gland-free cottonseed, McMichael (1959) found that through selection for low gland content of leaves and bolls, gossypol content could be reduced nearly to zero, while seeds were not affected materially. A chemical comparison of glanded and glandless cottonseed meals showed that total gossypol that is both gossypol "bound" to cellular constituents and "free" gossypol was reduced from 1.312% in the glanded seed to 0.02% in the gland-free seed. The percentage "free" gossypol was reduced from 0.595 in glanded to 0.006 in gland-free seed. Mattson et al. (1960) analyzed glandless cottonseed and concluded that this seed was essentially free of gossypol. It gave "free" gossypol values of 0.002% as compared to 0.40% in gland-containing cottonseed.

Milner (1966) stated that glandless cottonseed meals and flours were of excellent nutritive value. Amino acid composition of a protein was listed as the most important factor in determining the nutritive quality of a protein source (Bressani et al., 1966). Glanded and gland-free cottonseed protein reportedly had essentially the same amino acid

composition, with the exception of unbound or "free" lysine content, which was greater for glandless cottonseed protein. Glandless cottonseed protein was reported as significantly low in methionine and isoleucine when compared with the Food and Agriculture Organization reference protein (Smith et al., 1968).

In spite of the apparent deficiencies in growth-supporting amino acids, glandless cottonseed flours were reported to be safe and nutritious for humans (Graham, 1969) and animals (Milner, 1966; Watts, 1965; Johnson et al., 1964; Smart et al., 1960; Smith et al., 1968).

#### Use of Cottonseed Flours in Baked Products

While use of glanded cottonseed flour in baked products has been limited by its detrimental effects upon color and volume of those foods, the functional properties of this protein source lent it to certain domestic uses. Low levels of cottonseed flour used in bread, cookies, doughnuts, and other foods similar in nature, reduced dough stickiness, bound water, benefited machining properties, reduced fat absorption, and increased shelf life (Bressani, 1966; Bressani et al., 1966; Overman, 1951; Vix et al., 1968).

Levels of 1% to 6% glanded cottonseed flour added to wheat flour in baked products resulted in acceptable and marketable products (Bacigalupo, 1966; Verdery, 1968; Vix et al., 1968). Higher levels of added cottonseed flour resulted in less acceptable products.

Bacigalupo et al. (1967) demonstrated that oilseed flours mixed with wheat flour changed physical properties of absorption and dough strength of wheat. Extensive research dealing with substitution of oilseed flours for

wheat flour in breads has been performed. Yeast bread that contained 25% glandless cottonseed flour substituted for wheat flour had an unacceptable compact texture (Matthews, 1969). Vix et al. (1968) reported smaller loaf volumes, a dark color similar to that of whole wheat bread, and a dense gummy texture in breads that contained 17 to 20% glanded cottonseed flour.

Enrichment of Peruvian white bread with Protal, a commercially available cottonseed flour, yielded results characteristic of other baked products enriched with cottonseed flour (Bacigalupo et al., 1967). Bread became progressively more brown as enrichment with Protal increased. Breads enriched with 0, 5, 10, 15, 20, and 25% Protal were evaluated organoleptically for freshness. Breads that contained Protal at levels greater than 10% kept freshness 4 to 6 days whereas the control, containing no Protal, lost freshness 2 days after baking. Further objective evaluations of enriched breads evidenced: a significant (P = 0.01) positive linear correlation (r = 0.8465) between weight of enriched breads and Protal content; greater water retention with higher levels of cottonseed flour; and a significant (P = 0.01) negative linear correlation (r = 0.9656) between bread volume and Protal content. Protal at a 5% level of enrichment markedly improved bread volume whereas 10% Protal did not modify bread volume. Crumb texture and crust structure of breads with 0, 5, 10, and 15% levels of enrichment were scored high, whereas breads enriched at a 20% level were scored fair, and breads enriched at a 25% level were scored low.

Enrichment of crackers with a maximum of 10 to 15% cottonseed flour presented no difficulties. Crackers were not sensitive to specific rheological phenomena associated with use of this oilseed flour (Bacigalupo,

1969). However, cottonseed flour has not been recommended as a replacement for wheat flour since it has none of the functional properties of wheat flour. Dalby (1969) reported on enrichment of Syrian bread with cottonseed flours. Undesirable effects of cottonseed flours were first noticed when bread doughs became quite sticky and difficult to handle. Nonfunctional additives were thought to modify gluten structure to such an extent that the loaf did not rise in the oven. As temperature increased rapidly, steam that was generated escaped more readily through the surface of the enriched loaf.

Johnson (1969) and Dalby (1969) indicated that such nongluten additives as milk, sugar, and oilseed flours introduced a burden on volume-forming capacity and lowered gas retention capacity of gluten in doughs. Thus, the finished food product did not have the same physical characteristics as a product without an oilseed additive. Dalby (1969) further indicated that the maximum quantity of non-functional additives that could be added was approximately 5% based on the weight of the flour. In special cases, this level could reach 10%. When glandless cottonseed flour was added at a level of 6% to Syrian bread, a dark-colored loaf resulted.

Mottern (1968) reported that water-binding capacity of cottonseed flour adversely affected the volume of yeast bread. In general, bread doughs had a greater water-binding capacity with increased protein content (Lowe, 1955). Griswold (1962) stated that the correct amount of liquid in making bread dough was important. Too little water made gluten so strong that it would not stretch and as a result, volume of bread would be low.

Matthews (1969) suggested several principles related to use of

oilseed flours in enriching cereal products. She discussed the importance of particle size of oilseed flours when used in bread doughs. Better quality loaves of bread resulted when 25% coarse grind soy flour (60 to 80 mesh) was substituted for wheat flour than when a finer soy flour (100 mesh) was substituted at the same level. The coarser particles of soy flour in the mixture absorbed water more readily than did the finer soy flour. It was suggested that coarsely ground oilseed flours in bread dough might give better results than finely ground oilseed flours. Mouthfeel of foods containing oilseed flours is largely determined by particle size and the ability of the oilseed additive to soften in the presence of moisture (Johnson, 1969). Matthews (1969) indicated that thorough sifting of the oilseed flour with wheat flour was essential for even hydration and for smooth and uniform mixing of bread doughs with minimum mixing times.

Martinez et al. (1970) noted that functionally active protein supplements must contribute to or replace some desirable esthetic quality in the food such as appearance or texture. The total quantity of protein also should be increased. Functionally inert protein supplements must increase the level of protein without interferring with the normal structure, flavor and quality of the food. The authors further indicated that the manner in which the protein product functioned in the food was determined by both the protein and nonprotein components of the supplement and by the ionic environment of the end-use formulation.

Cottonseed flour has been shown to increase the protein level of bread. Dalby (1969) reported that 60%-protein cottonseed flour used in bread at a 7% level, based on flour, should produce a degree of protein supplementation close to that provided by 12% milk solids.

Bressani (1965) and Bressani et al. (1966) reported that the taste contributed to foods by cottonseed flours was either bland or slightly nutty. Glandless cottonseed flour was reported as having a low moderate flavor profile without any "off" notes (Martinez, 1969). Flavor profile may be classified as a descriptive analysis of the flavor and aroma characteristics of foods. A "low moderate" profile could indicate: a "low" or weak overall impression of aroma and flavor or body; a "moderate" or medium-strong impression of character notes or aroma and flavor.

#### Characteristics of Cookies

On the basis of consistency, flour mixtures are classified as doughs or batters. Doughs require handling or kneading on a board because they are too thick to be beaten, whereas batters are beaten during their preparation and are of a consistency to be dropped or poured (Griswold, 1962).

Matz (1962) discussed certain aspects of cookie texture. Cookies were classified as friable foods composed of small, usually irregular pieces rather loosely bound to one another. He states that on a macroscopic scale, cookies were non-homogenous and when masticated, they broke readily into their constituent pieces.

Cookie dough was described as being non-uniform with discrete globules of fat. After baking and cooling, the dough became brittle and hard. Ease with which cookies broke into pieces when pressure was applied was reportedly a function of the size and shape of gas pockets, the moisture content and other crumb characteristics. In general, a moisture content of 5% or less led to friable texture, while a moisture content

of 10% or more tended to cause a chewy or elastic effect (Matz, 1962).

## Evaluation of Cookies

Among other attributes, appearance of cookies is an important factor in acceptability. Finney et al. (1950) stated that a well-broken top containing numerous small islands was characteristic of good quality cookies.

