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THE UNIVERSITY OF ALBERTA

THE APPLICATION OF RESPONSE SURFACE METHODOLOGY
TO THE DEVELOPMENT OF RICE FLOUR YEAST BREADS

BY

GLADYS LORRAINE YLIMAKI

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH

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OF MASTER OF SCIENCE

IN

FOODS

FACULTY OF HOME ECONOMICS

EDMONTON, ALBERTA

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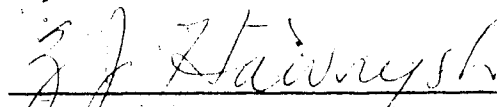
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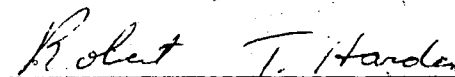
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in partial fulfilment of the requirements for the degree of Master of Science
in Foods.

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ABSTRACT

Gluten-free yeast breads, suitable for use by individuals with celiac disease, wheat allergies or dermatitis herpetiformis, were developed from rice flour (80%) and potato starch (20%). Using objective and sensory measurements as responses, response surface methodology (RSM) was utilized to find carboxymethylcellulose (CMC) - hydroxypropylmethylcellulose (HPMC) - water combinations which could successfully replace gluten in rice flour yeast breads made from each of three different locally available rice flours (A and B, medium grain rice; C, long grain rice). During ingredient screening experiments, the three variables (CMC, HPMC and water) were shown to influence the volume and loaf shape of rice breads. However, the actual levels of CMC and water used in the bread formulations had more of an effect on the objective and sensory responses measured than did the level of HPMC. The successful production of rice flour yeast breads was dependent upon the type of rice flour used and the amounts of gums (CMC and HPMC) and water incorporated into the formulation.

Using Flour A, rice bread formulations were found that produced breads which met wheat (white) bread reference standards for the objective measurements of specific volume, crust color 'L, a and b' values, crumb color 'L, a and b' values, Instron firmness, percent moisture and the sensory measurements of moistness, cohesiveness, yeasty flavor, adhesiveness, top crust color, crumb color, cell size uniformity and cell wall thickness. Flour A rice breads met the largest number of objective and sensory reference wheat bread standards

simultaneously (17 out of 25), as compared to 16 for Flour B and 12 for Flour C. A 42-member consumer panel (consisting of 23 celiacs and 19 non-celiacs) judged Flour A and B rice breads to be similar and more acceptable than Flour C rice breads. This study has provided valuable, much needed information about the objective and sensory evaluation of gluten-free breads. In addition, the successful application of RSM to the development of gluten-free rice flour yeast breads indicates the feasibility of its potential use in the production of other special diet products.

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I. Introduction

Bread is a popular menu item at all meals as well as a good source of nutrients and calories. To be utilized, a bread product must be accepted, consumed and enjoyed by consumers. Important quality attributes influencing bread acceptability include appearance, texture and flavor. The development of an acceptable gluten-free yeast bread, comparable to wheat flour yeast bread, for individuals who cannot tolerate gluten, is essential. However, attempts to develop such a product have thus far been unsuccessful.

At present, most gluten-free bread products available to consumers are made from wheat starch. However, many individuals sensitive to wheat gluten cannot tolerate even the very small amount of gluten present in wheat starch. Although some rice flour yeast breads are available commercially, the quality, especially the flavor, texture and appearance of these products is undesirable. Gluten replacements such as gums and surfactants may enhance the structure and textural properties of gluten-free yeast breads. However, published reports of the sensory (taste panel) and objective (instrumental) evaluations of the quality attributes of rice flour yeast breads, made with the addition of gums and surfactants, are lacking. In addition, response surface methodology (RSM), a statistical technique which is particularly appropriate for product formulation, has not been utilized for the development of gluten-free bread.

Therefore, the objective of this research was to develop an acceptable gluten-free rice flour yeast bread for individuals with

celiac disease, dermatitis herpetiformis and wheat allergies, through the use of response surface methodology. In addition, information on the gluten-free yeast breads currently being used by celiacs, and in Alberta hospitals, was obtained through mailed questionnaires.

LITERATURE REVIEW

Celiac disease

Celiac disease, also known as gluten sensitive enteropathy, is a chronic illness which occurs in adults and children (Anonymous, 1981). One in every 2,500 persons in the United States is affected by celiac disease (Hartsook, 1984). The incidence of celiac disease in Canada is similar. Although celiacs represent a small portion of the population, their illness is significant since dietary alterations must be followed for a lifetime.

Celiac disease is a disorder in which mucosal damage and dysfunction of the small intestine, primarily the proximal portion, is causally linked to the intake of gluten containing foods (Kowlessor, 1972; Booth, 1977; Chandra and Sahni 1981). Specifically, it is the gliadin of gluten which causes the intestinal mucosal damage. The villi of the small intestine, through which absorption of nutrients takes place, are usually absent in celiac disease, leaving the mucosa quite flat (Anderson et al., 1972; Hartsook, 1982). If the disease is untreated, symptoms of malabsorption and malnutrition, such as diarrhea, bloating, abdominal cramps, weight loss and anemia, may appear (Kasarda, 1972; Hartsook, 1982; Davidson, 1984). The severity of the symptoms differs among individuals; however, intestinal damage may occur even if there are no obvious symptoms (Campbell, 1984). Thus, strict adherence to a gliadin-free diet, including the removal from the diet of wheat, rye, barley and oats, is the only long-term treatment (Kasarda, 1978; Hartsook, 1984).

Since the gluten-free diet is a permanent treatment for celiac

disease, a correct diagnosis is important. Both an abnormal biopsy sample, taken from the lining of the small intestine, and a positive response to the gluten-free diet are necessary to diagnose celiac disease' (Davidson, 1984; Hartsook, 1984). Once gluten is eliminated from the diet, improvement in symptoms can occur within days. Other diseases and conditions can cause symptoms similar to those of celiac disease (Hartsook, 1984).

Dermatitis herpetiformis

Dermatitis herpetiformis, a gluten-related skin disease, appears as small, itchy blisters on the skin (Hartsook, 1982). Small intestinal biopsy samples of individuals with dermatitis herpetiformis often show damage similar to that seen in celiac disease (Hartsook, 1982). Gluten withdrawal clears the skin lesions and improves the damage to the intestinal mucosa (Andersson et al., 1984). In the United States, the incidence of dermatitis herpetiformis is not known, but is probably similar to that of celiac disease (Kasarda, 1978).

Wheat allergies

A wheat-free diet may also be necessary for a small number of individuals who suffer from wheat intolerance and allergies. These allergies are less well defined and less familiar than celiac disease (Kulp et al., 1974). The wheat albumins and globulins are the primary cause for symptoms of wheat allergies (Goodwin and Rawcliffe, 1983). The exact incidence of food allergies in individuals is unknown, but estimates range from 0.3% to 7.5% of the population (Taylor, 1984).

Wheat starch

Starch makes up approximately 70% of the wheat endosperm

(MacRitchie, 1980). Both large and small starch granules, ranging from 2-35 microns in diameter (Olkku and Rha, 1978), are present. The large amount of protein in wheat makes it difficult to extract the starch from wheat flour (Glicksman, 1969). Therefore, the protein content of wheat starch varies depending on the effectiveness of the separation process.

Wheat starch in Canada contains approximately 0.15% to 0.3% protein (Bell et al., 1981; Campbell, 1982). Some researchers (Kulp et al., 1974; Campbell, 1982) consider wheat starch to be acceptable for celiacs. However, among celiacs there is a large variation in tolerance to gluten. In some cases, ingestion of even minute amounts of gliadin by sensitive individuals can cause life threatening reactions (Hartsook, 1984). Locally, less than 30% of the membership of the Edmonton Celiac Association can tolerate wheat starch (Scott, 1984). Some hospitals no longer allow wheat starch on a gluten-free diet, since it may cause symptoms in sensitive individuals (Campbell, 1984). The Canadian Celiac Association, which originally allowed wheat starch on the gluten-free diet, plans to change their National Handbook and to list wheat starch as a non-allowable food on the diet (Friesen, 1987).

Low-gluten and gluten-free yeast breads

Bread is an important staple in the Canadian diet. Restricted use or omission of bread from the diet results in the loss of an important source of nutrients and calories (Sorenson, 1970). A bread product with an undesirable flavor or texture will not be consumed. Thus, an acceptable, palatable yeast bread replacement for those who cannot

tolerate wheat flour is of importance.

Since the gluten in wheat flour is mainly responsible for the light, open but rigid structure of bread (McGreer, 1967), gluten-free yeast breads need gluten substitutes for structural support and air retention. Gluten replacements should increase dough water binding capacity since starch granules lack the ability to bind sufficient water at the dough stage to achieve the proper degree of gelatinization during baking (Kulp et al., 1974).

Most research on the development of low-gluten and gluten-free yeast breads (Pearson, 1960; Steele et al., 1965; Landsman and Wills, 1968; Jongh et al., 1968; Sorenson, 1970; Smith, 1971, 1974; Christianson et al., 1974; Kulp et al., 1974; Ranhotra et al., 1975; Johnson and Penfield, 1976) has utilized wheat starch. Reports (McGreer, 1967; Sorenson, 1970) indicate that yeast breads made from wheat starch had a muffin-like appearance, a heavy, coarse crumb structure and a low specific volume. In general, wheat starch yeast breads were tasteless, crumbled easily and became stale quickly (Bell et al., 1981). Results of a questionnaire indicated that patients eating wheat starch bread did not find it to be a satisfactory substitute for regular bread (Johnson and Penfield, 1976). Pomeranz (1969) stated that the disparity between regular and gluten-free baked products was too great to even designate the latter as bread.

Rice

Rice naturally contains no gluten (Nishita, 1973; Campbell, 1982) and is acceptable to celiacs as well as most allergic individuals. Since most rice flours are made from broken grains of milled rice,

their chemical composition is the same as that of the whole rice (Deobald, 1972). The starch granules of rice are the smallest found in commercial starches, only 3-8 microns in diameter (Furia, 1968), and exhibit slow, restricted swelling during gelatinization (Glicksman, 1969). Rice varietal differences influence the gelatinization temperature, amylose content and gel properties of rice (Nishita et al., 1976), as well as the bread making qualities of the resultant flour.

Rice is similar to wheat in nutritional value, except for a lower protein content. While wheat flour is enriched with B-vitamins and iron to replace the vitamins and minerals lost during milling, rice flour is not enriched (Campbell, 1984).

Rice flour yeast bread

There are few published reports on gluten-free rice bread development. Although previous research (Nishita, 1973, 1977; Nishita et al., 1976; Nishita and Bean, 1979) provides a basis for product development, the flavor and texture of the rice flour yeast bread requires improvement. Moreover, duplication of gluten-free rice bread recipes is difficult because of varietal differences in rice, which affect bread quality. Flour made from white, short or medium grain rice produces bread with a better crumb texture (less dry and crumbly) than flour from long grain rice (Nishita, 1977).

Bean and Nishita (1983) and Nishita (1984) reported that the most acceptable crumb grain and texture in rice flour yeast breads was produced from rice flour with a low amylose content (<20%) and a low gelatinization temperature (<67°C). Nishita (1977) suggested that

brown rice flour, which was unsatisfactory in rice breads, may have contained some lipid materials which interfered with the methylcellulose gum included in the bread formula. In addition, the amount of water used in the rice bread formulation was critical (Nishita et al., 1976). Insufficient water produced a stiff dough that would not rise during proofing while excessive water caused dough overexpansion during baking (Nishita et al., 1976). However, the major problems associated with rice bread production were due to the absence of gluten and were thus similar to those encountered in wheat starch bread production (Nishita et al., 1976). Thus, wheat starch research is relevant and useful in rice bread development.

Potato starch

Potato starch is an efficient water retainer (Bennion, 1967). The potato starch granules are the largest of any of the common commercial starches, ranging from 15-100 microns in diameter (Furia, 1968). During heating and gelatinization, the potato starch granules swell rapidly and enormously (Glicksman, 1969).

Nishita (1973) found that the volume of gluten-free rice flour bread was improved by a 20% substitution of potato starch for rice flour and attributed the increased volume to the ability of potato starch granules to absorb more water and swell more than rice starch granules. In addition, potato starch softened the texture, reduced graininess and produced a bread with a more open cell structure than the 100% rice flour bread (Nishita, 1973). Potato starch pastes tend to disintegrate and become less viscous when cooled. In contrast, rice starch pastes increase in viscosity when cooled, due to the retrograda-

tion of linear amylose chains (Sanderson, 1981). The linear molecules in potato starch have a low tendency to retrograde (deMan, 1979), which may account for the softening of the texture of rice breads when potato starch is added. When combined with hydroxypropylmethylcellulose, potato starch produced a fine structure and improved the volume of sorghum flour breads (Hart et al., 1970).

Gums

Gums, or hydrocolloids, are complex polysaccharides frequently used to impart desirable textural and functional properties to food products without contributing to the nutritive value, taste or aroma of the finished product (Frost et al., 1984; Krumel and Sarkar, 1975). When dispersed in water, gums increase viscosity, which is the basis for their use as thickening and stabilizing agents (Glicksman, 1969).

There are a number of different types of hydrocolloids including plant seed gums (locust bean and guar gums), cellulose derivatives (methylcellulose, carboxymethylcellulose and hydroxypropylmethylcellulose) and fermentation gums (xanthan gum and dextrans). Each hydrocolloid type contains a family of chemical compounds, which vary in molecular weight, number and position of functional groups, as well as arrangements of the basic units (Balmaceda et al., 1973). Within these families are groups of gums which have a common background and structure, but with specific individual functions and distinctive properties which must be considered for effective use (Glicksman, 1969). For example, all cellulose gums are not the same and often cannot be used interchangeably. Variations between gums can include differences in viscosity, particle size and rate of hydration.

Two or three gums are often more effective than any of the individual gums alone (Glicksman, 1969). The use of gum combinations allows both the functional properties of the individual gums and the synergistic interactions between gums to be applied.

A number of gums have been employed as gluten substitutes in the development of low-gluten and gluten-free yeast breads. Gluten replacements which have had variable success in starch and non-wheat breads, when used alone or in combination, include sodium carboxymethylcellulose (McGreer, 1967; Kim and De Ruiter, 1968; Landsman and Wills, 1968; Smith, 1971, 1974; Kulp et al., 1974), hydroxypropylmethylcellulose (Kim and De Ruiter, 1969; Hart et al., 1970; Sorenson, 1970; Bradley, 1972; Nishita, 1973), xanthan gum (Christianson et al., 1974; Kulp et al., 1974; Ranhotra et al., 1975) and guar gum (Kim and De Ruiter, 1968; Smith, 1971).

Particularly relevant to starch breads is the fact that gums, such as sodium carboxymethylcellulose (CMC) and guar gum, can interact synergistically with some starches to increase viscosity (Carlson et al., 1962; Ganz, 1966). Kulp et al. (1974) found the consistency of the batter to be the most critical factor in the production of wheat starch breads. In gluten-free breads, sufficient batter viscosity is needed for gas retention, both during fermentation and the early stages of baking (Osman, 1975). Xanthan gum also interacts with starch, but rather than increasing viscosity, xanthan gum acts as a protective colloid, increasing the stability of starch to heat and minimizing the tendency of starch to retrograde and lose water (King, 1984). Amylograph curves have shown that guar, xanthan and CMC gum hastened the onset of initial paste viscosity and increased the final peak

viscosities of wheat starch during gelatinization (Christianson et al., 1981). During the initial stage of gelatinization (55-70°C) little interaction occurred between any of the gums and starch, and changes in the granule structure (i.e. swelling) were the primary causes for the viscosity achieved. However, significant differences in viscosity were evident at the second stage of gelatinization (80-94°C), during the 15-minute cooking period and during the cooling cycle. Each gum produced a distinct viscosity, indicating that each gum interacted with starch differently (Christianson et al., 1981). At 0.45% gum concentration, guar produced the solution with the highest peak viscosity (almost 10 times greater than that of the control without gum), and with a viscosity greater than that obtained with either 0.5% xanthan or CMC gum (Christianson et al., 1981).

Sodium carboxymethylcellulose. Sodium carboxymethylcellulose (CMC), or cellulose gum, is an anionic water soluble ether of cellulose (Furia, 1968). Over 250 types of CMC, of varying degrees of substitution, viscosity and particle size, are manufactured throughout the world (Ganz, 1966). The backbone of CMC is cellulose, a straight chain polymer of β -anhydroglucose units, each anhydroglucose unit having three hydroxyl groups (Stelzer and Klug, 1980). The anhydroglucose chains can vary in length from 100-200 units, the longer the chain, the higher the viscosity of the CMC derived from it (Keller, 1984).

Carboxymethylcellulose is soluble in hot and cold water and yields the fastest viscosity increase of any commercially available water soluble polymer (Anonymous, 1979; Keller, 1984). The viscosity of CMC

solutions is dependent on temperature, the viscosity decreasing with an increase in temperature (Glicksman, 1969; Balmaceda et al., 1973).

Carboxymethylcellulose has been used for the development of breads from wheat starch, yam-peanut flour and rice flour. Using a flour combination of 83% wheat starch and 17% potato flour with CMC, McGreer (1967) developed a gluten-free yeast bread that, although coarse in structure and lower in volume than typical wheat flour breads, was tender and sliced without crumbling. The level of CMC (0.5% flour wt) employed in the formulation markedly affected the quality and acceptability of the loaf. Kim and De Ruiter (1968) improved the volume of yam-peanut flour yeast breads (70% yam to 30% low-fat peanut flour) by the addition of 1% (flour wt) CMC. The water level in these breads was critical. CMC (1% flour wt), combined with 85% or 100% (flour wt) water, produced breads with volumes that were larger than that of the control bread without CMC, while a water level of 70% (flour wt) resulted in a bread volume that was lower than the control (Kim and De Ruiter, 1968). In rice flour breads, 2.1% (flour wt) CMC produced a compact, gummy bread (Nishita, 1973).

Hydroxypropylmethylcellulose. Hydroxypropylmethylcellulose (HPMC) is nonionic water-soluble methyl ether of cellulose (Furia, 1968). HPMC is prepared by treating alkali cellulose with methyl chloride and propylene oxide, resulting in methyl and hydroxypropyl substitution on the anhydroglucose units of cellulose (Glicksman, 1963). The properties of HPMC can be modified by changing the relative amounts of etherifying reagents and altering the ratios of methyl and hydroxypropyl substitutions (Greminger and Krumel, 1980). In this way, HPMC products can be produced with varying solubilities, thermal gelation

temperatures and gel textures ranging from firm to soft (Greminger and Krumel, 1980).

Hydroxypropylmethylcellulose has the ability to absorb up to 40 times its weight of water (Dow Chemical Company, 1982). Unlike most other commercial gums, HPMC is soluble in cold water, insoluble in hot water and gels at elevated temperatures. When a solution of HPMC is heated, the viscosity decreases until the HPMC thermal gelation temperature, 50-85°C (Anonymous, 1979), is reached. At the gelation temperature, HPMC viscosity rapidly increases and a three-dimensional gel structure is formed (Greminger and Krumel, 1980; Glicksman, 1969). An increase in the HPMC concentration of a solution will lower the gelation temperature (Dow Chemical Company, 1985).

Hydroxypropylmethylcellulose products have many functional properties that are advantageous for starch yeast breads. Some HPMC products have a gel point similar to the gelatinization temperatures of the starches being used to replace wheat flour (Glicksman, 1963). During the elevated baking temperatures, HPMC will gel and assist gas retention, as well as strengthen bread structure (Greminger and Krumel, 1980; Glicksman, 1969). The use of HPMC also allows more water to be included in a dough and permits more complete gelatinization of the starch (Glicksman, 1969).

Hydroxypropylmethylcellulose (0.8% flour wt) formed a fine stable cell structure in a wheat starch bread (Sorenson, 1970). Bradley (1972) incorporated an HPMC product (Methocel 65 HG 4000, Dow Chemical Company) into starch bread (98.5% wheat starch: 1.5% corn starch) with 99.7% (flour wt) water. Addition of 0.38% (flour wt) HPMC produced a fragile crumb with some large air cells and broken cell walls. As the

amount of HPMC in the formula was increased, the crumb became less fragile and more even-textured, with 0.76% HPMC producing small air cells and having somewhat breadlike texture. Beyond the 0.76% level, increased levels of HPMC produced no improvement in the bread crumb (Bradley, 1972). Glicksman et al. (1972) reported that a ratio of 5:3 by weight of HPMC to CMC (total gum weight of 4-5% flour wt) produced a starch bread with a sponge structure similar to that of wheat flour breads. The formulation also included 74% (flour wt) α -cellulose.

The volume and structure of rice flour yeast bread was improved by the addition of 3% (flour wt) HPMC (90 HG 4000) (Nishita et al., 1976). Rice bread made without HPMC did not retain gas during proofing, and after baking had the same volume as the original dough placed in the pan (Nishita et al., 1976). The type and level of HPMC was critical to the development of a 100% rice flour bread (Bean and Nishita, 1983). Eight types of HPMC, at a level of 3.0% of the rice flour weight, were studied but only two (Methocel 60 HG 4000 and 90 HG 4000, Dow Chemical Company) showed satisfactory dough strengthening action and desirable specific volumes (Nishita, 1973). All other HPMC products produced rice breads which had a low volume, poor grain and texture. When levels of 0, 1.5, 3.0, 4.5 and 7.0% (flour wt) Methocel 90 HG 4000 were compared in rice flour breads, 3.0% Methocel produced the optimum bread volume (Nishita, 1973).

In sorghum flour yeast bread, the inclusion of HPMC (2% flour wt) and 120% (flour wt) water increased gas retention and prevented the loaf from collapsing during baking (Hart et al., 1970). Compared to three other HPMC products, Methocel 4000 produced sorghum breads with the best volume (Hart et al., 1970). Sorghum breads with levels of

HPMC above 2%, rose more during proofing and had a finer crumb structure than breads with 2% HPMC, however, there was a corresponding weakening of loaf strength. The addition of 2% (flour wt) HPMC also produced a good texture in barley flour yeast bread (Hart et al., 1970).

Xanthan gum. Xanthan gum is a natural polysaccharide produced by fermentation of a pure culture of *Xanthomonas campestris* with glucose as the substrate (Rocks, 1971; Anonymous, 1979). The xanthan gum molecule has a backbone of 1,4 linked β -D-glucose units, which is identical to cellulose (Sanderson, 1982). The molecule is rendered water soluble by the presence of short side chains attached to the 3-position of every second glucose residue in the main chain (Sanderson, 1981). Two D-mannose units and a D-glucuronic acid unit make up the side chains of xanthan gum.

Xanthan gum dissolves in hot or cold water and binds water at a very high rate to produce highly viscous solutions at low concentrations. Xanthan gum viscosity is stable over a wide temperature range (King, 1984; Kelco, 1985), and thus is desirable for products that require stability during the baking process.

Xanthan gum has been incorporated into starch yeast breads with some success. In wheat starch and corn starch breads, xanthan gum improved the cohesion of starch granules and produced a bread comparable in appearance, mouthfeel, loaf volume and staling properties to a commercial wheat flour bread (Christianson et al., 1974). Christianson et al. (1974) observed that breads made from wheat starch, corn starch, high amylose corn starch and waxy corn starch, without

xanthan gum, did not rise and were brittle and coarse in texture. The addition of 6% (flour wt) xanthan gum improved the volume and texture of the breads. Lower levels of xanthan gum (3% and 4.5% flour wt) in wheat starch breads produced breads which had smaller volumes and coarser, more irregular crumb characteristics (Christianson et al. 1974). Kulp et al. (1974) found that pregelatinized starch, CMC and xanthan gum, when used individually in wheat starch breads, were satisfactory gluten replacements. However, the breads with 2.1% (flour wt) xanthan gum were superior in volume and flavor to the breads made with either pregelatinized starch or CMC. Using xanthan gum (2% flour wt) in a gluten-free, soy-fortified wheat starch bread, Ranhotra et al. (1975) also found xanthan gum to be a suitable dough stabilizer. For wheat starch bread, the use of 2% (flour wt) xanthan gum with 119% (flour wt) water has been recommended (Kelco, 1983).

In an attempt to improve the texture of starch breads, Bolam (1983) investigated the effect of separate hydration of xanthan gum prior to its incorporation into corn and wheat starch breads. Using a formula adapted from Christianson et al. (1974) (123-135% water [flour wt]), Bolam (1983) found that separate hydration of xanthan gum, with 55% of the water, altered the starch-xanthan matrix so that the starch granules gelatinized too much and were too fragile to prevent the expanding gas cells from coalescing. Prior xanthan gum hydration resulted in breads with a large cell size and irregular grain, which were hard and chewy. Bread volumes were unaffected by the prior hydration of xanthan gum (Bolam, 1983).

In rice flour breads, 2.6% (flour wt) xanthan gum with 75% (flour wt) water offered only a small increase in dough height during ferment-

tation and resulted in low baked bread volumes (Nishita, 1973).

Guar gum. Guar gum is a neutral galactomannan produced from the endosperm of the Indian cluster bean *Cyamopsis tetragonolobus* (Apling et al., 1978). Guar seed is grown in India, Pakistan and the United States (Daniel and Whistler, 1984). Guar gum has a linear backbone chain of β -D-mannose units linked 1,4 with single membered β -D-galactose units occurring as side chains on every other mannose unit (Seaman, 1980). The side branches are linked 1,6 with the guar gum backbone.

Guar gum can bind and immobilize large amounts of water. Guar can be hydrated rapidly in hot or cold water to give highly viscous solutions (Glicksman, 1969; Furia, 1968). Solutions of guar gum thin reversibly when heated and degrade irreversibly with time when an elevated temperature is maintained (Seaman, 1980).

In yeast breads made from yam and peanut flour, guar added at 1%, 4% and 8% of the flour weight with the addition of 100%, 120% and 140% (flour wt) water, respectively, greatly increased bread volume as compared to bread made without guar (Kim and De Ruiter, 1968). The two higher levels of guar produced breads with identical specific loaf volumes, but which were larger than that produced with 1% guar. Two percent guar (flour wt) added to sorghum flour bread, provided some gas retention and prevented the loaf from collapsing (Hart et al., 1970). However, bread volumes were very low. Guar (2% flour wt) also produced some improvement in the structure of barley bread (Hart et al., 1970).

Surfactants

Most surfactants, or emulsifiers, are hydrophobic fatty acid chains esterified to a hydrophilic polar group (Krog, 1977). The fatty acid chain length (from C_{12} to C_{20}) and the degree of unsaturation, which can differ according to the type of fats or fatty acids used in the manufacture of the surfactant, are both important to the functional properties of the surfactant (Krog, 1981). In food systems, surfactants perform a number of important functions such as stabilizing aerated systems, improving the texture and shelf life of starch containing products, and modifying the rheological properties of wheat doughs (Krog, 1977).

When a combination of surfactants is used in a food system, synergistic effects are often observed (Birnbaum, 1981). Multi-component emulsifier systems may be more effective than a single emulsifier system due to the geometry of the fat surface (Knightly, 1968). Because of the structural configuration, several surfactants may fit together more intimately on the surfaces than a single surfactant alone, forming a more closely packed protective layer with fewer interstitial voids (Knightly, 1968). Emulsifier systems also allow for the functional properties of each of the components to be utilized.

The interaction of surfactants with starch is very important in the production of bread. This interaction is responsible for starch-surfactant complex formations and influences the rate of starch gelatinization, the gelatinization temperature and the resulting gel strength (Schuster and Adams, 1984).

In low-gluten or gluten-free breads, the surfactants glyceroyl monostearate (GMS) (Steele et al., 1965; Jongh et al., 1968; Landsman

and Wills, 1968; Kim and De Ruiter, 1969; Hart et al., 1970; Smith 1971, 1974) and sodium-stearoyl-lactylate (SSL) (Kulp et al., 1974) have contributed desirable rheological properties when used alone or in combination with certain gums. In contrast, other researchers (Kulp et al., 1974; Nishita, 1973) have reported that surfactants had negative effects on starch and non-wheat flour breads. In rice flour doughs, surfactants (SSL and mono- and diglycerides) added with 3% (flour wt) Methocel 90 HG 4000 (HPMC), interfered with the gum-water-rice flour complex so that fermentation gases were not retained and no leavening occurred (Nishita, 1973). The use of emulsifiers (SSL and mono-diglycerides) in a xanthan gum wheat starch bread resulted in a low volume and a firm crumb (Kulp et al., 1974).

Glycerol monostearate. The production of mono- and diglycerides involves the interesterification or glycerolysis of fats and oils (Birnbaum, 1981). Glycerol monostearate (GMS) is produced from the reaction of stearic acid and glycerol. Typically, a GMS product contains approximately 50% monoglycerides, 40% diglycerides and 3% free glycerol (Kim and De Ruiter, 1968; Jongh et al., 1968).

The hydrophobic nature of monoglyceride crystals prevents the monoglycerides from absorbing water. Monoglycerides also affect water absorption of other food ingredients. In the presence of wheat flour, water absorption is decreased by the addition of certain monoglycerides which form a moisture barrier over the water absorbing flour components, particularly starch and protein (Knightly, 1968). A reduction in the rate of water absorption can also delay starch gelatinization. Longley and Miller (1971) found that monoglycerides

from long chain fatty acids affected the gelatinization of starch in this way.

Mono- and diglycerides are capable of functioning in starch complexing, protein interaction, aeration, emulsification and crystal modification (Birnbaum, 1981). In bread, monoglycerides prolong flavor, freshness and softness, improve texture, increase loaf volume and symmetry, give a finer, more uniform crumb grain and improve dough extensibility (De Renzo, 1975).

In starch breads, it is necessary to attract the starch granules to each other and to decrease their mobility (Kim and De Ruiter, 1968). This allows the doughs to more effectively retain gas and to produce breads with a greater volume, softer crumb and a more regular texture. Surfactants, such as GMS, act as starch binders (Kim and De Ruiter, 1968). When added to wheat starch doughs with 60% (flour wt) water, GMS (at levels of 0.05-1% flour wt) increased gas retention of the dough and improved overall bread quality (Jongh et al., 1968). The GMS (50% monostearate/40% distearate) was adsorbed onto the surface of the starch molecules, resulting in starch aggregation and formation of a coherent starch network. During dough mixing, air was distributed evenly throughout the small pores of the network (Jongh et al., 1968).

Thus, the wheat starch breads with GMS had a fine regular crumb structure compared to the irregular, coarse crumb of the wheat starch bread without GMS. Jongh et al. (1968) also observed that the level of GMS was important in wheat starch breads. An excess of GMS (i.e. 5.0% flour wt) resulted in a rigid dough that was unable to rise with developing gas pressure. Smith (1971) found that a combination of 1.5%

CMC, 2.5% guar gum and 2.2% GMS (flour wt) with either wheat or corn starch, produced an acceptable yeast bread. However, the soft, tender crumb had a more cake-like consistency than bread.

Kim and De Ruiter (1969) found that 1% (flour wt) GMS was needed to incorporate a large amount of air in a cassava-soy flour dough. The GMS product contained 50% monostearates, 40% distearates and 3% free glycerol (Kim and De Ruiter, 1968). Water representing 60-80% of the flour weight, depending on the water absorption of the flour, was included in the formulation (Kim and De Ruiter, 1969). The addition of GMS to the dough as a 10% emulsion (GMS to water 1:9) was necessary to obtain a light, structured bread as GMS added in the dry form did not significantly improve aeration. Although GMS (2% flour wt in a 10% emulsion) did not increase gas retention in either sorghum or barley breads, GMS improved the texture of sorghum bread and softened the crumb of barley bread (Hart et al., 1970). In sorghum flour bread, the incorporation of HPMC (2% flour wt) with GMS (2% flour wt) prevented collapse of the bread and produced a fine crumb structure (Hart et al., 1970).

Sodium-stearoyl-lactylate. Sodium-stearoyl-lactylate (SSL), an ionic surfactant, is the reaction product of lactic acid and stearic acid, partially neutralized to the sodium salt (Krog, 1981). SSL functions in starch complexing, protein interaction, aeration and foam stability (Birnbaum, 1981). In the baking industry SSL improves the mixing tolerance of doughs, produces more elastic doughs, softens crumb texture and increases the loaf volume of baked products (Schuster and Adams, 1984).

Smith (1974) found that SSL (0.5% flour wt) improved the texture of wheat starch breads. Kulp et al. (1974) studied various gum surfactant combinations in wheat starch breads. The incorporation of 0.8% (flour wt) SSL improved the volume of CMC (4.7% flour wt) breads but produced inferior crumb characteristics (Kulp et al., 1974). The use of SSL in conjunction with xanthan gum, however, failed to contribute to the structure of wheat starch bread and resulted in poor overall bread quality (Kulp et al., 1974).

Response surface methodology

The critical nature of ingredient levels and the synergistic interactions of some ingredients make the methodology of most previous research on gluten-free bread inappropriate. Some formulae were developed using a trial and error procedure (Johnson and Penfield, 1976). Generally, during gluten-free product development and formulation, one ingredient was altered at a time and only a limited number of ingredient levels were examined (Jongh et al., 1968; Nishita, 1973; Christianson et al., 1974; Ranhotra et al., 1975). Such a procedure does not permit examination of the effect of interactions between ingredients, requires many baking trials, is time consuming and expensive.

Response surface methodology (RSM) is a statistical technique that can consider several factors (ingredients) at different levels in a product, as well as the corresponding interactions among these factors and levels (Giovanni, 1983). This statistical procedure allows for the optimization of ingredient levels for specific desirable product characteristics (Johnson and Zabik, 1981). When using RSM with a

fractional factorial, central composite or rotatable experimental design, only a fraction of the treatment combinations is actually produced and examined (Dillon, 1977). An equation or mathematical model is developed from the actual experimental data and the model is subsequently used to predict variable conditions that were not tested (Henika and Palmer, 1976). Contour plots can be produced to estimate changes that will occur in the responses as the levels of ingredients are varied (Penfield and Axelson, 1984).

At the beginning of a project, screening studies can be conducted to identify which of many possible factors are the most important to the response being measured. Two-level fractional factorial designs can be utilized to identify important factors and indicate in which direction factors should be changed to improve the response (Bacon and Henson, 1971). The screening studies can be done sequentially, the result at each stage guiding the experimentation to be conducted at the next step (Box et al., 1978).

Recently, RSM has been used to optimize baked product formulations such as cakes (Johnson and Zabik, 1981; Lee and Hosney, 1982; Vaisey-Genser et al., 1987), cookies (Conner and Keagy, 1981), wheat flour chapaties (Ebeler and Walker, 1983) and for the development of a high protein bread (Henselman et al., 1974). No reports utilizing RSM for the development of a gluten-free yeast bread have been found in the literature.

Sensory evaluation of bread

Sensory evaluation utilizes humans and their senses as instruments to measure the quality attributes of food products. Difference and

descriptive sensory tests are generally employed by a trained taste panel to evaluate food quality attributes. Panelists are trained to know the evaluation techniques to be used and to detect differences in the quality characteristics of the products being evaluated. Panelists are also trained to make objective, precise and reproducible judgments. In contrast, untrained consumer panels utilize hedonic tests to determine food preference/acceptance. Consumer panelists score how much they like a product based on their own internal criteria (Hirsh, 1971). In the development of food products for special dietary needs, the use of both trained panels and untrained consumer panels is essential.

Important sensory attributes of bread include the appearance, odor, texture and flavor of the bread (Pomeranz and Shellenberger, 1971). The size, shape, symmetry and color of the loaf are included in the external appearance while internal attributes refer to the size and uniformity of the cells, the cell wall thickness and the crumb color. Bread texture includes the softness, elasticity and moistness of the crumb. The typically bland taste of wheat bread is a mixture of many interwoven flavors including salty, sweet and fermentative notes (Jackel, 1986). Odor, too, is a combination of many aromas such as yeasty and sweet. To obtain a complete overall impression of product quality, the inclusion of as many attributes as possible in the sensory evaluation of bread is important.

For many years, sensory evaluation has been used by the baking industry in the quality assurance of baked goods (Jackel, 1986). Products are scored for external, internal and organoleptic properties, however, the judgements are often made by one person, usually the baker

(Jackel, 1986). Although the baker may be very experienced at evaluating the products, his knowledge about the products and the expected outcome may result in biased judgements. In addition, all judgements may not be made by the same baker. Individuals differ considerably in their sensitivity and ability to judge differences (Amerine et al., 1965). Therefore, a large panel of trained judges is needed to provide accurate, reliable, unbiased results. The sensory evaluation of breads by trained panels, using unstructured line scales, is practiced by some researchers (Raidl and Klein, 1983; Brady and Mayer, 1985; Stroh et al., 1985).

Published reports of the sensory evaluation of yeast breads frequently involve the use of hedonic tests, with untrained panels, utilizing five or nine point hedonic scales (Klein et al., 1980; Seibel et al., 1983; Guy, 1986; Gayle et al., 1986) and triangle tests (Sosulski and Fleming, 1979; AACC [Method 33-50], 1983; Guy, 1986).