Texture of a friable food product depends upon the forces that hold the particles together. Mouthfeel is affected by the size, shape, consistency, and degree of uniformity of particles resulting when the piece breaks apart, whereas overall texture is affected by smoothness or roughness of surfaces and hardness of individual particles (Matz, 1962).

Effects of wheat flour granularity on sugar-snap cookie-baking potentialities were evaluated in terms of spread. Cookies made with fine, soft wheat flours, low in protein and ash content were larger than those made with coarse counterparts (Yamazaki, 1959a). Milling of a wheat mix, tempered to various levels, resulted in changes in flour granularities. Those changes were not associated with significant cookie diameter changes within reasonable limits of moisture contents of flours (Yamazaki, 1959b). Factors other than granularity were more important in determining cookie quality.

Matz (1968) indicated control of cookie spread as one of the most serious problems confronting commercial cookie producers. Hanson (1943) reported that a spread factor for a batch of cookies could be determined using six cookies and dividing the average diameter of three cookies by the average height of six cookies. If the result of this computation were large, a greater spread of cookies was indicated than if the result were

smaller.

Manufacturers recommended addition of one pound extra water for each added pound of PROFLO or CINACOA (commercially available cottonseed flours). Reportedly, that increase in water kept spread factors the same in cookies containing no cottonseed flour as in those containing cotton-seed flour (Anon., 1968a). Five per cent of the wheat flour in a sugar cookie was replaced with CINACOA 150. This reduced the spread of cookies about 5% as compared to cookies containing 2% cottonseed flour. In those tests, one pound extra water (in addition to the normal amount) was added for each pound of cottonseed flour. In those cookies containing 5% cottonseed flour, the diameter and thickness of the cookies increased although the spread factor decreased. Grind of flour used was reportedly 90% 200-mesh. Protein content was between 55-60% (Anon., 1968b).

# Proteins and Amino Acids of Glandless Cottonseed Flour

Cottonseed readily affords a flour containing 60 to 70% protein.

Native cottonseed flour proteins can be concentrated to 70% primarily by removal of the crude fiber and carbohydrate constituents. Those proteins can be classified as water-soluble and water-insoluble (Martinez et al., 1970).

Water soluble proteins were reported as being great in number, low in molecular weight, rich in lysine and sulfur amino acids, and having a minimum solubility at pH 4. Water-soluble proteins were predominantly the functional proteins of the cytoplasm. Those proteins represented 25 to 30% of the total nitrogen of defatted cottonseed flour.

Water-insoluble proteins were reported as few in number, high in

molecular weight, relatively low in lysine and the sulfur amino acids, and having a minimum solubility at pH 7 with maxima at pH 4 and 9. About 60 to 65% of the total nitrogen of defatted flour was represented by waterinsoluble or storage proteins (Martinez et al., 1970).

When cottonseed flour was compared with cereal flours, the former was relatively high in protein, fat, and ash contents and low in carbohydrates. Protein of cottonseed flour was reported to be globulins, pentose-containing protein, and glutelin (Pyler, 1952).

Both qualitative and quantitative aspects of protein supplementation must be considered in evaluating contributions to nutrition. Martinez et al. (1970) indicated that the protein quality of cottonseed is inherently high, but lysine will probably be the major deficiency in combinations of cottonseed and cereals.

Watts (1969) reported that cottonseed flour was deficient in five amino acids: lysine, methionine, leucine, threonine, and isoleucine.

Cottonseed contains a large proportion of nonessential amino acids (70%) and a smaller proportion of essential amino acids (30%) (Bressani, 1965).

Thus cottonseed is not a complete protein.

#### PROCEDURE

# Ingredients of Cookies

Ingredients for the cookies were purchased in bulk lots. Fine gland-less cottonseed flour (-100 mesh) was obtained from National Cottonseed Products Association, Inc. Coarse glandless cottonseed flour (-80 mesh) was obtained from Texas A&M Oilseed Products Research Center. Analyses of those flours are shown in Appendix, Table 9. All ingredients except

sugar, salt and cinnamon were kept refrigerated until used.

#### Recipes

A basic sugar cookie recipe was used as a control and as the basis for modifications with the cottonseed flours (Table 10, Appendix). Fine and coarse cottonseed flours were each substituted for 6 and 15% of the wheat flour. Products were prepared using a randomized balanced complete block design (Form 1, Appendix).

# Preparation

Before each baking period, shortening for each product was weighed into a Pyrex cup and covered securely with plastic wrap. Dry ingredients for each product were weighed, placed in a bowl, and covered securely with plastic wrap. After weighing, all ingredients were placed on trays and held at 21°C in a Labline incubator approximately 15 hr prior to mixing. All cookies were prepared according to the recipe in Table 10 (Appendix).

A daily record was kept of humidity in the laboratory. Relationships between humidity and tenderness and humidity and moisture were analyzed statistically.

Cookie dough was divided into two portions for baking. One baked portion was used for all objective and subjective evaluations except shortness; the other portion was used to measure shortness of cookies. One portion of the dough was placed between two sheets of wax paper. Two wooden guides, each 0.9 x 1.2 x 38 cm, were placed between the sheets on either side of the dough and the dough was rolled 9 mm thick. Six cookies of 1 1/2 in. diameter were placed on 3 aluminum cookie sheets and baked simultaneously in 3 preheated ovens at 400°F for 14 min (Figure 1,

Appendix). Immediately upon removal from the oven, cookies were placed on wire cooling racks and allowed to cool at room temperature for approximately 2 hr. At the end of this period, 5 cookies from each treatment were placed in small ceramic crocks and covered securely with plastic wrap until evaluated for moisture and pH. Clean towels were placed over the remainder of the cookies until products were evaluated approximately 4 hr later.

#### Evaluation

Objective and subjective measurements were made to evaluate the baked cookies. Three ovens were required to bake cookies from each product.

Thus, a randomized, predetermined pattern was used to select cookies for each objective measurement (Form 2, Appendix).

Tenderness (breaking strength). Tenderness of the cookies was determined by use of a 1000-g capacity shortometer. The instrument consisted of a modified spring balance and a container for fine lead shot. The shortometer was arranged so that the cookie was supported by two parallel bars, 1 3/8 in. apart, on the weighing pan of the spring balance. A movable third bar was suspended above the center of and parallel to the two supporting bars. A cup was positioned above this center bar. That cup was gradually filled with lead shot when a clamp was removed from a rubber siphon leading from the shot container. The cookie was placed across the two supporting bars, and the weight of shot, in grams, required to break the cookie with the third bar was recorded. Dough for cookies used to measure tenderness was rolled 4 mm thick, cut in four strips 1 1/2 x 5 1/2 cm and baked 8 min at 400°F in the same oven for each

replication (Figure 2, Appendix). Approximately 1 1/2 hr after baking, tenderness measurements were made on duplicate samples of each variable. A randomized, predetermined pattern was used to select samples (Form 3, Appendix).

Spread, volume, and specific volume. Finney et al. (1950) described a method for evaluating cookies in which diameter and thickness of all cookies were measured and a spread factor calculated. In this study, thickness and diameter of five cookies from each product were determined and a spread factor calculated. The same five cookies were used to determine volume by rape seed displacement and specific volume as described by Griswold (1962).

Moisture content. To determine moisture content of cookies, a

Brabender Rapid Moisture Tester (model SAS) was preheated to 120°C. Four

cookies from each product were reduced to crumb in a one-speed Waring

Blendor and duplicate 5 g samples placed in the oven chamber for 50 min.

At the end of that time, readings were obtained.

pH measurement. Measurements of pH were made on a slurry prepared by placing 50 ml distilled, demineralized water and 20 g cookie crumbs, in that order, in a one-speed Waring Blendor and blending for 2 min. A magnetic stirring device was placed with the slurry in a beaker and the slurry stirred for 30 sec prior to measuring pH. Duplicate readings of pH of each treatment were made to one decimal place with a Beckman pH meter. The beaker was turned 90° clockwise, the slurry stirred for 30 sec before the second pH reading was taken.

Color. Color differences, reflectance, redness, and yellowness, of cookies was determined by use of a Gardner Automatic Color Difference

Meter (model #AS-2A, series 200). A ceramic tile with the following calculated values was used to standardize the color-difference meter: Rd (reflectance), 37.6; a (redness), 6.2; b (yellowness), 14.9. Immediately prior to obtaining readings from each product, the instrument was standardized. One cookie from each product was placed in such a manner that the top center of the cookie was directly over and completely covering the aperture of the color-difference meter. Two sets of readings were taken on each cookie by obtaining an initial set of readings; then rotating the cookie 90° in a clockwise direction, and taking another set.

Amino acid analyses. Amino acid analyses for samples of baked cookies that contained no cottonseed flour, 6% fine cottonseed flour, and 6% coarse cottonseed flour were made. Methionine and tryptophan were analyzed according to the microbiological method of Henderson et al. (1948). Amounts of all other amino acids were analyzed according to the method of Spackman et al. (1958). Calculated protein values of cookies and of cookie samples analyzed are shown in Table 11 (Appendix).

<u>Subjective evaluation</u>. Sensory evaluation for quality of cookies was performed by a 7-member taste panel. Cookies evaluated for appearance were placed on a white background beneath a fluorescent ceiling light. A score card (Form 4, Appendix) was used to record subjective evaluations.

Frequency of use for descriptive terms appearing on the score card was computed. Those data were not subjected to statistical analyses.

# Analyses of Data

Data for each measurement made to evaluate the cookies were subjected to the following two-way analysis of variance:

Source of Variation	<u>D/F</u>
Treatment	4
Replication	9
Error	36
Total	49

Orthogonal comparisons were computed to determine effects of grind and level of cottonseed flours on the quality of cookies. The treatment sums of squares were broken down as follows:

Source of Variation	D/F
Grind of cottonseed flour (G)	1
Level of cottonseed flour (L)	1
G x L	1
Control vs. cookies containing	
cottonseed flour	_1_
	4

When replications were nonsignificant, the replication and error DF were pooled to increase the power of the test. After adjusting error for replications, this same analysis was used when replications were significant. When it was necessary to explain interactions, least significant differences (LSD, P = 0.05) were calculated. Correlation coefficients were computed for selected paired variates.

## RESULTS AND DISCUSSION

Treatment means for objective and subjective evaluations appear in Tables 1 and 3, respectively. Orthogonal comparisons of objective and subjective evaluations appear in Tables 2 and 4, respectively. The analyses of variance appear in Appendix, Tables 12 and 13. Throughout the discussion, 6 and 15% refer to levels of substitution of fine (F) and coarse (C) cottonseed flour (CSF) for wheat flour.

# Objective Evaluation

Tenderness (breaking strength). Since proteinaceous materials have great water absorbing or binding abilities (Lowe, 1955), effects of the presence of CSF on wheat gluten hydration and consequent tenderness were investigated. It was believed that water binding properties of CSF protein affected not only tenderness but also other factors such as: height, width, spread, volume, specific volume, and moisture.

Tenderness of cookies varied significantly. As evidenced by mean treatment values (Table 1) and orthogonal comparisons (Table 2), increasing levels of CSF within a grind of flour resulted in significantly less tender cookies (Table 1).

Cookies that contained C- at both levels were less tender than corresponding cookies that contained F-CSF. Orthogonal comparisons indicated that differences in mean tenderness scores among cookies containing CSF might be attributed to effects of some interaction of grind and level of CSF (Table 2). LSD values employed to elucidate interactions supported the statistical evidence of orthogonal contrasts (Table 1). Differences in significance of effects of grind and level of CSF as a possible source of interaction were noted also. Differences in tenderness between cookies containing 6% F- and 6% C-CSF were nonsignificant. Cookies containing 15% F- were significantly more tender than cookies containing 15% C-CSF. As indicated by orthogonal comparisons and mean treatment values, control cookies were significantly more tender than all other cookies.

Variations in tenderness of cookies may be attributed to grind and/or level of CSF. Cottonseed flour contains no gluten; thus, it does not contribute to the wheat-gluten structure of baked products. Possibly

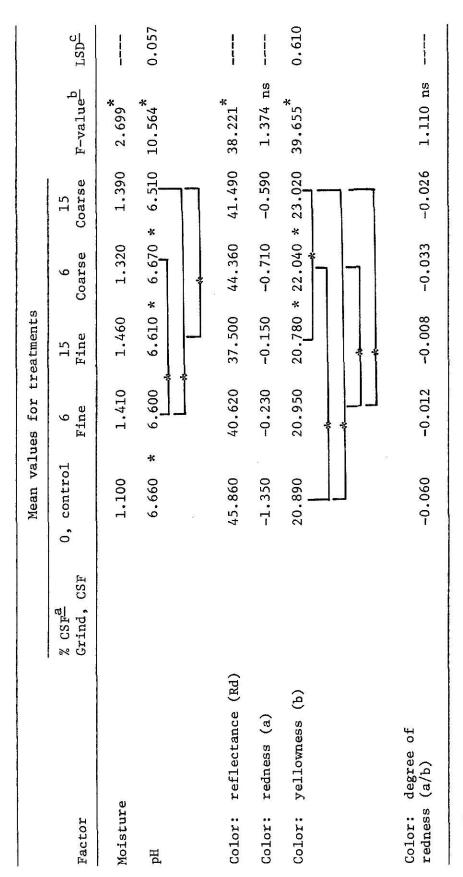
THIS BOOK CONTAINS NUMEROUS PAGES WITH DIAGRAMS THAT ARE CROOKED COMPARED TO THE REST OF THE INFORMATION ON THE PAGE.

THIS IS AS
RECEIVED FROM
CUSTOMER.

Treatment means, F-values, and LSD values between means for objective evaluation of cookies. Table 1.

		Mean values	for	treatments				
Factor	% CSF <sup>a</sup> Grind, CSF	0, control	6 Fine	15 Fine	6 Coarse	15 Coarse	F-value	LSDC
Tenderness (breaking strength)		373.00 *	537.80 * 607.00	607.00	589.30 * 780.10	780.10	73.116*	61.501
						77		
Width		12.310	12.144	12.174	11.911	11.741	8.693*	! ! !
Height		2.300	2.175	2.025	2.063	2.038	9.387*	
Spread (width/height)		5.380	5.590 *	0,000 *	5.800	5.770	4.526	0.314
		<u>֚֚֚֚֚֚֚֚֚֚֚֓֞</u>			7			
Volume		165.00	155.00	146.50	144.50	140.50	28.042	 
Weight		\$7.40 *	06.90	61.10	* 02.09	63.60	20.333*	1.361
			* *					
Specific volume (volume/weight)		2.880	2.570	2.390	2.420	2.200	2.200 41.890*	

Table 1. (concl.)



a Cottonseed flour.

 $<sup>^{</sup>b}$  \*, P = 0.05; ns = not significant.

 $<sup>^{</sup>c}$  P = 0.05; means separated by \* are significantly different.

Table 2. Effects of grind and/or level of cottonseed flour on objective evaluations as shown by orthogonal comparisons, F-values and significance.  $\frac{\Delta}{2}$ 

			0	Orthogonal comparison	omparison			
Factors	6 and 15% fine CSF vs. 6 and coarse grind	% fine grind 6 and 15% grind CSF	6% fine and c CSF vs. 15% and coarse	and coarse 15% fine arse CSF	Interaction	ction	Control vs. cookies containing CSF	d vs. ies ng CSF
	F-value	Signif.	F-value	Signif.	F-value	Signif.	F-value	Signif.
Tenderness (breaking strength)	43.163	*	57.837	*	12.651	*	178.797	*
Width	19.180	*	0.842	ns	1,728	ns	13.986	*
Height	1.846	su	5.654	*	2.885	su	29.908	*
Spread (width/height)	0.074	ns	3,628	su	4.739	*	11.609	*
Volume	19.173	*	11.004	*	1.426	ns	76.088	*
Weight	5.795	<b>-</b> K	10.527	*	7.986	*	61,101	*
Specific volume (volume/weight)	18.526	*	25.641	*	0.256	ns	120.628	*
Moisture	0.758	ns	0.427	ns	0.012	ns	8.249	*
hф	0.553	*	13.832	*	17.766	*	7.684	*
Color: reflectance (Rd)	39.445	*	23.684	*	0.041	ns	50.045	*
Color: redness (a)	0.987	ns	0.047	ns	0.002	ns	3.226	su
Color: yellowness (b)	116.089	*	6.868	ns	13.844	*	21.843	*
Color: degree of redness $(a/b)$	0.989	ns	0.087	su	0.005	ns	3.366	ns

 $\frac{a}{2}$  \*, P = 0.05; ns = not significant.

competition of CSF proteins for moisture limited the hydration of gluten.