There is very little published research which includes the sensory evaluation of low-gluten or non-wheat flour breads. Many investigations (Pearson, 1960; Jongh et al., 1968; Kim and De Ruiter, 1969; Hart et al., 1970;) have not employed any type of sensory evaluation of the breads. Some studies (Steele et al, 1965; McGreer, 1967; Landsman and Wills, 1968; Sorenson, 1970; Kulp et al., 1974) have included informal evaluations in which the researcher, patients or accessible subjects (i.e. students) determined if the products were satisfactory. Other researchers (Smith, 1971, 1974; Bradley, 1972; Nishita, 1973; Christianson et al., 1974; Johnson and Penfield, 1976) have held more formal evaluations, with patients or accessible subjects, and have utilized a hedonic scale to rate breads for

preference or acceptance. However, only Bolam (1983) employed a trained panel to evaluate the texture of wheat and corn starch-xanthan breads. A 6-member trained panel utilized a texture profile method and a 15.0 cm unstructured line scale to evaluate the visual appearance, springiness, hardness, crumbliness, chewiness, crumb feel and moistness of the starch breads. Although Ranhotra et al. (1975) described the appearance, crust color, crumb color, grain, texture, flavor and mastication of a wheat starch bread, the bread attributes were assessed by a single baking technologist, rather than by a trained taste panel.

Thus, there is a need for research focusing on the development of gluten-free yeast breads which incorporates appropriate sensory techniques and trained taste panels. In addition, consumer panels employing the criteria and methods used most often by consumers in evaluating bread acceptability should be included in studies which are concerned with developing special diet breads.

Objective evaluation of bread

Objective evaluation includes chemical and instrumental measurements of the quality characteristics of bread. As with sensory evaluation, aspects of the appearance, odor, texture and flavor of bread can be determined using a number of different objective methods.

Loaf volume, an essential aspect of loaf appearance, is one of the most common objective measurements conducted on bread. Loaf volume is easily and reproducibly evaluated, by rapeseed displacement, and is a good indicator of bread crumb grain, softness and overall consumer acceptance (Pomeranz, 1969). This is particularly important if only one or two measurements of bread quality can be made. Color is another

important component, of bread appearance which can be easily measured. The crust and crumb colors of bread have been measured instrumentally using a Hunter Color Difference Meter (Shogren et al., 1979, 1981; Chen and Rasper, 1982) or a Gardner Color Difference Meter (Raidl and Klein, 1983).

Permanent records of the appearance of the bread loaf and the crumb of individual slices can be made by photographing the breads (Harrel, 1930; Cooley and Davis, 1936). To study bread cell structure, the slices can be photocopied. Photocopies provide a record of the actual size, shape and grain of a slice of bread (Campbell et al., 1979).

The odor and flavor of bread can be studied instrumentally by separating volatile compounds using headspace gas chromatography and identifying them by mass spectrometry (Hironaka, 1986; Stollman, 1986). However, since many individual components comprise the odor and flavor of bread, the correlation between the specific components identified by gas chromatography and the human response of bread odor and flavor may be difficult (Amerine et al., 1965). Amerine et al. (1965) suggest that to have a bearing on flavor, the chromatograph must have its response analyzed by and correlated with sensory flavor or odor tests. If only one test can be done, a sensory panel would probably provide the most useful information for the evaluation of bread odor and flavor.

The most commonly measured textural characteristic of bread is the crumb firmness (Kilborn et al., 1983). A variety of instruments have been used to objectively measure the maximum force and deformation of bread samples (Hibberd and Parker, 1985). These instruments include

the Baker Compressimeter (Lorenz and Disaver, 1982; Kamel et al., 1984; Kamel and Rasper, 1986) and the Instron Universal Testing Machine (Bashford and Hartung, 1976; Elgedaily et al., 1982; Kamel et al., 1984; Ghiasi et al., 1984; Soulaka and Morrison, 1985; Redlinger et al., 1985; Baker and Ponte, 1986; Baker et al., 1986).

Comparisons of the results of many published papers measuring textural characteristics of bread are difficult since specific details of the experimental methods and sampling procedures are not presented and often differ between papers (Hibberd and Parker, 1985). Although there is a standard method (Method 74-10) for the Baker Compressimeter (AACC, 1983), there is no standard procedure for the Instron Universal Testing Machine. Thus, the report of Baker and Ponte (1986), presents a much needed standard method for measuring the crumb firmness of bread with the Instron.

Published research on the objective evaluation of low-gluten and non-wheat flour bread is limited. Bread volume (Smith, 1971, 1974; Christianson et al., 1974; Ranhotra et al., 1974; Nishita et al., 1976) is the most frequent objective measurement made. Smith (1974), Ranhotra et al. (1975), and Nishita et al. (1976) also measured bread moisture. Christianson et al. (1974) provided the only report of the instrumental measurement of bread crumb firmness for starch breads. Since gluten is an important structure forming protein in wheat bread, gluten-free breads typically have low volumes and coarse textures (Kulp et al., 1974). Therefore, objective measurement of the quality characteristics of gluten-free breads, particularly texture, is important for the production of acceptable gluten-free breads.

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II. A Survey of Celiacs and Hospital Dietary Personnel

in Edmonton and Surrounding Communities:

The Gluten-Free Diet and the Use of Gluten-Free Yeast Breads

INTRODUCTION

Celiac disease is a lifelong condition which requires strict adherence to a gluten-free diet (Hartsook, 1984; Cooke and Holmes, 1984). The elimination of wheat, rye, barley, oats and their derivatives from the diet is essential (Hartsook, 1982), but not always easy. One of the most difficult aspects of the dietary treatment of celiac disease is the maintenance of a strict diet after the severe phase of the illness is over (Fry et al., 1982; Davidson, 1984). Additional problems of the disease include the identification of foods which unknowingly contain gluten (Kasarda, 1978; Hartsook, 1984) and the replacement of staple wheat flour products, such as yeast bread, in the diet.

Thus, a survey was conducted to better understand the needs of individuals following a gluten-free diet and to facilitate future research on the development of a gluten-free yeast bread. Specifically, the objectives of the survey were to determine: 1) the dietary modifications necessary on a gluten-free diet; 2) the types of gluten-free yeast breads being used by celiacs at home and for patients in hospitals, including methods of purchase, use and storage; 3) celiac and hospital personnel satisfaction with products and problems users encountered with gluten-free yeast breads; 4) the quality attributes of

gluten-free yeast bread considered important by gluten-free yeast bread users; and 5) the need for a new gluten-free yeast bread.

METHODS °

Celiac survey

Two hundred members of the Edmonton Celiac Association were surveyed by mail in February of 1985. Each respondent received a letter (Appendix 1) and a questionnaire (Appendix 2). To ensure the confidentiality of respondents, the letter requesting participation and the survey were addressed and mailed to all members by individuals on the Edmonton Celiac Association executive. The questionnaire (Appendix 2) consisted of 16 questions requesting information about the gluten-free diet and the use of gluten-free yeast breads. Demographic information about the respondents, as well as duration of celiac disease was also solicited to describe the sample population. The respondents were notified of the results of the survey in a summary (Appendix 3) that appeared in the July/August, 1985 issue of the Edmonton Celiac Association Newsletter, which was mailed to all members.

Hospital survey

Dietary departments of twenty hospitals in Edmonton and surrounding communities were also surveyed by mail in February of 1985. Addresses of hospitals were obtained from the Canadian Hospital Directory (1984); only hospitals with dietary department managers or directors were sent the letter (Appendix 4) and questionnaire (Appendix

5). The questionnaire (Appendix 5) consisted of 13 questions requesting information regarding the gluten-free diet and the use of gluten-free yeast breads. Demographic information related to hospital size and position of the respondent was also obtained to describe the sample population. The respondents were notified of the results of the survey via a summary letter (Appendix 6), which was sent to participants.

RESULTS AND DISCUSSION

Celiac survey

Response and demographics. One hundred and twenty-two of the two hundred questionnaires mailed were returned, representing a 61% response rate. Most of the respondents (70%) were female. This finding corresponds with the 1985 membership of the Edmonton Celiac Association, which consisted of 65% females (Friesen, 1987). A survey conducted by the Quebec Celiac Foundation also reported that 70% of those surveyed were female (Meunier, 1987). Age groups represented in the present survey sample (Table II.1) ranged from under 18 to over 65 years of age. Respondents indicated that they had suffered from celiac disease for a duration ranging from 2 months to 73 years. Approximately 50% of the celiac respondents reported having the disease for under 5 years, while the other 50% had celiac disease for over 5 years. While the duration of the disease was usually based on the length of time since diagnosis, many respondents reported that they had suffered from celiac disease for many years prior to diagnosis. Eleven percent of the respondents indicated that their households included two family

Table II.1 - Age distribution of celiacs participating in the survey

AGE GROUP	NUMBER OF RESPONDENTS	PERCENT
Under 18	20	16.4
19-25	8	6.6
26-35	25	20.5
36-50	29	23.8
51-65	30	24.6
Over 65	9	7.4
No response	1	0.8

members with celiac disease. Ten percent of Quebec Celiac Foundation members (Meunier, 1987) indicated that another member of their family had celiac disease.

Diet modifications. Eating out was the most frequently (33%) reported problem encountered by individuals on the gluten-free diet. This finding agrees with that of Barry et al. (1978) who interviewed 62 patients attending a celiac clinic. Fifty-eight percent of the patients who coped adequately with their gluten-free diet in a home environment found that they experienced considerable difficulty when away from home (Barry et al., 1978).

In the present survey, 21 celiacs (17%) indicated that the gluten-free diet affected their food choices 'quite a bit' and 10 respondents (8%) suggested that the gluten-free diet restricted the variety of foods in their diet. Other problems with the diet volunteered by the respondents were: all food labels must be read (5%), the diet is time consuming (2%) and there are some nutritional/fibre concerns (2%). However, some respondents (17%) reported positive aspects of the gluten-free diet. These celiacs noted that they were eating nutritious, well-balanced meals. Some respondents reported eating fewer sweets and junk food and more meat, fruit and vegetables than they ate prior to having the disease symptoms. Another 16% of the participants reported that the gluten-free diet presented no problems when eating at home. Bell et al. (1981) suggested that, compared with other therapeutic diets, the preparation of a gluten-free diet need not be difficult and demanding. Nutritionally, this is true since the gluten-free diet is easily modified. The protein of wheat, rye, oats and barley may be easily replaced with rice and corn, which

are only slightly lower in protein, or with dried legumes such as soybeans, beans, peas and lentils, which are even better sources of protein than the cereals being replaced (Campbell, 1984). Wheat fibre may also be replaced with rice bran, rice polishings, corn bran and raw fruits and vegetables (Bell et al., 1981; Campbell, 1984). The B vitamins can be provided by including meat (particularly liver), green leafy vegetables and milk in the diet. However, there are many problems and challenges with the gluten-free diet, such as hidden sources of gluten in foods, a restricted variety of gluten-free bread products and the poor quality of many gluten-free baked products, which cannot be ignored.

Wheat starch. Almost half of the survey respondents (46%) indicated an inability to tolerate wheat starch in their diet. Another 15% of the survey participants stated that they were either unsure of their tolerance to wheat starch or had never tried to eat wheat starch. Of the 102 respondents who knew their tolerance to wheat starch, 59% made their determination through personal assessment, 22% had the determination made by a medical doctor and 16% used both a medical doctor and personal assessment. Personal assessment of the disease was frequently decided by the presence of diarrhea, cramps, gas, stomach bloating or abdominal discomfort when wheat starch was included in the diet.

Only 10% of the respondents actually reported using wheat starch in their cooking and baking. According to the respondents, rice flour was the most popular flour for baking and cooking. Ninety-three percent of the respondents reported using rice flour, while 73% and 72% used potato flour and corn starch, respectively. Other flours and

starches used by the respondents for food preparation included soya flour (53%), corn flour (43.4%), tapioca starch (19%), potato starch (12%), cornmeal (7%), pea and bean flours—(7%), arrowroot (3%) and buckwheat flour (2.5%). Only 2 (1.6%) respondents reported using oats in their baking and cooking.

Gluten-free yeast bread. Results of the celiac survey indicated that 72% of the 122 respondents regularly consumed a gluten-free yeast bread, and that 80% of these individuals consumed one or more slices per day. The majority of the consumers (70%) ate commercially prepared bread, while others either made their own bread from recipes (39%) or used a commercial dry mix (23%). Many respondents used two or three different types of bread. Of the respondents who regularly used commercially prepared bread, 64% purchased Woodward's brown rice bread and 31% purchased IGA white rice bread. Approximately 50% of the respondents purchased commercially prepared bread weekly (39% purchased 1 to 2 loaves per week; 10% purchased 3 or more loaves per week); the other 50% purchased gluten-free bread monthly (13% purchased 1 to 2 loaves per month; 36% purchased 3 or more loaves per month). The most popular commercial dry mixes were Celimix (65% of dry mix users) and Juvela (25%), which both contained wheat starch. Sixteen percent of the respondents who made gluten-free yeast bread from a recipe used recipes that included xanthan gum as a gluten replacement.

Wheat bread was also substituted with other products by 88% of the survey respondents. Among the 107 respondents who made bread substitutions, 68% used rice crackers, 69% used rice cakes and 36% substituted potatoes. Cooked rice (13%), muffins and quickbreads (10%), as well as cereals (8%) were also used as bread substitutes.

The need for toasting gluten-free yeast bread was evident as 80 of the 88 respondents who regularly ate gluten-free bread reported eating gluten-free bread toasted. Although more than half of the survey participants (59%) ate gluten-free bread as sandwiches, the sandwich bread was often toasted. Only 11 individuals ate gluten-free bread plain. Some respondents commented that plain gluten-free yeast bread tasted awful, crumbled and was hard to digest. Suggestions offered by the respondents for improving the palatability of gluten-free bread were grilling and microwave cooking the bread and making french toast, cinnamon buns and breadcrumbs from the bread or dough.

Frozen storage was the most popular method of storing gluten-free bread (for 91% of the respondents who regularly consumed gluten-free yeast bread). The length of time for freezer storage of gluten-free bread varied from one to three weeks (25%), one to two months (42%) to over three months (26%). Another 35% of the respondents who regularly consumed yeast bread, stored gluten-free bread in the refrigerator for periods of up to two weeks (61% of these individuals stored breads for 1 to 3 days, 87% indicated bread storage for 7 to 10 days and 3% stored breads for 2 weeks). Room temperature storage for periods of 2 to 4 days was used by only 6 respondents. Plastic bags were the most common storage container (86% of regular bread consumers), followed by plastic wrap (18%) and aluminum foil (2%).

Forty-three percent of the celiac respondents were satisfied with the available gluten-free yeast breads, 42% were not satisfied, and 10% were undecided. Many problems with gluten-free breads were noted by both satisfied and dissatisfied respondents. Table II.2 lists respondent volunteered problems in order of frequency. The major

difficulties encountered in gluten-free breads were the crumbliness, poor flavor and dryness. Bell et al. (1981) also reported that the gluten-free baking mixes, baked bread and cookies produced by several firms were often tasteless, crumbled easily and became stale quickly.

Forty-three percent of the respondents were satisfied with the available gluten-free yeast breads. Paulus (1986) reported that individuals requiring a special diet adapt themselves to specific nutritional necessities and are more inclined to accept products of lower sensory quality. However, in the present study almost all of the respondents (94%) indicated a willingness to try a new product. Thus, there appears to be a need and a potential market for a new gluten-free yeast bread.

According to the celiac respondents, the most important quality characteristics desired in gluten-free yeast bread were flavor (ranked the most important by 51% of the respondents) and texture (ranked the second most important by 42% of the respondents). These attributes were followed by concerns about odor, visual characteristics, shelf-life, price, nutrition and availability. Martinsen and McCollough (1977) and Rognerud et al. (1983) also reported that flavor or taste was the most important quality factor consumers considered when selecting wheat bread. In a survey of 600 Sacramento residents, Schutz et al. (1986) found that the importance ratings for sensory attributes of 15 foods, including bread, were far higher than those of price or nutrition.

Table II.2 - Problems cited for gluten-free yeast breads by celiac survey participants

PROBLEM	NUMBER OF RESPONDENTS
Crumbles easily	49
Lacks flavor/poor flavor	25
Dry	16
Heavy/poor texture	13
Stales/molds quickly	11
Impossible to make a sandwich	9
Sticks to wrapper	9
Must be toasted	8
Too expensive	4
Big air holes	4
Unreliable availability	4
Hard to slice	3
Inconsistent product	2

Hospital survey

Response and demographics. Fifteen of the 20 questionnaires mailed to hospital dietary departments were returned, representing a .75% response rate. However, two of the hospitals that responded could not complete the questionnaire since they had never had the opportunity to use gluten-free bread products. Thus, the results presented are based on the information provided by 13 hospitals.

The hospital questionnaires were completed by hospital dietary department dietitians (8), directors (3), coordinators (1) or supervisors (1). The size of the hospitals ranged from 30 beds to 1000 beds; 8 hospitals had 125 beds or less and the remaining 5 had more than 125 beds.

Gluten-free diet recommendations. Diet recommendations were made to patients requiring a gluten-free diet by 11 of the 13 hospitals, and all 11 hospitals provided pamphlets and diet sheets for the patients. The majority of hospitals (10) used some form of the gluten-free diet pamphlet, prepared by the Edmonton Hospitals Diet Therapy Committee and distributed by the University of Alberta Hospitals, for counselling and distribution to patients. The pamphlet is a comprehensive guide to the gluten-free diet which describes the diet, defines gluten, instructs patients to read labels, lists food ingredients to avoid, provides names of some flours which may be used as a substitute for wheat flour in recipes, lists the foods recommended and foods to avoid in 14 food categories and provides some recipes. A number of notes, such as warnings to patients that wheat starch contains some gluten and may not be tolerated by all celiacs, that oats should be avoided in the diet initially and that some common medications may contain gluten, are also

included in the pamphlet. Two hospitals also provided additional gluten-free recipes to patients. Two other hospitals provided a gluten-free products list while one hospital supplied patients with information and application forms for the Edmonton Celiac Association. Since the acceptance of a gluten-free diet by patients is largely conditioned by the initial explanation of the need for the diet and its implications in practical terms (Cooke and Holmes, 1984), the diet recommendations made by dietitians must be accurate, thorough and complete. Organizations like the Edmonton Celiac Association provide another source of information, recipes, comfort and support for celiacs which should be made known to newly diagnosed celiacs.

Gluten-free yeast bread. Three of the 13 hospitals listed gluten-free breads on the hospital menu daily. Nine hospitals never included gluten-free bread on the menu, but 6 hospitals would provide gluten-free bread daily at all meals if a patient required it. Commercially prepared gluten-free bread was used by 46% of the hospitals (Woodward's brown rice bread and a Co-op bread), while 15% used commercial dry mixes (Celimix and Juvela) and 31% of the hospitals made gluten-free bread from recipes. Hospitals purchasing commercially prepared gluten-free bread either procured from 1 to 6 loaves per month or purchased the bread when a patient required it.

Seventy-seven percent of the hospitals substituted other products for wheat bread on the menu for patients requiring a gluten-free diet. Replacement products included muffins/cookies/quickbreads (10 hospitals), potatoes (9), rice cakes (5), rice crackers (2), rice (2) and cereals (1).

In hospitals, gluten-free bread was served plain (10 hospitals),

toasted (10) and as a sandwich (7). Gluten-free bread was stored in the freezer by 9 hospitals for periods of 1 to 3 months. One hospital reported refrigerating gluten-free bread for one week while another hospital stored gluten-free bread at room temperature for two days. Plastic bags were the most common storage container for gluten-free bread (5 hospitals). The use of plastic wrap and plastic wrap plus a plastic bag were also reported.

Five of the hospitals were satisfied with the available gluten-free breads, while 4 were not. Problems identified in gluten-free yeast breads by dietary personnel were similar to those listed earlier by celiacs. These problems included dryness, crumbliness, a heavy and coarse texture, an inconsistent product, poor availability, quick staling and that gluten-free bread required toasting for desirable quality. Only one hospital reported patient complaints about the gluten-free breads served, and the complaints included the dry, crumbly and heavy texture of the breads. Eighty-five percent of the hospitals indicated a willingness to try a new gluten-free bread if it became available, suggesting a potential market for a new product.

The most important quality characteristics desired in gluten-free yeast bread by the hospital dietary personnel, were an acceptable texture (ranked the most important by 5 of the hospitals), desirable flavor (ranked the second most important by 4 of the hospitals) and satisfactory visual characteristics (ranked the third most important by 4 of the hospitals). Odor, price and shelf life were also considered important quality attributes of gluten-free breads.

CONCLUSIONS

The gluten-free diet presents a number of challenges and difficulties for individuals who must adhere to the diet. These challenges include the necessity for appropriate food selection when eating out, the limited variety of gluten-free foods available, the time required for reading all food labels, the planning and preparation of gluten-free meals. The fact that the gluten-free diet must be followed for a lifetime makes the challenge all the more difficult.

Commercially prepared gluten-free breads are used most frequently by celiacs and hospital dietary departments. Commercially prepared gluten-free breads are more popular than dry mixes or recipes for home use. However, only two brands of commercially prepared breads are being used by the majority of local celiac consumers.

Presently, many gluten-free breads are being consumed toasted, usually to improve the flavor and the texture, and to make the breads more palatable. A gluten-free yeast bread that could be eaten plain and used to make sandwiches, would be a welcome addition to the breads currently available.

Although many celiacs and hospital dietary personnel serving a gluten-free diet are satisfied with the gluten-free yeast breads that are available, there are numerous problems with these products which must be overcome. These problems include crumbliness, dryness and poor flavor of the gluten-free breads. Thus, the need for a quality gluten-free yeast bread, that more closely resembles wheat flour yeast bread, is obvious.

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III. The Development of Standardized Formulations for Rice Flour Yeast Bread Production

INTRODUCTION

A gluten-free yeast bread comparable to wheat flour yeast bread is essential for individuals who cannot tolerate wheat flour. However, the manufacture of bread without gluten presents considerable difficulty (Kulp et al., 1974). McGreer (1967) noted that wheat starch bread was quite sensitive to ingredient proportions and that accurate measurement of ingredients was important. Strict adherence to the water requirements of a wheat starch batter was imperative for the production of good quality bread (Ranhotra et al., 1975). Since a wheat starch bread system was very sensitive to baking and proofing conditions, the results were difficult to reproduce (Bradley, 1972).

In rice flour yeast breads the proportion of rice flour, methylcellulose and water was critical (Nishita, 1977), as was the fermentation time and the proofed dough structure (Nishita, 1973). Varietal differences in rice can affect the baking quality of rice flours. Compositional differences in rice can influence the viscometric properties, starch gelatinization temperatures and the rate and amount of water absorption of rice flours (Deobald, 1972). Rice flour particle size affected water absorption and the overall character of the resultant bread (Nishita, 1984). Thus, one brand or type of rice flour often cannot be replaced with another.

Nishita's (1973) rice bread somewhat resembled wheat bread in volume and grain characteristics, but lacked the flavor and texture associated with white bread. Flavor and texture were the most important quality attributes desired in gluten-free bread by consumers and hospital dietary personnel (Chapter II). Thus, adaptations to Nishita's formula that would allow for recipe replication and improve the flavor and/or texture of the rice bread would be advantageous.

The objective of this investigation was to develop a reliable, reproducible standard formulation for the production of rice flour yeast breads, which could be utilized in future research. Nishita's (1977) rice bread formula was adapted to permit the use of locally available rice flours, from different sources, and to incorporate techniques which could improve the flavor and texture of the resultant rice breads.

MATERIALS AND METHODS

The ingredients used in the rice bread formulations were: white rice flours, A, Dainty Foods (medium grain/finely ground), B, Ener-G Foods (medium grain/coarsely ground) and C, Woodward's (long grain/finely ground); potato starch (Casco Potato Flour), Canada Starch Co.; hydroxypropylmethylcellulose (Methocel K4M), Dow Chemical Co.; iodized table salt (Windsor), Canadian Salt Co.; granulated sugar, Alberta Sugar Co.; canola oil, Canbra Foods; and active dry yeast (Fleischmann's Fast Rising), Standard Brands Canada Ltd. Distilled water was the liquid for all breads.

Rice flour and potato starch analysis

The three rice flours and the potato starch were analyzed for

percent moisture, protein and lipid by standard AOAC (1980) methods 14.003, 14.026 and 14.019, respectively.

Gelatinization temperatures and pasting characteristics of the rice flours and potato starch were determined with a Brabender amylograph. Complete amylograms were obtained by using the method of Halick and Kelly (1959), as modified by Juliano et al. (1985). Ten percent slurries were utilized for the rice flours and 5% slurries for the potato starch. Amylograph gelatinization temperatures were determined by the method of Juliano et al. (1985), using 20% slurries.

Gelatinization temperatures were also determined by observing the swelling of the rice and potato starch granules microscopically. Thin slurries of each sample were placed on glass slides and heated on a Mettler FP5/FP52 melting point apparatus and viewed on a Zeiss microscope.

Baking

The rice bread formula and procedure (Nishita, 1977) was employed and subsequently adapted using Flour C (Woodward's). The adapted formulation was standardized and modified for use with Flours B and A (Ener-G and Dainty, respectively).

Rice bread doughs were prepared using a 10-speed Hobart KitchenAid mixer (Model K45SS), equipped with a 4.5 qt stainless steel bowl (K45) and whip attachment. Both the flat paddle and dough hook attachments were tested. The whip was selected because it enhanced air incorporation into the dough and produced breads with large volumes.

A Cres-Cor (Crescent Metal Products Inc., Cleveland, Ohio) proofing cabinet (30°C, 95% humidity) was used to proof the doughs (let

them rise). The cabinet was altered to ensure a constant internal temperature and humidity. The large single door was reconstructed into 3 separate doors, each with a locking handle. The three individual doors on the proofing cabinet facilitated placement or removal of single pans of dough, without lowering the cabinet internal temperature. A small hole was drilled into the middle at the back of the cabinet to allow insertion of six thermocouples into the cabinet. The thermocouples were attached to a Honeywell recording potentiometer to monitor the temperature within the proofing cabinet. Only the upper half of the cabinet, partitioned with three metal shelves, was utilized to proof the rice breads. Two copper constantan thermocouples measured the temperature of the cabinet at each of the shelves. The temperatures ranged from 30°C at the top and middle shelves to 31.5°C at the bottom shelf. Temperatures were maintained, within 2°C, throughout, a 9-hour period. Initially, to attain a temperature of 30°C, the water reservoir in the proofer was filled and the temperature dial (10 settings) was set to 8. When a temperature of 28°C-30°C was reached, the cabinet temperature dial was turned down to a setting of 7.5. During bread production, one pan of dough was proofed on the center of each shelf.

Rice breads were individually baked in one of three household ovens (Kenmore, Mark 3). Each bread was placed in the middle of a rack positioned in the center of the oven. The oven and proofing cabinet shelves used for each loaf of bread were randomized using a 3x3 Graeco latin square design (Box et al., 1978), by randomizing rows and columns.

After baking, breads were cooled on racks for 15 minutes at room

temperature (23°C), removed from the pans and cooled for an additional hour. Then, bread weights and volumes were recorded. Bread volume was determined by rapeseed displacement using a National Loaf Volumeter (National Mfg. Co., Lincoln, Nebraska). Breads were placed in the volumeter right-side up, unless the bottom surface of the loaf had a large indentation, in which case the loaf of bread was placed in the volumeter upside-down. Cooled breads were wrapped in plastic wrap, labelled, put into plastic bags and frozen (-29°C). A bench top sensory evaluation of the appearance, flavor and texture of some of the rice breads, fresh or frozen and thawed, was conducted by the researcher and/or members of the Department of Foods and Nutrition.

Consumer panel evaluation

Breads prepared from each of the three rice flours (A, B and C), using the standard formulations, were evaluated by a consumer panel. Thirty-two members of the Edmonton Celiac Association (20 celiacs and 12 non-celiacs) participated in the consumer panel pre-test. Each participant evaluated three bread samples (a slice from each rice flour bread) for aroma, appearance, firmness, moistness, flavor and overall acceptability. The characteristics were scored on a 6-point category scale where 6 represented a very desirable aroma, appearance or flavor and a very soft, very moist or very acceptable sample. A score of 1 represented a very undesirable aroma, appearance or flavor and a very firm, very dry or very unacceptable sample. The ballot is shown in Appendix 7.

The results of the consumer pre-test were tabulated and means calculated over all of the participants.

RESULTS AND DISCUSSION

Rice flour and potato starch analysis

The percentages of fat, moisture and protein for each of the three rice flours (Table III.1) tended to be similar. Flour A had a slightly higher percentage of protein; Flour B was slightly higher in moisture than the other two flours. The percentages of fat and protein in potato starch (Table III.1) were lower and the percentage of moisture was higher than those in the rice flours. Composition differences in rice can influence the viscometric properties and gelatinization temperatures of rice flours (Nishita et al., 1976; Luh and Liu, 1980) which can, in turn, affect the flour baking properties. Thus, flour differences should be identified and related to the production of acceptable rice yeast breads.

The amylograph gelatinization temperatures of the rice flours (Table III.2) differed. Both Flours A and B, (medium grain) had lower gelatinization temperatures (64.5°C and 65.0°C, respectively) than Flour C, the long grain rice flour (72.5°C). Rice flour gelatinization temperatures obtained by granule swelling (Table III.2) were slightly higher than those obtained for comparable flours using the amylograph. The potato starch gelatinization temperatures (Table III.2) were identical and lower than that determined for any of the rice flours.

Granule swelling can be used as a criteria of gelatinization for very small granules, or those showing weak birefringence (Zobel, 1984), such as rice starch granules. Halick et al. (1960) took the gelatinization temperature of 20 rice varieties as the temperature obtained

Table III.1 - Composition of rice flours and potato starch

	% FAT	% MOISTURE	% PROTEIN
Flour A	1.2	10.84	6.7
Flour B	0.9	12.80	5.9
Flour C	0.8	10.82	6.2
Potato Starch ^a	0.03	13.84	0.06

^a Casco potato flour, Canada Starch Company

Table III.2 - Gelatinization temperatures and pasting characteristics of rice flours and potato starch

	GRAIN LENGTH	GELATINIZATION TEMP.		VISCOSITY (BU)		
		AMYLOGRAPH (°C)	GRANULE SWELLING (°C)	PEAK	COOL to 50°C	SETBACK
Rice Flour A	Medium	64.5	66	960	840	-120
Rice Flour B	Medium	65.0	66	840	720	-120
Rice Flour C	Long	72.5	74	900	970	70
Potato starch		61.0	61	1420	560	-860

when the individual granules had swollen to the bursting point. The gelatinization temperature obtained in this manner correlated highly and significantly ($r=0.955$) with the amylograph determination of gelatinization temperature (Halick et al., 1960).

Milling methods can also affect the gelatinization temperature, since the amount of starch damage will differ (Bean, 1986). In the present study, however, the gelatinization temperatures of the fine (Flour A) and coarse (Flour B) medium grain rice flours were very similar.

According to the classification system of Nishita and Bean (1979) and amylograph gelatinization temperatures, rice Flour A had a low (<65°C), Flour B an intermediate (65°C - 70°C) and Flour C a high gelatinization temperature (>70°C). Typically, long grain rice has a higher gelatinization temperature than medium grain rice (Halick and Kelly, 1959; Nishita and Bean, 1979) and Flour C, from long grain rice, followed that pattern. Using the granule swelling gelatinization temperatures, both medium grain flours (A and B) had identical intermediate gelatinization temperatures. Flour C had a high gelatinization temperature. On the basis of granule swelling gelatinization temperatures, only the classification of Flour A differed from that determined using the amylograph. Halick et al. (1960) suggested that the amylograph determination provided more accuracy than the granule swelling procedure.

Starch gelatinization is one of the most characteristic processes occurring during bread baking (Jongh et al., 1968). Baked products set (gelatinize) or reach a temperature at which the dough can no longer expand under the gas pressure generated by the increasing temperature

(Hoseney, 1986). Wheat starch reaches its gelatinization temperature at about 53°C; rice starch gelatinizes at a higher temperature (Hoseney, 1986). Thus, rice flour doughs must attain a higher temperature, during baking, than wheat flour doughs, before they set. Since rice flour dough is gluten-free, starch gelatinization is even more important to the structural development of rice bread than it is to wheat bread.

Table III.2 presents the pasting characteristics of the three rice flours and potato starch. Flour B had the lowest peak viscosity and cooled viscosity. Flour C (long grain rice) had an intermediate peak viscosity, the highest cooled viscosity and a high setback viscosity. Deobald (1972) noted that long grain rice usually has a relatively low peak viscosity and forms a rigid gel on cooling, producing a high setback viscosity. The setback is mainly due to retrogradation of the linear amylose chains (Sanderson, 1981). In contrast, short or medium grain rice starches usually have high peak and low setback viscosities. Potato starch had a very high peak viscosity, much higher than that of the rice flours, but it also showed a very low setback. Sanderson (1981) and Zobel (1984) also reported that potato starch granules had high peak viscosities and readily disintegrated with an accompanying loss in viscosity.

Formula adaptations

Initial attempts to duplicate Nishita's (1977) rice bread formula, using locally purchased rice flour, were unsuccessful. The loaves were small and flat, did not rise during proofing or baking and had a dense, gummy texture. Increasing the water levels from 198 ml to 210 ml and

227 ml allowed the doughs to rise during proofing, but the loaves shrank in the oven and had low volumes. Thus, adaptations to the formula were examined in preliminary baking trials, in order to produce a rice bread with optimum quality that could be replicated and adapted for use with other rice flour varieties. Flour C (Woodward's) was used in the preliminary trials.

Inclusion of potato starch. Twenty percent of the rice flour was replaced with potato starch. The addition of potato starch to rice breads produced a good loaf volume, whitened and softened the crumb and gave a mild, less ricey flavor. Nishita (1973) reported that replacement of 20% of the rice flour in rice bread, by potato, tapioca or wheat starch, increased the resemblance between the rice bread and conventional wheat bread. The starches improved volume, softened texture, reduced graininess and produced a more open cell structure (Nishita, 1973). Nishita (1973) suggested that the textural improvement of breads with the potato or tapioca starches may have been due to the ability of cooked dispersions of either starch to remain as a fluid sol rather than to gel on cooling. Potato starch, when combined with hydroxypropylmethylcellulose, also produced a finer structure and a better volume in sorghum breads when compared to sorghum breads without potato starch (Hart et al., 1970).

Remix method of dough preparation. The one-step method of dough preparation of Nishita (1977) was replaced with a remix method similar to that used by Ranhotra et al. (1975) to produce wheat starch breads. Table III.3 shows the effects of the dough mixing method and proof time on the volume of rice breads. The remix method incorporated an additional mixing (5 min) and proofing period into bread preparation.

Table III.3 - Effect of dough mixing method and proof time on rice bread^a volume

BAKING TRIAL	MIXING METHOD	PROOF TIME (MIN)	VOLUME (CC)
1	1-step	45	1116.7
2	remix	30/30 ^b	1025.0
3	remix	30/50 ^b	1280.0
4	remix	30/60 ^b	1685.0 ^c

^a Woodward's Flour, 185 ml water, baked 40 min at 190°C

^b First proof/second proof

^c Over-proofed, expanded excessively in oven and fell over

In Trial 2, an initial 30 min proof of the dough in the mixing bowl was followed by a 5 min mix. The dough was panned and proofed for a second time (30 min). With this 30 min second proof, the bread volume was lower than that produced with the one-step method (Trial 1). In Trial 3, a 50 min second proof produced rice breads with a good volume (larger than the one-step loaf) and good loaf shape. A 60 min second proof (Trial 4) caused the dough to overproof in the proofer and to expand excessively and partially fall over in the oven. Although the volume of the bread was high, the crumb had many large holes. A milder rice flavor was noted in the remixed rice breads. Remixing eliminates waste products (Bennion, 1967), possibly by allowing for the volatilization of some gases and offodors which, otherwise, would remain in the bread dough. Thus, both an additional mixing period and additional proofing time were needed to produce breads acceptable in volume, appearance and flavor.

Nishita (1973) noted no advantage to rice bread volume with a remix method. However, her remix method incorporated only 60 sec of mixing, which may have been insufficient to redistribute the gas and yeast cells, and did not allow for additional proofing time. Nishita (1973) found that rice flour doughs had to be proofed to just less than the maximum height so that during oven spring, the dough reached its maximum height just as the starch started to gel and set. Remixing non-wheat flour doughs for 5 min before the final proof produced breads with a finer crumb texture and a more tender crumb than similar breads without a remix (Kim and De Ruiter, 1968). However, contrary to the present study, Kim and De Ruiter (1968) reported that extending the final proof of non-wheat flour doughs did not increase volume. In

addition, gas cells within the dough coalesced and produced a bread with a coarse, firm crumb with thick cell walls (Kim and De Ruiter, 1968).

Yeast addition. In this study, four grams of the total sugar were dissolved in the warm, yeast pre-soak water, prior to yeast addition. McGreer (1967) and Bolam (1983) also allowed the yeast to activate in a sugar-water solution. Kim and De Ruiter (1968) suggested that rapid gas production was advantageous for gluten-free doughs. Rapid gas production reduced the time before baking and lowered the chance of gas cells coalescing and escaping (Kim and DeRuiter, 1968).

The effect on loaf volume of decreasing the yeast in the rice bread formulation was examined (Table III.4) in an attempt to lower the incidence of over-proofing and reduce rice bread yeasty flavor. Reductions in yeast, from 7 g to 4 g, resulted in breads with low volumes and strong rice flavors. The strong rice flavors were less acceptable than the yeasty flavor notes present in the rice bread with 7 g of yeast. No effect of decreased yeast level on crumb appearance was noted. Thus, the 7 g of yeast was retained in the formula. Nishita (1973) suggested that excessive yeast action could produce holes and large spaces in rice bread crumb.

Fat can retard yeast action by coating the cells. In the present formula the yeast was pre-mixed into the flour (15 sec) before the oil was poured into the batter.