Thus, less hydrated and less developed gluten did not stretch during baking. As such, unstretched gluten would contribute to toughness of the baked product.

Matthews (1969) reported that coarse ground soy flour absorbed water more readily than finer soy flour when substituted in bread. In that study, the coarser sample of soy flour gave better results. In the present study, it appeared that C- absorbed water more readily than F-CSF when substituted for wheat flour. Thus, it was believed that less water was available for gluten hydration. Cookies containing C- were less tender than those containing F-CSF at comparable levels of substitution. Possibly, some of the differences in water absorption of cottonseed flours was attributable to differences in the moisture content of those flours (Table 9, Appendix). Moisture content of F-CSF reported by the manufacturer was not found to be as high at the time of testing as when reported.

Width. Mean measurements of width were significantly different (Table 1), but means of all treatments were not significantly different from each other (Table 2). Use of F- and C-CSF in cookies possibly contributed to significant differences in mean widths (Tables 1 and 2). Cookies containing F-CSF were significantly wider than cookies containing C-CSF; whereas, the level of CSF used did not affect width significantly. Mean width of control cookies was significantly greater than mean widths of cookies containing CSF. Again, the proteinaceous nature of CSF may have affected width of cookies containing CSF.

Height. Variations in height of cookies may be attributable to competition of CSF and wheat gluten for water. Gluten of wheat flour probably did not hydrate and stretch during baking; thus, mean height of cookies containing CSF was significantly lower than that of control cookies. As level of CSF, thus protein content, increased, mean height of cookies decreased (Table 1). Cookies containing 6% CSF had significantly greater height than cookies containing 15% CSF (Table 2). As indicated by orthogonal comparisons, grind of CSF substituted for wheat flour did not contribute to significant differences in heights of cookies.

Spread (width/height). Spread of cookies containing CSF was affected by some interaction of grind and level of CSF (Table 2). Mean values for spread indicated that within each of the two grinds of CSF, spread of cookies did not respond in the same manner to increasing levels of CSF (Table 1). As shown by LSD values, cookies containing 6% F-CSF spread significantly less than those containing 15%; whereas, cookies containing 6 and 15% C-CSF did not differ significantly. Thus, effects of substitution of CSF for wheat flour on spread of cookies appeared to be dependent upon grind of CSF used.

The spread of control cookies differed significantly from that of cookies containing CSF (Table 2). Control cookies had the greatest width and height, yet they had the lowest calculated spread. From examination of mean values for width and height (Table 1), it appeared that calculated spread values were not indicators of width alone.

Effects of F-CSF on spread of cookies varied from those reported by PROFLO manufacturers when fine cottonseed flour was substituted for wheat flour. It was reported that increasing levels of PROFLO resulted in decreased spread of cookies. However, when an additional 1% water for each 1% CSF was used, an increase in both diameter and thickness, but a

decrease in spread was encountered. Addition of water to cookies prepared in the present study might have lessened the detrimental effects of substituting CSF for wheat flour.

It seemed reasonable to expect lower spread values for cookies containing C-CSF, since those cookies exhibited smaller mean widths and lower heights. However, cookies containing F-CSF had less width per unit height than those cookies containing C-CSF; thus, cookies containing F-CSF spread less.

<u>Volume</u>. Volume of cookies differed significantly (Table 1); both grind and level of CSF significantly affected volume of cookies (Table 2). Volume of control cookies was significantly greater than volume of cookies from any other treatment. Mean volumes of cookies containing C-CSF were significantly lower than volumes of cookies containing F-CSF (Tables 1 and 2). Mean volumes of cookies containing 6% CSF were significantly greater than those of cookies containing 15%.

Decrease in volume may be attributed to grind and level of CSF substituted for wheat flour. Water binding properties of CSF protein perhaps limited expansion of gluten during baking. Dalby (1969) commented that increased levels of nonfunctional additives in baked products decrease volume and result in more compact products. It appeared that such was the case with increasing levels of CSF in the present study.

Cookies containing C-CSF had the smallest volumes. Thus it appeared that C- absorbed greater amounts of water than F-CSF and prevented gluten hydration and consequent expansion during baking.

Weight. Variation in mean weights of cookies may be attributed to water absorbing and binding abilities of high-protein CSF. Even though an

identical amount of egg was added to cookies in each treatment, varied amounts of CSF protein from different sources perhaps affected amounts of water held by cookies during baking. Thus, weights of cookies varied after baking.

Weight of cookies was affected by some interaction of grind and level of CSF (Table 2). Mean weight of cookies increased as the level of each grind of CSF substituted increased from 6 to 15% (Table 1). As shown by LSD values, those differences were significant only between cookies containing 6 and 15% C-CSF. Mean weights of cookies containing 6 or 15% F-CSF did not differ significantly. At the 15% level of substitution, cookies containing C-CSF were significantly heavier than all other cookies. Control cookies were significantly lighter in weight than cookies from other treatments (Tables 1 and 2).

Lowe (1955) stated that bound water was so closely associated with the molecule with which it was combined that it no longer had the characteristics of ordinary water. Possibly CSF bound water so effectively in cookies that it was not driven off during baking. Control cookies contained no CSF and were lightest in weight. It appeared that when 15% CSF was substituted, protein from C-CSF bound more water or bound it more securely than did protein from F-CSF. Thus, those cookies containing 15% C-CSF had highest mean weights (Table 1). At levels of 6%, grind of flour made no significant difference in mean weight values.

Greater water holding and binding abilities of CSF reportedly contribute to prolonged freshness of baked products. However, if water is bound preferentially by CSF with resultant limited hydration of wheat gluten, undesirable qualities may result.

Specific volume (volume/weight). Fortification of baked products with protein-rich oilseed supplements reportedly lowered the volume and increased the weight of those products. Thus, specific volume of cookies was measured in an effort to determine whether substitution of either grind and/or level of CSF for wheat flour would result in a product with a lower specific volume.

Both grind and level of CSF, when substituted in cookies for wheat flour, significantly affected specific volume (Table 2). As level of CSF increased, specific volume of cookies decreased. Cookies containing both levels of F-CSF had significantly greater specific volume than cookies containing corresponding levels of C-CSF (Tables 1 and 2). However, control cookies had the greatest specific volume.

It appeared that C-CSF absorbed and held more rather than less moisture. The lower specific volume of cookies in this study may be attributed to use of CSF and/or increasing level of CSF. Decrease in specific volume was thought to have been associated with certain undesirable qualities noted in cookies during the course of this study. Decrease in tenderness has already been discussed.

Moisture. Mean moisture content of cookies was not affected significantly by grind and/or level of CSF (Tables 1 and 2). Such a lack of measurable differences in moisture content may be attributable to binding of water by CSF proteins. Control cookies had significantly lower moisture content than those with CSF. Control cookies did not contain as much proteinaceous material as cookies containing CSF.

After consideration of mean weight and specific volume, it appeared that cookies containing either 15% F- or C-CSF and those containing both

levels of C-CSF should have had a greater moisture content since they were greatest in mean weight and lowest in specific volume. However, this conjecture was not supported by experimental evidence. Possibly most volatiles and moisture not bound securely by CSF protein in cookies were driven off during baking.

pH. Mean pH values were significantly different (Table 1). Increasing levels of F- or C-CSF were associated with significant variations in mean pH values (Tables 1 and 2). Mean pH values were affected significantly by an interaction of effects of grind and level of CSF. The pH of cookies containing the two levels of F-CSF did not differ significantly from each other; whereas, cookies containing 6% C-CSF had significantly greater mean pH values than those containing 15% (Table 1). The mean pH values of cookies containing C-CSF were significantly different from those of cookies containing F-CSF. Thus, the effects of CSF on mean pH values appeared to be dependent upon grind of CSF used. Cookies containing no CSF or 15% C-CSF.

Mean pH of control cookies differed significantly from mean pH of cookies containing CSF. Under the conditions of this study, changes in pH could not be attributed to a particular factor. However, it is possible that some of the water soluble proteins discussed by Martinez et al. (1970) affected pH of cookies.

Color: reflectance (Rd). Substitution of glanded CSF in baked products altered the reflectance (darkness and lightness) and color of those products (Dalby, 1969). One of the improved aspects of glandless CSF was reportedly its lack of detrimental effect on color of baked products.