Effect of water level. Table III.5 shows the decrease in Flour C bread volume that resulted from a 15 ml water reduction. For Flour C breads, 170 ml of water produced a satisfactory volume and a desirable loaf shape. These breads are shown in Plate III.1. The critical

Table III.4 - Effect of yeast level on rice bread^a volume

BAKING TRIAL	YEAST (g)	VOLUME (cc)
1	7	1606.7
2	6	1321.7 ^b
3	5	1153.3 ^b
4	4	1110.0 ^b

^a Woodward's Flour, 185 ml water, remix method, 30 min first proof 50 min second proof, baked 5 min at 230°C and 30 min at 180°C

^b A stronger rice flavor was noted in the breads with reduced yeast levels

Table III.5 - Effect of water level on rice bread^a volume

BAKING TRIAL	WATER (ml)	VOLUME (cc)
1	185	1450.0 ^b
2	170	1123.3

^a Woodward's Flour, remix method, 30 min first proof, 50 min second proof, baked 5 min at 215°C and 30 min at 180°C

^b Loaf over-expanded in oven, resulting in a mushroom shaped loaf with a large hole under the crust

nature of the water level in non-wheat and starch breads has been reported (Kim and De Ruiter, 1968; Kulp et al., 1974; Osman, 1975; Ranhotra et al., 1975; Johnson and Penfield, 1976; Nishita et al., 1976).

In rice breads, flour varietal differences can result in different water absorption rates and make water addition to rice bread doughs difficult. All three flours of the present study had different water requirements. Nishita et al. (1976) observed that insufficient water in rice breads produced stiff doughs that did not rise; excessive water caused over-expansion during baking which resulted in large volumed loaves containing large holes and weak crust walls. Similar results were obtained in the present research.

Effect of baking temperature. In the preliminary trials, crust fissuring or cracking was a major problem. Crust cracking was detrimental to rice bread appearance and quality as well as to volume measurement replicability. To prevent top crust cracking, oven temperature combinations of: 1) 205°C for 10-20 min followed by 177°C - 191°C for 15-25 min (Pearson, 1960; McGreer, 1967), and 2) 177°C for 5 min followed by 255°C - 268°C for 25 min (Sorenson, 1970; Johnson and Penfield, 1976) were evaluated. When bread baking began in a cool oven, cracks were produced almost immediately after the loaf was put in the oven. The extended baking at a high temperature also caused a blackened crust. If the water level and proof time were optimal, baking the rice breads at 215°C for 5 min, followed by 40 min at 180°C, allowed for the gelatinization and setting of the rice breads and prevented the rice breads from cracking. The effects of baking temperature on rice breads are shown in Plate III.2.

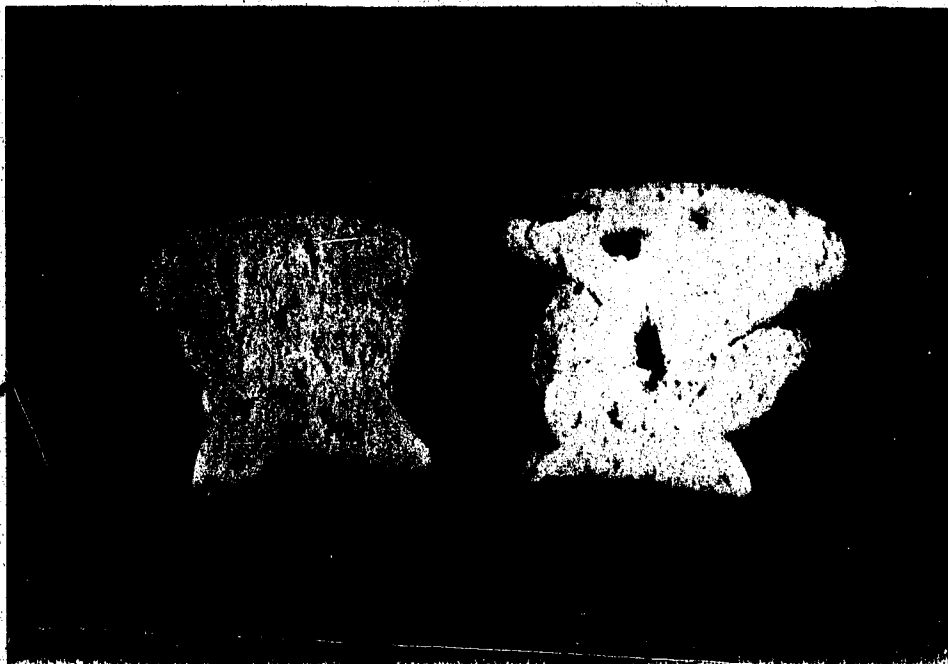


Plate III.1 - Effect of water on Flour C rice breads.
 The bread on the left was prepared with 170 ml water and
 the bread on the right was prepared with 185 ml water.



Plate III.2 - Effect of baking temperature on Flour C rice breads.
 The bread on the left was baked at 205°C (30 min), the
 center loaf was baked at 177°C (5 min) and 218°C (25 min),
 while the loaf on the right was baked at 218°C (5 min)
 and 177°C (30 min).

Table III.6 - Standard rice bread formula^a

INGREDIENTS ^b	WEIGHT (g)	% FLOUR WEIGHT
Rice Flour	182	80.2
Potato Starch	45	19.8
Salt	6	2.6
Sugar, granulated	24	10.6
Yeast	7	3.1
Oil	13	5.7
Hydroxypropylmethylcellulose	7	3.1
Water ^c	170	74.9

^a Adapted from Nishita, 1977

^b See the method for brand names

^c Flour C (Woodward's)

For Flour A (Dainty Foods) use 205 g water

For Flour B (Ener-G Foods) use 163 g water

Standard formulation

The standard rice bread formula for Flour C is shown in Table

III.6. The preparation procedure is as follows:

1. Dissolve 4 g sugar in 50 ml warm water (43°C). Stir in yeast and soak for 10 min.
2. Mix rice flour, potato flour, salt, remaining sugar (20 g) and methylcellulose at speed 1 (Hobart KitchenAid Model K45SS) for 2 min, using the whip beater attachment.
3. Add yeast mixture and remaining water (40°C) and mix at speed 2 for 15 sec.
4. Add oil and mix at speed 4 for 15 sec, followed by 2.5 min speed 6.
5. Scrape the sides of bowl and mix at speed 6 for 2.5 min.
6. Scrape sides of bowl, pushing dough to the bottom of the bowl and proof dough in bowl for 30 min in proofing cabinet (30°C/95% humidity).
7. Remix dough at speed 6 for 5 min.
8. Place 400 g dough in a greased pan (18.7 cm x 9.2 cm x 5.7 cm) using metal & rubber spatulas. Press dough into the pan corners and sides, eliminating air pockets and flattening the top surface, using an oiled rubber spatula. Using the same oiled spatula, push the edges of the dough towards the center, away from the pan sides, rounding the top edges of the dough.
9. Proof panned dough for 50 min (30°C/95% humidity).
10. Bake in preheated oven (215°C) for 5 min. Lower the oven temperature to 180°C and continue baking the bread for 40 additional min.
11. Cool the bread at room temperature (23°C) for 15 min. Remove the bread from the pan. Cool bread on a rack for 1 hr before packaging.

When the rice flour doughs are panned, it is important that the dough is pressed well into the bottom of the pan and that all air spaces are removed. Rounding the top edges produced a loaf of bread.

with a rounder top surface. Nishita (1973) noted the frequent appearance of large empty spaces in the crumb structure of rice breads or an indentation on the bottom of the loaves and suggested that the holes could result from air entrapment during dough transfer to the pan. In the present study, removal of air spaces during panning reduced large air holes in the crumb of rice breads, however, the bottom surface indentation mentioned by Nishita (1973) was still present in many rice breads.

Replication. The standard rice bread formula was replicated twice (using all proofer shelf and oven combinations, 3x3=9 loaves per rep) with Flour C. A 3x3 Graeco latin square design (Box et al., 1978) was used. Analysis of variance for loaf volume was computed. No significant effects were found for proofer shelf, oven, replication, order of bake or time of day of bake. Analysis of variance data appear in Appendix 8. Bread volumes ranged from 1075.0 cc to 1172.9 cc with a mean of 1125.1 cc.

Ingredient and method modifications for other rice flours

The medium grain Flour A required more water in the rice bread formulation (205 ml) than the long grain Flour C (170 ml). However, Flour B breads, from a medium grain rice with a gelatinization temperature similar to Flour A (Table III.2), had the lowest (163 ml) water requirement. Halick and Kelly (1959) found that medium grain rice varieties, with low gelatinization temperatures (64.5°C - 67.5°C), absorbed more water than the long-grain rice types. Flour B also had the highest percent moisture (Table III.1) and was coarsely ground, compared to the finely ground Flours A and C. Both of these factors

may have reduced Flour B's rice bread water requirements. The more coarsely ground rice Flour B would also have less damaged starch than the finely ground Flours A and C. Nishita (1973) suggested that fine rice flour particles absorbed considerably more water than coarse rice flour particles. Later, Nishita and Bean, (1982) found that water absorption capacity was greater for fine rice flours than coarse rice flours, the fine flours having more damaged starch.

A breads also needed a shorter second proof (40 min) than Flour B and C breads, which required 50 min.

Consumer evaluation of the standard rice bread products

Data from a consumer evaluation of the standard rice breads are given in Table III.7. All three breads were judged to be slightly acceptable to acceptable. However, Flour A and Flour B breads received slightly higher overall acceptability scores than Flour C rice breads. For appearance and flavor, the rice breads received scores of 4.0 to 5.0 out of 6.0, indicative of slightly desirable to desirable appearance and flavor. Rice bread scores for aroma, firmness and moistness ranged from 3.2 to 4.9. The breads were slightly undesirable to desirable in aroma, firmness and moistness. Flour C breads received lower firmness, moistness and flavor scores than Flour A and B rice breads.

CONCLUSIONS

A rice bread formula (Nishita, 1977) was adapted for use with three different local rice flours. Techniques that improved the appearance, volume, grain, flavor and reproducibility of the

Table III.7 - Results of the consumer panel pre-test for standard rice flour breads^a

CHARACTERISTICS	A - DAINTY	B - ENER-G	C - WOODWARDS
AROMA	4.2 ^b	3.9	4.2
APPEARANCE	4.0	5.0	4.6
FIRMNESS	3.9	3.4	3.2
MOISTNESS	4.9	4.0	3.4
FLAVOR	4.6	4.6	4.1
OVERALL ACCEPTABILITY	4.7	4.7	4.3

^a Average scores N=32 (aroma, appearance, firmness)
N=31 (moistness, flavor, overall acceptability)

20 were celiacs, 1 had a wheat allergy and 11 were on no special diet

^b Maximum score is 6

procédure were incorporated into the bread formulation. The three rice flour doughs required a remix method of preparation, two baking temperatures and specific water levels, proof times and baking times. Flour A and B breads were more moist, less firm, and more acceptable in flavor and overall acceptability than Flour C breads.

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IV. The Application of Response Surface Methodology
to the Development of Rice Flour Yeast Breads -
Ingredient Screening Experiments

INTRODUCTION

Response surface methodology (RSM) may be used to determine an optimal combination of gums and/or surfactants for gluten replacement in gluten-free breads. At the initiation of RSM research, screening experiments can be used to find the few effective variables from a large number of potential variables which may be affecting the response(s) of interest (Watson, 1961). The objective is to select those variables which have an important effect on the response rather than to examine the relationship between the variables, their interactions and the response (Mullen and Ennis, 1985). Ideally, the number of variables can be reduced to the 2 or 3 most important ones and then RSM designs can be utilized to study the relationships between the key variables and the responses in more detail (Mullen and Ennis, 1985).

Two-level fractional factorial designs can be used in screening experiments to locate the most important variables and the levels needed to optimize the response(s). A complete factorial design of seven variables at two levels would require 128 runs; however, a fractional factorial design can be used to obtain the desired information in fewer runs.

Typically, one response is observed in screening experiments. In breadmaking, loaf volume is the simplest, most reproducible

quantitative parameter. to nearly all other parameters are related (Chung et al., 1978). K and De Ruiter (1968) used loaf volumes as the most important criterion for the baking quality of non-wheat flour breads. Rice bread volume was related to the texture, grain and overall appearance of the bread, with larger loaf volumes producing the best rice bread quality (Nishita, 1973). Wheat bread loaf volume was a reliable indicator of emulsifier effectiveness in breadmaking (Schuster and Adams, 1984). Thus, loaf volume may be an important and relevant indicator for the determination of overall gluten-free bread quality.

Gums and surfactants, used as gluten replacements for the development of gluten-free yeast breads, have included carboxymethylcellulose (CMC) (McGreer, 1967; Glicksman et al., 1972; Kulp et al., 1974), hydroxypropylmethylcellulose (HPMC) (Sorenson, 1970; Bradley, 1972; Glicksman et al., 1972; Nishita, 1977), xanthan gum (Christianson et al., 1974; Kulp et al., 1974; Ranhotra et al., 1975; Bolam, 1983), guar gum (Smith, 1971), sodium-stearoyl-lactylate (SSL) (Smith, 1974; Kulp et al., 1974), and glycerol monostearate (GMS) (Jongh et al., 1968; Smith, 1971). Although no one gum or surfactant has proven to be an ideal gluten-replacement, a gum and/or surfactant combination may be more effective than either a single gum or surfactant (Knightly, 1968; Glicksman, 1969). However, research on the use of blends of gums and/or surfactants for the production of gluten-free yeast breads is limited. Thus, the determination of which gums and/or surfactants could be utilized for gluten-free breads, and at which levels, is warranted.

Rice flour is suitable for gluten-free yeast breads. However, the levels of gums and/or surfactants (variables) that are optimum for one

rice flour may differ for another rice flour, due to varietal and compositional differences among rice flours.

In the present study, RSM screening experiments were conducted to determine which variables (gums, surfactants and water), either singly or together, influenced the volume and loaf shape of rice breads sufficiently to merit further detailed study. The levels or range of levels of these variables, to be used in future RSM experiments, were also determined.

MATERIALS AND METHODS

Rice bread preparation

Sequential screening experiments were conducted independently for each of the three rice flours (A, B and C) described in Chapter III. Rice breads were prepared from the standardized formulations and ingredients (Chapter III), and were frozen (-29°C) 2-3 hr after baking.

Variables and their levels

A total of seven variables, CMC (cellulose gum, Hercules Inc.), HPMC (Methocel K4M, Dow Chemical Co.), xanthan gum (Keltrol, Merck and Co., Inc.), guar gum (FG 60-70, Hercules Inc.), SSL (Top-Scor S, Breddo), GMS (GMS-90, Breddo) and water, were altered in the screening experiments. The gums and surfactants were in a dry, powdered form except for the GMS, which was a hydrated (77% water) product. The actual level of GMS-90 used, was adjusted to compensate for the water content of the hydrate.

In the present study the four gums and two surfactants, which replaced the 7 g of HPMC, were incorporated into the standard rice bread formulations (Chapter III) with the dry ingredients. The actual variable levels utilized in the screening experiments were rounded and weighed to two decimal points. Specific water levels were incorporated so that 50 ml water was used to hydrate the yeast and the remainder was added to the dry ingredients, along with the hydrated yeast mixture (Chapter III).

The range (maximum and minimum) of gum and surfactant levels was determined by considering past research, previous baking trials, Canadian Food and Drug Regulations and the experimental design. Previous levels of gums used singly were too high to be used in a blend of all six gums and surfactants. A level of 7 g HPMC, the optimum level of HPMC in Nishita's (1977) formula, was taken as the initial maximum HPMC level, rather than the ideal (center point) level, because additional gums and emulsifiers might enhance HPMC effects. The ratio of 5 parts of HPMC to 3 parts CMC for wheat starch bread (Glicksman et al., 1972) was also considered. Canadian Food and Drug Regulations for SSL in bread permit a maximum of 3750 ppm of flour or 0.375% flour wt (The Food and Drugs Act and Regulations, 67-2A, Item S.1, 1981). The maximum level of use for the other variables is 'Good Manufacturing Practice' (unstandardized foods).

For a combination of gums and surfactants, the maximum level found in a gluten-free yeast bread formulation was 16.5 g of gum and surfactant with 265 g of wheat starch or 6.2% flour wt (Smith, 1971). Thus, a total gum/surfactant level of approximately 16.5 g was considered appropriate for the present research.

Water levels in the standard bread formulations were too low to accommodate the total gum levels used in the screening experiments. Thus, water levels were increased until the very thick and dry doughs could be mixed. The largest gum level was usually pre-tested with the highest water level to determine if the dough could be easily mixed. Generally, each flour only required one pre-test for water levels.

The initial water levels for Flour C, the first flour screened, were calculated by setting the water level in the standard formulation (170 ml) as the low water level. The maximum water level (208 ml) was set at 85% of the flour and gum weight combined (244 g). Smith (1971) added water to a wheat starch dough at the level of 85% of the weight of wheat starch plus the gum and surfactant levels combined. However, in this research, the water level of Smith (1971) was too low and resulted in an unmixable dough. The water was adjusted to 220 ml (90% of flours plus gum/surfactant wt) and 244 (100% of flours plus gum/surfactant wt) for the low and high water levels, respectively.

The initial maximum and minimum water levels for Flours A and B were determined by adjusting the Flour C water levels to reflect the water differences in the standard formulations for each flour. For example, the standard formula for rice Flour A required 205 ml water while that of Flour C needed 170 ml. Since Flour C initial water levels were 220 and 244 ml, to estimate the water levels for Flour A, 35 ml of water were added to both the initial high (279 ml) and low (255 ml) levels.

Once the maximum and minimum levels for each of the variables were selected, the levels of the variables utilized in each run or rice bread formulation were determined in accordance with the requirements

of the experimental design. In some screening trials, particular variables were deliberately held constant to evaluate the effects of other variables.

Rice bread quality measurements

Rice bread quality was determined on the basis of volume and loaf shape. Bread volume was measured by rapeseed displacement using a National Loaf Volumeter (National Mfg. Co., Lincoln, Nebraska). Three volume measurements were made per bread. Generally, each bread was placed in the volumeter right-side up. If a loaf had a large indentation in the bottom surface, the bread was placed in the volumeter upside-down. The five attributes of bread loaf shape were each scored on a 6-point category scale by the researcher. Loaf shape (maximum score = 30) was the sum of the scores for loaf symmetry, roundness of loaf top surface, size of loaf top-side indentation, straightness of loaf sides and flatness of loaf bottom surface. The loaf shape score-card is shown in Appendix 9. Both volume and loaf shape measurements were made on rice breads 75 min after baking.

Bench top sensory evaluations were conducted on most of the rice breads by the author and other members of the Department of Foods and Nutrition, University of Alberta. Rice breads were scored for crust color, crumb color, average cell size, cell size uniformity, cell wall thickness, aroma intensity, aroma desirability, firmness, moistness, gumminess, flavor intensity, flavor desirability, aftertaste intensity and aftertaste desirability on an 8-point category scale (Appendix 10). Evaluations were made on a single slice (1.25 cm thick) of thawed bread, equilibrated to room temperature (23°C).

Experimental design

A two-level fractional factorial design was used for preparing breads for each of the screening trials. The experimental design was a 1/16 fraction of the full 2^7 (two-level, seven variables) factorial (Box et al., 1978). The eight rice bread formulations (runs) of the design are shown in Table IV.1. The design was of resolution III with no main effects confounded with any other main effect, but certain main effects were confounded with two-factor interactions (Box, 1963). The confounding pattern for the main effects and the two-factor interactions is given in Appendix 11. The eight runs in the design (Table IV.1) were prepared in a random order. Oven order and proofer shelf position were also randomized according to a 3x3 Graeco latin square design (Box et al., 1978). As variables were eliminated during screening trials, only as many columns of the design as there were variables, were used. For the statistical analyses, the two levels (maximum and minimum) of each of the seven variables were coded as +1 and -1.

Statistical analyses

To estimate effects of variables on the responses (volume or loaf shape), the design and response data were fitted to a linear regression model. Regression equations were developed that included all the variables tested. As variables were eliminated from the design, the number of coefficients and variables in the regression equation was also reduced. The size and sign of the regression equation coefficients indicated variable importance and suggested the direction in which to change variables in order to improve the response. A large

Table IV.1 - The eight-run experimental design

RUN	VARIABLES ^a						
	x ₁	x ₂	x ₃	x ₄	x ₅	x ₆	x ₇
1	-1 ^b	-1	-1	+1	+1	+1	-1
2	+1 ^c	-1	-1	-1	-1	+1	+1
3	-1	+1	-1	-1	+1	-1	+1
4	+1	+1	-1	+1	-1	-1	-1
5	-1	-1	+1	+1	-1	-1	+1
6	+1	-1	+1	-1	+1	-1	-1
7	-1	+1	+1	-1	-1	+1	-1
8	+1	+1	+1	+1	+1	+1	+1

^a When all 7 variables were included in the design, x₁=CMC, x₂=xanthan gum, x₃=HPMC, x₄=guar gum, x₅=SSL, x₆=GMS, x₇=water

^b Minimum variable level

^c Maximum variable level

coefficient had a greater effect on the response than a small coefficient. A negative coefficient suggested that reducing the variable level would improve the response; a positive coefficient indicated that increasing the level of the variable would improve the response. The results of the screening trials were sequential, with the outcome of each trial indicating which variables to eliminate or in which direction to change the variable levels. Each screening trial was analyzed separately and/or together with all other trials.

RESULTS

The results for each flour will be discussed in the order that screening experiments were conducted.

Flour C

Six trials were completed during the screening of Flour C rice breads. The response (volume and loaf shape) regression equation coefficients for the variables tested in each trial are given in Table IV.2. Because of their importance, loaf shape responses were measured after Trial 4.

The bread volumes in Trial 1 were low (567-827 cc). The size and sign of the regression coefficients (Table IV.2) suggested that reducing the levels of CMC (x_1), xanthan gum (x_2) and HPMC (x_3) would increase loaf volume. The other variables [guar gum (x_4), SSL (x_5), GMS (x_6) and water (x_7)] had less of an effect on loaf volume. For Trial 2 the levels of CMC, xanthan gum, HPMC and guar gum were reduced as the negative coefficients suggested. Two less important variables

Table IV.2 - Response regression equation coefficients for variables tested - Flour C rice breads

TRIAL	VARIABLE	LEVELS (g)		COEFFICIENTS	
		-	+	VOLUME	LOAF SHAPE
1	CMC (x_1)	1.0	3.5	-57.29	
	xanthan (x_2)	1.0	3.5	-57.29	
	HPMC (x_3)	4.5	7.0	-40.11	
	guar (x_4)	1.0	3.5	-3.64	
	SSL (x_5)	0.6	0.8	-4.16	
	GMS (x_6)	1.0	3.5	19.79	
	water (x_7)	220.0	244.0	28.64	
2	CMC (x_1)	0.5	0.7	-11.99	
	xanthan (x_2)	0.5	0.7	4.69	
	HPMC (x_3)	3.0	4.0	12.51	
	guar (x_4)	0.5	0.7	-32.29	
	water (x_7)	220.0	244.0	-41.16	
3	CMC (x_1)	0.5	0.7	18.25	
	xanthan (x_2)	0.5	0.7	-51.55	
	HPMC (x_3)	3.0	4.0	1.05	
	guar (x_4)	0.5	0.7	-25.00	
	water (x_7)	170.0	194.0	272.90	
4	CMC (x_1)	1.0	3.5	-251.29	
	xanthan (x_2)	1.0	3.5	-112.24	
	HPMC (x_3)	4.5	7.0	18.49	
	guar (x_4)	1.0	3.5	-57.04	
	water (x_7)	220.0	244.0	112.24	
5	CMC (x_1)	0.7	1.1	-25.00	0.38
	xanthan (x_2)	0.5 ^a			
	HPMC (x_3)	0.0	4.0	240.62	-1.62
	guar (x_4)	0.5 ^a			
	water (x_7)	182.0	188.0	240.62	0.12
6	CMC (x_1)	0.7 ^a			
	xanthan (x_2)	0.0	0.5	-20.84	0.38
	HPMC (x_3)	2.0	4.0	32.81	-0.12
	guar (x_4)	0.0	0.5	-6.79	0.88
	water (x_7)	182.0 ^a			

^a Levels held constant throughout the trial

(SSL and GMS) were eliminated from the design. The water level of Trial 1 was retained.

Trial 2 rice breads had very large volumes (1302-1502 cc) and appeared to contain too much water (breads over-expanded in the oven). Water had the most effect on loaf volume (Table IV.2), water levels were reduced to improve loaf shapes.

The rice breads of Trial 3, had volumes that ranged from 725 to 1506 cc. The large regression coefficients (Table IV.2) for water, xanthan gum and guar indicated some influence on bread volume. However, the negative effects of both xanthan gum and guar gum suggested that a reduction in levels of both these gums might improve loaf volumes. HPMC level had little effect on loaf volume. Water levels were increased to affect loaf volume positively.

In Trial 4, the earlier elimination (Trial 2) of the surfactants was confirmed by re-running Trial 1, without GMS and SSL, to ascertain surfactant effects on the regression equation coefficients. If the surfactants produced only negligible changes in loaf volume, the regression coefficient trends should remain the same, with the surfactants omitted from the design. Flour C bread volumes (Trial 4) were much larger (525-1398 cc) than those of rice breads produced during Trial 1 (567-827 cc). Therefore, SSL and GMS were detrimental to bread volumes. The Trial 4 coefficients (Table IV.2) suggested that CMC, xanthan and guar levels should be reduced and water levels increased to improve bread volume. Although the sizes of the coefficients were larger in Trial 4, their effects were the same as those obtained in Trial 1. Level of HPMC had little effect on loaf volume in Trial 4.

To test the effect of GMS in rice breads, additional breads were baked with 1.0 and 3.5 g of GMS. The rice breads also contained 0.7 g CMC, 0.5 g xanthan gum, 0.5 g guar gum and 4.0 g HPMC with either 182 or 188 ml water. The eight loaves which were baked all had very low volumes (602 to 752 cc). GMS was obviously harmful to rice bread volume and not suitable for the formulations. Breads without GMS had higher crumb and crust color scores (brownier crust and whiter crumb) and more desirable aroma, flavor and aftertaste scores than rice breads with GMS.

Trial 5 evaluated the need for HPMC in the Flour C bread formulation. CMC levels were reduced to 0.7 g and 1.1 g; xanthan and guar gum levels were each held at 0.5 g. HPMC levels in the breads were set at either 0.0 g or 4.0 g. Water levels were reduced to allow for lower total gum levels in the formulations. Flour C breads without HPMC were very low in volume (700-775 cc). The breads with 4.0 g HPMC had volumes of 1060-1277 cc. The HPMC coefficient (Table IV.2) for loaf volume indicated that HPMC levels should be increased to improve volume. Thus, HPMC was an important component of the Flour C bread formulation if a large volume was desired. The volume coefficient for CMC suggested that the CMC level could be further reduced. However, the positive, but small loaf shape coefficient indicated that CMC level had a role in the maintenance of rice bread loaf shape. The loaf volume coefficient for water was positive and large.

In Trial 6, the importance of xanthan gum and guar gum to the volume and loaf shape of rice breads was observed. The bread volume coefficients (Table IV.2) for both xanthan gum and guar gum were

negative. Although the loaf shape coefficients suggested guar gum might improve loaf shapes, xanthan gum and guar gum were omitted from Flour C bread formulations, since the levels of 0.0 g and 0.5 g were already as low as possible.

Flour A

Five trials were completed during the screening of Flour A rice breads. Since SSL and GMS had detrimental effects on the loaf volumes of Flour C rice breads, they were not screened for Flour A breads. The initial gum levels utilized for Flour A breads were the same as those for Trial 1 Flour C breads. However, water levels differed as specified earlier. The Flour A response regression equation coefficients for the variables tested in each trial are given in Table IV.3.

In Trial 1, Flour A rice bread volumes ranged from 596 cc to 1125 cc. Loaf shape scores of 13 to 22 were obtained. The size and sign of the loaf volume regression coefficients (Table IV.3) suggested that CMC, xanthan gum and guar gum levels should be reduced to improve loaf volume. However, the positive CMC coefficient for loaf shape indicated that increased CMC levels could have a desirable effect on loaf shape. Additional HPMC was needed to improve both loaf volumes and shapes. An increase in water level would improve loaf volume but would be detrimental to rice bread loaf shape.

Trial 2 replicated Trial 1. Since a replication study (Chapter III) was conducted for the standardized formula of Flour C rice breads, but not for Flour A and B formulas, Trial 2 estimated the reproducibility of the Flour A rice bread standard formula. The results

Table IV.3 - Response regression equation coefficients for variables tested - Flour A rice breads.

TRIAL	VARIABLE	LEVELS (g)		COEFFICIENTS	
		-	+	VOLUME	LOAF SHAPE
1	CMC (x_1)	1.0	3.5	-163.02	1.50
	xanthan (x_2)	1.0	3.5	- 64.08	-1.50
	HPMC (x_3)	4.5	7.0	33.85	1.00
	guar (x_4)	1.0	3.5	- 57.30	0.75
	water (x_5)	255.0	279.0	65.62	-1.25
2	CMC (x_1)	1.0	3.5	-182.04	1.38
	xanthan (x_2)	1.0	3.5	- 55.46	-0.38
	HPMC (x_3)	4.5	7.0	33.59	1.12
	guar (x_4)	1.0	3.5	- 54.94	0.62
	water (x_5)	255.0	279.0	59.11	-1.38
3	CMC (x_1)	3.5 ^a			
	xanthan (x_2)	0.0	1.0	- 21.36	-0.38
	HPMC (x_3)	7.0	9.0	7.29	0.38
	guar (x_4)	0.0	1.0	- 11.99	-0.88
	water (x_5)	279.0 ^a			
4	CMC (x_1)	3.5 ^a			
	xanthan (x_2)	0.0	1.0	- 10.66	0.50
	HPMC (x_3)	7.0	9.0	- 2.86	1.00
	guar (x_4)	0.0	1.0	- 13.79	0.25
	water (x_5)	279.0 ^a			
5	CMC (x_1)	0.0	1.0	- 90.09	-2.75
	HPMC (x_3)	5.0	7.0	- 6.76	1.50
	water (x_5)	200.0	215.0	97.91	-1.25

^a Levels held constant throughout the trial

obtained for Trial 2 were similar to those for Trial 1; thus, Flour A formulation reproducibility appeared to be good.

Detailed examination of the CMC effects on loaf volume in Trials 1 and 2 (Table IV.4) indicated some interaction between xanthan, HPMC and/or guar at the high CMC level. All rice breads with high (+) CMC levels had low volumes except for the bread from one run (Run #6). In this run, bread volumes were high. Since this occurrence was noted in both Trials 1 and 2, Trial 3 further examined this gum interaction before the CMC level was decreased.

In Trial 3 (Table IV.3), CMC and water levels were held constant, xanthan gum and guar gum were reduced, HPMC levels were increased. Bread volumes ranged from 1031 cc to 1138 cc, loaf shape scores of 20-25 were obtained. No direct HPMC, guar, xanthan effect was evident and no reason for the high loaf volumes of Trial 1 and 2, high CMC breads was found. Coefficients (Table IV.3) for volume and loaf shape indicated that xanthan and guar gum levels should be reduced to improve volume and shape. The Flour A breads were slightly moist inside and some of the top crusts were wrinkling upon cooling. Thus, the Trial 3 breads needed extra baking.

Trial 4 replicated Trial 3, but the breads were baked for 55 min rather than a 45 min. Coefficients were checked to determine the effects of the longer baking on the coefficients of Trial 3. Rice bread volumes of 1000 cc to 1073 cc were produced. Good loaf shape scores of 20-25 were also obtained. The 55 min bake improved the bread so that the crumb was less moist and the crust had less wrinkling. Trial 4 confirmed Trial 3 results. The regression coefficients (Table IV.3) suggested that xanthan and guar gum should be reduced to improve

Table IV.4 - The effect of variable levels on Flour A rice bread volumes

RUN	VARIABLES ^a					VOLUME cc	
	x ₁	x ₂	x ₃	x ₄	x ₇	Trial 1	Trial 2
1	-1	-1	-1	+1	+1	1125	1173
2	+1	-1	-1	-1	-1	740	735
3	-1	+1	-1	-1	+1	1125	1104
4	+1	+1	-1	+1	-1	596	577
5	-1	-1	+1	+1	-1	1075	1054
6	+1	-1	+1	-1	+1	1038	983
7	-1	+1	+1	-1	-1	1048	1121
8	+1	+1	+1	+1	+1	696	700

^a x₁=CMC, x₂=xanthan gum, x₃=HPMC, x₄=guar gum, x₇=water

loaf shape. HPMC had little effect on volume, but a positive effect on loaf shape.

In the final trial (Trial 5), xanthan gum and guar gum were omitted from the Flour A bread formulation. CMC, HPMC and water levels were reduced. Bread volumes ranged from 779 cc to 1238 cc; loaf shape scores were 13 to 25. Regression coefficients (Table IV.3) for CMC indicated that CMC levels should be reduced to increase volumes, but increased to improve loaf shapes. HPMC level had little effect on volume, but showed a positive effect on loaf shape. The coefficient suggested that water levels should be increased to improve bread volume and decreased to improve loaf shape.

Flour B

The response (volume and loaf shape) regression equation coefficients for the variables tested in each of the three screening trials for Flour B breads appear in Table IV.5. For Trial 1, the gum levels used were the same as those used in the Flour C and A bread screening experiments. Trial 1 water levels were adjusted as described in the Methods section of this chapter. SSL and GMS were not screened. Flour B bread volumes ranged from 550 cc to 1288 cc. The loaf shape scores were 17 to 22. The CMC effect (Table IV.5) on loaf volume was the largest, suggesting CMC levels should be decreased to increase loaf volumes. Volume regression equation coefficients also indicated that xanthan and guar levels should be reduced and water levels increased. Water level and xanthan gum levels had a negative effect on loaf shape.

Trial 2, a repeat of Trial 1, tested the reproducibility of the

Table IV.5 - Response regression equation coefficients for variables tested - Flour B rice breads

TRIAL	VARIABLES	LEVELS (g)		COEFFICIENTS	
				VOLUME	LOAF SHAPE
1	CMC (x_1)	1.0	3.5	-227.86	0.12
	xanthan (x_2)	1.0	3.5	- 68.49	-0.38
	HPMC (x_3)	4.5	7.0	14.31	0.38
	guar (x_4)	1.0	3.5	- 60.69	0.12
	water (x_7)	213.0	237.0	87.76	-1.12
2	CMC (x_1)	1.0	3.5	-232.80	0.88
	xanthan (x_2)	1.0	3.5	- 78.65	0.12
	HPMC (x_3)	4.5	7.0	16.65	1.88
	guar (x_4)	1.0	3.5	- 34.38	-0.12
	water (x_7)	213.0	237.0	69.28	-1.38
3	CMC (x_1)	0.0	3.5	-236.72	1.58
	xanthan (x_2)	0.0	3.5	- 66.40	0.25
	HPMC (x_3)	4.5	7.0	31.50	0.67
	guar (x_4)	0.0	3.5	- 60.68	-0.50
	water (x_7)	213.0	237.0	70.58	-0.92

Flour B bread standard formula. Good repeatability was determined, particularly for bread volume. The regression coefficients and trends of Trial 2 were similar to those of Trial 1.

In Trial 3, the effects of low (0.0 g) and high (3.5 g) levels of CMC, xanthan gum and guar gum were evaluated. HPMC and water levels remained as in Trials 1 and 2. Results showed that the levels of CMC, xanthan and guar had to be reduced to increase bread volume. CMC also had a strong, positive effect on loaf shape. A high CMC level was needed to improve loaf shapes. Xanthan and guar gums were deleted from the Flour B bread formulation; CMC was retained to improve loaf shape. The HPMC effect was stronger (larger coefficients) in Trial 3 than in Trials 1 and 2. A strong water effect indicated that high water levels would increase volume but decrease loaf shape scores.

Determination of variable levels for a 5-level central composite design

The screening experiments facilitated the determination of the variables and levels of variables to include in the subsequent 5-level central composite design. RSM would be conducted to examine the variables, their interactions and their effects on a larger number of responses.

The actual variable levels for further study (Table IV.6) were chosen by considering 1) the screening trial results, 2) the combinations of CMC, HPMC and water (Table IV.7) that produced the best loaves during screening and 3) the proposed 5-level experimental design with an α -value of 1.633, which was to be used in subsequent studies. Levels of gums and water which could be easily weighed (i.e., 2 decimal

Table IV.6 - Variable levels selected for study

		LEVELS				
CODED LEVELS		-1.633	-1	0	1	1.633
CMC (X ₁) (g)	Flour A	0.37g	1.0g	2.0g	3.0g	3.63g
	Flour B	0.37	1.0	2.0	3.0	3.63
	Flour C	0.37	1.0	2.0	3.0	3.63
HPMC (X ₂) (g)	Flour A	6.37	7.0	8.0	9.0	9.63
	Flour B	5.37	6.0	7.0	8.0	8.63
	Flour C	3.05	4.0	5.5	7.0	7.95
WATER (X ₃) (g)	Flour A	194.74	215.0	247.0	279.0	299.26
	Flour B	171.34	184.0	204.0	224.0	236.66
	Flour C	169.97	182.0	201.0	220.0	232.03

Table IV.7 - Combinations of variable levels that produced the most desirable volume and loaf shape in rice breads during ingredient screening experiments

FLOUR	TRIAL	LEVELS (g)			VOLUME	LOAF SHAPE
		x ₁ (CMC)	x ₂ (HPMC)	x ₃ (WATER)		
A	1	1.0	7.0	255	1075	22
A	3	3.5	9.0	279	1135	25
A	4	3.5	9.0	279	1073	24
A	5	1.0	7.0	215	1092	25
B	1	1.0	7.0	213	1098	22
B	2	1.0	7.0	213	1106	22
B	3	3.5	7.0	237	1008	25
C	5	3.5	7.0	244	1125	-
C	5	1.0	7.0	220	1231	-
C	6	0.7	4.0	182	1225	22
C	6	1.1	4.0	188	1277	22
C	7	0.7	4.0	182	1206	22

points) were selected. The design variable levels were also chosen so that the optimal levels from the screening studies were placed close to the center with a moderate range between levels. Smith and Rose (1963) indicated that the center of the experimental design (0,0,0) was chosen so that it represented the set of conditions which, according to experience, was the best at the present time. Thus, the variable levels that produced the best breads during screening in the present experiment (Table IV.7) were positioned around the center point. Once the two +1 and -1 levels were chosen, the other three levels were calculated from the design format. For example, the 0 level is half-way between the -1 and +1 levels and the difference between the +1 and the +1.633 levels is 63.3% of the difference between the 0 and the +1 levels.

Water levels in each of the three (Flour A, B and C) bread formulations were pre-tested. The water levels of Flour B were reduced to 171, 184, 204, 224 and 237 ml. The original water levels chosen (204, 217, 237, 257 and 270 ml) were too high and the resulting loaves overexpanded in the oven.

DISCUSSION

Although volume was a good indicator of rice bread quality, it did have limitations. Ideally, a yeast bread should have a large loaf volume. However, as this study has shown, volume alone was an insufficient indicator of bread quality. Yeast breads also require a good loaf shape with a flat loaf bottom surface, straight loaf sides, round and symmetrical loaf top surface and a loaf top-side indentation

which is not too large. Large loaf volumes are not desirable if the breads have ballooned in the oven, are lopsided and have large air pockets under the crust. Loaf volume is the most powerful single and quantitative yardstick of breadmaking potential, but its limitations must be recognized (Pomeranz, 1981).

Of the seven variables tested, CMC, HPMC and water were the most important to rice bread volume and loaf shape. CMC was not effective in increasing bread volume, but did improve loaf shape. Nishita (1973) found that 2.1% (flour wt) CMC, the equivalent of 4.8 g in the present study, produced rice breads that had very small volumes and had textures which were compact and gummy. In Nishita's (1973) research, only one level of CMC was used and the water level was not adjusted. Thus, the low volumes may have been due to insufficient water in the bread formulations.

Rice bread volume was not dependent upon the actual HPMC level included in the formulations; however, HPMC was necessary for desirable loaf volume. Nishita et al. (1976) also noted that rice bread without HPMC had a very low volume.

Water level was dependent on the total gum level incorporated into the rice bread formulation. Since gums attract water, more water was needed as gum levels increased. High water levels also produced rice breads with large volumes. However, this was only advantageous up to a point. If water levels were too high, the breads expanded excessively in the oven and had irregular loaf shapes. The water effect on rice bread loaf shape was also demonstrated in Chapter III.

At the levels utilized in the present screening studies, xanthan gum and guar gum were ineffective in increasing loaf volumes or

improving the loaf shapes of rice breads. Thus, neither xanthan gum nor guar gum were considered for future experimentation.

Sodium-stearoyl-lactylate and GMS were detrimental to the loaf volume of rice flour breads. Nishita (1973) also found that any type of surfactant, including SSL, depressed rice bread loaf volumes severely. The surfactants may have interfered with the gum-water-rice flour complex such that no fermentation gases were retained and, thus, no leavening occurred (Bean and Nishita, 1983). Addition of monoglycerides to wheat starch breads was detrimental to bread crumb structure (Bradley, 1972). Bradley (1972) reported that monoglycerides decreased wheat starch bread volume and produced a crumbly, cake-like texture.

CONCLUSIONS

Using a two-level fractional factorial design, a series of screening experiments were successfully conducted to determine which gums and surfactants to include in rice bread formulations. CMC, HPMC and water were important variables for the production of rice breads with desirable loaf volumes and loaf shapes. A range of gum and water levels, for incorporation into the rice bread formulations was determined. The range of CMC levels was the same for Flours A, B and C. Specific levels of HPMC and water were required for each of the three flours.

For rice flour breads, bread volume alone was not a reliable measure of bread quality. Excessively large volumes could be obtained

in breads which were undesirable' in terms of overall loaf shape. A large volume and a good loaf shape, with a rounded top, straight sides and a flat bottom, are desired in rice breads.

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V. The Application of Response Surface Methodology
to the Development of Rice Flour Yeast Breads -
Objective Measurements¹

INTRODUCTION

There is an important need for an acceptable gluten-free yeast bread for individuals who suffer from celiac disease, wheat allergies and dermatitis herpetiformis. Although gluten-free breads are available commercially, their quality is inferior to that of regular wheat flour bread. Recently, Bell et al. (1981) reported that commercial wheat starch breads and baking mixes were tasteless and crumbled easily.

Most research on the development of gluten-free yeast breads has utilized wheat starch as a replacement for wheat flour (Pearson, 1960; McGreer, 1967; Jongh et al., 1968; Smith, 1971, 1974; Kulp et al., 1974; Ranhotra et al., 1975). However, many individuals sensitive to the gliadin fraction of gluten protein cannot tolerate even the very small amount of this protein in wheat starch. The protein content of wheat starch is approximately 0.15-0.3 percent (Bell et al., 1981; Campbell, 1982). In contrast, rice flour is naturally gluten-free (Nishita, 1973; Campbell, 1982) and relatively non-allergenic (James and McCaskill, 1983). Although published research focusing on the

¹ A version of this chapter has been submitted for publication. Ylimaki, G., Hawrysh, Z.J., Hardin, R.T. and Thomson, A.B.R. 1987. J. Food. Sci.

development of rice flour yeast bread (Nishita, 1973, 1977; Nishita et al., 1976; Nishita and Bean, 1979) is limited, researchers indicate that rice flours from short and medium grain rice generally have better baking qualities than rice flours from long grain rice (Nishita and Bean, 1979). In 100% rice flour breads, coarse rice flours functioned better than finely ground rice flours (Nishita and Bean, 1982).

Gluten-free breads require a gluten replacement to provide structure and gas retaining properties in the bread dough. A number of gums have been used, individually and in combinations, as gluten replacements with varying degrees of success (McGreer, 1967; Smith, 1971; Nishita, 1973; Kulp et al., 1974; Ranhotra et al., 1975). In the present study, three variables (hydroxypropylmethylcellulose [HPMC], carboxymethylcellulose [CMC] and water) were chosen as important structural components of rice flour yeast breads based on data for loaf volume and loaf shape obtained from the preliminary screening experiments described in Chapter IV.

Published research utilizing instrumental evaluation of gluten-free or starch breads is limited. Although the volume of gluten-free breads has been determined (Smith, 1971, 1974; Christianson et al., 1974; Ranhotra et al., 1975; Nishita et al., 1976) and measurements of bread crumb firmness (Christianson et al., 1974) and percent moisture (Smith, 1974) have been made, reports of determinations of loaf shape, crumb and crust color in gluten-free yeast breads have not been found.

Response surface methodology (RSM) is a statistical technique particularly appropriate for product development. Successful use of RSM in the development of baked goods such as cakes (Johnson and Zabik, 1981; Lee and Hosney, 1982; Neville and Setser, 1986; Vaisey-Genser et

al., 1987), cookies (Conner and Keagy, 1981) and high protein breads (Henselman et al., 1974) have been reported. However, published research describing the use of RSM for development of gluten-free breads is lacking. Thus, the objective of this study was to develop gluten-free rice flour yeast breads comparable to wheat flour (white) bread. Response surface methodology was utilized to find a combination of gums and water which could successfully replace gluten in breads made from several types of locally available rice flours.

MATERIALS AND METHODS

Rice flours

Three different local rice flours were selected for bread preparation - A, a medium grain/finely ground flour; B, a medium grain/coarsely ground flour; C, a long grain/finely ground flour. Flours were stored at 4°C. The % fat, % moisture, % protein of the flours, determined by standard AOAC (1980) methods 14.003, 14.026 and 14.019, respectively, and amylograph gelatinization temperatures, obtained by the method of Juliano et al. (1985) using 20% slurries, are shown in Table V.1.

Variables

Two gums, carboxymethylcellulose (CMC), obtained from Hercules Incorporated, and hydroxypropylmethylcellulose (HPMC), obtained from The Dow Chemical Company, and water were the three variables chosen for study on the basis of data from ingredient screening experiments (Chapter IV).

Rice bread preparation

A rice bread formula (Nishita, 1977) was adapted and standardized for each of the three rice flours (Chapter III). The basic rice bread formula, which included 20% potato flour, is shown in Table V.2. To prepare the rice breads, 4 g of sugar was dissolved in 50 ml warm water (43°C). The yeast was added and soaked for 10 min. The rice flour, potato starch, salt, remaining sugar, HPMC and CMC were premixed for 2 min at no. 1 speed in a Hobart KitchenAid K45SS 10-speed mixer using a stainless steel bowl (4.5 qt) and whip attachment. The yeast mixture and remaining water were added to the dry ingredients and mixed for 15 sec at no. 2 speed. The oil was added and mixed for 15 sec at no. 4 speed and then 5 min at no. 6 speed. The sides of the bowl were scraped down halfway through the 5 min mix. The resultant batter was proofed in the bowl for 30 min (30°C and 95% humidity) and then remixed for 5 min at no. 6 speed. The batter (400 g) was placed into a greased aluminum loaf pan (18.7 x 9.2 x 5.7 cm) and flattened with an oiled spatula, rounding the upper edges of the batter. The panned batter was proofed (Flour A - 45 min; Flours B and C - 50 min) at 30°C/95% humidity and baked at 215°C for 5 min, then at 180°C for an additional 50 min (Flour A) or 40 min (Flours B and C). Breads were cooled for 15 min and removed from their pans.

Measurements of bread weight, volume, specific volume and loaf shape score were made 75 minutes after baking. Crust color, crumb color, percent moisture and Instron firmness were determined on bread which had previously been frozen (-29°C) for 4-6 days and thawed (21°C) 1.5-2 hours before sampling.

Table V.1 - Composition of rice flours

RICE FLOUR	% FAT	% MOISTURE	% PROTEIN	GELATINIZATION TEMPERATURE (°C)
Flour A	1.2	10.84	6.7	64.5
Flour B	0.9	12.80	5.9	65.0
Flour C	0.8	10.82	6.2	72.5

Table V.2 - Basic rice flour bread formula^a

INGREDIENTS	WEIGHT (g)	% FLOUR WEIGHT
Rice Flour	182	80.2
Potato Starch	45	19.8
Salt	6	2.6
Sugar	24	10.6
Yeast	7	3.1
Oil	13	5.7
Hydroxypropylmethylcellulose ^{b,c}		
Carboxymethylcellulose ^{b,d}		
Water ^b		

^a Adapted from Nishita, 1977

^b The levels of hydroxypropylmethylcellulose, carboxymethylcellulose and water were varied according to flour type and the experimental design as shown in Table V.4

^c Methocel K4M, The Dow Chemical Company

^d Hercules Cellulose Gum, Hercules Incorporated

Reference bread

A commercial wheat flour (white) bread served as the reference for all objective measurements. The bread was purchased within 4 hr of baking and measurements of bread weight, volume, specific volume and loaf shape were made. Crust color, crumb color, percent moisture and Instron firmness were determined on bread which had previously been frozen (-29°C) for 1-5 days, and thawed (21°C) for 1.5-2 hours before sampling. Eighteen loaves of bread were purchased in five different lots over a six-week period.

Objective measurements

Objective measurements were made on all rice and reference breads. Bread volume was determined by rapeseed displacement using a National Loaf Volumeter (National Mfg. Co., Lincoln, Nebraska). Specific volume (cc/g) was calculated as volume divided by weight. The loaves were scored for loaf shape (symmetry and roundness of loaf top surface; size of loaf top-side indentation, straightness of loaf sides and flatness of loaf bottom surface) by the researcher using a 6-point category scale, (Appendix 9), where six represented the highest quality for each attribute (maximum loaf score = 30). Samples for objective measurements of crust color, crumb color, percent moisture and Instron firmness were taken from the same relative position in each thawed loaf of bread. Measurements were made on samples equilibrated to room temperature (22°C). Percentage moisture was determined by a modification of the two-stage standard AACC method 44-15A (1983). The air-drying and oven-drying stages were extended to 96 hr and 24 hr, respectively, for the moist rice flour breads. Bread firmness was tested using an Instron Food Testing System (Model 4201) and the

testing conditions recommended by Baker et al. (1986). Firmness was recorded as the maximum force (kg) after one compression. For Instron firmness determinations, rounds 2.5 cm in diameter were removed from the centers of each of ten slices (1.25 cm). A total of five firmness measurements were made on each loaf of bread using two rounds (2.5 cm in diameter) from adjacent slices for each measurement. Crust and crumb colors were determined with a Hunterlab Color/Difference Meter D25-2. The meter was standardized using the white standard tile (C2-8692) with values of $L=92.7$, $a=-1.0$, $b=0.3$. Two rounds of crust (2.5 cm in diameter) were taken for crust color determinations. Samples (2 rounds from each of 2 slices) were taken from the center top crust for crumb color determinations. Two crumb color measurements were made on each loaf of bread by using two rounds from adjacent slices to form one color sample.

Experimental design

For each rice flour, a central composite design, with three blocks and with six replications of the center point (Meyers, 1971), was used. The experimental design (Table V.3) consisted of a three-variable (CMC, HPMC and water), five-level pattern with twenty runs (rice bread formulations) prepared over the three blocks (consecutive days). The design was replicated twice for each flour. For the statistical analyses, the five levels of each of the three variables were coded as -1.633, -1, 0, +1, +1.633 (Table V.4).

Statistical analysis

For each objective response, analysis of variance (ANOVA) was

Table V.3 - Experimental design^a

VARIABLES	CODED LEVELS		
	X ₁ (CMC)	X ₂ (HPMC)	X ₃ (WATER)
BLOCK 1	1	1	1
	1	-1	-1
	-1	1	-1
	-1	-1	1
	0	0	0
	0	0	0
BLOCK 2	1	1	-1
	1	-1	1
	-1	1	1
	-1	-1	-1
	0	0	0
	0	0	0
BLOCK 3	-1.633	0	0
	1.633	0	0
	0	-1.633	0
	0	1.633	0
	0	0	-1.633
	0	0	1.633
	0	0	0
	0	0	0

^a Blocks and treatment combinations within a block were randomized.

Table V.4 - Variable levels

		LEVELS				
CODED LEVELS		-1.633	-1	0	1	1.633
CMC (X ₁) (g)	Flour A	0.37g	1.0g	2.0g	3.0g	3.63g
	Flour B	0.37	1.0	2.0	3.0	3.63
	Flour C	0.37	1.0	2.0	3.0	3.63
HPMC (X ₂) (g)	Flour A	6.37	7.0	8.0	9.0	9.63
	Flour B	5.37	6.0	7.0	8.0	8.63
	Flour C	3.05	4.0	5.5	7.0	7.95
WATER (X ₃) (g)	Flour A	194.74	215.0	247.0	279.0	299.26
	Flour B	171.34	184.0	204.0	224.0	236.66
	Flour C	169.97	182.0	201.0	220.0	232.03

conducted to determine significant differences among the 15 different treatment combinations. Also, data were analyzed using multiple regression procedures. To estimate CMC, HPMC and water effects on each objective response, the design was fitted to the second-order regression equation:

$$Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$$

including linear, quadratic and interaction effects. Coefficients of determination (R^2) were computed. The adequacy of the models was tested by separating the residual sum of squares into pure error and lack of fit. Replicate and block effects, as well as their contributions to R^2 , were removed from the equations; all other terms were retained.

For each response, contour plots (generated by Statgraphics, STSC, Inc., 1986) were produced from the equations, by holding the variable with the least effect on the response equal to a constant value, and changing the other two variables. Each contour plot was compared to reference wheat flour bread data. Areas on each plot that represented bread formulations which met reference standards were identified. For each flour, contour plots for all responses were superimposed to locate rice bread formulations which met reference standards for all objective responses.

RESULTS AND DISCUSSION

For each flour, ANOVA of the fifteen rice bread treatments showed significant differences for all responses except for rice bread crumb color 'a' values for Flours B and C, and crust color 'a' and 'b' values

for Flour B breads. The lack of a significant difference between treatments, for a response, indicated that HPMC, CMC and water had no effect on the response. Thus, for those responses further statistical analysis was not required. Mean values, taken over the 40 design points (20 design points x 2 reps), for Flour B bread crust color 'a' and 'b' values and crumb color 'a' values, and for Flour C bread crumb color 'a' values, were compared to REF wheat bread standards (Table V.5). REF standards were met by Flour B breads for crust color 'a' values and crumb color 'a' values. Flour C breads met REF standards for crumb color 'a' values. Flour B breads were unable to meet crust color 'b' value REF standards as they were slightly more yellow (20.8 compared to REF standards of 15.3 - 20.2) than the REF wheat bread.

Twenty-six regression equations were computed for the responses which had shown significant differences in the ANOVA (Flour A, 10 responses; Flour B, 7 responses; Flour C, 9 responses). Each regression equation was tested for lack of fit. Responses for loaf shape (Flour C), specific volume (Flours A, B and C), crust color 'L' (Flour B), Instron firmness (Flours A, B and C) and percent moisture (Flour C) showed significant lack of fit. Although significant lack of fit may indicate inadequacy of the regression equation, this test may not be appropriate in the present study. Pure error, utilized to calculate lack of fit, was estimated from the data of the 12 center points of the replicated experimental design. In the present research, center point rice breads were easier to sample and test instrumentally than some of the treatment breads. Thus, the pure error obtained in sampling and testing the center point rice breads may not be representative of the overall error in the other treatment combinations.

Table V.5 - Reference wheat bread objective response measurement standards

RESPONSE	MEAN ^a	RANGE
Loaf shape ^b	26.4	24-28
Specific volume, cc/g	5.5	4.5-6.2
Crust color 'L'	41.3	35.9-44.7
'a'	13.2	12.4-14.4
'b'	17.7	15.3-20.2
Crumb color, 'L'	64.2	57.2-67.8
'a'	-1.2	-0.8-(-1.6)
'b'	7.9	7.0-9.0
Instron firmness, kg	.0036	.020-.084
Percent moisture	37.1	35-40

^a n=18 loaves

^b Maximum score = 30

The regression coefficients and R^2 values for loaf shape scores of breads from each of the three flours are shown in Table V.6. The size and significance of the regression equation coefficients indicate the relative importance of the variables (Mullen and Ennis, 1979). Because the size and importance of the linear, quadratic and interaction effects for loaf shape differed among the three rice flours in the present study, flour differences are suggested. Loaf shape R^2 values also differed among the three rice flours. Regression equations explained 80% and 72% of the variation in loaf shape scores for Flours B and C, respectively, but only 57% of the variation for Flour A. Regression equation coefficients and R^2 differences among the three flours were apparent for many of the other objective responses measured. Overall, the twenty-six regression equations explained 53% to 94% of the response variation. Regression equation coefficients and R^2 values for Flour A, B and C responses are reported in Appendices 12, 13 and 14, respectively. For each flour, regression coefficients showed that water and CMC affected the responses most; HPMC had the least effect. Thus, to study the response surfaces within each rice flour, contour plots were produced for the 26 responses by holding HPMC levels at -1.633, 0 and 1.633. Contour plots for all objective responses appear in Appendices 18 to 20 and should be referred to in the discussion of the results pertaining to each of the three flours. Each contour plot was compared to reference wheat bread (REF) response data (Table V.5). Areas on the contour plots that represented rice bread formulations which met the range of REF response values for that specific response were identified. In the case of crumb color 'L'

Table V.6 - Regression equation^{ab} coefficients and R² values
for loaf shape scores

COEFFICIENT	FLOURS		
	A	B	C
b ₀	20.576	22.960	22.917
b ₁ (CMC)	1.154**	2.494***	1.686**
b ₂ (HPMC)	-0.596	0.634	0.725*
b ₃ (WATER)	-2.142***	-1.416***	-1.134**
b ₁₁	-0.368	-1.904***	-1.219*
b ₂₂	0.007	0.159	-0.188
b ₃₃	-0.087	-1.341***	-1.031
b ₁₂	0.438	-0.125	-0.312***
b ₁₃	0.938	2.000***	0.688
b ₂₃	-1.188*	-0.125	-0.312**
R ^{2b}	.57	.80	.72

^a $Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$
where $x_1 = \text{CMC}$, $x_2 = \text{HPMC}$, $x_3 = \text{water}$.

^b Replication and block effects removed.

*, **, *** Significant at $P < 0.05$, $P < 0.01$ and $P < 0.001$, respectively.

values, rice bread responses within the REF range or higher (whiter) were considered to have met the REF standards.

Flour A

Only Flour A rice breads with the lowest HPMC level (-1.633) met REF standards (Table V.5) for loaf shape. Water levels below -1.0 and any CMC level gave loaf shape scores of 24 or greater. As the water levels in the breads were increased above -1.0, loaf shape scores fell. Nishita (1973) noted that rice breads with excess water were asymmetrical in shape. However, in the present study, loaf shape scores approached those of the REF at water levels above -1.0, if the level of CMC was increased.

HPMC had no effect on the specific volume of Flour A breads. Compared to the REF, rice breads tended to have lower specific volumes. For specific volume responses similar to those of the REF, Flour A breads with water levels above -1.0 and low CMC levels were needed. In contrast, Nishita (1973) reported that the specific volumes of 100% rice flour breads increased from 1.5 ml/g at 0% HPMC to 5.3 ml/g at 3% HPMC (which corresponds to -1 levels in the present study), and decreased with each subsequent level. This suggests an optimum level of HPMC which was not demonstrated in the present research.

Rice bread crust color 'L' value contour plots representing the three HPMC levels, appear in Fig. V.1. The contour plots are saddle-shaped and show a region of minimum 'L' that diagonally crosses each figure, with 'L' values increasing peripherally. The rice bread region meeting the REF crust color 'L' (lightness) value standards decreased as the HPMC level increased. To retain a constant 'L' value

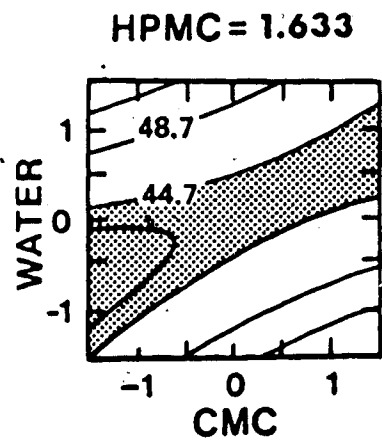
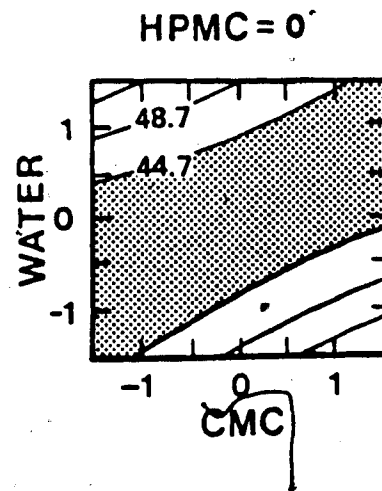
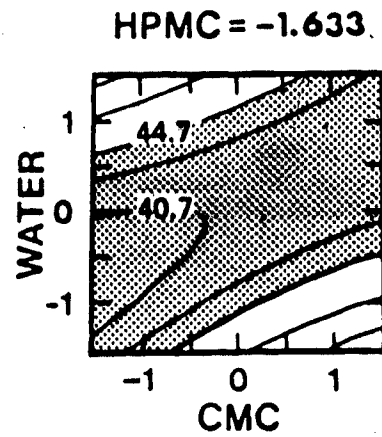


Figure V.1 Contour plots for Flour A rice bread crust color 'L' values. Levels of HPMC, water and CMC are coded values. The shaded regions met crust color 'L' reference standards of 35.9 to 44.7

in rice breads at each HPMC level, extra water was needed as CMC was increased.

Flour A bread crust color 'a' (redness) values were affected slightly by HPMC level. Except for high water-low CMC levels, all rice breads met REF crust color 'a' standards.

The REF standards for crust color 'b' (yellowness) values were most often met by Flour A breads with -1.633 HPMC. However, for rice breads with 1.633 HPMC, low water levels (-1.4 to 0.0) and CMC levels less than 1.0 were needed to meet crust color 'b' value REF standards.

Rice bread crumb color 'L' value contour plots at all HPMC levels were similar, indicating no HPMC effect. All Flour A breads had a whiter crumb color than the REF. A bright white crumb color is desirable in a white bread, therefore, a crumb color that was as white or whiter than the REF was considered to have met the REF standards. The contour plot of crumb color 'L' values at -1.633 HPMC showed that the highest rice bread 'L' values (whitest bread crumb) were obtained at high water, low CMC levels.

HPMC had only a slight effect on Flour A bread crumb color 'a' values. Flour A breads from all water-CMC combinations met REF standards for crumb color 'a' values, except at the 1.633 HPMC level. In these breads (1.633 HPMC), low water-high CMC levels gave rice breads with crumb color 'a' values higher than the REF.

Rice bread crumb color 'b' values were not affected by HPMC level. To keep rice bread crumb color 'b' values within the REF standards, water levels between -1.1 and 0.6 with any CMC level could be used.

At all HPMC levels, Flour A bread contour plots for Instron firmness had a minimum which diagonally crossed each plot, with

increasing firmness values on either side. The rice bread region meeting Instron firmness REF standards enlarged as the HPMC level increased. Rice breads were softest at water levels of -1.0 to 1.0 and low CMC levels, and at water and CMC levels above 0.

One of the most undesirable characteristics of gluten-free breads is a firm, crumbly crumb. Therefore, it is essential to include measures of crumb firmness when attempting to improve the quality of gluten-free breads. However, published research on the objective measurement of bread firmness is noticeably lacking. In the present study, the moist rice breads presented some sampling difficulties not found in the REF wheat bread. If the rice breads were very moist and gummy, the samples were unavoidably compressed to a slight extent during sampling and did not regain the initial 1.25 cm thickness. Thus, when these samples were tested for firmness, using an Instron pre-set to compress a 1.25 cm sample 25%, they could not be compressed as much as the samples which were 1.25 cm thick. This problem might be eliminated by the use of whole slices of bread instead of rounds, and by adjusting the position of the compression plunger for each sample, such that the plunger barely touched the surface of the bread before compression (Baker et al., 1986). Although this sampling difficulty only occurred in breads from one of the rice flours (Flour A) in the present study, it should be considered if a very moist bread were to be studied.

No HPMC effect on percent moisture of Flour A rice breads was found. To meet percent moisture REF standards, any water level with CMC levels below -0.5 was necessary in rice bread formulations. Most rice breads were moister than the REF. This is understandable since

75-132% (flour wt) water was added to the rice flour breads while wheat bread formulas typically include a maximum of 60% water on a flour basis (Kim and De Ruiter, 1968). Nishita (1973) found that 75% (flour wt) water was the optimal level for 100% rice flour breads. Lower water levels produced a dense rice bread with a low volume (Nishita, 1973). In the present study, the high starch content of the rice breads and the addition of water binding gums both increased the water requirements of the rice breads over those of wheat breads. Ranhotra et al. (1975) also found that gluten-free wheat starch bread formulas required more water than wheat bread formulas.

The results obtained when contour plots for all responses for Flour A breads were superimposed, suggested that, at an HPMC level of -1.633, Flour A rice breads with water levels between -0.85 and 0.6, and CMC levels below -1.0 would meet all REF standards except loaf shape (Fig. V.2). To meet loaf shape REF standards at -1.633 HPMC, water and CMC levels less than -1.0 were needed in Flour A rice breads.

Flours B and C

Since HPMC and water levels differed in the bread formulations for each of the three rice flours (Table V.4), direct comparison of the breads resulting from the flours was not feasible. Since the ANOVA for crumb color 'a' values of breads from rice Flours B and C, and crust color 'a' and 'b' values of breads from Flour B were not significant, regression equations and response contour plots were not produced for these responses. However, REF standards were met by Flour B rice breads for crust color 'a' values and crumb color 'a' values and by Flour C rice breads for crumb color 'a' values. Examination of the

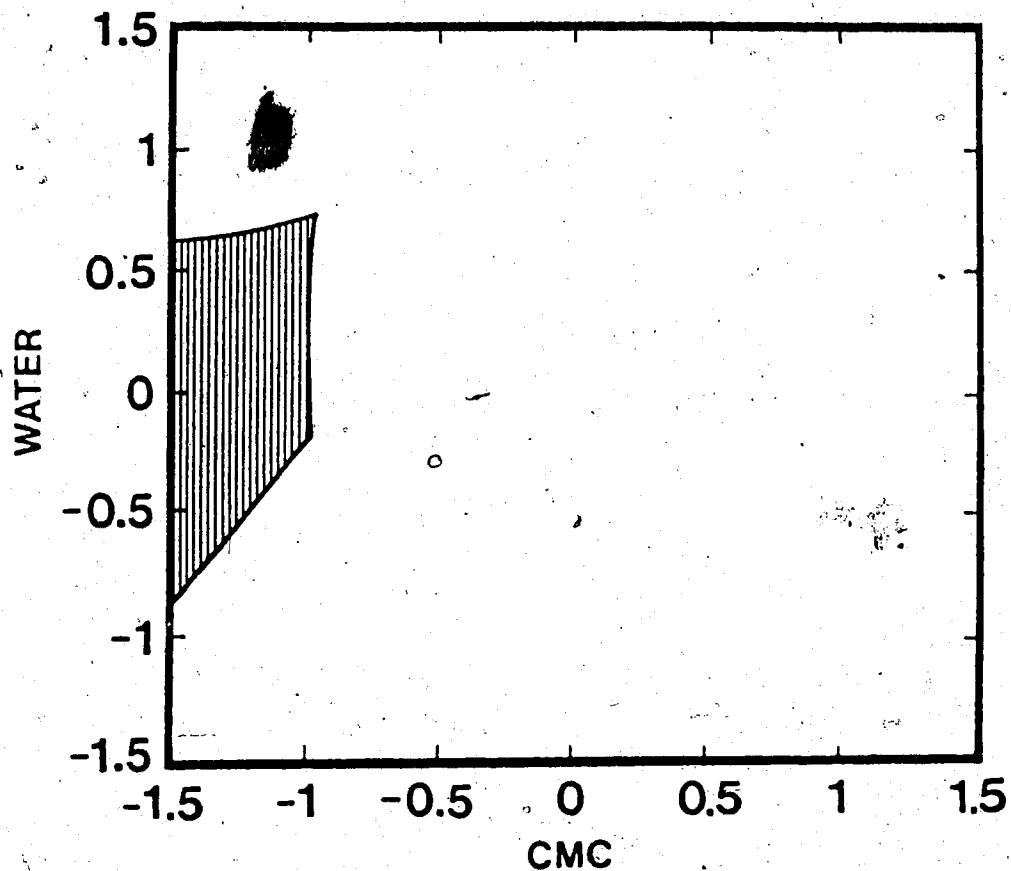


Figure V. Superimposed contour plot region for Flour A rice bread objective responses at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for specific volume, crust color 'L, a and b' values, crumb color 'L, a and b' values, Instron firmness and percent moisture.

contour plot trends for all other responses gave some insight into response differences among the rice breads from specific flours.

None of the rice breads from Flours B and C met the REF standards for loaf shape. However, as for Flour A, the loaf shapes of breads from rice Flours B and C were affected by HPMC level. Rice breads made from Flour C showed maximum loaf shapes at the -1.633 and 0 levels of HPMC; Flour B rice breads displayed maximum loaf shapes at the 1.633 HPMC level. CMC effects were also evident for Flours B and C. At each HPMC level, water levels between 0.5 and -0.5 with CMC levels above 0.0 were needed to attain the maximum loaf shape scores for each of the flours (B and C). In contrast, this CMC effect was not determined for Flour A rice bread loaf shape scores unless the water level was above -1.0.

The specific volume contour plots for breads from Flours B and C were similar to those of Flour A, representing rising ridge surfaces. While Flour B breads showed no HPMC effect and met the REF standards at all HPMC levels (as did Flour A), Flour C rice bread formulations met the REF standards more often at the -1.633 level of HPMC than at 1.633. For breads from all three flours, specific volumes were similar to the REF at water levels above 0 and at low CMC levels (less than -1.0).

The crust color 'L' values of flour B rice breads were affected by HPMC level. Crust color 'L' values for Flour B breads met the REF standards most often at 1.633 HPMC. Flour C bread crust color 'L' values were only affected slightly by HPMC level. All Flour C rice breads had a lighter crust (higher 'L' values) than the REF. Crust color 'a' values for Flour C breads showed an HPMC effect. At 1.633 HPMC, Flour C rice breads met the crust color 'a' REF standards at all

water-CMC combinations except at water levels below -0.3 and CMC levels above 0.6. Flour C rice breads did not meet REF standards for crust color 'b' values at any HPMC, water or CMC level.

Although each of the flours (B and C) resulted in bread crumb color 'L' contour plots which differed in appearance from those of Flour A, Flour B and C rice breads, like Flour A breads, had a whiter crumb than the REF. Crumb color 'b' contour plots for breads from both Flours B and C were similar to those of Flour A. While HPMC had no effect on crumb color 'b' values for Flour A rice breads, HPMC did affect the crumb color 'b' values for Flour B and C rice breads. In both Flour B and C breads, the REF standards were met most often at 1.633 HPMC.

Flour B and C contour plots for Instron bread firmness showed strong HPMC effects and were similar to the plots for Flour A Instron bread firmness. However, in contrast to Flour A, the Flour B rice bread region meeting the REF standards decreased as the HPMC level increased. Flour C breads only met REF standards for Instron firmness at the 1.633 HPMC level.

The HPMC effects on the percentages of moisture of Flour B and C rice breads were slight. Rice breads from both of these flours (B and C) were noticeably drier than Flour A rice breads. Although breads from both Flours B and C met REF percent moisture standards more frequently than Flour A rice breads, Flour C breads attained the REF standards most often.

Response contour plots for rice breads from Flours B and C, respectively, were superimposed. For Flour B rice breads a -1.633 HPMC, specific water (-0.3 to 0.85) and CMC (<-1.1) combinations

met REF standards for specific volume, crust color 'a' values, crumb color, 'L, a and b' values, Instron firmness and percent moisture (Fig. V.3). For Flour C rice breads at -1.633 HPMC, specific water (0.1 to 1.5) and CMC (<-0.8) combinations met REF standards for specific volume, crust color 'a' values, crumb color 'L, a and b' values and percent moisture (Fig. V.4).

In the present study, the rice flours selected for use were a medium grain, finely ground flour (A); a medium grain, coarsely ground flour (B) and a long grain, finely ground flour (C). At the -1.633 HPMC level, rice breads from Flour A had the largest number of water-CMC combinations which met REF standard objective responses (9 out of 10) determined in breads. Flour A was also the only rice flour to produce breads which met loaf shape REF standards. Breads from Flours B and C, with -1.633 HPMC, had fewer water-CMC combinations meeting the REF standard responses than Flour A breads. Breads from Flours B and C met seven (Flour B) and 6 (Flour C) of the ten objective REF standards simultaneously. Thus, on the basis of objective measurements, the medium grain, finely ground rice flour (Flour A) produced rice breads that were more like wheat flour breads than either the medium grain, coarsely ground (Flour B) or the long grain, finely ground (Flour C) rice flours. Nishita and Bean (1979) also found that flours from medium grain rice had better baking qualities than flours from long grain rice. However, Nishita and Bean (1982) reported that coarse rice flours functioned more effectively than finely ground rice flours in their 100% rice flour breads.