Both grind and level of CSF affected reflectance values significantly (Table 2). Control cookies had significantly greater mean reflectance values than cookies containing CSF. In fact, control cookies appeared creamy white in color as did the flour from which they were made. As level of CSF increased, mean reflectance values of cookies decreased. That decrease may be attributed to shade and color of the CSF used. When viewed with the naked eye, F-CSF appeared slightly greenish and darker than C-CSF which appeared slightly yellow and lighter in color. Thus, as level of CSF was increased in cookies, a certain amount of change in reflectance occurred. Cookies containing 15% F- or C-CSF had significantly lower mean reflectance values than those cookies containing 6% F- or C-CSF.

Cookies containing C-CSF had significantly greater mean reflectance values than those containing F-CSF (Tables 1 and 2). Perhaps color differences between the two flours could be attributed to the type of seed and/or milling procedures used.

Color: redness (a). Even though flours used in cookies varied in color, those color differences did not contribute to significant differences in redness or greenness of baked cookies. All mean (a) values were actually negative, indicating that absence of redness, or greenness, was measured. In light of the fact that F-CSF appeared slightly green, it is interesting to note the lack of detectable significant differences among cookies with respect to (a) measurements (Tables 1 and 2).

Dalby (1969) reported excessive browning in bread containing glandless CSF. However, no excessive browning was noted in cookies made in this study even though relatively high proportions of sugar and protein

constituents were available for browning or Maillard reactions. The relatively short baking period was most probably a factor in the absence of excessive browning of cookies.

Color: yellowness (b). Significant differences among (b) values may be attributed to color differences among flours used. Yellowness of control cookies varied significantly from that of cookies containing CSF; differences among mean (b) values of the latter were attributed to some significant interaction of effects of grind and level of CSF used (Tables 1 and 2).

Grind of CSF was one of the factors contributing to significant (b) differences (Table 2). As shown by LSD values, cookies containing either level of C-CSF had significantly higher mean (b) values than all other cookies (Table 1). Only mean (b) values of cookies containing C-CSF increased significantly as level of CSF increased. Mean (b) values of cookies containing 6 and 15% F-CSF varied significantly from cookies containing C-CSF at corresponding levels. Thus, the manner in which CSF affected mean (b) values depended upon grind used. The apparent variations in yellowness of F- and C-CSF were detectable in the baked product.

Color: degree of redness (a/b). Mean (a/b) values of cookies as measured did not differ significantly. Perhaps a more sensitive measure would have detected color differences.

## Subjective Evaluation

Ultimately, the test of a product is its acceptance by consumers.

Thus, a panel of judges evaluated selected characteristics for quality and acceptability of cookies. Frequency of use of selected descriptive

terminology is shown in Table 5.

Appearance: surface. A desirable surface appearance for sugar or snap-type cookies was defined as "well broken, containing numerous small islands" (Finney et al., 1950). On the basis of that criteria, mean scores for control cookies were significantly lower than those for all cookies containing CSF, although surface appearance of all cookies was judged acceptable (Tables 3 and 4). Control cookies were almost smooth in appearance, whereas cookies containing CSF had relatively more well defined islands on their surfaces. Panel members checked "unbroken surface" as a descriptive term for control cookies 19.4% of the time, whereas that term was indicated only 0.1% of the time for cookies containing 15% F-CSF (Table 5).

Mean appearance scores for cookies containing CSF were not significantly different (Tables 4 and 5). Surface appearance of all cookies containing CSF was affected in the same manner by the presence of CSF regardless of grind or level. Development of numerous small islands might be attributed to varying degrees of underhydration of wheat gluten and consequent inability of gluten to stretch and rise during baking. Another possible factor in formation of islands on the surface of cookies might have been contraction or coagulation of proteins upon exposure to heat.

Appearance: shape. Neither grind nor level of CSF noticeably affected shape of cookies (Table 4). All cookies remained essentially round during baking. However, edges of cookies containing CSF were labeled uneven and pitted more often than those of control cookies (Table 5).

Appearance: color. In contrast to objective measurements of color

Table 3. Treatment means and F-values for subjective evaluation of cookies.

			Mean sc	Mean scores— for treatments	treatments			
Factor		% CSF <sup>b</sup> Grind, CSF	0, control	6 Fine	15 Fine	6 Coarse	15 Coarse	F-value
Appearance:	surface		3.020	3.560	3.600	3.780	3.750	2,662*
Appearance:	shape		3.460	3.070	3.160	3.390	2.940	1.369 ns
Appearance:	color		3.300	3.350	2.700	3.370	2.870	4.659*
Aroma			3.730	3.270	2.780	3.360	2.930	15.252*
Flavor		N	3.770	3.260	2.690	3.070	2.640	18.152*
Texture			3.860	3.480	2.840	3.090	2.430	21.507*
Acceptability	٨		2.630	2.240	1.670	2.030	1,530	21.360*

 $\frac{a}{}$  Range: 5 (Highly desirable) to 1 (Unacceptable).

 $<sup>\</sup>frac{b}{}$  Cottonseed flour.

 $<sup>\</sup>frac{c}{c}$  \*, P = 0.05; ns = not significant.

Table 4. Effects of grind and/or level of cottonseed flour on subjective evaluations as shown by orthogonal comparisons, F-values, and significance. $\frac{a}{a}$ 

			0	Orthogonal comparison	omparison			
Factor	6 and 15% fine CSF vs. 6 and coarse grind	fine grind and 15% grind CSF	6% fine a CFS vs. and coa	% fine and coarse CFS vs. 15% fine and coarse CSF	Interaction	ction	Control vs. cookies containing C	Control vs. cookies containing CSF
	F-value	Signif.	F-value	Signif.	F-value	Signif.	F-value	Signif.
Appearance: surface	1.053	ns	0.001	su	0.038	នព	10.483	*
Appearance: shape	0.081	su	1.046	ns	2.353	su	2.644	su
Appearance: color	0.441	su	16.160	*	0.275	su	2.024	ns
Aroma	1.186	ns	17.430	*	0.074	su	27.415	*
Flavor	1.095	ns	19.007	*	0.373	ពន	44.462	*
Texture	11.191	*	29.550	*	0.007	ns	45.332	*
Acceptability	3.820	su	35.699	*	0.153	su	58.021	*

and a not significant; \*, P = 0.05.

Table 5. Frequency of use (%) of selected descriptive terms for each treatment.

				Des	scripti	Descriptive terminology used	nology	nsed				
Treatment	Uneven Unbroken pitted surface edges	Uneven pitted edges	Green exterior	Beany, grassy een Green straw- erior interior Nutty like	Nutty	Beany, grassy, straw- like	Oily	Bitter	Tough	Hard	Oily Bitter Tough Hard Tender	Unevenly cooked
Control	19.4	17.9	0.0	0.0 14.9	14.9	0.0	0.0	0.1	0.0	0.1 40.2	40.2	11.9
6% fine CSFª	0.0	31.3	0.1	16.4	13.4	11.9	10.4	0.0	0.1	17.9	19.4	0.1
15% fine CSF	0.1	34.3	41.7	49.2	0.0	17.9	20.8	14.9 28.3	28.3	49.2	0.0	0.1
6% coarse CSF	0.0	34.3	10.4	29.8	0.1	16.4	11.9	10.4 11.9	11.9	38.8	0.1	0.1
15% coarse CSF	F 0.0	38.8	25.3	53.7	0.1	17.9	23.8		11.9 35.8	73.1	0.0	0.1

a Cottonseed flour.

or degree of redness (a/b), panel members perceived significant differences among colors of cookies (Table 3). Mean color scores of control cookies did not differ significantly from those of cookies containing CSF (Table 3). Panel judges perceived color differences between cookies containing 6 and 15% CSF. As level of each grind of CSF increased from 6 to 15%, color of cookies became significantly less acceptable (Tables 3 and 4). Possibly the green shade of F-CSF and the yellow shade of C-CSF became more pronounced and objectionable with increased levels of those flours.

As level of F- and C-CSF increased in cookies, panel members indicated that cookies appeared to have greener appearing exteriors and interiors (Table 5). Green color was noticed more frequently in the interior than on exterior surfaces of cookies containing CSF.

The ideal color of cookies was described as golden brown, a color in few of the cookies. During preliminary work, panel members indicated that cookies baked until a golden brown colored surface resulted were often too brown on the bottom and had an objectionable bitter flavor. Thus, a shortened cooking period was necessary.