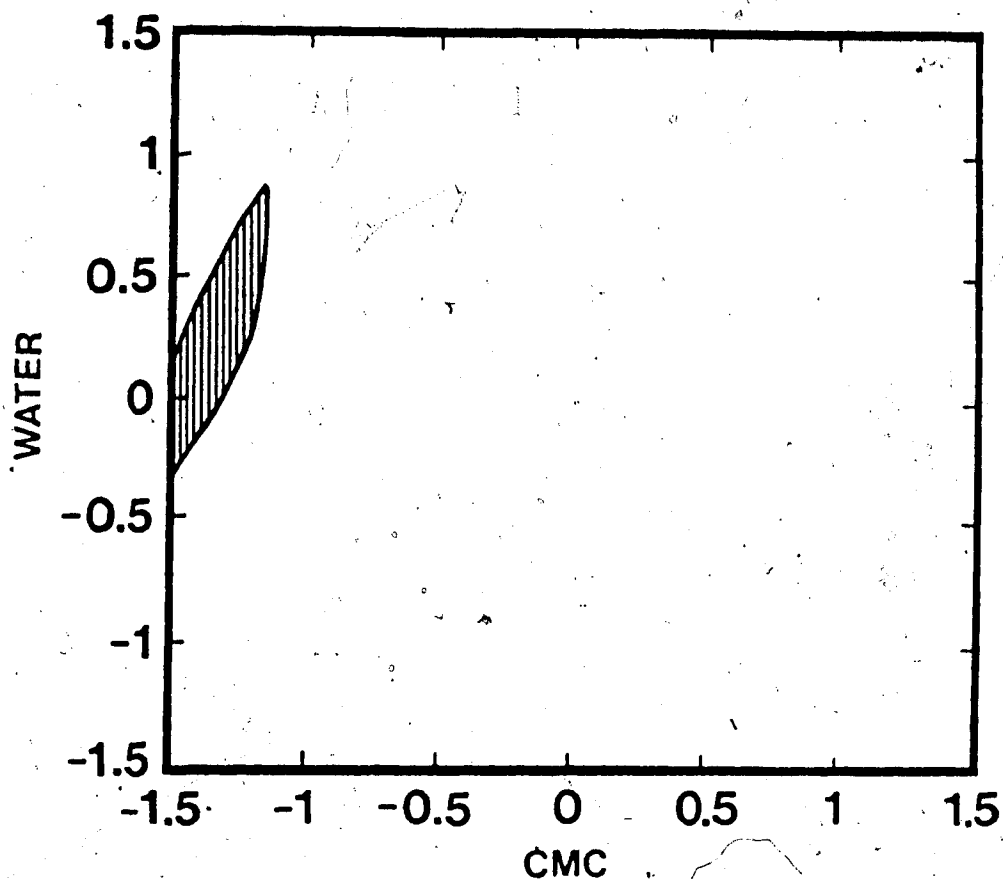


Figure V.3 Superimposed contour plot region for Flour B rice bread objective responses at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for specific volume, crust color 'a' values, crumb color 'L, a and b' values, Instron firmness and percent moisture.

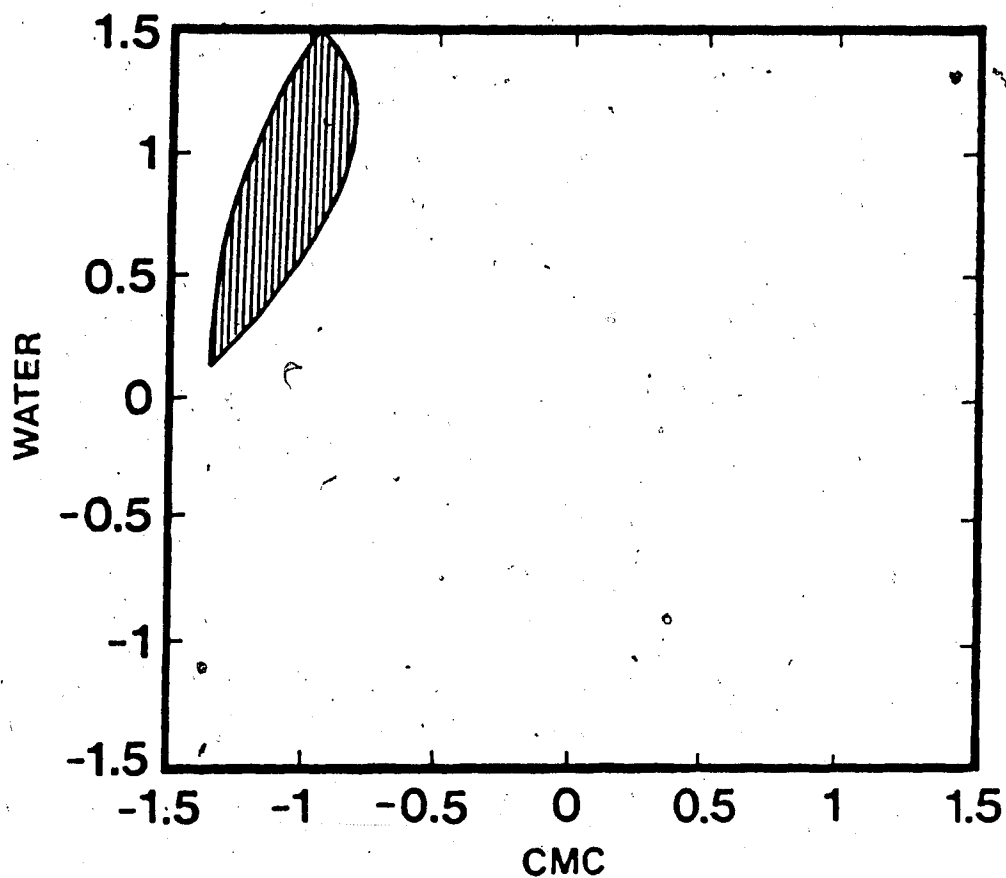


Figure V.4 Superimposed contour plot region for Flour C rice bread objective responses at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for specific volume, crust color 'a' values, crumb color 'L, a and b' values, and percent moisture.

CONCLUSIONS

This study has resulted in the development of formulations for acceptable rice flour yeast breads for individuals with celiac disease, wheat allergies and dermatitis herpetiformis. Several optimum combinations of HPMC, CMC and water were found that could result in rice breads with a specific volume, crust color ('L, a and b' values), crumb color ('L, a and b' values), Instron firmness and percent moisture comparable to that of a reference wheat (white) bread. As illustrated in this research, development of rice breads comparable to the reference wheat bread, at particular HPMC levels, was dependent on the type of rice flour and on the levels of water and CMC used in the formulation. Thus, it is suggested that response surface methodology can be successfully applied to the development of acceptable special diet products, such as gluten-free rice bread.

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VI. The Application of Response Surface Methodology
to the Development of Rice Flour Yeast Breads -
Trained and Consumer Panel Sensory Evaluation

INTRODUCTION

Special diet products, such as gluten-free breads, frequently present difficulties in providing the sensory quality that consumers expect (Paulus, 1986). To ensure that gluten-free bread is accepted, enjoyed and consumed regularly, gluten-free yeast breads which have organoleptic (odor, flavor and texture) and visual characteristics that are similar to those of wheat flour yeast bread are essential.

Research on gluten-free yeast breads which has included sensory evaluation by a trained taste panel (Bolam, 1983) is very limited. There are also few published studies on the consumer acceptability of gluten-free breads. Some researchers have reported gluten-free bread preference/acceptability with 5-57 member panels (Smith, 1971; Bradley, 1972; Nishita, 1973; Christianson et al., 1974; Johnson and Penfield, 1976). Others (Steele et al., 1965; McGreer, 1967; Landsman and Wills, 1968; Kulp et al., 1974; Smith, 1974) have mentioned bread acceptability but have provided very little information about who evaluated the products or about the type of test.

Response surface methodology (RSM) has been successfully used for trained taste panel evaluation of baked products such as pie crusts (Smith and Rose, 1963), high protein bread (Henselman et al., 1974) and cakes (Vaisey-Genser et al., 1987). However, the use of both RSM

and trained panel sensory evaluation for the development of gluten-free breads is noticeably absent.

The purpose of this study was to develop gluten-free rice flour yeast breads comparable to wheat flour (white) breads in appearance, odor, flavor, texture and aftertaste, using RSM. Consumer acceptability of selected breads was determined with a consumer panel consisting of celiac and non-celiac members.

MATERIALS AND METHODS

Detailed information about the rice flours, variables and experimental design of the study is given in Chapter V.

Rice bread preparation

Yeast breads were prepared from three different rice flours using the formulations and procedures given in Chapter V.

Reference bread

A commercially prepared wheat flour (white) bread served as the reference (REF) for all trained panel sensory evaluations.

Sensory evaluation - trained panel

Screening of panelists. Panelists were chosen during a preliminary screening of 19 volunteers (female graduate students and staff in the Department of Foods and Nutrition, University of Alberta). Panelists were screened using a method similar to that used by Cross et al. (1978). Panelists completed a series of 16 rice bread triangle

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tests over a nine day period. During each triangle test, panelists picked the odd sample with respect to firmness and/or moistness and indicated the degree and direction of difference. Samples used in the tests were thawed rice bread rounds (2.5 cm in diameter, 1.25 cm high) taken from breads prepared during ingredient screening experiments (Chapter IV). Samples, made from each of the three rice flours, covered a wide range of firmness and moistness. Triangle test difficulty increased over the screening period. Panelists were briefly familiarized with basic tasting procedures (rinsing the mouth with water prior to tasting and between samples, placement of samples in the mouth and order of tasting samples) and given definitions for the firmness and moistness of breads:

FIRMNESS: The perceived force required to gently compress the sample between the molar teeth (first bite).

MOISTNESS: The perceived degree of moisture in the sample (during mastication).

During screening, panelists correctly identified the odd samples 83% of the time, with a range of 35-100% over the 16 triangle tests. Sixteen panelists were selected for training on the basis of their ability to correctly identify the odd sample at least 70% of the time, interest in the study and availability for the duration of the study.

Background information on potential panelists was obtained through a questionnaire. The questionnaire provided demographic information on the panel and suggested the suitability of potential panelists.

Training of panelists. Training sessions were held four to five times per week for 11 weeks. During each week, three to four days were devoted to individual evaluation of the sensory properties in three to

six bread samples per day. Samples represented a number of variations of rice and wheat flour yeast breads and were similar to those in the actual study. Replicate bread samples were also included. Before each session, a brief description of the previous day's results and procedures to be followed were provided by the panel leader. Round table panel discussions were held at least one day per week. Previous results were discussed and panelists freely asked questions, commented on problems and made suggestions. Panelists described how they evaluated the samples and defined the descriptive terms they used. The vocabulary for the sensory characteristics of the rice breads, the ballot and the procedure for assessing rice bread quality were developed, agreed upon and refined. Ten organoleptic attributes (yeasty odor, rice odor, firmness, moistness, cohesiveness, yeasty flavor, rice flavor, adhesiveness, graininess and aftertaste) and five visual attributes (top crust color, crumb color, predominant cell size, cell size uniformity and cell wall thickness) were selected for the assessment of rice breads. Discussion of results and comments by panelists during the sessions developed panel consistency and increased panelists' understanding of the methods and rice bread attributes being studied.

During the first week, panelists were introduced to basic panel procedures and to sensory methods. Panelists individually rated the firmness and moistness of rice breads, using an eight-point descriptive category scale. A value of 8 indicated extremely soft and extremely moist, and a value of 1 represented extremely firm and extremely dry. Preliminary instruction sheets with definitions were provided. Panelists also described the flavor, texture and after-effect of the

samples, using descriptive adjectives.

The REF wheat bread sample was introduced with the rice breads in the second week. Yeasty flavor and rice flavor were added to the ballot and assessed. A value of 8 represented extremely intense yeasty and rice flavors while a value of 1 illustrated extremely weak yeasty and rice flavors. Dry yeast, rice flour and wheat flour samples were provided to familiarize the panelists with their flavors and odors.

During the third week of training, the unstructured linescale technique was introduced, as some panelists were not comfortable with the category scale. Each panelist was instructed to place a vertical line across a 15 cm horizontal line at the point which best described her impression of each of the rice bread characteristics. The line scale was anchored 1.3 cm from each end with appropriate descriptors. A value from 0.0 to 15.0 was assigned to each rating by converting the mark on the line to a one decimal numerical score (0-15). Rice flour slurries (5 parts rice flour to 7 parts water) and cooked rice pastes (1 part rice to 1 part water, cooked to differing degrees of gelatinization and blended with an additional equal volume of water) were provided to further identify rice flavor and odor.

In the fourth week, the five visual bread characteristics were introduced on a second ballot. In addition, rice odor, yeasty odor and aftertaste were added to the first ballot. Graininess, cohesiveness and adhesiveness were introduced in week five to complete the ballots. For tasting prior to the panel assessments, rice pastes were provided daily to reinforce rice flavor and odor. Rice flour slurries with added dry yeast (0, 5 or 15 g) were cooked (3 min at a medium setting) in a microwave oven (Kenmore, Model No. 99231) to provide panelists

with cooked ricey, yeasty odor and flavor references similar to those in the rice breads. Water, if needed, was added to the samples after cooking to bring all samples to the same consistency.

Additional bread samples were evaluated in the following weeks. The attributes of the REF wheat flour breads were temporarily positioned on the line scales. For training, REF scores for each bread attribute were obtained from the data of previous panels. REF scores were mean values calculated over all the panelists. Rice paste samples were discontinued once panelists were comfortable with scoring rice and yeasty odors and flavors. Apple slices dipped in lemon water (400 ml water to 15 ml lemon juice) were provided to the panelists to help clear the mouth and teeth between samples. The final ballots for the organoleptic and visual evaluations of breads are shown in Fig. VI.1 and VI.2 respectively. Instruction sheets for the organoleptic and visual evaluations are given in Appendices 15 and 16. Final ballot REF scores were obtained from the scores given to coded wheat bread samples included in the second panel performance evaluation. To ensure that the REF scores reflected the judgements of the panelists, means were taken for data over the four REF samples and the eight final panelists.

Performance evaluations. After the sixth and tenth week of training, panelist performance evaluations were conducted using the procedure described by Cross et al. (1978). For the first evaluation, four replications of six treatments were used to determine panelists' ability to discriminate among samples and to measure consistency in replicate judgments. Panelists scored the ten organoleptic characteristics of the samples presented. Judges were ranked for each

Judge: _____

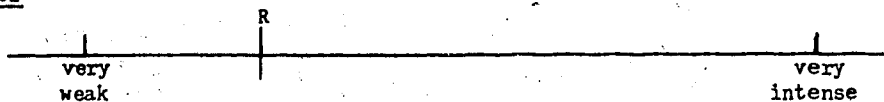
Date: _____

RICE BREAD EVALUATION

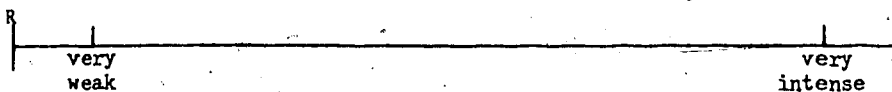
Sample #: _____

Instructions: For each characteristic listed, place a vertical line across the horizontal line at the point that best describes that characteristic in the sample.

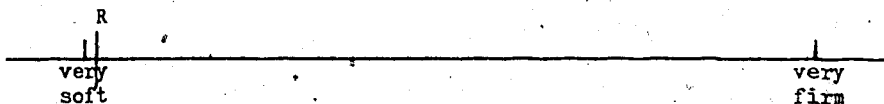
Yeasty Odor



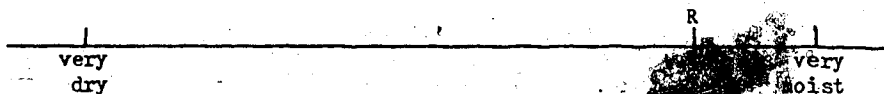
Rice Odor



Firmness



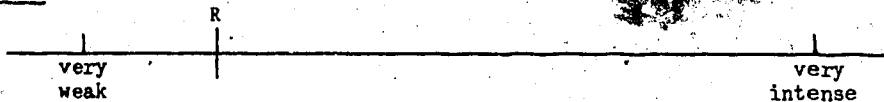
Moistness



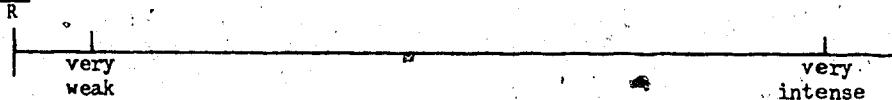
Cohesiveness



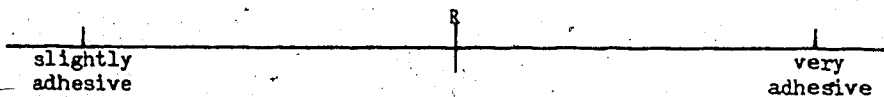
Yeasty Flavor



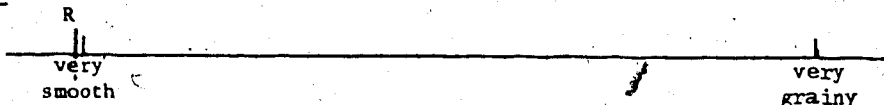
Rice Flavor



Adhesiveness



Graininess



Aftertaste

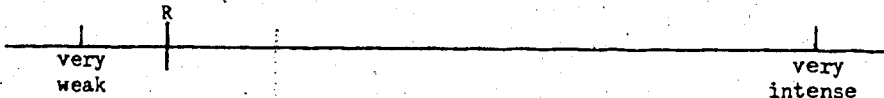


Figure VI.1 Organoleptic evaluation ballot

Judge: _____

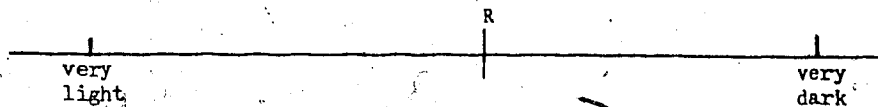
RICE BREAD EVALUATION

Sample # _____

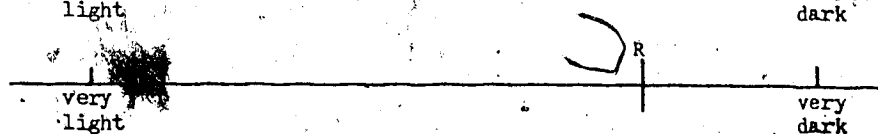
Date: _____

Instructions: For each characteristic listed, place a vertical line across the horizontal line at the point that best describes that characteristic in the sample.

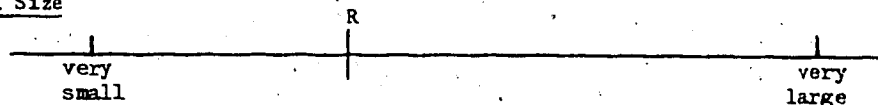
Top Crust Color



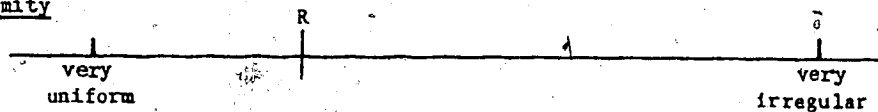
Crumb Color



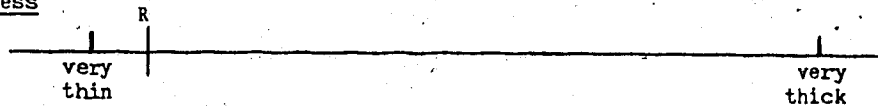
Predominant Cell Size



Cell Size Uniformity



Cell Wall Thickness



COMMENTS:

Figure VI.2. Visual evaluation ballot

characteristic on the basis of their F-value from a one-way analysis of variance. A panelist with a high F-value was more consistent and more discriminating than a panelist with a lower F-value. The evaluation data revealed the bread characteristics panelists were having problems with as well as attributes that required additional work. All panelists continued with further training. The second evaluation was used to confirm that further training was not required and to select panelists for the study. Four replications of six treatments (two were duplicates) were included and eight panelists were chosen.

Sample preparation and presentation. In accordance with the experimental design (Table V.3) one hundred and twenty breads (20 design points x 2 reps x 3 flours) were evaluated by each of the trained panelists. Fifteen sensory responses (10 organoleptic and 5 visual) were measured on all rice breads. All evaluations were made in comparison to a REF wheat bread.

Samples for sensory evaluation were taken from the same relative position in each thawed loaf of bread. Measurements were made on samples equilibrated to room temperature (22°C). For organoleptic evaluation, two or three rounds (2.5 cm in diameter) were removed from the lower half of each of 8 slices (1.25 cm thick) of bread. For each rice bread, two rounds from the same slice of bread, were presented to each panelist. Each panelist also received 3 rounds of the REF wheat bread. The position of samples presented to panelists was rotated among panelists so that each panelist tasted samples from each of the eight slices of bread. The two rounds of bread per treatment (3 rounds of REF bread) were placed in coded plastic petri dishes with lids and all the petri dishes for one set of samples were put in plastic bags

with twist-ties, in the randomized order selected for each panelist. As they began their evaluations, panelists removed the dishes from the bags. Samples were evaluated in the same 3-block central composite design used for baking, with the flours and blocks within flours randomized.

Organoleptic evaluations were conducted in individual booths in an atmospherically controlled sensory panel room. Samples were evaluated under red lights to mask any sample color differences. Water and apple slices were provided to clean the mouth before sampling and between the evaluation of different treatments. Toothpicks were also available to remove any bread particles remaining in the mouth after tasting. Panelists evaluated 6-8 rice bread samples plus a REF wheat bread sample at each session.

Visual evaluations were conducted in a Macbeth skylight booth (Model No. BBX-826, two 1,000 watt bulbs) which provided a constant, consistent source of daylight. The entire center slice (1.25 cm thick), from each loaf of bread, was used for visual evaluation. The top crust was removed for top crust color evaluations. The remaining slice and detached top crust section were placed in a plastic petri dish, which had the bottom lined with a round of black felt, and covered. The petri dishes were kept in plastic bags with twist-ties until evaluation. For evaluation, the samples (6-8 plus a REF) were placed in the Macbeth booth. The center position of the booth was marked with a black round having dimensions similar to the petri dish. Panelists scored the samples randomly by bringing the sample's petri dish to the black circle and removing the lid. Instructions were provided to the judges, along with a visual reference card which was

hung on the back wall of the booth. The visual reference card consisted of pictures and air-dried bread samples to illustrate the line scale anchor points for top crust color, crumb color, predominant cell size, cell size uniformity and cell wall thickness (Plate VI.1). Judges made their visual evaluations individually, immediately after the organoleptic evaluation.

Sensory evaluation - consumer panel

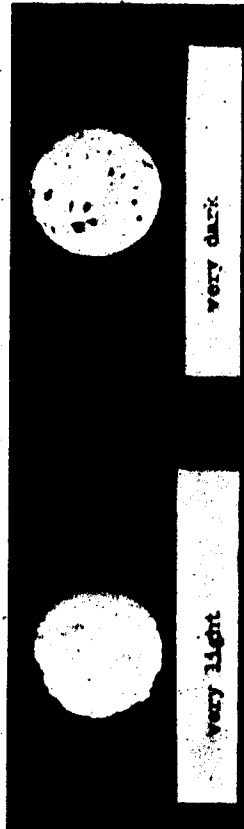
Three rice breads, one from each of the Flours A, B and C, were evaluated during the consumer panel. Each bread was prepared from the center point formulation (0 coded levels of HPMC, CMC and water) of the experimental design for each flour. Since all of the rice breads of the study could not be evaluated by the consumer panel, the center point breads were chosen as being representative of typical breads from each flour. The center point breads from Flours A, B and C (Plate VI.2) had a good appearance and flavor and were reproducible.

A total of four loaves of bread from each flour was required for sampling. The breads were prepared the day prior to the panel, frozen (-29°C) for 24-28 hr and thawed (21°C) 1.5-2 hr before sampling. The breads were sliced (1.25 cm); the crusts and end slices closest to the crust were discarded. Individual slices were immediately placed in coded plastic bags and closed with twist-ties.

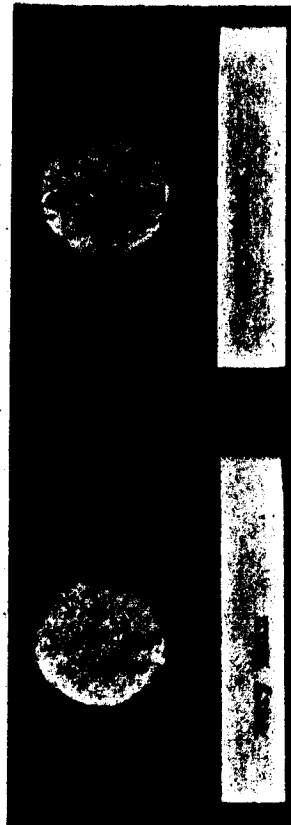
The consumer panel consisted of 42 individuals (23 celiacs and 19 non-celiacs) who had attended the Edmonton Celiac Association Annual Meeting, October 25, 1986. Members had been encouraged to attend through a letter which appeared in the September/October issue of the 1986 Edmonton Celiac Association Newsletter. Each participant



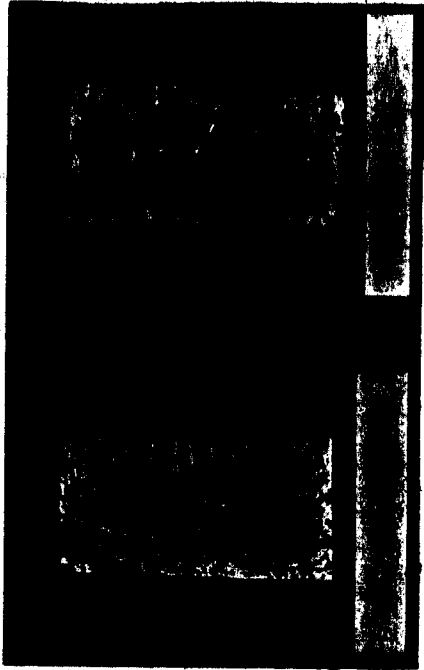
TOP CRUST COLOR



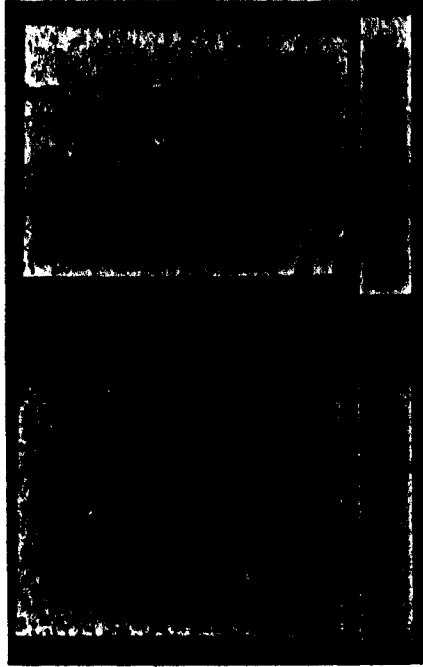
CRUMB COLOR



CELL WALL THICKNESS



PREDOMINANT CELL SIZE



CELL SIZE UNIFORMITY

Plate VI.1 - Visual reference card line scale anchor points

evaluated 3 bread samples (1 slice from each type of rice flour bread). The samples were presented in random order on a tray with a ballot (Fig. VI.3), questionnaire (Appendix 17), pencil, napkin and a glass of water. Using a six-point category scale, participants evaluated the bread samples for aroma (6 = very desirable to 1 = very undesirable), appearance (6 = very desirable to 1 = very undesirable), firmness (6 = very soft to 1 = very firm), moistness (6 = very moist to 1 = very dry), flavor (6 = very desirable to 1 = very undesirable) and overall acceptability (6 = very acceptable to 1 = very unacceptable). The panelists were instructed to evaluate the samples individually, in the order presented, and to take a drink of water between samples. An enlarged ballot was utilized to explain the scoring system. Participants were asked to evaluate the bread crumb for firmness (measured by touch, not by mouth), moistness and flavor.

Statistical analysis

A central composite design, consisting of a three-variable (carboxymethylcellulose gum [CMC], hydroxypropylmethylcellulose gum [HPMC] and water), five-level pattern with 20 rice bread formulations was used. The design was replicated twice for each flour and the five levels of each of the three variables were coded as -1.633, -1, 0, +1, +1.633.

For each sensory characteristic (response) measured by the trained panel, ANOVA was conducted to determine significant differences among the 15 treatment combinations. Also, data were analyzed using multiple regression procedures. To estimate CMC, HPMC and water effects on each sensory response, the design was fitted to a second-order regression

RICE BREAD EVALUATION

DATE: _____

Instructions: For each characteristic listed, rate each of the three bread samples by placing a number (your score) from 1 to 6 in the box under the appropriate sample number.

CHARACTERISTIC	SAMPLES					
	6	5	4	3	2	1
Aroma	very desirable	desirable	slightly desirable	slightly undesirable	undesirable	very undesirable
Appearance	very desirable	desirable	slightly desirable	slightly undesirable	undesirable	very undesirable
Firmness	very soft	soft	slightly soft	slightly firm	firm	very firm
Moistness	very moist	moist	slightly moist	slightly dry	dry	very dry
Flavor	very desirable	desirable	slightly desirable	slightly undesirable	undesirable	very undesirable
Overall Acceptability	very acceptable	acceptable	slightly acceptable	slightly unacceptable	unacceptable	very unacceptable

Compared to the bread I usually eat:

Sample _____ is more acceptable _____; equal in acceptability _____; or less acceptable _____.

Sample _____ is more acceptable _____; equal in acceptability _____; or less acceptable _____.

Sample _____ is more acceptable _____; equal in acceptability _____; or less acceptable _____.

● Figure VI.3 Consumer evaluation ballot

equation (Chapter V). Coefficients of determination (R^2) were computed. The adequacy of the models was tested by separating the residual sum of squares into pure error and lack of fit.

For each response, contour plots were produced from the equations, by holding the variable with the least effect on the response equal to a constant value, and changing the other two variables. Each contour plot was compared to REF data. Areas on each plot, that represented bread formulations which met REF standards, were identified. For each flour, contour plots for all responses were superimposed to locate rice bread formulations which met REF standards for all sensory responses.

The results of the consumer panel were tabulated and means calculated over all participants, celiac participants and non-celiac participants. For each attribute evaluated by the consumer panel (all participants) ANOVA was conducted to determine significant differences among the three rice breads. Duncan's Multiple Range test was used to establish significant differences.

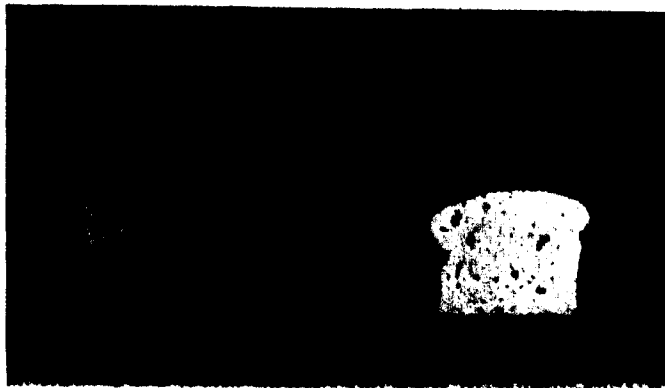
RESULTS AND DISCUSSION

Sensory evaluation - trained panel

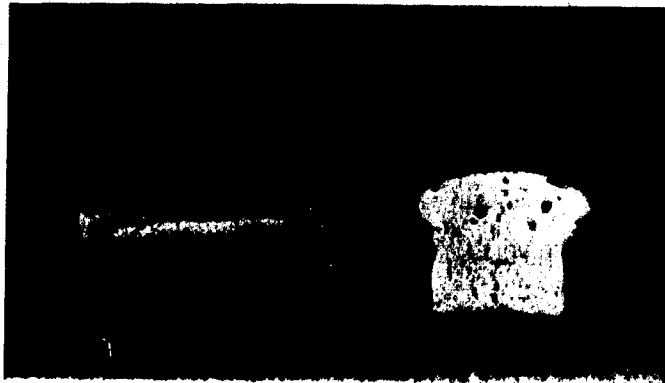
For each flour, ANOVA of the fifteen rice bread treatments showed significant differences for the firmness, moistness, crumb color and cell wall thickness of the breads. Significant differences among the 15 treatments were also determined for top crust color and predominant cell size of Flour A breads; the graininess, predominant cell size and cell size uniformity of Flour B breads; and for cohesiveness, adhesiveness, graininess and top crust color of Flour C breads. The



Flour A



Flour B



Flour C

Plate VI.2 - Rice breads (center point formulations) evaluated by the consumer panelists

lack of significant differences among treatments, for the sensory responses (Table VI.1), indicated that HMPC, CMC and water level had no effect on the response and that further statistical analysis was not required.

Mean values for sensory responses with no significant treatment differences (Table VI.1), calculated over the 40 design points (20 design points x 2 replications) per flour, were compared to the REF standards (Table VI.2). REF standards were met by rice breads for cohesiveness (Flours A and B), yeasty flavor (Flours A, B and C), adhesiveness (Flours A and B), aftertaste (Flour B), top crust color (Flour B) and cell size uniformity (Flours A and C). However, Flour A, B and C bread response means for yeasty odor, rice odor and rice flavor were larger than the REF standards. Thus, breads from each of the rice flours had a stronger yeasty odor, rice odor and rice flavor than the REF. Rice bread response means for graininess (Flour A) and aftertaste (Flours A and C) were also higher than REF standards. Flour C rice bread response means for predominant cell size were lower than REF standards.

Twenty-one regression equations were computed for the responses which had shown significant differences in the ANOVA (Flour A, 6 responses; Flour B, 7 responses; Flour C, 8 responses). The regression coefficients and R^2 values for the sensory responses of breads from Flours A, B, and C are shown in Tables VI.3, VI.4 and VI.5, respectively. Each regression equation was tested for lack of fit. Flour A and C bread response equations did not show lack of fit. Responses for moistness and predominant cell size (Flour B) showed significant lack of fit. The lack of fit of the regression equation

Table VI.1 - Mean values of sensory attribute responses with no significant treatment differences^a

FLOUR	RESPONSE	MEAN ^b	REFERENCE STANDARDS ^c MET
A	Yeasty odor	7.6	No
	Rice Odor	7.4	No
	Cohesiveness	12.6	Yes
	Yeasty Flavor	7.0	Yes
	Rice Flavor	6.6	No
	Adhesiveness	9.8	Yes
	Graininess	3.7	No
	Aftertaste	5.9	No
	Cell Size Uniformity	6.9	Yes
B	Yeasty Odor	7.4	No
	Rice Odor	7.5	No
	Cohesiveness	11.2	Yes
	Yeasty Flavor	6.5	Yes
	Rice Flavor	6.7	No
	Adhesiveness	9.0	Yes
	Aftertaste	5.6	Yes
	Top Crust Color	7.0	Yes
C	Yeasty Odor	7.5	No
	Rice Odor	7.8	No
	Yeasty Flavor	7.1	Yes
	Rice Flavor	8.4	No
	Aftertaste	6.8	No
	Predominant Cell Size	2.9	No
	Cell Size Uniformity	6.9	Yes

^a Determined by analysis of variance

^b Means calculated over the 15 treatments, 8 panelists and 2 replications of the design

^c Reference standards are given in Table VI.2

Table VI.2 - Reference wheat bread sensory response measurement standards

RESPONSE	MEAN ^a	RANGE ^b
Yeasty odor	4.3	1.6-6.6
Rice odor	0.0	0.0
Firmness	1.5	0.7-2.2
Moistness	11.6	9.5-13.1
Cohesiveness	12.4	11.0-13.4
Yeasty Flavor	3.6	1.6-7.4
Rice Flavor	0.0	0.0
Adhesiveness	7.6	3.8-10.7
Graininess	1.2	0.2-1.7
Aftertaste	2.8	0.6-5.8
Top crust color	8.0	6.3-10.8
Crumb color	10.7	7.7-13.4
Predominant Cell Size	5.7	3.1-8.4
Cell size uniformity	4.9	2.9-7.4
Cell wall thickness	2.3	1.8-2.9

^a Mean value is the reference score positioned on the ballots.
N = 4 treatments x 8 panelists = 32.

^b The range of values obtained for 4 replicate treatments.

Table VI.3 - Regression equation^a coefficients and R² values for Flour A rice bread sensory responses

COEFFICIENT	RESPONSE						
	FIRMNESS	MOISTNESS	TOP CRUST COLOR	CRUMB COLOR	PREDOMINANT CELL SIZE	CELL WALL THICKNESS	
b ₀	4.018	12.421	7.908	4.317	3.459	4.132	
b ₁ (CMC)	1.312***	-0.286***	-0.359*	0.947***	0.305*	1.582***	
b ₂ (HPMC)	0.234	0.016	0.129	0.070	0.296*	0.393*	
b ₃ (WATER)	-2.351***	1.018***	0.159	-1.474***	-0.016	-2.134***	
b ₁₁	0.366	0.035	-0.309	0.365**	0.018	0.719***	
b ₂₂	-0.282	-0.108	-0.168	-0.010	0.033	0.046	
b ₃₃	1.547***	-0.198***	-1.212***	0.544***	-0.557***	1.599***	
b ₁₂	-0.209	-0.070	-0.175	0.226	0.030	-0.296	
b ₁₃	-1.573***	0.142	1.448***	-0.830***	0.698***	-1.882***	
b ₂₃	0.135	0.059	-0.044	0.010	-0.070	0.137	
R ²	.88	.71	.61	.88	.66	.92	

^a $Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$
 where x₁=CMC, x₂=HPMC, x₃=water.

^b Replication and block effects removed.

*, **, *** Significant at P<0.05, P<0.01 and P<0.001, respectively.

Table VI.4 - Regression equation coefficients and R² values for Flour B rice bread sensory responses

COEFFICIENT	RESPONSE						
	FIRMNESS	MOISTNESS	GRAININESS	CRUMB COLOR	PREDOMINANT CELL SIZE	CELL WALL UNIFORMITY	CELL WALL THICKNESS
b ₀	5.099	10.386	5.844	4.069	3.046	5.128	4.396
b ₁ (CMC)	1.547***	-0.287**	0.210	1.288***	0.477***	-0.742***	1.990***
b ₂ (HPMC)	0.464**	-0.100	0.234*	0.018	-0.045	0.247	0.226
b ₃ (WATER)	-2.484***	0.793***	-0.502***	-1.090***	0.055	0.646***	-1.679***
b ₁₁	0.760***	-0.034	0.137	0.567***	-0.082	0.102	1.029***
b ₂₂	-0.041	0.011	-0.004	-0.002	-0.026	0.100	0.070
b ₃₃	1.163***	-0.193*	0.243*	0.590***	0.074	-0.091	1.250***
b ₁₂	-0.334	0.234*	0.081	-0.024	0.001	0.223	0.045
b ₁₃	-1.581***	-0.127	-0.225	-0.865***	0.520***	0.201	-1.571***
b ₁₂	-0.058	0.116	-0.094	0.037	0.229	0.174 ^e	0.282
R ²	.94	.36	.19	.94	.55	.56	.95

$$a \quad Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$$

where x₁=CMC, x₂=HPMC, x₃=water.

b Replication and block effects removed.