Color variations in the flour itself and in the baked product should be considered when supplementing baked products with CSF. Possibly color variations such as those encountered in this study could be masked through use of chocolate or other naturally dark flavoring substances.

Aroma. Prior to incorporation into cookies, both F- and C-CSF had distinctive nutty and slightly grassy aromas. Aroma of cookies was less acceptable at increased levels of substitution than aroma of cookies containing a lower level or no CSF.

Aroma scores for control cookies were significantly higher than those of any cookies containing CSF (Tables 3 and 4). The level of substitution of CSF appeared to have a significant effect on desirability of aroma. As level of F- or C-CSF increased, mean aroma scores decreased from acceptable to fair. Grind of CSF used had no significant effects on aroma of cookies (Table 4).

Variations in aroma may be attributed to the presence of CSF and to the level of CSF substituted for wheat flour. Possibly undesirable effects of CSF on aroma could be masked through additional and/or stronger flavoring agents. The benefits of including such flavoring agents in a food formula would depend upon the relative acceptability of other quality factors affected by use of CSF.

<u>Flavor</u>. Flavor and aroma components of a food are often difficult to separate. Flavor of control cookies was scored significantly higher than that of cookies containing CSF (Tables 3 and 4). As with aroma, flavor was significantly affected by level of CSF (Table 4). As level of CSF increased, mean flavor scores decreased to a quality judged fair by panel members (Table 3).

Indications of other than desirable flavor notes increased as level of CSF increased (Table 5). Flavor of control cookies and of cookies containing 6% F-CSF were considered nutty, whereas only flavors of cookies containing CSF were considered beany, grassy, or straw-like, oily, and bitter. Nutty, grassy and associated flavors, and bitter flavors may be attributed to the flavor of CSF; whereas, oily flavors may be attributed to effects of CSF on gluten structure of cookie dough. Possibly gluten strands did not hydrate sufficiently; thus, fat did not have an opportunity

to form lakes and pools among gluten strands. Perhaps those conditions contributed to localization of large amounts of fat or to a more pronounced concentration of fat in the dough.

The rather bland flavor of the control cookie might have served to amplify any flavor differences caused by substitution of CSF. Such differences might prove unacceptable; but, they might be masked through use of other flavoring agents. Cinnamon used in the present study appeared to have little effect in masking flavor of CSF in cookies, although it did serve to mask black specks noted in F-CSF.

Texture. As with objective values for tenderness, subjective scores for texture indicated that texture of cookies varied significantly (Table 3). Texture of cookies was significantly affected by grind and level of CSF (Tables 3 and 4); as level of CSF increased, mean texture scores of cookies decreased. Increasing levels of CSF accompanied by water binding and underhydrated gluten may have contributed to a decrease in acceptability of cookie texture. Texture of cookies containing F-CSF were judged fair (Table 3). Texture of all cookies containing CSF were described as tough and hard, although cookies containing C-CSF were described as tough and hard more frequently than cookies containing F-CSF (Table 5).

Texture of control cookies was scored significantly higher than that of cookies containing CSF even though all were judged acceptable (Tables 3 and 4). "Tender" was the descriptive term checked most frequently for control cookies (Table 5). In a few instances, control cookies were labeled as unevenly baked.

Acceptability. The test of success of a new or modified food product is in the consumer's willingness to purchase and consume that product

repeatedly. Thus, a descriptive scale was designed to assess acceptability in those terms.

There were significant differences in preference for cookies (Tables 3 and 4). As the level of CSF substituted in cookies was increased, a significant decrease in acceptability was noted. Panel members indicated that they would neither purchase nor consume cookies containing 15% CSF. However, they indicated that they would consume but not purchase cookies containing 6% CSF. Considering that mean acceptability scores for control cookies were slightly but significantly higher than those for cookies containing CSF, a level of 6% CSF might prove to be acceptable in baked products.

Supplementation of 15% CSF might result in acceptable products through alteration of cookie formula or through use of another type of baked product. Addition of more moisture in the form of water or milk and of leavening agents might possibly improve some of the relatively undesirable products encountered in this study.

### Amino Acid Analyses

Recommended essential amino acid intake and analyzed and calculated amino acid content of cookies are shown in Tables 6 and 7, respectively. CSF contributed to increased levels of all essential amino acids. In most instances, cookies containing C-CSF had a greater quantity of each essential amino acid than did cookies containing F-CSF (Table 7). Absence of heat in processing C-CSF might explain the higher levels of amino acids in that flour.

Each of the samples analyzed evidenced a ratio of nonessential to

Table 6. Recommended essential amino acid intake for adult males.

	Recommended dai	ly intake (mg/day)
	Minimum dai	1y allowances
Amino acid	FAO <u>a</u>	usa <sup>b</sup>
Isoleucine	676	700
Leucine	644	1100
Lysine	572	800
Phenylalanine		
without tyrosine in presence of 1100 mg tyrosine/day	864 	1100 300
Methionine		lates mad spire
without cystine in presence of 810 mg cystine/day in presence of cystine	  852	1100 200 
Threonine	242	500
Tryptophan	188	250
Valine	572	800

Amino acid requirements were calculated on the basis of a reference man of 65 Kg (NAS, 1968) and on the basis of the amino acid requirements listed by Altschul (1965) in mg/Kg/day.

 $<sup>\</sup>frac{b}{}$  Williams, 1959.

Table 7. Analyzed amino acid content of cookies, amino acid content per cookie, and distribution of amino acids in cookies.

	Analyses (mg/g sample)	mg/g sa	mple)	Amino a	Amino acid content cookie (mg) <sup>a</sup>	int per	Dist	Distribution of amino acids	of Jo
Cookies	辺	뒏	al.	д	ĒΉ	ט	д	ĽΉ	ນ
Amino acid									
Isoleucine	2.10	2.26	2.37	24.15	27.57	29.15			
Leucine	3.92	4.23	4.47	45.10	51.60	54.98			
Lysinef	0.79	1.03	1.10	9.03	12.57	13.53			
Phenylalanine	2.28	2.69	2.65	26.20	32.81	32.60			
Methionine	1.28	1.30	1.32	14.70	15.86	16.24			
Threonine	1.58	1.79	1.94	18.17	21.84	23.86			
Tryptophan	0.38	0.52	0.57	4.40	6.32	7.02			
Valine	2.57	2.86	2.98	29.56	34.89	36.65			
Arginine	1.71	3.12	3.34	19.76	38.06	41.08			
Aspartic acid	3.26	4.41	4.61	37.50	53.80	56.70			
Alanine	2.25	2.64	2.72	25.88	32.31	33.46			
Histidine	0.52	0.45	0.42	00.9	5.50	5.12			
Glutamic acid	16.60	17.20	17.90	190.90	209.84	220.17			
Glycine	1.49	1.50	1.58	17.13	18.30	19.43			
Proline	4.49	4.24	4.30	51.64	51.73	52.89			
Serine	2.72	2.89	3.25	31.28	35.26	39.98			
Tyrosine	1.55	1.75	1.78	17.82	21.35	21.89			
% essential amino acids							30.1	31.3	30.3
% nonessential amino acids							6.69	9.89	9.69
			_	(					-

 $\frac{a}{}$  Average weight of one cookie was: P = 11.5g; F = 12.2g; C = 12.3g.

 $<sup>^{</sup>m b}$  Based on the amino acid content of one cookie.

 $<sup>\</sup>frac{c}{c}$  Control.

 $<sup>\</sup>frac{d}{d}$  Fine cottonseed flour substituted for 6% wheat flour.

e Coarse cottonseed flour substituted for 6% wheat flour.

 $<sup>\</sup>pm$  Lysine was reported as total lysine (bound and epsilon amino free lysine).

essential amino acids similar to that noted by Bressani (1965) for CSF.

Non-supplemented cookies had a similar ratio of nonessential to essential amino acids (Table 7). Thus, it appeared that supplementation with CSF did little to alter that ratio in favor of essential amino acids. Although the significance of such a ratio is not fully understood, Scrimshaw et al. (1966) stated that it might be more important than previously thought.

From inspection of amino acid requirements and amounts of amino acids in cookies, it appeared that a cookie supplemented at the 6% level could make a small contribution to amino acid requirements (Tables 6 and 7).

#### Selected Correlations

Correlation coefficients between selected factors apparently varied from treatment to treatment. Variations for those selected factors were not significant. Few of the correlation coefficients calculated were significant (Table 8). Of those that were significant, all were in the low or moderate range.