*, **, *** Significant at P<0.05, P<0.01 and P<0.001, respectively.

Table VI.5 - Regression equation coefficients and R² values for Flour C rice bread sensory responses

COEFFICIENT	RESPONSE							
	FIRMNESS	MOISTNESS	COHESIVENESS	ADHESIVENESS	GRAININESS	TOP CRUST COLOR	CRUMB COLOR	CELL WALL THICKNESS
b ₀	4.704	5.249	4.923	7.592	9.338	8.059	4.221	4.276
b ₁ (CMC)	2.104***	-0.108	0.318*	0.197	0.322**	-0.474***	1.327***	1.885***
b ₂ (HPMC)	0.398**	0.736***	1.062***	0.576***	-0.395***	0.406**	-0.085	0.136
b ₃ (WATER)	-2.553***	0.364*	-0.520**	-0.413**	-0.511***	0.736***	-1.165***	-1.911***
b ₁₁	0.691***	-0.141	0.232	0.060	0.038	-0.509***	0.702***	1.024***
b ₂₂	-0.142	-0.043	-0.084	0.060	0.004	-0.285*	-0.038	-0.082
b ₃₃	-1.192***	-0.091	0.321*	0.175	0.042	0.753***	0.584***	0.987***
b ₁₂	-0.488**	0.034	-0.116	0.011	-0.106	-0.398*	-0.038	-0.026
b ₁₃	-1.512***	-0.004	-0.550**	-0.264	-0.016	-1.253***	-0.987***	-1.524***
b ₂₃	0.046	0.376	0.277	0.062	-0.066	-0.016	-0.182	0.084
R ²	.96	.54	.74	.38	.54	.67	.94	.98

a $Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$
 where $x_1 = \text{CMC}$, $x_2 = \text{HPMC}$, $x_3 = \text{water}$.

b Replication and block effects removed.

*, **, *** Significant at P<0.05, P<0.01 and P<0.001, respectively.

for the moistness of Flour B breads may indicate inadequacy of the equation. The reason for the lack of fit is not known, but perhaps, as mentioned in Chapter V, the center point rice breads used to calculate lack of fit, produced breads that had a more uniform cell size and moistness throughout the slice of bread and were easier to evaluate than the other treatment breads. In that case, the pure error obtained by evaluating the center point breads may not be representative of the overall error in the other treatment combinations.

Flour A rice bread regression equations (Table VI.3) explained 61% to 92% of the response variation. For Flour A breads, CMC and water affected the sensory responses the most; HPMC had the least effect.

The regression equations for Flour B rice breads (Table VI.4) explained 19% to 95% of the variation in the sensory responses. The low R^2 values of 0.19 and 0.36 for graininess and moistness, respectively, suggest that factors other than the levels of CMC, HPMC and water affected the graininess and moistness of the Flour B breads. Both responses had significant replication effects which, when added to the models, changed the R^2 values to 0.87 (graininess) and 0.92 (moistness), and thus explained a large proportion of the variation in graininess and moistness of these breads. For all Flour B bread responses, water and CMC had the most effect while HPMC had the least effect.

The eight regression equations (Table VI.5) of Flour C breads explained 38% to 98% of the response variation. As for Flour B, the low R^2 value (0.38) for adhesiveness of Flour C breads could be increased to 0.76 by incorporating the effect of replication into the model. Thus, replication differences accounted for a large portion of

the variation in the adhesiveness of Flour C rice breads. Unlike breads from Flours A and B, the level of HPMC significantly affected the moistness, cohesiveness, adhesiveness and top crust color of Flour C rice breads. All Flour C bread responses were affected by water level while many responses (firmness, cohesiveness, graininess, top crust color, crumb color and cell wall thickness) were also affected by CMC level.

To study the response surfaces of breads within each rice flour, contour plots were produced for the 21 responses by holding HPMC levels at -1.633, 0 and 1.633. Contour plots for all sensory responses appear in Appendices 21 to 23 and should be referred to in the discussion of the results pertaining to each of the three flours. Each contour plot was compared to REF sensory response data (Table VI.2). Areas on the contour plots that represented rice bread formulations which met the range of REF response values for that specific response were identified. In the case of crumb color, rice bread responses within the REF range or lower (lighter) were considered to have met the REF standard.

Flour A

For Flour A breads, CMC, HPMC and water levels had no effect on the sensory measurements of yeasty odor, rice odor, cohesiveness, yeasty flavor, rice flavor, adhesiveness, graininess, aftertaste and cell size uniformity. Thus, regression equations and contour plots were not produced for these responses.

None of the Flour A breads met REF standards (Table VI.2) for firmness; all breads were more firm than the reference. The softest Flour A breads were found at the 1.633 HPMC level with water levels

between -0.5 and 1, and CMC levels less than 0.5. These results differ from those found for the objective measurement of bread firmness (Chapter V). Flour A breads met Instron firmness REF standards at all HPMC levels. This finding suggests that the trained panel evaluated a different aspect of bread firmness than that measured by the Instron. When measuring the hardness of rye and french wheat breads, Brady and Mayer (1985) also found that the trained panel results differed from the Instron data. They (Brady and Mayer, 1985) suggested that the panel and the Instron may have been measuring different bread characteristics or that the Instron may have been more sensitive to slight differences in sample hardness than the panelists.

HPMC had very little effect on the moistness of Flour A breads. At all HPMC levels, rice breads with water levels below 0 and any CMC level met moistness REF standards. Water levels between 0.0 and 1.2 could produce Flour A breads which met REF standards for moistness, if appropriate CMC levels were used (higher CMC levels were needed as the water level increased). However, water levels above 1.2 resulted in Flour A breads which were moister than the REF. Similar results were obtained for the HPMC effect on the measurement of percent moisture in Flour A rice breads (Chapter V). No HPMC effect on percent moisture was found in Flour A rice breads. However, water levels were less important and CMC levels were more important to percent moisture, than they were to the panelists' evaluation of moisture in Flour A breads.

Top crust color of Flour A breads was affected by HPMC level. More Flour A breads met REF standards for top crust color at the -1.633 HPMC level than at the 1.633 HPMC level. At all HPMC levels, high water-low CMC level combinations and low water-high CMC level

combinations produced Flour A breads with top crust colors which were lighter than those of the REF. The sensory data for top crust color (very light to very dark), were very similar to the crust color 'L' values determined with the Hunterlab Color/Difference Meter (Chapter V). For the Flour A breads, a strong HPMC effect was noted for the objective response of crust color 'L' values and crust color 'L' value REF standards were met most often at the -1.633 HPMC level (Chapter V).

The effects of HPMC level on the crumb color of Flour A breads were small. All Flour A breads were as light (white) or lighter than the REF and met REF standards for crumb color. The crumb color of Flour A breads was darkest at low water - high CMC combinations. These sensory findings agree with the instrumental crumb color 'L' value results (Chapter V). All Flour A rice breads met objective REF standards for crumb color 'L' values and had a whiter crumb than the REF (Chapter V).

Flour A bread predominant cell size contour plots representing the three HPMC levels appear in Fig. VI.4. The contour plots show that the region of rice bread formulations meeting the REF standards for predominant cell size increased as the HPMC level increased. To meet cell size REF standards, Flour A breads at the -1.633 HPMC level needed water levels greater than -0.3 and CMC levels greater than 0.3. At the 1.633 HPMC level, most Flour A breads met REF standards for predominant cell size except at combinations of high water (> 0.3) - low CMC (< 0.3) level and at combinations of low water (< -0.9) - high CMC (> -0.5) levels.

Only Flour A breads with the lowest HPMC level (-1.633) met REF

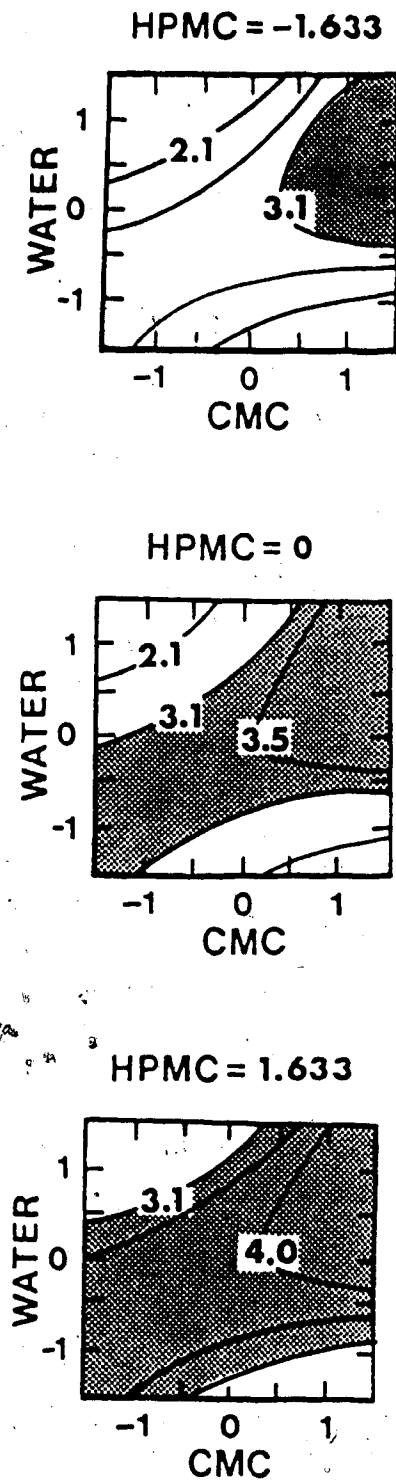


Figure VI.4 Contour plots for Flour A rice bread predominant cell size scores. Levels of HPMC, water and CMC are coded values. The shaded regions met predominant cell size reference standards of 3.1 to 8.4

standards for cell wall thickness. Rice breads from formulations with -1.633 HPMC, water levels between -0.4 and 0.6, and CMC levels less than -0.3 had cell walls with a thickness similar to that of the REF bread. All other Flour A breads had thicker cell walls than the REF wheat bread.

The results obtained when the contour plots for all sensory responses for Flour A were superimposed and suggested that, at an HPMC level of -1.633, Flour A breads with water levels between -0.3 and 0.8, and CMC levels greater than 0.3 would meet REF standards for moistness, cohesiveness, yeasty flavor, adhesiveness, top crust color, crumb color, predominant cell size and cell size uniformity (Fig. VI.5).

Flours B and C

Since HPMC and water levels differed in the bread formulations for each of the three rice flours, direct comparison of the breads resulting from the flours was not feasible. However, examination of the contour plot trends for all responses gave some insight into sensory differences among the rice breads from specific flours.

Discussion of the sensory attributes for Flour B and C will follow the order of the attributes given in Fig. VI.1 and VI.2.

Eight Flour B bread sensory measurements (yeasty odor, rice odor, cohesiveness, yeasty flavor, rice flavor, adhesiveness, aftertaste and top crust color) and seven Flour C bread sensory measurements (yeasty odor, rice odor, yeasty flavor, rice flavor, aftertaste, predominant cell size and cell size uniformity) did not show significant differences following ANOVA. Therefore, regression equations and

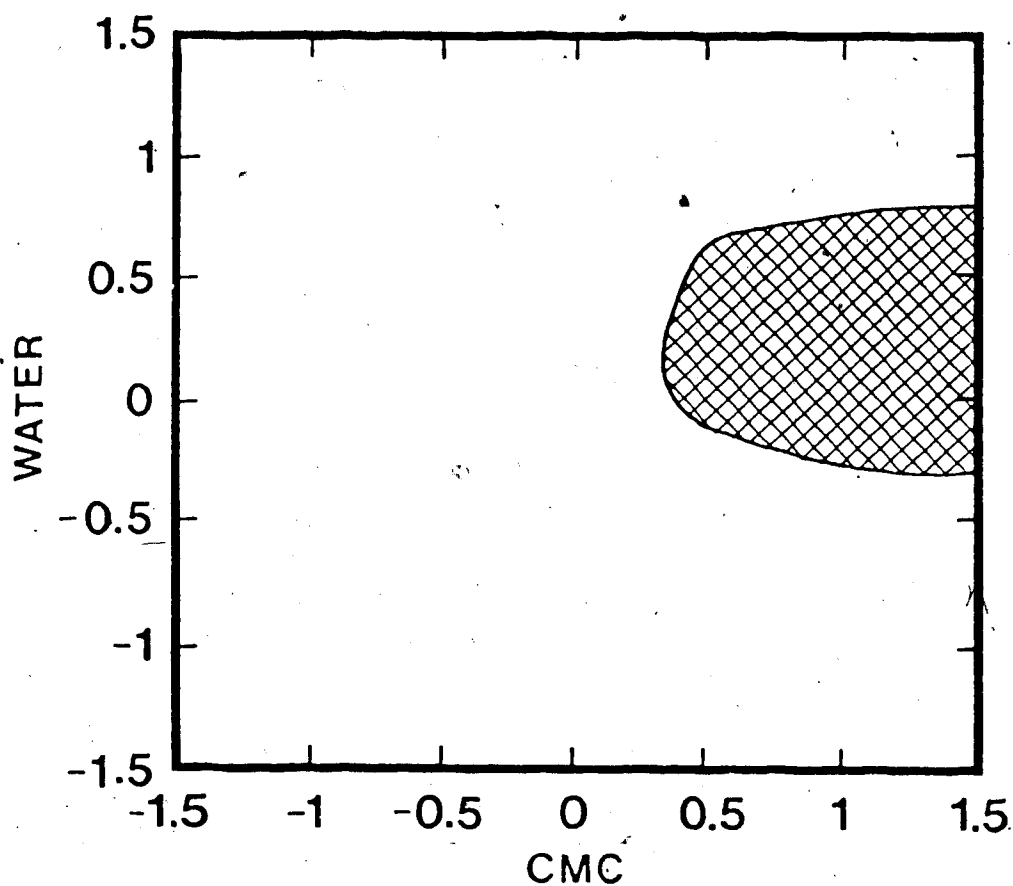


Figure VI.5

Superimposed contour plot region for Flour A rice bread sensory responses at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for moistness, cohesiveness, yeasty flavor, adhesiveness, top crust color, crumb color, predominant cell size and cell size uniformity.

response contour plots were not produced for these responses.

The Flour B bread firmness contour plots were similar to those of Flour A breads. All Flour B breads were firmer than the REF wheat bread. None of the Flour B breads met REF firmness standards. In contrast, at all HPMC levels, Flour C breads had a ridge of water (0.5 to 1.3) - CMC (all levels) combinations that met firmness REF standards. Flour C bread formulations with water levels above 1.3 produced breads with a softer crumb than the REF and breads with water levels below 0.5 produced breads with a firmer crumb than the REF bread. As with Flour A, the panelists' evaluations of the firmness of Flour B and C breads differed from the data obtained for the instrumental firmness of rice bread (Chapter V). Flour B breads met Instron firmness REF standards at all HPMC levels; Flour C rice breads only met the objective firmness REF standards at the 1.633 HPMC level (Chapter V).

The moistness of Flour B breads, like Flour A breads, was affected slightly by HPMC level. Flour B breads met moistness REF standards at all HPMC levels if water levels above 0.0, with any CMC levels were chosen. In contrast, Flour A breads met moistness REF standards at all HPMC levels only if water levels below 0, with any CMC level, were used. Contour plots for percent moisture of Flour B breads (Chapter V) differed from the sensory moistness response plots and showed a much stronger CMC effect and a weaker effect of water level.

None of the Flour C breads met REF moistness standards; all Flour C breads were drier than the REF wheat bread. An HPMC effect was evident as the most moist Flour C breads were those found at high HPMC levels (1.633) with water levels above 0.6 and CMC levels between -1.5

and 0.7. This finding differs from data for percent moisture (Chapter V) in Flour C breads. Flour C breads met the percent moisture REF standards at all HPMC levels. Although some Flour C breads could attain a percent moisture similar to the REF bread, this percent moisture in Flour C breads was not detected by the panelists as sensory moistness.

The cohesiveness of Flour C breads was affected by HPMC level. The least cohesive Flour C breads were found at the -1.633 HPMC level with water levels above -0.3 and CMC levels above -1.1. None of the Flour C breads met cohesiveness REF standards. All Flour C breads were less cohesive than the REF wheat bread. The mean cohesiveness values for Flour B and A breads met REF standards for cohesiveness. Since Flours B and A were from medium grain rice and Flour C was from long grain rice, the findings of the present research are in agreement to those of Nishita (1977). Nishita (1977) found that breads from medium grain white rice flour had a better crumb texture than breads from long grain rice flour, which produced a crumbly (less cohesive) crumb.

Contour plots for Flour C bread adhesiveness indicated an HPMC effect. The most adhesive Flour C rice breads were found for formulations with 1.633 HPMC, water levels below -1.3 and CMC levels above 1.1. However, all Flour C breads met REF standards for adhesiveness.

Flour B and C breads were more grainy in texture than the REF wheat bread and rice bread graininess was affected by HPMC level. For both Flours (B and C) at all HPMC levels, maximum graininess was obtained in breads with combinations of low water + high CMC levels. However, HPMC showed a weaker effect on Flour B bread graininess than

on Flour C breads. Flour C breads were the most grainy at -1.633 HPMC. Flour C breads were also much more grainy (contour lines ranged from 8.25-11.25) than breads from either Flour B (contour lines ranged from 5.5-8) or Flour A (mean graininess = 3.7). Flours A and B were ground from medium grain rice; Flour C was made from long grain rice. Nishita and Bean (1979) also found that long grain rice flour produced breads with a more sandy, grainy texture than medium grain rice flour breads.

Contour plots for the top crust color of Flour C breads showed an HPMC effect with the region of rice bread formulations meeting REF top crust color standards enlarging as the level of HPMC decreased from 1.633 to -1.633. All Flour C breads met REF standards for top crust color except those at high water - low CMC level and low water - high CMC level combinations. These findings are similar to top crust color results for Flour A breads.

Rice breads from both Flours B and C had a lighter crumb color than the REF wheat bread. Flour B and C breads met crumb color REF standards at all HPMC/CMC/water combinations. Crumb color contour plots for breads from both Flours (B and C) were similar, with rice breads with the darkest crumb colors produced from formulations with low water and high CMC level blends.

The predominant cell size of Flour B breads was affected strongly by HPMC level. Flour B breads met predominant cell size REF standards most often at the 1.633 HPMC level, as did Flour A breads, with bread cell size becoming larger as the HPMC level in the formulation increased. At -1.633 HPMC, only Flour B breads with water levels above -0.3 and CMC levels above 0.0 had cells large enough to meet REF

standards for predominant cell size.

The cell size uniformity of Flour B breads was affected slightly by HPMC level. All Flour B breads met cell size uniformity REF standards except at the 1.633 HPMC level. At 1.633 HPMC, water (>-0.7) and CMC (<-0.6) combinations produced breads with a more irregular cell size than the REF wheat bread.

Cell wall thickness contour plots for breads from both Flours (B and C) were similar and showed only slight HPMC level effects. Like Flour A breads, Flour B breads only met REF standards for cell wall thickness at the -1.633 HPMC level (with water levels between -0.7 to 1; CMC levels below 0). All other Flour B breads and all Flour C breads had thicker cell walls than the REF bread.

Response contour plots for rice breads from Flours B and C were also superimposed. For Flour B breads at -1.633 HPMC (Fig. VI.6), specific water (above -0.2) and CMC (above 0) combinations met REF standards for moistness, cohesiveness, yeasty flavor, adhesiveness, aftertaste, top crust color, crumb color, cell size uniformity and predominant cell size. For Flour C breads at -1.633 HPMC (Fig. VI.7), specific water (0.7 to 1.4) and CMC (all levels) combinations met REF standards for firmness, yeasty flavor, adhesiveness, crumb color, top crust color and cell size uniformity.

Sensory evaluation - consumer panel

The consumer panel consisted of 23 celiac members and 19 non-celiac members ranging from under 18 to over 65 years of age. Seventy-four percent of the celiac members and 68% of the non-celiac members were female. The celiacs were all on a gluten-free diet and

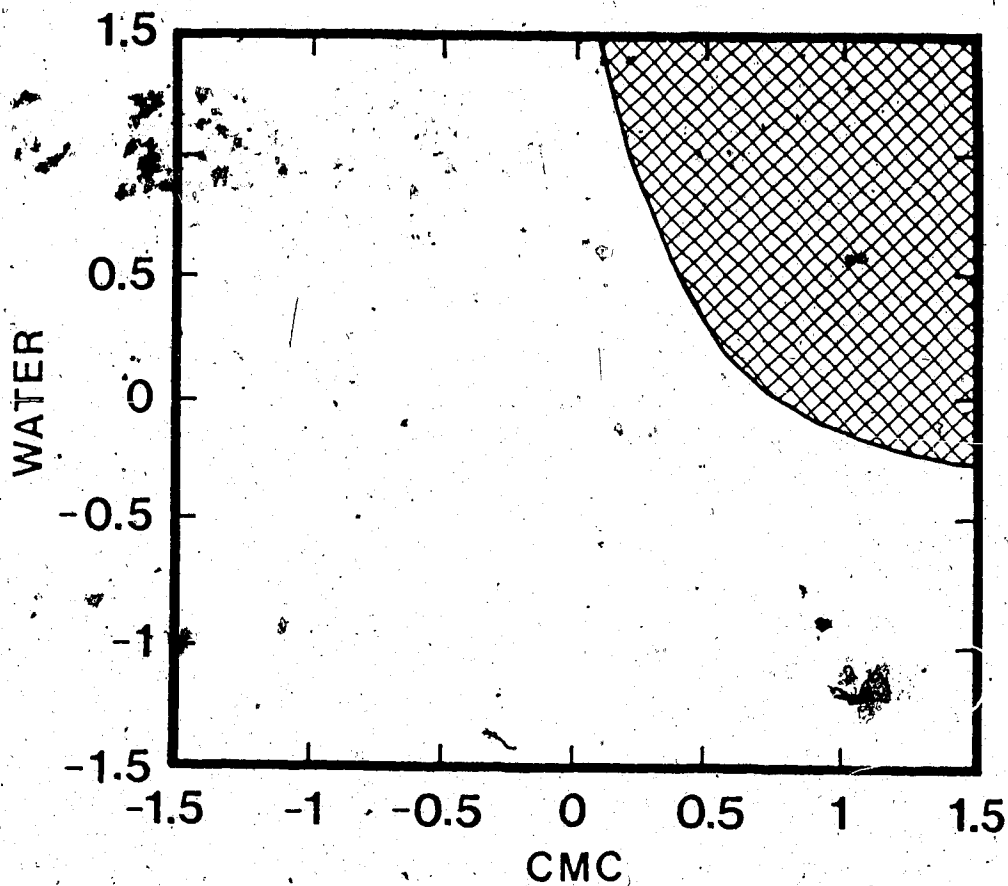


Figure VI.6 Superimposed contour plot region for Flour B rice bread sensory responses at the -1.633 HPMC level. Levels of HPMC, Water and CMC are coded values. The shaded region met reference standards for moistness, cohesiveness, yeast flavor, adhesiveness, aftertaste, top crust color, crumb color, cell size uniformity and predominant cell size.

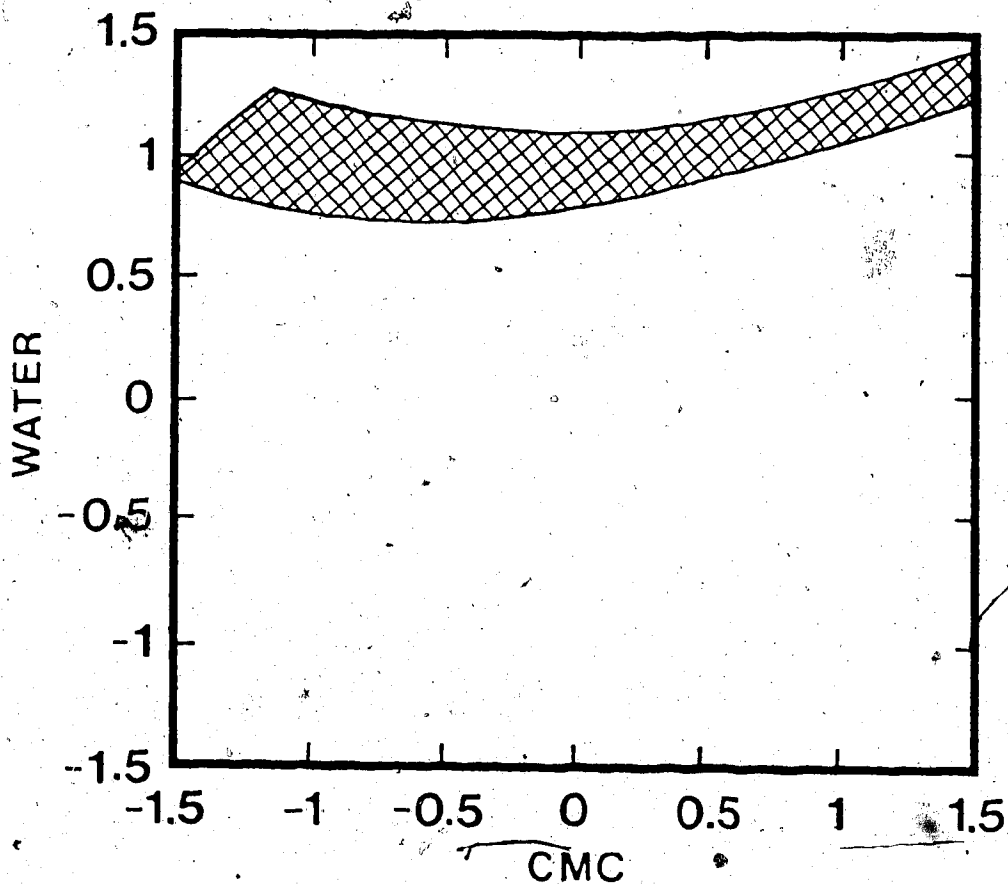


Figure VI.7 Superimposed contour plot region for Flour C rice bread sensory responses at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for firmness, yeasty flavor, adhesiveness, crumb color, top crust color, and cell size uniformity.

65% of them regularly ate from 1-4 slices of gluten-free bread per day. The non-celiac members were not on a special diet and did not regularly consume gluten-free bread.

All three rice bread samples were generally rated as acceptable by the consumer panelists. Overall acceptability mean scores of 4.8, 5.0 and 3.9 out of 6, were given to breads from Flours A, B and C, respectively (a score of 5 = acceptable, 4 = slightly acceptable and 3 = slightly unacceptable). Thus, the breads from Flours A and B were considered more acceptable than Flour C rice breads.

Consumer panel data (Table VI.6) for the aroma, appearance, firmness, moistness, flavor and overall acceptability of rice breads from Flours A and B indicated that these breads were similar and very good (scores of 4.6 and higher). For all characteristics except aroma and appearance, Flour C breads received significantly lower scores than the Flour A and B breads.

A comparison of results from the celiac and non-celiac panel members (Table VI.6) revealed few differences from the total panel data. However, non-celiac members did score the Flour C rice bread slightly lower than the celiacs. Compared to the bread they usually eat (Table VI.7), the non-celiacs found the rice breads to be less acceptable than did the celiac members. Nishita (1977) suggested that people with wheat allergies or celiacs, who have a limited choice of baked products, may not be as critical of rice breads as individuals who can regularly eat wheat bread. Paulus (1986) also reported that individuals requiring special diets are more inclined to accept products of lower sensory quality. In the present study, the celiac members compared the rice breads to other gluten-free breads, while the

Table VI.6 - Mean^a consumer panel scores^b for rice bread characteristics

FLOUR	TOTAL PANEL			CELIAC MEMERS			NON-CELIAC MEMBERS		
	A	B	C	A	B	C	A	B	C
Aroma	4.6x	4.9x	4.3x	4.5	4.9	4.5	4.7	4.9	4.0
Appearance	4.7x	5.2x	5.1x	4.6	5.2	5.3	4.8	5.2	4.9
Firmness	5.0x	4.6x	3.9y	4.7	4.7	4.0	5.4	4.5	3.9
Moistness	5.3x	5.0x	3.5y	5.3	5.0	3.7	5.4	5.0	3.3
Flavor	4.7x	4.9x	3.6y	4.7	5.0	3.8	4.7	4.8	3.4
Overall Acceptability	4.8x	5.0x	3.9y	4.8	5.0	4.1	4.7	5.0	3.7

^a Total panel N=42, celiac members N=23, Non-celiac members N=19

^b Maximum score = 6

xy Means within the same row sharing a common letter are not significantly different at $P < 0.05$.

Table VI.7 - Consumer panel evaluation of rice bread acceptability, in comparison to bread usually eaten

PANEL MEMBERS	FLOUR	MORE ACCEPTABLE	EQUAL IN ACCEPTABILITY	LESS ACCEPTABLE
Celiacs	A	12	4	5
	B	12	7	2
	C	6	8	7
Non-Celiacs	A	2	4	9
	B	4	6	6
	C	2	1	13

non-celiacs compared the rice breads to wheat bread. Overall, both Flour A and Flour B breads were judged to be similar in acceptability by both celiac and non-celiac consumer panelists. The Flour C bread was the least acceptable. Typical panelists' comments included: all rice breads were very good, a great improvement over the currently available gluten-free breads, good flavor, good all-purpose bread.

CONCLUSIONS

An eight-member trained sensory panel evaluated the organoleptic and visual quality of rice breads using an unstructured line scale. Breads were prepared with three rice flours; A and B from medium grain rice and C from long grain rice. At the -1.633 HPMC level, breads from Flour B met REF wheat bread standards for the largest number of sensory responses (9 out of 15), while Flour A breads met 8 response REF standards and Flour C breads met 6 REF standards. A 42-member consumer panel also judged the medium grain rice flour (Flours A and B) breads as more acceptable than the Flour C breads.

The development of formulations for rice breads comparable to a REF wheat (white) bread was dependent on the type of rice flour and on levels of CMC, HPMC and water. Several optimum combinations of CMC, HPMC and water were found that could result in rice breads with a moistness, cohesiveness, yeasty flavor, adhesiveness, aftertaste, top crust color, crumb color, cell size uniformity and predominant cell size comparable to a REF wheat bread.

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VII. General Discussion and Conclusions

DISCUSSION

Chapter I of this thesis reviewed literature pertinent to the development of gluten-free rice flour yeast breads. The review suggested that for yeast bread, rice flour was a more appropriate substitute for wheat flour than wheat starch. However, wheat starch is typically used in gluten-free breads. In addition, previously developed gluten-free bread products were not comparable, in quality, to the wheat flour breads that they replaced. Thus, further research on the development of a rice flour yeast bread was justified. Lack of research utilizing sensory evaluation and objective measurements for the development of gluten-free breads, indicated the need for incorporation of both these methods in future studies.

Past research suggested that there were a number of gums and/or surfactants which were appropriate as gluten substitutes in gluten-free breads. Since response surface methodology (RSM) provides an efficient means to examine a number of variables at different levels and to determine those most appropriate for a particular use, RSM appeared to be ideal for finding a gluten replacement gum combination.

Results of the questionnaires completed by celiacs and hospital dietary personnel (Chapter II) confirmed the necessity for a gluten-free bread comparable to wheat flour bread. The questionnaires permitted an understanding of the needs of individuals following a gluten-free diet. By describing the types of gluten-free yeast breads in current use, the disadvantages and benefits of the products and the

most important quality attributes desired in gluten-free breads, questionnaire respondents contributed to the direction of the present research. Consideration of questionnaire responses also made it possible to address the problems, concerns and interests of the consumers of gluten-free breads.

Standardized formulations for the production of rice flour yeast breads from three locally available rice flours were developed in Chapter III. Methods that improved the appearance, volume, flavor and reproducibility of the standard rice breads were incorporated into the formulations. This stage of the research proved to be difficult and time consuming. In future studies of this type the use of sequential RSM experiments would be beneficial at this stage. The problems encountered in obtaining quality, consistent rice flours and information about the flours from the suppliers were not anticipated. The many modifications to the original rice bread formulation (Nishita, 1977) were not expected. In addition, each rice flour had individual formulation requirements and produced a unique rice bread.

Four gums, two surfactants and water, at various levels and combinations, were tested during the ingredient screening experiments of Chapter IV. Using bread volume and loaf shape as indicators of rice bread quality, hydroxypropylmethylcellulose (HPMC), carboxymethylcellulose (CMC) and water were chosen for more detailed examination in Chapter V. The other gums and both surfactants had either no effect or a detrimental effect (glycerol monostearate) on rice bread volume.

In Chapter V, rice flour yeast breads (80% rice flour/20% potato starch) were developed that were comparable to a wheat flour yeast

bread. The rice flour breads met objective REF standards for specific volume, crumb color, crust color, Instron firmness and percent moisture. Of the three variables which were examined in the design, CMC and water levels had the most effect on the objective measurements of the rice breads, HPMC levels had the least effect. Rice breads made from Flour A (Dainty Foods - medium grain) met more REF standards than either Flour B (Ener-G Foods - medium grain) or Flour C (Woodward's - long grain) breads. Flour A breads were also the only breads to meet loaf shape REF standards.

The sensory characteristics of rice breads were examined by both [redacted] and consumer panels in Chapter VI. Rice flour yeast breads were produced that were comparable to a REF wheat bread in moistness, cohesiveness, yeasty flavor, adhesiveness, aftertaste, top crust color, crumb color, cell size uniformity and predominant cell size. Flour B breads met the largest number of trained panel sensory REF standards (9 compared to 8 and 6 for Flours A and C, respectively). Consumer panelists also judged breads from Flours A and B to be more acceptable than Flour C breads.

By combining the results of Chapters V and VI, rice bread formulations that simultaneously met objective and sensory REF standards were determined. For breads from each flour, contour plots from both studies were superimposed, and the region of rice bread formulations that met the maximum number of both objective and sensory REF standards was found. The superimposed contour plot regions (objective and sensory results combined) for Flour A, B and C breads, at the -1.633 HPMC level, are shown in Fig. VII.1, VII.2 and VII.3, respectively. For breads from each flour, the shaded region differs

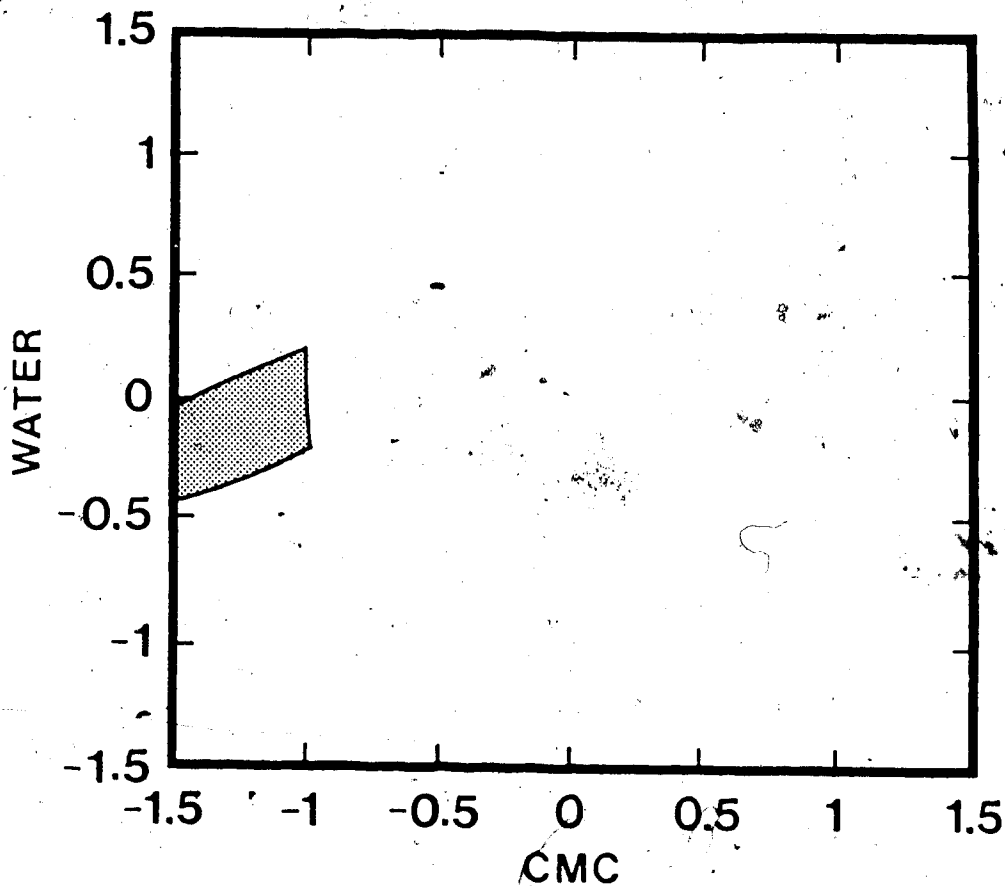


Figure VII.1 Superimposed contour plot region (objective and sensory responses combined) for Flour-A rice breads at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for the objective measurements of pH, crumb color, L^* and b^* values, crumb color and a^* values, Instron firmness, percent moisture and the sensory measurements of moistness, cohesiveness, yeasty flavor, adhesiveness, top crust color, crumb color, cell size uniformity and cell wall thickness.

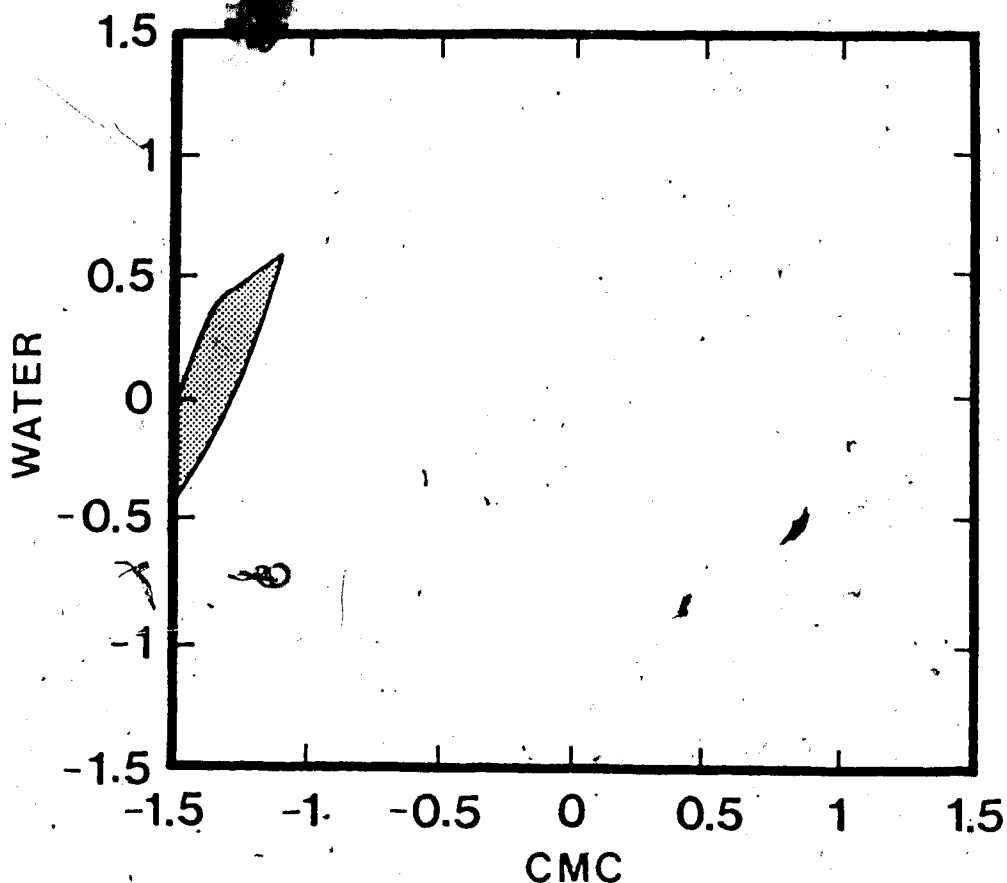


Figure VII.2 Superimposed contour plot region (objective and sensory responses combined) for Flour B rice breads at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for the objective measurements of specific volume, crust color 'a' values, crumb color 'L, a and b' values, Instron firmness, percent moisture and the sensory measurements of moistness, cohesiveness, yeasty flavor, adhesiveness, after-taste, top crust color, crumb color, cell size uniformity and cell wall thickness.

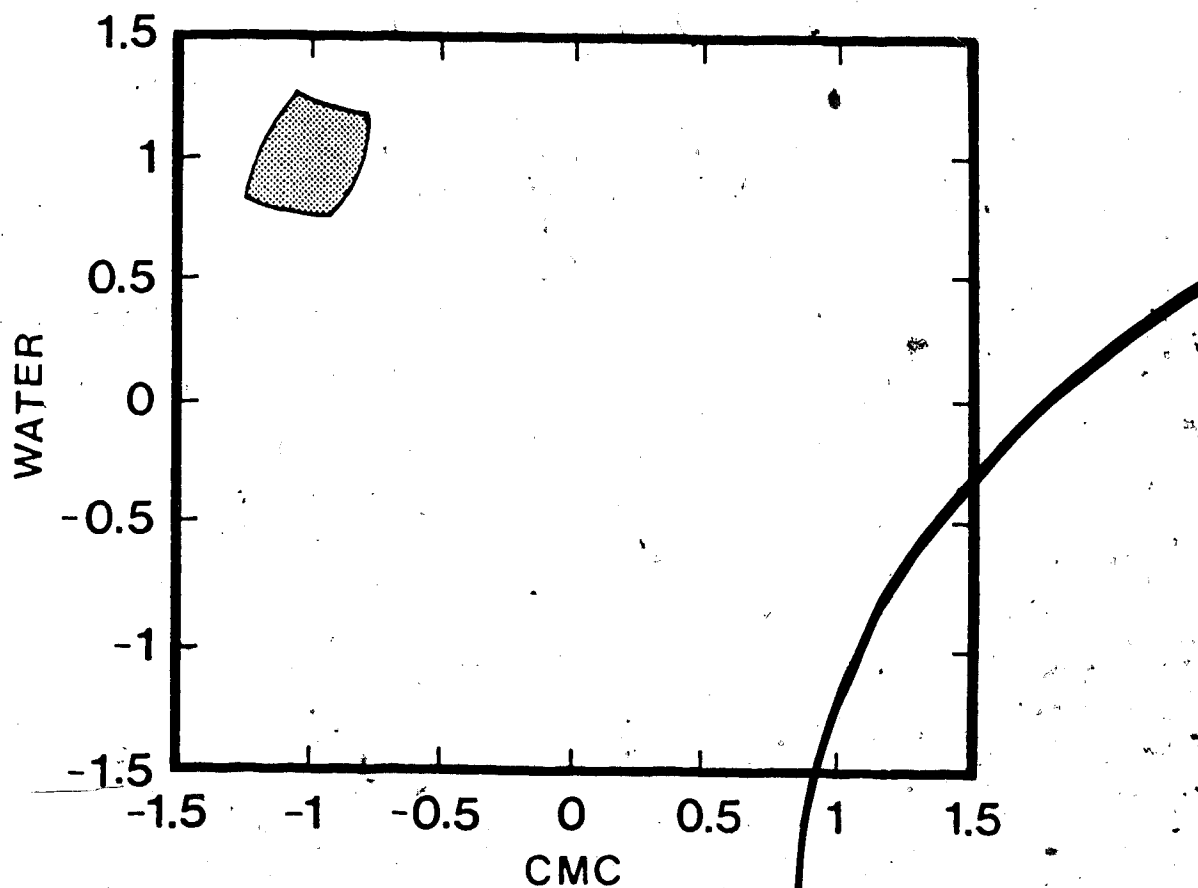


Figure VII.3 Superimposed contour plot region (objective and sensory responses combined) for Flour C rice breads at the -1.633 HPMC level. Levels of HPMC, water and CMC are coded values. The shaded region met reference standards for the objective measurements of specific volume, crust color 'a' values, crumb color 'L, a and b' values, percent moisture and the sensory measurements of firmness, yeasty flavor, adhesiveness, top crust color, crumb color and cell size uniformity.

from the region meeting the largest number of objective REF standards (Chapter V), and that meeting the largest number of sensory REF standards (Chapter VI). Rice breads, made from bread formulations from each of the shaded regions (Fig. VII.1, VII.2 and VII.3), are shown in Plate VII.1. The Flour A bread (Plate VII.1) met REF standards for specific volume, crust color 'L, a and b' values; crumb color 'L, a and b' values, Instron firmness, percent moisture, moistness, cohesiveness, yeasty flavor, adhesiveness, top crust color, crumb color, cell size uniformity, and cell wall thickness. The bread from Flour B (Plate VII.1) met REF standards for specific volume, crust color 'a' values, crumb color 'L, a and b' values, Instron firmness, percent moisture, moistness, cohesiveness, yeasty flavor, adhesiveness, aftertaste, top crust color, crumb color, cell size uniformity and cell wall thickness. Flour C breads (Plate VII.1) met REF standards for specific volume, crust color 'a' values, crumb color 'L; a and b' values, percent moisture, firmness, yeasty flavor, adhesiveness, crumb color, top crust color and cell size uniformity. Thus, the rice breads resembled the wheat bread REF not only in volume and grain characteristics, but also in some color, flavor and texture attributes. In contrast, the rice bread of Nishita (1977) resembled wheat bread somewhat in volume and grain characteristics, but lacked the flavor and texture associated with wheat bread.

Rice bread formulations, giving the actual levels of HPMC, CMC and water, for Flour A, B and C breads, respectively, that met the largest number of objective and sensory REF standards are given in Table VII.1. Since these formulations approximate the center point of the shaded regions shown in Figures VII.1, VII.2 and VII.3, they should allow for

Table VII.1 Rice bread formulas^{ab}.

INGREDIENTS	WEIGHT (g)		
	FLOUR A	FLOUR B	FLOUR C
Rice flour ^c	182.0	182.0	182.0
Potato Starch ^d	45.0	45.0	45.0
Salt	6.0	6.0	6.0
Sugar	24.0	24.0	24.0
Yeast	7.0	7.0	7.0
Oil	13.0	13.0	13.0
Hydroxypropyl- methylcellulose ^e	6.4	5.4	3.0
Carboxymethyl- cellulose ^f	0.8	0.7	1.0
Water	240.0	207.0	220.0

^a Adapted from Nishita, 1977.

^b Rice bread preparation procedure is given in Chapter V

^c Flour A - medium grain, finely ground, Dainty Foods

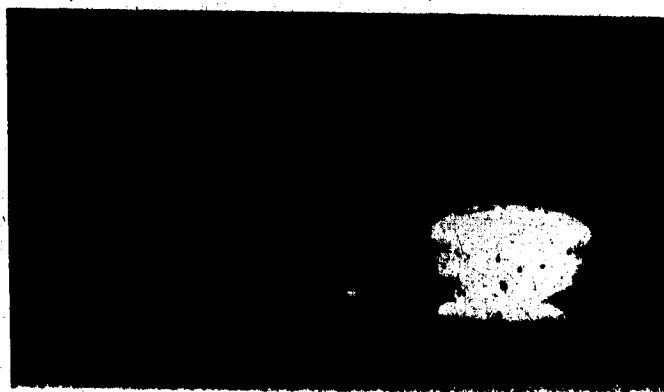
Flour B - medium grain, coarsely ground, Ener-G Foods.

Flour C - long grain, finely ground, Woodwards

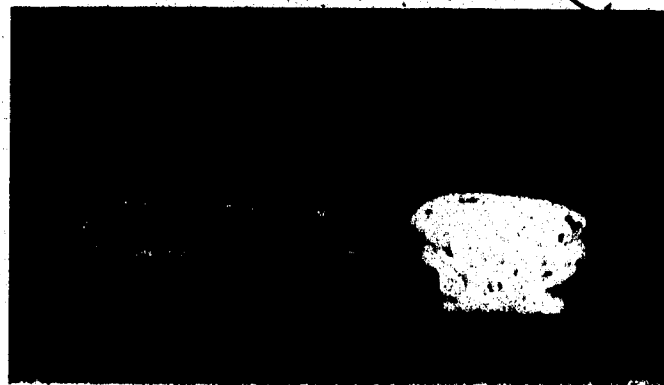
^d Casco potato flour

^e Methocel K4M, The Dow Chemical Company

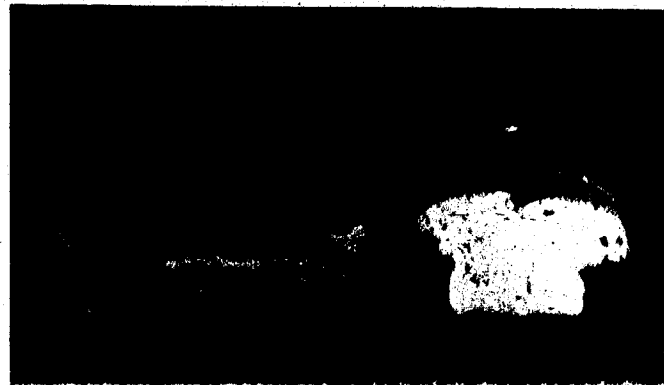
^f Hercules Cellulose Gum, Hercules Incorporated.



Flour A



Flour B



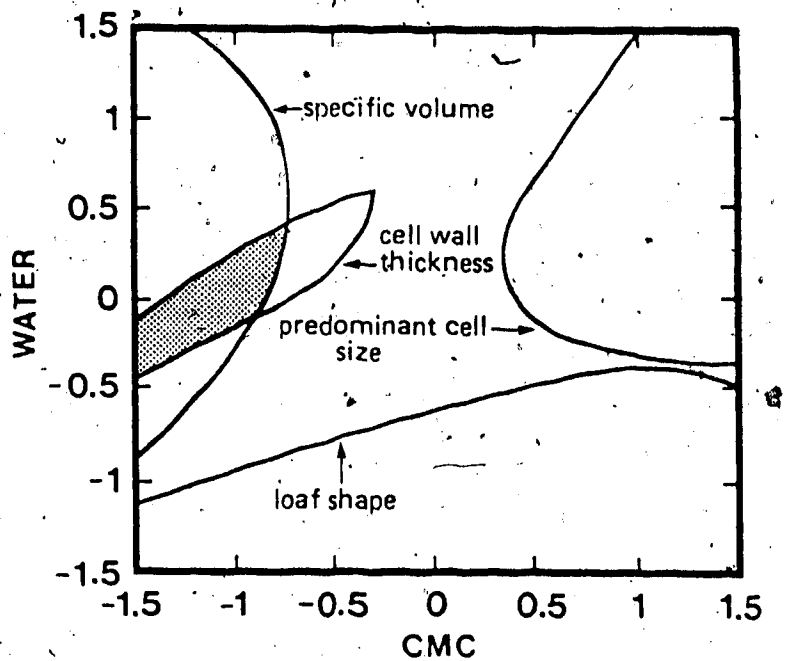
Flour C

Plate VII.1 - Rice breads meeting the maximum number of objective and sensory responses

variability that might be encountered in the preparation of these breads.

Evaluations of the objective (Chapter V) and sensory (Chapter VI) responses of rice breads show how each attribute can be considered to locate formulations that meet specific standards for an attribute. Flour A data for rice bread appearance (objective and sensory) at the a) -1.633 and b) 1.633 HPMC levels are shown in Fig. VII.4. At -1.633 HPMC, Flour A breads met REF standards for specific volume, cell wall thickness and cell size uniformity (no contour plots; mean met REF standards); rice breads at the 1.633 HPMC level met REF standards for specific volume, predominant cell size and cell size uniformity (no contour plots; mean met REF standards). Similarly, the Flour A bread color measurements (objective and sensory) at a) -1.633 and b) 1.633 HPMC levels (Fig. VII.5) can be considered. At the -1.633 HPMC level, a large number of water/CMC combinations would result in Flour A breads which met crust color 'L, a and b' values, crumb color 'L, a and b' values (no contour lines for crumb color 'L' and 'a' values - all formulations met REF standards), top crust color and crumb color (no contour line - all formulations met REF standards) REF standards. However; at the 1.633 HPMC level, only a few specific water/CMC (-1.3 to 0/-0.9) combinations met the objective and sensory color REF standards. The moisture and texture responses of Flour A breads at a) -1.633 and b) 1.633 HPMC levels, can be observed in Fig. VII.6. The region of Flour A bread formulations which met REF standards for Instron firmness, percent moisture, moistness, cohesiveness (no contour plots; mean met REF standards) and adhesiveness (no contour plots; mean met REF standards) was larger at the 1.633 HPMC level than at the

a) HPMC = -1.633



b) HPMC = 1.633

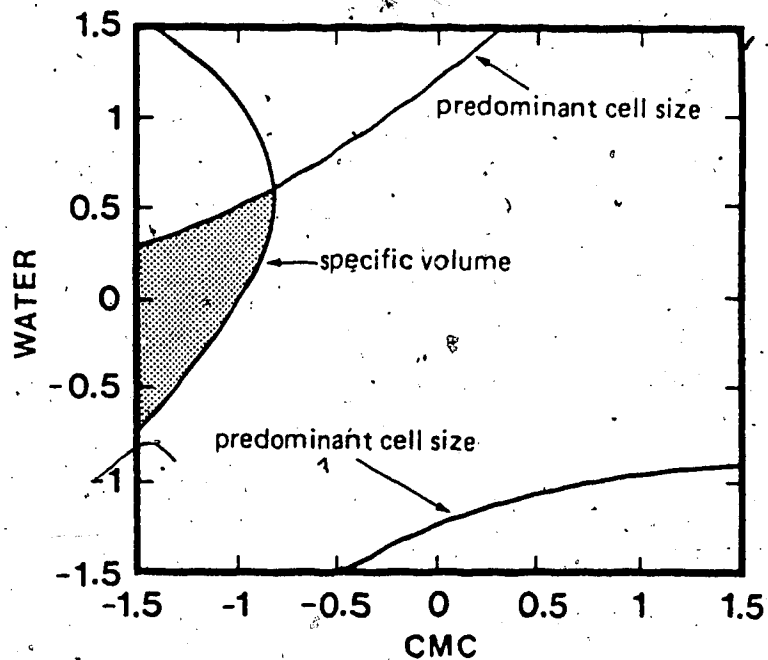
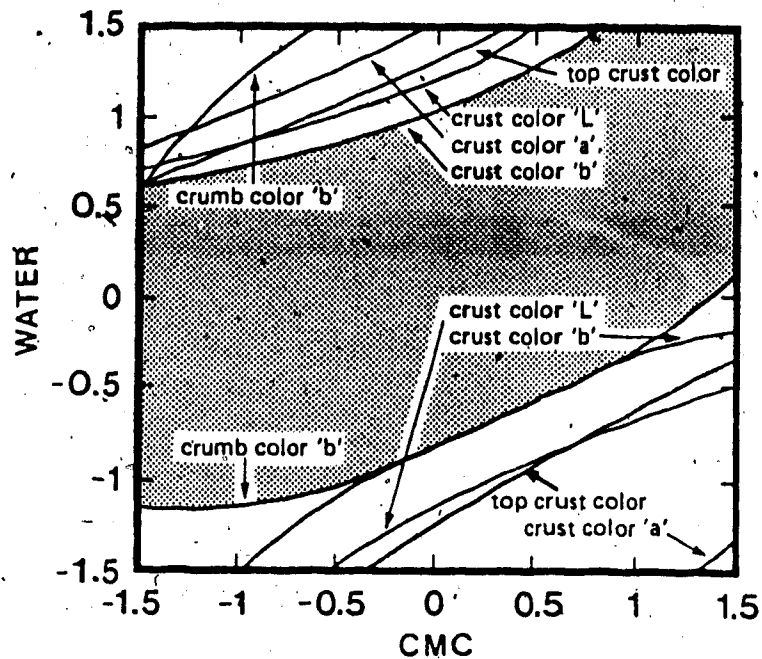


Figure VII.4 Superimposed contour plots (appearance) for Flour A rice breads at the a) -1.633 HPMC and b) 1.633 HPMC levels. Levels of HPMC, water and CMC are coded values. The contour lines represent the division between regions that met and regions that did not meet reference standards. The shaded region of plot a) met reference standards for specific volume, cell wall thickness and cell size uniformity. The shaded region of plot b) met reference standards for specific volume, predominant cell size and cell size uniformity.

a) HPMC = -1.633



b) HPMC = 1.633

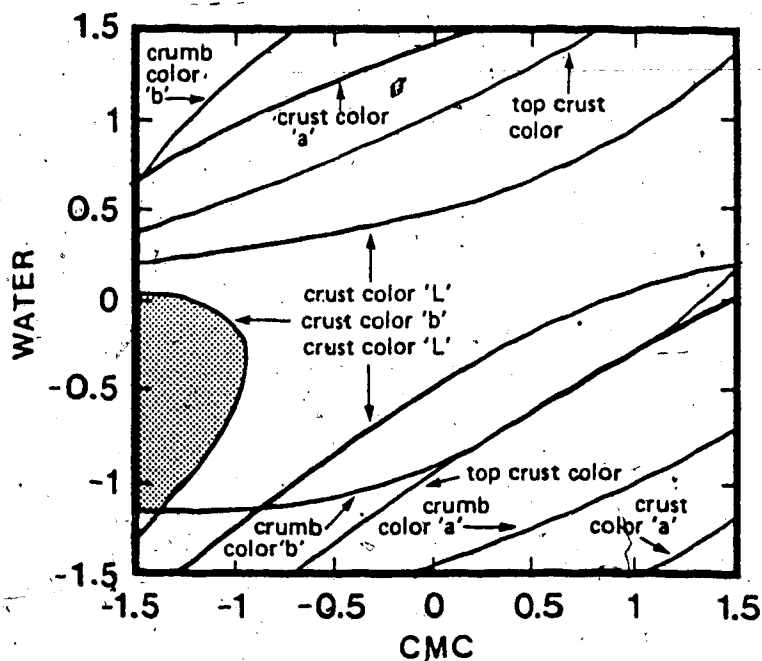
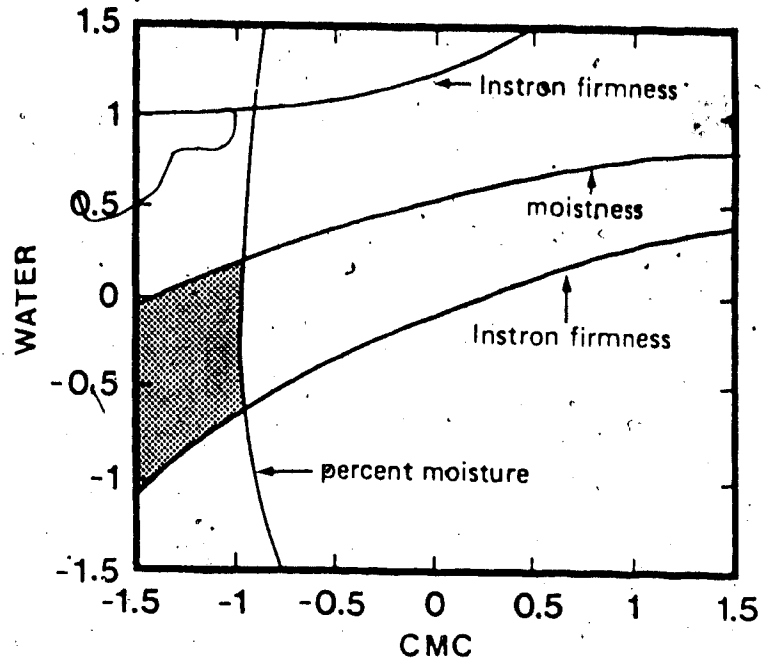


Figure VII.5. Superimposed contour plots (color) for Flour A rice breads at the a) -1.633 HPMC and b) 1.633 HPMC levels. Levels of HPMC, water and CMC are coded values. The contour lines represent the division between regions that met and regions that did not meet reference standards. The shaded region of plots a) and b) met reference standards for crust color 'L, a and b' values, crumb color 'L, a and b' values, top crust color and crumb color.

a) HPMC = -1.633



b) HPMC = 1.633

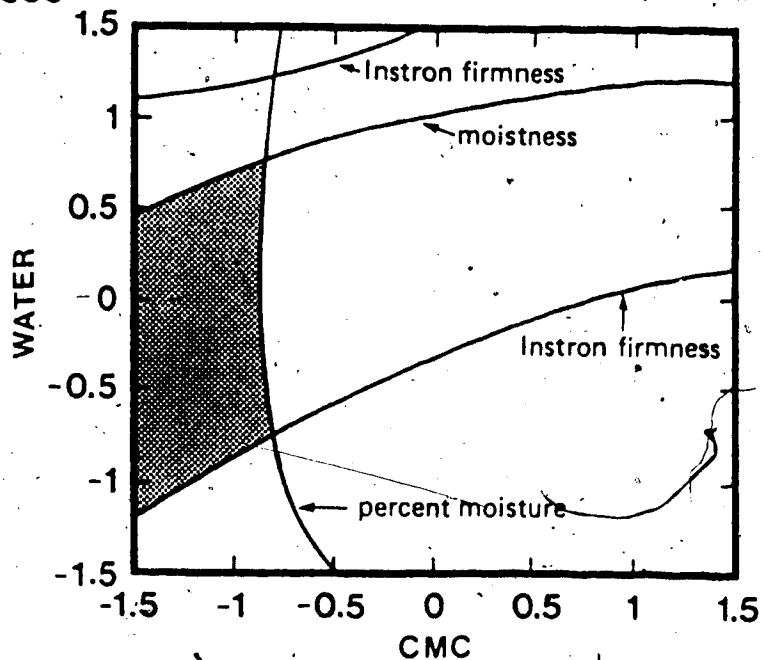


Figure VII.6 Superimposed contour plots (moisture and texture) for Flour A rice breads at the a) -1.633 HPMC and b) 1.633 HPMC levels. Levels of HPMC, water and CMC are coded values. The contour lines represent the division between regions that met and regions that did not meet reference standards. The shaded region of plots a) and b) met reference standards for Instron firmness, percent moisture, moistness, cohesiveness and adhesiveness.

-1.633 HPMC level. By considering the bread quality attributes singly, it is possible to choose those attributes that are the most important for product acceptability and to locate rice bread formulations that meet those specific attributes. Of the 25 attributes examined in the study, a maximum of 16 (Flour B) were found that met REF standards.

Thus, specifications for the final bread product must be set and then formulations determined to meet those specific standards. The end product should satisfy the user's requirements, wishes, requests and expectations (Folich, 1981). Both objective and sensory characteristics of a bread must be considered when the final bread formulation is determined. While instrumental measurements, such as bread volume, are a good indication of baking quality, the sensory qualities of the breads are also of significance. The nutritional value and the flavor may be parameters of more vital importance (Frolich, 1981), particularly to the quality of special diet products such as gluten-free bread. Good sensory quality represents a decisive stimulant for the regular use of dietetic foods (Paulus, 1986).

FUTURE RESEARCH NEEDS

The successful use of response surface methodology (RSM) in the present research suggests the applicability of RSM for the development of other special diet products. The appropriateness of RSM to the evaluation of the sensory and objective attributes of rice breads was illustrated and should be explored further.

Research on rice flour yeast breads should continue. Although the

present study, has produced acceptable rice bread formulations, these rice bread formulations must be brought up to scale, from laboratory conditions, to home use or commercial conditions, and refined. Difficulties will be encountered when a procedure, developed on a laboratory scale, must be scaled up (Kim and De Ruiter, 1968). However, acceptable rice bread formulations should be available to meet the special diet needs of the consumers.

In the present research, a number of difficulties were encountered in obtaining commercial rice flours. Thus, the use of a flour mill to grind whole grain rice may be more desirable. This would ensure knowledge of rice type and would allow for a more accurate comparison of rice varieties. Whole grain rice in bulk is also more readily available and more economical than bulk rice flour.

There is a need for the food industry to develop products which have equivalent nutritional value to the staples they replace (Campbell, 1984). Since wheat flours and breads are usually enriched with iron and B-vitamins, enrichment of rice breads is suggested.

The incorporation of brown rice flour and/or rice bran into the standard formulations could improve the nutritional value and fiber content of the rice breads. Although Nishita (1977) found the total replacement of white rice flour, with brown rice flour, to be unsuccessful in 100% rice breads, the partial replacement of white rice flour with brown rice flour has not been examined.

Additional research on the sensory evaluation of rice breads would be advantageous. The present study provides a beneficial introduction to the sensory characteristics of importance in rice breads. But, a detailed examination of rice bread flavor and texture could result in

further improvements in rice bread quality. Although the RSM design of the present study necessitated the presentation of 7-9 rice bread samples to the trained panelists, in the future, fewer rice bread samples should be evaluated at one sitting. Since the present study has provided information on rice bread sensory attributes, fewer sensory tasks should be given to the trained panelists. This would prevent panelist fatigue.

A technique which may be useful for the examination of rice bread flavor would be to study the flavor volatiles of the bread using gas liquid chromatography and mass spectrometry. This, combined with data from a rice bread flavor profile panel, could provide much needed information on the components of rice bread flavor and, perhaps, lead to improvements in rice bread flavor.

Optimization of the storage practices and packaging methods of rice breads, to prolong shelf life, would be another area of interest to study. The use of preservatives, such as calcium and sodium propionates which are typically added to wheat bread to inhibit mold growth, may also be appropriate for rice breads to enhance shelf life. Modified atmosphere packaging is often used to extend the shelf life of foods and may have possibilities for rice bread storage. For whole wheat and white bread, carbon dioxide delayed the firming of bread and growth of mold over a 5-15 day storage period (Knorr and Tomlins, 1985).

CONCLUSIONS

This research has resulted in the development of acceptable rice

flour yeast breads for individuals with celiac disease, wheat allergies or dermatitis herpetiformis. Since yeast breads represent an important aspect of the diet, the production of rice flour yeast breads for individuals who cannot tolerate gluten is warranted.

The ~~collective~~ data of these studies indicate that response surface methodology (RSM) can be applied to the development of rice flour yeast breads. Rice breads that are comparable to wheat breads are desired by gluten-free bread consumers and can be made to meet pre-set sensory and instrumental standards. However, successful production of rice flour yeast breads is dependent upon the type of rice flour used and the amounts of gums (HPMC and CMC) and water incorporated into the formulation. Specific levels of HPMC, CMC and water were required in bread formulations for each of the Flours A, B and C. The use and appropriateness of RSM in this research could result in the application of RSM to the development of other special diet products.

In addition, this study has provided valuable, much needed information about the sensory and instrumental evaluation of the gluten-free breads. Methods for the training of panelists for rice bread sensory evaluations, as well as suggestions for the selection of rice bread quality attribute descriptors, were determined. Procedures for the instrumental measurement of bread firmness were also studied.

Future research on rice flour yeast breads should include the refinement of rice bread formulations for commercial and/or home production, the use of whole-grain rice flours and the enrichment of rice breads with iron and B-vitamins. Improvements in the shelf stability of rice breads through the addition of preservatives, as well

as the use of packaging methods and storage practices that prolong the shelf life of rice breads, should also be examined in detail.

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Appendix 1 - Edmonton Celiac Association survey
covering letter

I require your assistance for a study that I am conducting with Dr. Zenia Hawrysh, Department of Foods and Nutrition, University of Alberta. The study will focus on the development of a gluten-free rice flour yeast bread. You are asked to provide some information on gluten-free products you now use. Later, I will require your assistance in evaluating some breads resulting from my research. The evaluation of breads will be conducted during a Celiac Association general meeting. All information collected will be held in confidence and used only for research purposes. You are free to refuse to answer any questions or to decline bread sample evaluations. A summary of results from this questionnaire will appear in your Newsletter, after March 15, 1985, and will not reveal information about any given individual. If you have any questions, please contact me at the University (432-4925) or at home (437-6858). Your help would be greatly appreciated in this research project.

Sincerely,

Gladys Ylimaki
Graduate Student

Appendix 2 - Edmonton Celiac Association survey
questionnaire

1. Can you tolerate wheat starch in your diet?
_____ yes _____ no
2. Has your ability to tolerate wheat starch been determined by your medical doctor or your own personal assessment?
3. What type(s) of starch do you use in cooking and baking?
4. a) Do you substitute products such as rice cakes, rice crackers and potatoes for bread in your diet? _____ yes _____ no
b) If yes, please specify substitute product(s):

5. a) Do you regularly consume a gluten-free yeast bread product? _____ yes _____ no
b) If yes, what type? (Please provide brand names)
Commercially prepared bread _____
Commercial dry mix _____
Made from a recipe _____
Other _____
6. a) If you purchase commercially prepared gluten-free bread, how many loaves would you purchase per week? _____ loaves/week
b) If not weekly, how many loaves would you purchase per month? _____ loaves/month

7. a) Please indicate the number of slices of gluten-free bread you eat per day?
 _____ slice(s)/day
- b) If not daily, how many slices per week?
 _____ slice(s)/week
8. How do you generally eat gluten-free bread?
- plain (ie. as bread) _____
- toasted _____
- as a sandwich _____
- other _____
9. How do you store gluten-free bread?
 (Please answer for a and b)
- a) Plastic bag _____ b) Room temperature _____
- Aluminum foil _____ Refrigerated _____
- Plastic wrap _____ Frozen _____
- Other _____
10. In general, how long would you store a loaf of gluten-free bread:
- at room temperature _____
- in the refrigerator _____
- in the freezer _____
11. a) In general, are you satisfied with the gluten-free product(s) you use? _____ yes _____ no
- b) If no, what problems or difficulties have you encountered?
12. Would you be willing to try a new gluten-free yeast bread if it became available? _____ yes _____ no

13. List the 5 quality characteristics that you consider to be the most important in a gluten-free yeast bread. List them in order of importance, the first one being the most important. (Quality characteristics of food products include flavor characteristics, odor characteristics, visual properties and textural properties.)

Most important 1. _____
 2. _____
 3. _____
 4. _____
 Least important 5. _____

14. How does your diet modification affect your food choices?

15. a) Are you the only member of your household with celiac disease? yes no

b) If no, how many other household members have celiac disease? _____

16. How long have you had celiac disease?

ADDITIONAL COMMENTS:

Age (years): Under 18 _____ Sex: Male _____
 19-25 _____ Female _____
 26-35 _____
 36-50 _____
 51-65 _____
 Over 65 _____

Appendix 3 - Summary of the results of the Edmonton
Celiac Association survey

In February, with the help and support of Joyce Friesen and other members of the Edmonton Celiac Association, you were sent a questionnaire. I would like to thank each of you who answered the questionnaire. Your kind support and encouragement is also sincerely appreciated.

The results of the questionnaire ascertained the gluten-free products currently being used and determined the problems encountered by product users and the quality attributes in gluten-free bread considered important by gluten-free bread users. The questionnaire was necessary to ensure that, in my research, I would address the problems and areas of interest which you, the major consumers of the product, felt were important. We also wanted to determine the extent of the need for another gluten-free yeast bread and the potential market for a new, gluten-free yeast bread product. Thus, your responses have helped in the planning of the subsequent stages of my research, which is to develop an acceptable gluten-free rice flour yeast bread, using response surface methodology (RSM). RSM, a statistical technique, is particularly appropriate for product formulations and recipe development since it can consider several ingredients at different levels in a product, as well as the corresponding interactions among these ingredients and levels. However, thus far, RSM has not been utilized for the development of gluten-free bread products.

The response to the survey was extremely encouraging. Two hundred questionnaires were mailed and 122 people responded, representing a 61% response rate. Most of the respondents (70%) were female; respondent age groups ranged from under 18 to over 65. All but 8 respondents indicated having celiac disease for a period of from 2 months to 73 years, based on the time since diagnosis of the disease. However, many respondents reported that they suffered from celiac disease for a long time prior to diagnosis.

Overall, many respondents on the gluten-free diet appear to be eating well-balanced, nutritious meals. Some respondents reported eating fewer sweets and junkfood and more meat, fruit and vegetables. Since whole grain wheat breads have been eliminated in a gluten-free diet, some respondents were concerned about their intake of B vitamins and fiber. Thus, it is important to note that milk, meat (especially liver),

green leafy vegetables and legumes are excellent sources of B vitamins. Fruits, such as prunes and apples, and vegetables, particularly raw, are readily available sources of fiber. There are also some bran products available, such as rice bran, rice polishings and oat bran (if oats are tolerated) which are good sources of fiber. These products are quite acceptable when added to baked goods and entrees such as meat loaf.

Almost half of the respondents (46%) indicated an inability to tolerate starch in their diet. Another 15% of the participants stated that they were unsure of their wheat starch tolerance, or that they had never tried to eat wheat starch. In addition, many of the respondents who can tolerate wheat starch reported they never or seldom use it. In fact, less than 10% of the respondents actually reported using wheat starch in their cooking and baking. Therefore, no wheat starch will be included in my gluten-free bread formulation. The major flour component will be rice flour, which also presents fewer problems for individuals with allergies. According to the respondents, rice flour was the most popular flour for baking and cooking. Ninety-three percent of the respondents reported using rice flour while 73% and 72% used potato flour and corn starch, respectively. Since potato flour is second most popular, after rice flour, a small percentage of potato flour will be incorporated into my rice bread formulation to improve the quality characteristics. Other flours and starches used by the respondents are soya flour (53%), corn flour (43%), tapioca starch (19%), potato starch (12%), pea and bean flour (7%), corn meal (7%), wheat starch (7%) and arrowroot (3%).

It was encouraging to note that almost 75% of the respondents regularly consumed a gluten-free yeast bread. The majority (70%) ate commercially prepared bread, while others used commercial dry mixes (23%) or made their own bread from recipes (39%). Many respondents used two or three different types of bread. Overall, bread is still a popular staple; most respondents (74%) consumed at least 1-4 slices of gluten-free yeast bread per day.

The need for toasting many gluten-free yeast breads was evident as 80 of the 88 respondents, who regularly eat gluten-free bread, reported eating gluten-free bread toasted. Although more than half of the respondents ate gluten-free bread as sandwiches, the sandwich bread was often toasted. Other suggestions offered by the respondents for eating gluten-free bread were grilling and microwaving the bread and making french toast, cinnamon buns and breadcrumbs from the bread or dough.