Volume of cookies was moderately correlated (r = +0.649) with calculated specific volume. A larger calculated specific volume indicated more volume per unit weight. Thus, as the specific volume of cookies increased, volume increased. Variations in specific volume may be attributable to level and grind of CSF substituted for wheat flour in cookies.

Surface appearance of cookies was slightly correlated with texture (r = +0.332) and acceptability (r = +0.375), while flavor and texture were moderately correlated (r = +0.452). Presence of CSF was a factor affecting surface appearance, texture, and flavor of cookies (Tables 3 and 4). It is plausible that acceptability of cookies was also related to the use

Table 8. Correlation coefficients and significance  $\frac{a}{a}$  among selected objective and subjective measurements.

Factors	Tender- ness <u>b</u>	Width	Height	W/H <sup>C</sup>	Volume	Weight	v/w <u>d</u>	Moisture
Tenderness	1.000							
Width		1.000						
Height			1.000					
W/H <sup>C</sup>				1.000				
Volume	-0.128				1.000			
Weight	0.111					1.000		
$V/W^{\underline{d}}$	-0.217	0.209	0.080		0.649*	-0.210	1.000	
Moisture	-0.240	-0.236	0.170	-0.223	0.016	0.014	-0.003	1.000
рН	% <b></b>							
(b) <u>e</u>								
Humidity	0.051							-0.117
Appearance: Surface					<b></b>	som was con term		
Shape						mont mine table think		
Color								
Aroma								
Flavor								
Texture	-0.094	0.090	-0.018	0.043	-0.013	-0.172	0.020	
Accept- ability					-0.139	<u> </u>		

 $<sup>\</sup>frac{a}{}$  \*, P = 0.05.

 $<sup>\</sup>frac{b}{}$  (breaking strength).

 $<sup>\</sup>frac{c}{}$  Spread (width/height).

 $<sup>\</sup>frac{d}{d}$  Specific volume (volume/weight).

 $<sup>\</sup>frac{e}{}$  Color: yellowness (b).

Table 8. (concl.)

( <del>************</del>			Арр	earance			<del></del>		
pН	(b) <u>e</u>	Humidity	Surface	Shape	Color	Aroma	Flavor	Texture	Accept- ability

1.000 1.000 1.000 0.084 -0.071 1.000 ---- -0.324 1.000 1.000 -0.012 -0.163 1.000 1.000 0.452\* 0.332\* 1.000 -0.0150.375 \* 0.586 \* 0.560 \* 0.439 \* 0.724 \* 1.000 -0.023

of CSF. Five factors were significantly correlated with acceptability of cookies (Table 8). Relative desirability of all characteristics except shape appeared to be affected by the presence of CSF (Tables 3 and 4).

#### SUMMARY

Effects of substitution of glandless cottonseed flour (CSF) on quality, acceptability, and amino acid content of a plain sugar cookie were investigated. Fine, -100-mesh (F), and coarse, -80-mesh (C), CSF were each substituted at levels of 6 and 15% for wheat flour.

Throughout the study, C-CSF in cookies appeared to absorb more water and/or to bind water more securely than F-CSF. Significant decreases were noted in tenderness, height, volume and specific volume; and significant increases in weight with increasing levels of CSF in cookies. It was believed that water binding properties of CSF protein adversely affected tenderness, width, height, spread, volume, weight and specific volume of cookies. Detectable moisture content was not affected significantly by grind or level of CSF.

On the basis of subjective evaluations, it appeared that use of CSF in cookies at a level of 6% was more acceptable than use at a level of 15%. Significantly lower scores for color, aroma, flavor, texture, and acceptability, but significantly higher scores for surface appearance resulted when 6% CSF was substituted for wheat flour and when the level of CSF was increased to 15%. Texture of cookies containing C-CSF was scored significantly lower than that of cookies containing F-CSF.

Few of the correlation coefficients calculated were significant. Of those that were significant, all were in the low or moderate range.

Supplementation with CSF did not alter appreciably the ratio of essential to nonessential amino acids. However, amounts of all amino acids were increased through supplementation of cookies with 6% CSF.

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APPENDIX

Table 9. Analyses of cottonseed flours.

	Grind of cotto	onseed flour
Factor	Fine a	Coarse b
2	(%)	(%)
Moisture <sup>C</sup>	9.5	2.69 <u>d</u>
Nitrogen	8.75	9.28
Protein	54.7	58.00
Protein (MFB) e	60.5	dial 999 dais Gas
Crude fiber	3.3	2.05
Gossypol-free	0.04	0.0199
Gossypol-total	0.06	0.0339
Lipids	2.4	0.45 <sup>£</sup>
Ash	6.7	
N-solubility	95.7	99.73
Particle size	-100.0 mesh	-80.0 mesh

 $<sup>\</sup>frac{a}{}$  Smith (1969).

 $<sup>\</sup>frac{b}{}$  Lawhon (1970).

c Moisture determined at time of use: fine, 3.8%; coarse, 2.7%.

 $<sup>\</sup>frac{\mathrm{d}}{\mathrm{m}}$  Moisture and volatiles.

e Moisture free basis.

 $<sup>\</sup>frac{f}{}$  oil.

Table 10. Experimental recipes.

Ingredients	Control	Varia	bles
		6%, Fine or coarse CSF	15%, Fine or coarse CSF
	(g)	(g)	(g)
Hydrogenated shortening	94.0	94.0	94.0
Egg	48.0	48.0	48.0
Sugar (granulated cane)	150.0	150.0	150.0
Cream of tartar 🖟	3.1	3.1	3.1
Soda	1.9	1.9	1.9
Salt	0.8	0.8	0.8
All-purpose flour	151.0	141.9	128.3
Glandless cottonseed flour	programme and allow	9.1	22.7

All mixing was done in a 4-qt mixing bowl with a Kitchen-Aid 10-speed electric mixer. Six fresh eggs were blended 1 min at speed 1 (low) and 48 g of total mixture weighed for each batch of cookies. Flour, soda, cream of tartar, and cinnamon were sifted together 4 times. Shortening, sugar, and egg were creamed 2 min at speed 2. The sides and bottom of the mixing bowl and beater were scraped with a rubber spatula, and the mixture creamed 1 min more at speed 6. The sifted dry ingredients were folded into the creamed mixture for 30 sec at speed 1. Again, sides and bottom of the bowl and beater were scraped with a rubber spatula, and mixing continued for 1 min at speed 2. The beater was scraped clean of dough and the dough transferred to wax paper for rolling.

Form 1. Randomized complete block design for preparation of products.

Day		Tre	eatment	s <u>a</u>	
1	5	1	2	3	4
2	1	2	4	3	5
3	4	1	2	3	5
4	1	3	4	5	2
5	4	5	2	3	1
6	1	2	3	4	5
7	2	3	5	4	1
8	4	1	5	3	2
9	5	1	2	3	4
10	4	1	3	5	2

a Treatments:

- 1 = Control product, using all wheat flour.
- 2 = Fine cottonseed flour substituted for 6% wheat
- 3 = Fine cottonseed flour substituted for 15% wheat flour.
- 4 = Coarse cottonseed flour substituted for 6% wheat flour.
- 5 = Coarse cottonseed flour substituted for 15% wheat flour.

## Back of ovens

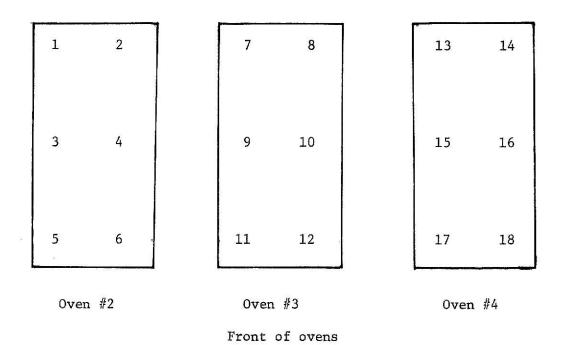
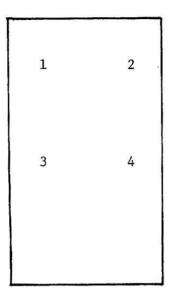


Figure 1. Oven design used in baking cookies for all tests except tenderness.

Form 2. Randomized design used to select cookies for objective and subjective evaluations.

 $<sup>\</sup>frac{a}{c}$  Numbers represent cookies so designated in the oven design (Figure 1, Appendix).

Back of oven



Oven #1

Front of oven

Figure 2. Oven design used in baking cookies for tests of tenderness.

Form 3. Randomized design used to select cookies tested for tenderness (breaking strength).

Day	Cook:	ies <u>a</u> b
1	1	4
2	4	2
3	4	3
4	2	1
5	1	4
6	1	3
7	3	2
8	3	4
9	1	2
10	1	4

 $<sup>\</sup>frac{a}{c}$  Numbers represent cookies so designated in the oven design (Figure 2, Appendix).

 $<sup>\</sup>frac{b}{c}$  Cookies tested for tenderness.

Table 11. Calculated protein value of cookies

					Protein
Treatment	Ingredient	% protein	Ingredient (g)	g/batch	g/baked cookie
Control	All-purpose wheat flour	10.5ª	151.0	15.9	
	Whole egg	12.9ª	48.0	4.2	0.056 <u>b</u>
Fine cottonseed flour (CSF) 6%	All-purpose	ال 10	6,171	6 71	
	Whole egg	12.9ª	48.0	6.2	
	GSF	60.5 <sup>c</sup>	9.1	5.5	Q.
P. P.	٥٠٠٠ - ٢١٨				0.065
%CT	All-purpose wheat flour	$10.5^{a}$	128.4	13.4	
	Whole egg	12.9ª	48.0	6.2	
	CSF	<del>5</del> 60.96	22.6	13.7	6
					0.082

Table 11. (concl.)

					Protein
Treatment	Ingredient	% protein	Ingredient (g)	g/batch	g/baked cookie
Coarse CSF					
%9	All-purpose wheat flour	10.5ª	141.9	14.9	
	Whole egg	12.9ª	48.0	6.2	
	CSF	58.0 <del>d</del>	9.1	5.3	ą
					0.064
15%	All-purpose wheat flour	10.54	128.4	14.9	
	Whole egg	12.9ª	48.0	6.2	
	CSF	58.0 <del>4</del>	22.6	13.1	

a Watt et al. (1963).

b Samples analyzed for amino acids.

 $\frac{c}{}$  Moisture free basis, Smith (1969).

 $\frac{d}{}$  Moisture containing basis, Lawhon (1970).

Form 4. Score card.

Factors	Desired traits	Descriptive terms		San	Sample code	ode	
		4	425	378	314	136	535
Appearance: Surface	Well-broken top containing numerous small islands						
		Unbroken surface					
Shape	Evenly rounded						
		Uneven, pitted edges					
Color	Golden brown						
		exterior					
		Green interior (Crumb)					
Palatibility:	:						
Aroma	Pleasant, light, no "off"						
	notes						
Flavor	Sweet, pleasing; no pro-						
	nounced oil itavors						
		Nutty					
		Beany, grassy, straw-					
		like					
		Oily					
		Bitter					
Texture (crispness)	Crisp, crumbly, not soggy, chewey, tough, or elastic						
	× ×	Tough					
		Hard					
		Tender					
		Unevenly cooked					
Acceptability							
7		1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1			, 10		
score key ior appearance	oo	ceptability:		,	comments:	··	
and paralibility	L	would purchase and consume occasionally.					
Highly desirable	ble 5 Would purchase and	and consume, but less					

Would consume cookies if they were around, Would neither purchase nor consume but would not purchase them. frequently than 4. cookies. 1 2 3 4 Unacceptable Acceptable Desirable Fair

Please check descriptive terms which you feel apply to the cookies.

 $\mathfrak{C}$ 

2

Table 12. Analyses of variance for objective and subjective evaluation of cookies attributable to treatments and replications.

Source	Analyses of variance				
	D/F	MS	F-value	Sig.	
Objective measurements			*9		
Tenderness (breaking					
strength)					
Treatments	4	213631.250	73.116	*	
Replications	9	11624.691	3.979	*	
Error	36	2921.799			
Width					
Treatments	4	0.516	8.693	*	
Replications	9	0.051	0.865	ns	
Error	36	0.059			
Height					
Treatments	4	0.136	9.387	*	
Replications	9	0.010	0.660	ns	
Error	36	0.015	0.000	110	
EIIOI	30	0.013			
Spread (width/height)					
Treatments	4	0.609	4.526	*	
Replications	9	0.069	0.514	ns	
Error	36	0.135			
Volume					
Treatments	4	955.749	28.042	*	
Replications	9	41.167	1.208	ns	
Error	36	34.082			
Weight					
Treatments	4	48.730	20.333	*	
Replications	9	1.824	0.761	ns	
Error	36	2.397	01701		
Specific volume					
(volume/weight)					
Treatments	4	0.644	41.890	*	
Replications	9	0.017	1.076	ns	
Error	36	0.017	1.0/0	us	
ELIUI	30	0.013			
Moisture		¥ 272	x 2x=	<b>22</b> 7	
Treatments	4	0.199	2.699	*	
Replications	9	0.127	1.714	ns	
Error	36	0.074	ii.		

Table 12. (cont'd.)

Source	Analyses of variance					
	D/F	MS	F-value	Sig. <u>a</u>		
pН						
Treatments	4	0.041	10.564	*		
Replications	9	0.005	1.304	ns		
Error	36	0.004				
Color: reflectance (Rd)						
Treatments	4	107.194	38.221	*		
Replications	9	7.718	2.752	*		
Error	36	2.805				
Color: redness (a)						
Treatments	4	2.285	1.374	ns		
Replications	9	4.072	2.448	*		
Error	36	1.663				
Color: yellowness	(b)					
Treatments	4	9.471	39.655	*		
Replications	9	1.339	5.606	*		
Error	36	0.239				
Color: degree of redness (a/b)						
Treatments	4	0.004	1.110	ns		
Replications	9	0.010	2.712	*		
Error	36	0.004				
Palatability scores						
Appearance: surfac	e					
Treatments	4	0.094	2.662	*		
Replications	9	0.212	0.560	ns		
Error	36	0.353				
Appearance: shape						
Treatments	4	0.474	1.369	ns		
Replications	9	1.630	0.470	ns		
Error	36	0.347				
Appearance: color				0.40		
Treatments	4	0.997	4.659	*		
Replications	9	0.193	0.931	ns		
Error	36	0.207				

Table 12. (concl.)

Source	Analyses of variance				
	D/F	MS	F-value	Sig.	
Aroma					
Treatments	4	1.399	15.252	*	
Replications	9	0.240	2.616	*	
Error	36	0.092			
Flavor					
Treatments	4	2.135	18.152	*	
Replications	9	0.187	1.591	ns	
Error	36	0.118			
Texture					
Treatments	4	3.076	21.507	*	
Replications	9	0.143	0.997	ns	
Error	36	0.143			
Acceptability					
Treatments	4	1.958	21.360	*	
Replications	9	0.034	0.373	ns	
Error	36	0.092			

 $<sup>\</sup>frac{a}{}$  \*, P = 0.05; ns = not significant.

# EFFECTS OF VARYING GRIND AND LEVEL OF GLANDLESS COTTONSEED FLOUR ON QUALITY AND AMINO ACID CONTENT OF PLAIN SUGAR COOKIES

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Effects of substitution of glandless cottonseed flour (CSF) on quality, acceptability, and amino acid content of a plain sugar cookie were investigated. Fine, -100~mesh (F), and coarse, -80-mesh (C), CSF were each substituted at levels of 6 and 15% for wheat flour.

Throughout the study, C-CSF in cookies appeared to absorb more water and/or to bind water more securely than F-CSF. Significant decreases were noted in tenderness, height, volume and specific volume; and significant increases in weight, with increasing levels of CSF in cookies. It was believed that water binding properties of CSF protein adversely affected tenderness, width, height, spread, volume, weight and specific volume of cookies. Detectable moisture content was not affected significantly by grind or level of CSF.

On the basis of subjective evaluations, it appeared that use of CSF in cookies at a level of 6% was more acceptable than use at a level of 15%. Significantly lower scores for color, aroma, flavor, texture, and acceptability, but significantly higher scores for surface appearance resulted when 6% CSF was substituted for wheat flour and when the level of CSF was increased to 15%. Texture of cookies containing C-CSF was scored significantly lower than that of cookies containing F-CSF.

Few of the correlation coefficients calculated were significant. Of those that were significant, all were in the low or moderate range.

Supplementation with CSF did not alter appreciably the ratio of essential to nonessential amino acids. However, amounts of all amino acids were increased through supplementation of cookies with 6% CSF.