Although 43% of the respondents were satisfied with the available gluten-free yeast breads, almost all of the respondents (94%) indicated a willingness to try a new product. Thus there appears to be a need for and a potential market for a new gluten-free yeast bread. The major problems reported by the respondents who were not satisfied with the available gluten-free breads were crumbliness, dryness and a poor or bland flavor. Similarly, the most important quality characteristics desired in bread by respondents were flavor and texture. These attributes were followed by odor, visual characteristics, shelf-life, price, nutrition and availability. I will attempt to solve some of these problems, most importantly the texture. Although flavor was the most important quality characteristic reported by the majority of the respondents, the flavor and texture of the product are interrelated. Methods of improving bread texture will inevitably enhance bread flavor, as well as the visual appearance, loaf size and odor. Initially I will keep the number of ingredients to a minimum so that the resulting gluten-free bread can also be consumed by individuals with other intolerances and allergies, such as for milk and eggs. I hope to counteract the many problems by using gluten-free gums and emulsifiers. Gums, such as xanthan gum, which many respondents reported using in gluten-free bread recipes, are important as gluten replacements to stabilize or give structure to gluten-free breads. While gums may work effectively individually, many work more effectively when a number are combined. Using response surface methodology I hope to find a combination of gums which will work together to produce a better product; one which is less dry and crumbly and more like a wheat flour bread.

At present, I am baking bread and will be measuring the quality characteristics, such as texture, volume and color, using a variety of instruments. The evaluation of bread quality will also be obtained from trained and consumer panels. I hope to have some samples for you to taste and evaluate at the October meeting, so I hope you will attend.

In conclusion, thank you for your support and assistance, I can be reached in the lab (432-3833) or at home (437-6858) if you have any further questions or suggestions.

Gladys Ylimaki, Graduate Student
Department of Foods and Nutrition
University of Alberta

¹ Appeared in the Edmonton Celiac Association Newsletter
July/August, 1985

Appendix 4 - Hospital survey covering letter

I require your assistance for a study that I am conducting with Dr. Zenia Hawrysh, Department of Foods and Nutrition, University of Alberta. The study will focus on the development of a gluten-free rice flour yeast bread. You are asked to provide some information on your hospital's use of gluten-free products. All information collected will be held in confidence and used only for research purposes. You are free to refuse to answer any questions. If you desire, a summary of results of the questionnaire will be available after March 15, 1985. The summary will not reveal details about any given hospital. If you have any questions, please contact me at the University (432-4925) or at home (437-6858). Your help would be greatly appreciated in this research project.

Sincerely,

Gladys Ylimaki.
Graduate Student

Appendix 5 - Hospital survey questionnaire

1. a) What diet recommendations do you make to patients requiring a gluten-free diet?

b) Do you have pamphlets or diet sheets available for such patients? _____yes _____no
c) If yes to 1b, describe. (Provide copies if possible)

2. How often does a gluten-free yeast bread appear on the menu for patients requiring a gluten-free diet?

3. How is gluten-free bread generally served?
plain (ie: as bread) _____
toasted _____
as a sandwich _____
other _____

4. What type(s) of gluten-free yeast bread does your hospital use? (Please provide brand names and/or type of starch used.)
Commercially prepared bread _____
Commercial dry mix _____
Made from a recipe _____
Other _____

5. a) If commercially prepared gluten-free bread is purchased, how many loaves would be purchased per week? _____loaves/week
b) If not weekly, how many loaves would be purchased per month? _____loaves/month

6. a) Do you substitute products such as rice cakes, rice crackers and potatoes for bread on the menu, for patients requiring a gluten-free diet? yes no

b) If yes, please specify substitute product(s):

7. How do you store gluten-free bread? (Please answer for a) and b)

- a) Plastic bag b) Room temperature
- Aluminum foil Refrigerated
- Plastic Wrap Frozen
- Other

8. In general, how long would a loaf of gluten-free bread be stored:

- at room temperature _____
- in the refrigerator _____
- in the freezer _____

9. a) In general, are you satisfied with the gluten-free product(s) you use? yes no

b) If no, what problems or difficulties have you encountered?

10. a) Have you had patient complaints concerning the quality of gluten-free bread served? yes no

b) If yes, please describe.

11. Would your hospital be willing to try a new gluten-free yeast bread if it became available?
 _____ yes _____ no
12. List the 5 quality characteristics that you consider to be the most important in a gluten-free yeast bread. List them in order of importance, the first one being the most important. (Quality characteristics of food products include flavor characteristics; odor characteristics, visual properties and textural properties.)
- Most important 1. _____
 2. _____
 3. _____
 4. _____
 Least important 5. _____
13. Would you like a copy of a summary of the results of this questionnaire? _____ yes _____ no

ADDITIONAL COMMENTS:

NAME OF HOSPITAL: _____

ADDRESS: _____

SIZE (NUMBER OF BEDS): _____

POSITION OF RESPONDENT
 (DIETARY DEPARTMENT) _____

Appendix 6 - Summary of the results of the hospital survey

In February 1985, you were sent a questionnaire. I would like to thank you for answering and returning the questionnaire and helping me with my research. Your kind support and encouragement is sincerely appreciated.

The results of the questionnaire determined the gluten-free products currently being used in hospitals in Alberta, the problems encountered with the products and the quality attributes in gluten-free bread considered important by dietary personnel. The questionnaire was necessary to ensure that, in my research, I would address the problems and areas of interest which you felt were important. We also wanted to determine the extent of the need for a gluten-free rice flour yeast bread and the potential market for a new, gluten-free yeast bread product. Thus, your responses have helped in the planning of the subsequent stages of my research, which is to develop a gluten-free rice flour yeast bread, comparable to wheat flour (white) bread, using response surface methodology (RSM). RSM, a statistical technique, is particularly appropriate for product formulation and recipe development since it can consider several ingredients at different levels in a product, as well as the corresponding interactions among these ingredients and levels. However, thus far, RSM has not been utilized for the development of gluten-free bread products.

The response to the survey was extremely encouraging. Twenty questionnaires were mailed to the dietary departments of hospitals in Edmonton and surrounding areas in Alberta, and 15 hospitals responded, representing a 75% response rate. Two of the hospitals that responded, however, could not complete the questionnaire since they had not had the opportunity to use gluten-free bread products.

Overall, the hospitals that had used gluten-free bread products for patients on gluten-free diets appeared to be making appropriate recommendations. The majority of hospitals (77%) used some form of the gluten-free diet pamphlet prepared by the Edmonton Hospitals Diet Therapy Committee and distributed by the University of Alberta Hospitals, for distribution to patients. Three hospitals provided additional gluten-free recipes to patients and three provided

information to patients about where to obtain gluten-free recipes, products and additional information. One hospital provided information to patients about the Edmonton Celiac Association, distributing an application form for membership.

Most of the hospitals did not have gluten-free breads on the menu daily but 69% of the dietary departments would provide gluten-free bread daily if a patient required it. Commercially prepared gluten-free bread was used by 54% of the hospitals, while others used commercial dry mixes (23%) or made their own recipes (31%).

The need for toasting many gluten-free yeast breads was evident as 11 of the 13 hospitals, who used gluten-free breads, reported serving gluten-free bread toasted. Ten of the hospitals served gluten-free bread plain and seven served the bread as sandwiches. Seventy-seven percent of the dietary departments also substituted rice cakes, potatoes, muffins, rice and rice crackers for bread on the menu for patients requiring a gluten-free diet.

Five of the respondents were satisfied with the available gluten-free yeast breads and four hospitals were not satisfied. Almost all of the respondents (93%) indicated a willingness to try a new product. Thus there appears to be a need for and a potential market for a new gluten-free yeast bread.

The major problems in gluten-free yeast breads reported by the dietary personnel were dryness and crumbliness. The most important quality characteristic desired in bread by respondents was texture, followed closely by flavor. Odor, visual characteristics, shelf-life and price were also considered important quality attributes of gluten-free breads. I will attempt to solve some of the problems, most importantly the texture. The flavor and texture of the product are interrelated and methods of improving bread texture will also inevitably enhance bread flavor, as well as the visual appearance and odor. Initially I will keep the number of ingredients to a minimum so that the resulting gluten-free bread can also be consumed by individuals with other intolerances and allergies, such as for milk and eggs. Since rice flour is lower in protein than wheat flour, rice flour bread can also be used for patients requiring a reduced protein intake. I hope to counteract the many problems by using gluten-free gums and emulsifiers. Gums, such as xanthan gum, which some

hospitals reported using in gluten-free bread recipes, are important as gluten replacements to stabilize or give structure to gluten-free breads. While gums may work effectively individually, many work more effectively when a number are combined. Using response surface methodology I hope to find a combination of gums which will work together to produce an acceptable product; one which is not dry and crumbly and more like a wheat flour (white) bread.

At present, I am baking breads and will be measuring the quality characteristics, such as texture, volume and color, using a variety of instruments. The evaluation of bread quality will also be obtained from trained and consumer panels. I hope to have an acceptable rice bread product by December, 1986.

In conclusion, thank you for your support and assistance. I can be reached in the lab (432-3833) or at home (437-6858) if you have any further questions or suggestions.

Gladys Ylimaki
Graduate Student
Department of Foods and Nutrition
University of Alberta

Appendix 7 - Consumer panel ballot

Instructions: For each characteristic listed, rate each of the three gluten-free bread samples by placing a number (your score) from 1 to 6 in the box under the appropriate sample number.

CHARACTERISTIC	SAMPLES					
	6	5	4	3	2	1
Aroma	very desirable	desirable	slightly desirable	slightly undesirable	undesirable	very undesirable
Appearance	very desirable	desirable	slightly desirable	slightly undesirable	undesirable	very undesirable
Firmness	very soft	soft	slightly soft	slightly firm	firm	very firm
Moistness	very moist	moist	slightly moist	slightly dry	dry	very dry
Flavor	very desirable	desirable	slightly desirable	slightly undesirable	undesirable	very undesirable
Overall Acceptability	very acceptable	acceptable	slightly acceptable	slightly unacceptable	unacceptable	very unacceptable

Are you on a special diet? YES _____ NO _____

If yes, specify. CELIAC _____ OTHER _____

SEX: MALE _____ FEMALE _____

AGE(years): Under 18 _____

19-25 _____

26-35 _____

36-50 _____

51-65 _____

Over 65 _____

COMMENTS: 2

Appendix 8 - Analysis of variance table for a replicated 3 x 3
 Graeco-Latin square design -
 Flour C standard rice bread formula

SOURCE OF VARIATION	DEGREES OF FREEDOM	SUM OF SQUARES	MEAN SQUARE	F VALUES
Proofer shelf	2	2281.82	1140.91	1.89
Oven	2	3958.16	1979.08	3.28
Replications	1	956.30	956.30	1.59
Baking order	2	821.27	410.63	.68
Time of day	2	3001.25	1500.63	2.49
Residual	8	4825.11	603.14	
TOTAL	17	15843.91		

Appendix 9 - Scorecard use for rice bread loaf shape

LOAF SHAPE

LOAF NUMBER: _____

DATE: _____

	6	5	4	3	2	1
LOAF BOTTOM SURFACE	flat	very sl concave	sl concave	concave	very concave	ext concave
LOAF SIDES	straight	very sl caved	sl caved	caved	very caved	ext caved
LOAF TOP SURFACE	ext round	very round	round	sl round	flat	caved
TOP SURFACE SYMMETRY	very symmet	symmet	sl symmet	sl unsymmet	unsymmet	very unsymmet
LOAF TOP-SIDE INDENTATION	no indent	very sl indented	sl indented	mod indented	very indented	ext indented

LOAF SHAPE SCORE _____

Appendix 10 - Ballot used for bench top sensory evaluation of rice breads

Judge _____
 Date _____
 Sample Number _____

Instructions: For each characteristic listed, circle the descriptive word or phrase which best describes the sample.

CHARACTERISTIC	8	7	6	5	4	3	2	1
Crust Color	extremely dark brown	very dark brown	dark brown	slightly dark brown	slightly light brown	light brown	very light brown	extremely light brown
Crumb Color	extremely bright white	very white	white	slightly white	slightly grey	grey	very grey	extremely grey
Average Cell Size	extremely small	very small	small	slightly small	slightly large	large	very large	extremely large
Cell Size Uniformity	extremely uniform	very uniform	uniform	slightly uniform	slightly irregular	irregular	very irregular	extremely irregular
Cell Wall Thickness	extremely thin	very thin	thin	slightly thin	slightly thick	thick	very thick	extremely thick
Aroma	Intensity	extremely intense	intense	slightly intense	slightly weak	weak	very weak	extremely weak
	Desirability	extremely desirable	very desirable	desirable	slightly undesirable	undesirable	very undesirable	extremely undesirable
Firmness	Intensity	extremely soft	very soft	soft	slightly firm	firm	very firm	extremely firm
	Desirability	extremely moist	very moist	moist	slightly dry	dry	very dry	extremely dry
Gumminess	Intensity	no gumminess	trace of gumminess	very slight gumminess	slight gumminess	moderately gummy	very gummy	extremely gummy
	Desirability	extremely intense	very intense	intense	slightly intense	weak	very weak	extremely weak
Flavor	Intensity	extremely desirable	very desirable	desirable	slightly undesirable	undesirable	very undesirable	extremely undesirable
	Desirability	extremely intense	very intense	intense	slightly weak	weak	very weak	extremely weak
Aftertaste	Intensity	extremely desirable	very desirable	desirable	slightly undesirable	undesirable	very undesirable	extremely undesirable
	Desirability	extremely desirable	very desirable	desirable	slightly undesirable	undesirable	very undesirable	extremely undesirable

COMMENTS:

Appendix 11 - Confounding pattern for main effects and two-factor interactions^a

VARIABLES ^b	CONFOUNDING PATTERN
x_1	1 + 24 + 35 + 67
x_2	2 + 14 + 36 + 57
x_3	3 + 15 + 26 + 47
x_4	4 + 12 + 56 + 37
x_5	5 + 13 + 46 + 27
x_6	6 + 23 + 45 + 17
x_7	7 + 34 + 25 + 16

^a Adapted from Box et al., 1978, pg. 392

^b When all 7 variables were included in the design, x_1 =CMC, x_2 =xanthan gum, x_3 =HPMC, x_4 =guar gum, x_5 =SSL, x_6 =GMS, x_7 =water

Appendix 12 - Regression equation coefficients and R² values for objective rice bread responses - Flour A rice breads

Coefficient	RESPONSE										
	Loaf Shape	Specific Volume	Crust Color 'L'	Crust Color 'a'	Crust Color 'b'	Crumb Color 'L'	Crumb Color 'a'	Crumb Color 'b'	Instrument Firmness	Moisture	X
b ₀	20.576	3.793	42.433	13.630	19.755	78.714	-1.105	7.884	0.037	42.512	
b ₁ (CMC)	1.154**	-0.762***	0.536	0.076	0.286*	0.926***	0.029	0.220	0.107***	2.277***	
b ₂ (HPMC)	-0.596	-0.077	0.046	0.000	0.020	0.238	0.018	0.156	-0.002	-0.218	
b ₃ (WATER)	-2.142***	0.608***	-0.223	-0.280***	-0.233	0.512*	-0.062***	-0.815***	-0.230***	0.687***	
b ₁₁	-0.368	0.019	0.387	-0.151*	0.026	-0.312	0.019	0.434**	0.003	-0.480	
b ₂₂	0.007	0.021	0.138	-0.030	0.054	-0.340	-0.014	-0.142	-0.015	-0.008	
b ₃₃	-0.087	-0.385***	2.946***	-0.386***	0.968***	0.053	0.047**	0.669***	0.192***	-0.217	
b ₁₂	0.438	0.054	0.778	-0.094	0.225	0.153	0.047*	-0.134	-0.004	0.127	
b ₁₃	0.938*	0.209**	-2.897***	0.338***	-1.006***	-0.216	-0.034	-0.784***	-0.156***	0.708**	
b ₂₃	-1.188*	-0.032	0.028	0.069	0.044	0.222	0.022	0.116	0.000	-0.238	

Equation Significance	.57	.90	.78	.78	.76	.54	.55	.70	.90	.89
R ² b	***	***	***	***	***	**	***	***	***	***

Y = b₀ + b₁x₁ + b₂x₂ + b₃x₃ + b₁₁x₁² + b₂₂x₂² + b₃₃x₃² + b₁₂x₁x₂ + b₁₃x₁x₃ + b₂₃x₂x₃ where x₁ = CMC, x₂ = HPMC, x₃ = water.

b Replication and block effects removed.

*, **, *** Significant at P<0.05, P<0.01 and P<0.001, respectively.

Appendix 13 - Regression equation^a coefficients and R² values for objective rice bread responses - Flour B rice breads

Coefficient	RESPONSE							
	Loaf Shape	Specific Volume	Crust Color 'L'	Crumb Color 'L'	Crumb Color 'b'	Instron Firmness	Z Moisture	
b ₀	22.960	3.528	44.816	77.878	7.476	0.110	40.830	
b ₁ (CMC)	2.494***	-0.747***	1.508**	-1.490***	0.290**	0.209***	2.581***	
b ₂ (HPMC)	0.634	-0.008	-0.338	0.109	0.009	0.032	0.057	
b ₃ (WATER)	-1.416***	0.530**	-0.040	0.556*	-0.624***	-0.232***	0.206	
b ₁₁	-1.904***	-0.002	1.721**	0.323	0.507***	0.106***	-0.726***	
b ₂₂	0.159	-0.007	-0.215	-0.024	-0.042	0.018	0.171	
b ₃₃	-1.341***	-0.245***	0.844	0.290	0.600***	0.104***	-0.709***	
b ₁₂	-0.125	-0.056	0.638	0.100	-0.153	0.043	-0.230	
b ₁₃	2.000***	0.140	-3.594***	-0.175	-0.728***	-0.247***	1.070***	
b ₂₃	-0.125	0.067	-0.844	0.062	-0.091	-0.063	0.170	

R ² b	.80	.90	.66	.60	.87	.88	.87
Equation Significance	***	***	***	***	***	***	***

^a $Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$ where x_1 = CMC, x_2 = HPMC, x_3 = water.

^b Replication and block effects removed.

*, **, *** Significant at P<0.05, P<0.01, and P<0.001, respectively.

Appendix 14 - Regression equation^a coefficients and R² values for objective rice bread responses - Flour C rice breads

Coefficient	RESPONSE									
	Loaf Shape	Specific Volume	Crust Color 'L'	Crust Color 'a'	Crust Color 'b'	Crumb Color 'L'	Crumb Color 'b'	Firmness	Instroton	% Moisture
	22.917	3.496	46.456	13.246	21.680	78.927	7.606	0.247	40.176	
MC	1.686***	-0.686***	0.791	-0.323***	0.106	-0.897***	0.720***	0.408***	2.432***	
b ₁ (MC)	0.725*	-0.075	-0.831	0.172	-0.275*	0.450*	0.092	-0.008	0.385	
b ₃ (WATER)	-1.134**	0.615***	-1.193**	0.048	-0.396**	0.270	-0.974***	-0.516***	0.872***	
b ₁₁	-1.219***	-0.056	1.334**	-0.180*	0.279*	-0.595**	0.333***	0.149***	-0.690**	
b ₂₂	-0.188	-0.009	0.134	-0.030	0.091	-0.520**	-0.177	0.007	0.305	
B ₃₃	-1.031**	-0.224***	0.884*	-0.162	0.255	-0.337	0.469***	0.291***	-0.775**	
b ₁₂	-0.312	0.098	0.638	-0.106	0.253	-0.494*	-0.178	-0.059	-0.469	
b ₁₃	0.688	0.091	-2.225***	0.369**	-0.522**	0.094	-0.553***	-0.445***	0.786**	
b ₂₃	-0.312	-0.100	0.094	0.100	0.103	0.044	0.041	0.000	0.048	

R ^{2b}	.72	.94	.63	.53	.54	.65	.92	.93	.84
Equation Significance	***	***	***	**	**	***	***	***	***

^a $Y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_{11}x_1^2 + b_{22}x_2^2 + b_{33}x_3^2 + b_{12}x_1x_2 + b_{13}x_1x_3 + b_{23}x_2x_3$ where $x_1 = \text{MC}$, $x_2 = \text{HPMC}$, $x_3 = \text{water}$.

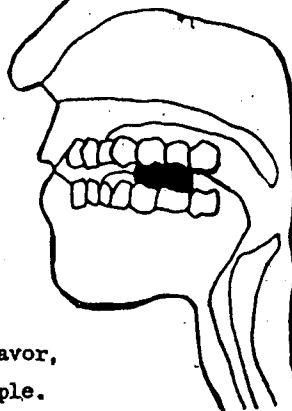
^b Replication and block effects removed.

*, **, *** Significant at P<0.05, P<0.01, and P<0.001, respectively.

Appendix 15 - Organoleptic evaluation instruction sheet

RICE BREAD EVALUATIONS

1. Eat a piece of apple. Then, rinse your mouth with water, swooshing the water around in your mouth.
2. Evaluate the bread samples in the order indicated on your ballot.
3. Evaluate the yeasty and rice odors of the first sample by bringing the petri dish up to your nose, lifting the lid at one side and taking 2-3 short sniffs.
4. Evaluate the firmness of the sample by inserting a toothpick into the side of one round of the sample and positioning the sample between your molar teeth.



FIRMNESS: The perceived force required to gently compress the sample between the molar teeth (DO NOT BITE RIGHT THROUGH).

5. Chew the sample, moving it around in your mouth, and evaluate the moistness, cohesiveness, yeasty flavor, rice flavor, adhesiveness and graininess of the sample.

MOISTNESS: The perceived degree of moisture in the sample after 3-4 chews.

COHESIVENESS: The degree to which the sample holds together during chewing.

ADHESIVENESS: The degree to which the sample sticks to the teeth during the later stages of chewing. (Not mouthcoating or graininess)

GRAININESS: The perceived graininess in the mouth at the end of chewing.

6. After swallowing the sample and before rinsing, evaluate the aftertaste of the sample.
7. Between each sample (petri dish), eat a piece of apple to clear out your mouth and your teeth and rinse with water, swooshing it around in your mouth. Evaluate the remaining samples, one at a time, waiting 30 sec. between each. Swallow the bread samples if possible.
8. When you have evaluated all the samples, move on to the MacBeth Booth in the lab to evaluate the appearance of some bread slices.

Appendix 16 - Visual evaluation instruction sheet

RICE BREAD VISUAL EVALUATIONS

1. View the visual reference card which illustrates the line scale anchor points for top crust color, crumb color, predominant cell size, cell size uniformity and cell wall thickness.
2. Visually evaluate the bread slices in the order indicated on your ballots.
3. Bring the first sample to the position in the booth marked by a black circle. Remove the lid of the petri-dish to view the sample, leaving the dish flat on the black circle. Try not to touch the bread sample as it may crumble. Evaluate the top crust color, crumb color, predominant cell size, cell size uniformity and cell wall thickness of the sample.

PREDOMINANT CELL SIZE: The cell size that makes up the largest percentage of the slice.

CELL SIZE UNIFORMITY: The cell size uniformity is dependent on the number of different cell sizes present within the bread slice. If the cells are all the same size the slice is very uniform. As the number of different cell sizes within a sample increases, the sample becomes more irregular.

CELL WALL THICKNESS: If a sample is so compact that the cell walls are indistinguishable (you cannot tell if they are very thick or very thin), score the cell wall thickness at the right endpoint of the scale (15).

4. Please comment on any irregularities in the bread slices or about any difficulties you may have had evaluating the characteristics.
5. Replace the lid on the petri-dish, so the samples do not dry out and return the sample to its original position in the booth. Move the second sample to the viewing position and evaluate it, continuing until all the samples have been visually evaluated.

Appendix 17 - Consumer evaluation questionnaire

Rice Bread Consumer Panel

1. Are you on a special diet? Yes _____ No _____

If yes, please specify. Celiac _____ Other _____

2. Do you regularly eat gluten-free bread? Yes _____ No _____

If yes, how many slices of bread would you eat per day?

Sex: Male _____

Female _____

Age (years): under 18 _____

19-25 _____

26-35 _____

36-50 _____

51-65 _____

over 65 _____

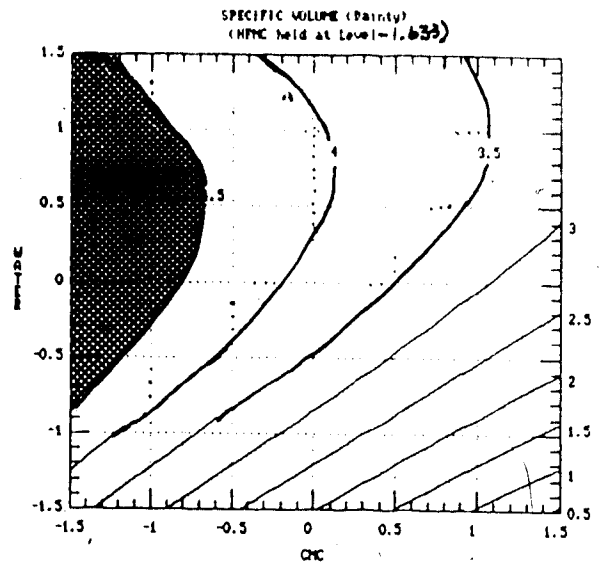
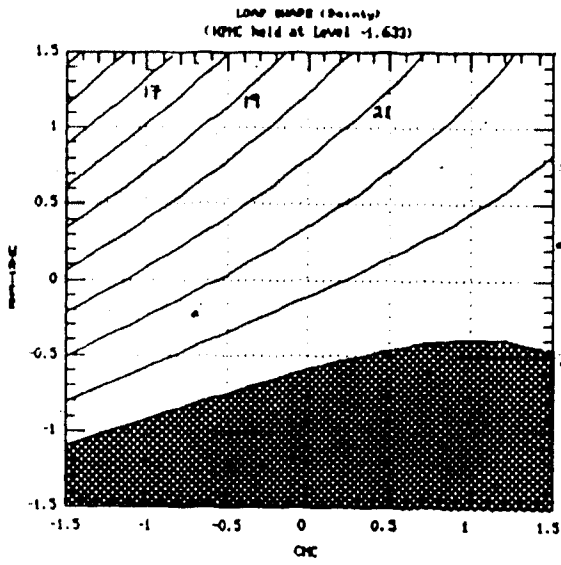
COMMENTS:

Appendix 18 - Contour plots for Flour A rice bread objective responses. Levels of HPMC, water and CMC are coded values. The shaded regions met reference standards.

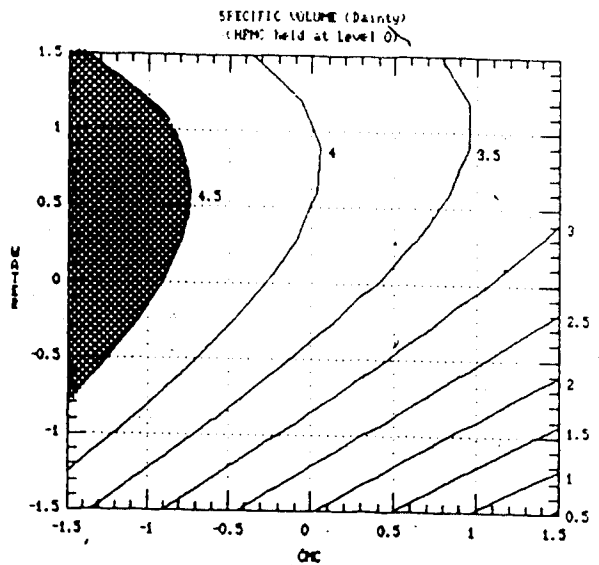
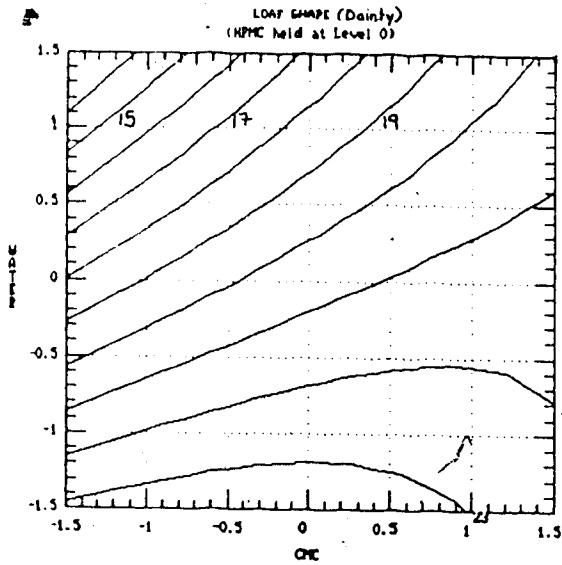
LOAF SHAPE

SPECIFIC VOLUME

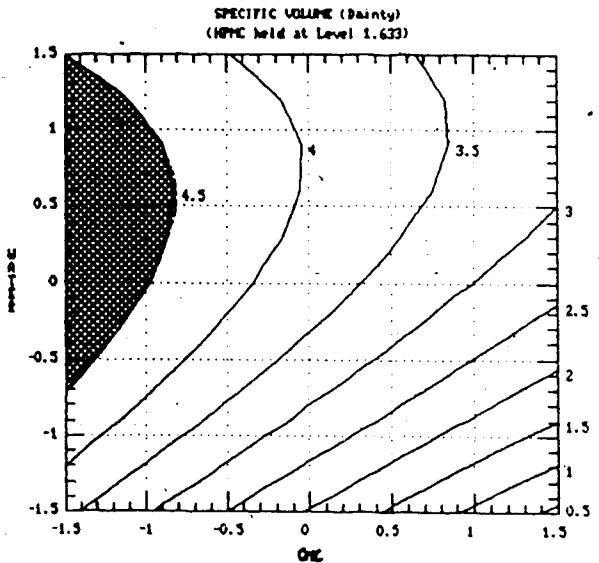
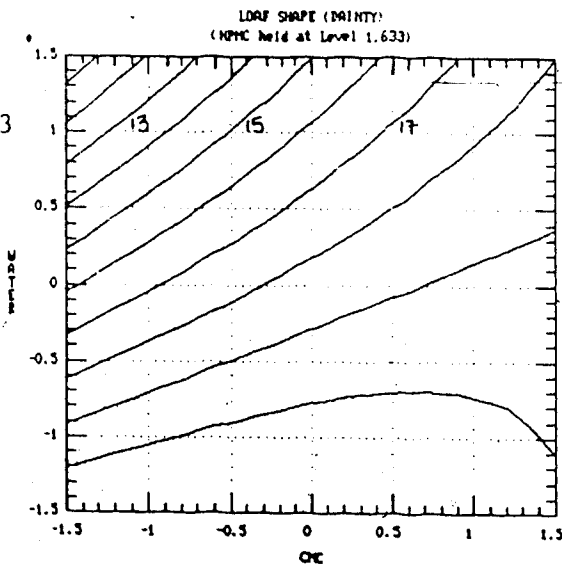
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HPMC



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HPMC



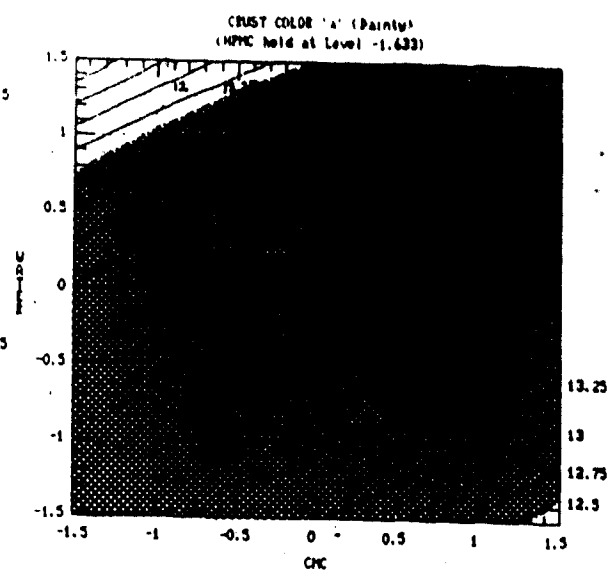
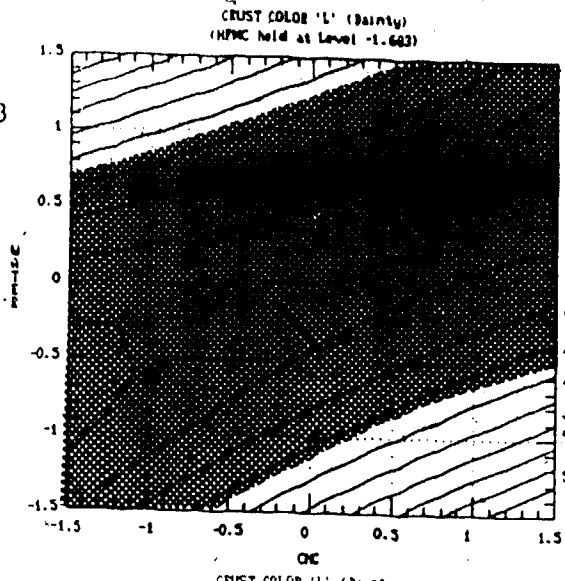
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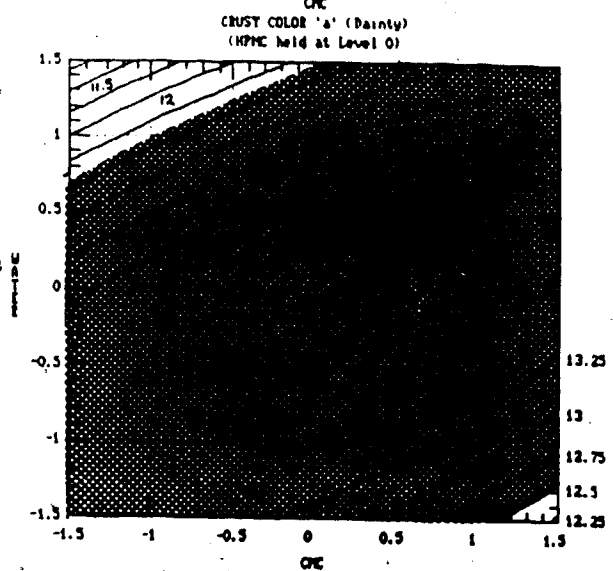
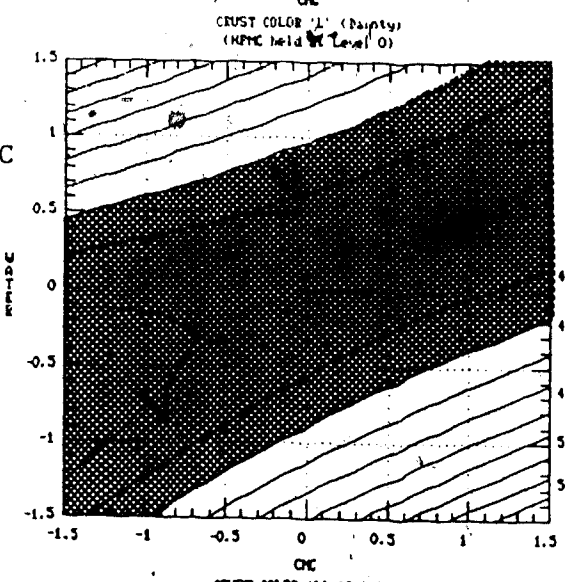
CRUST COLOR "L"

CRUST COLOR "a"

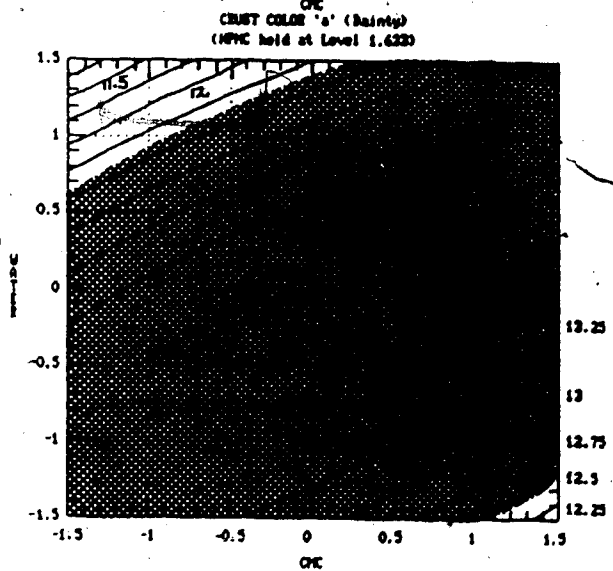
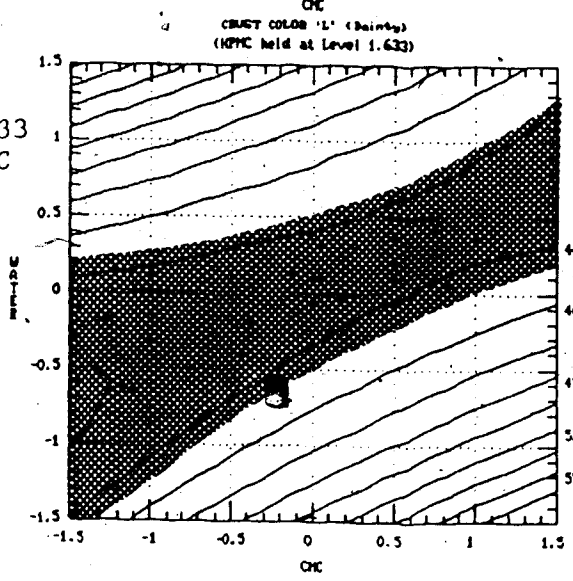
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HPMC



0
HPMC



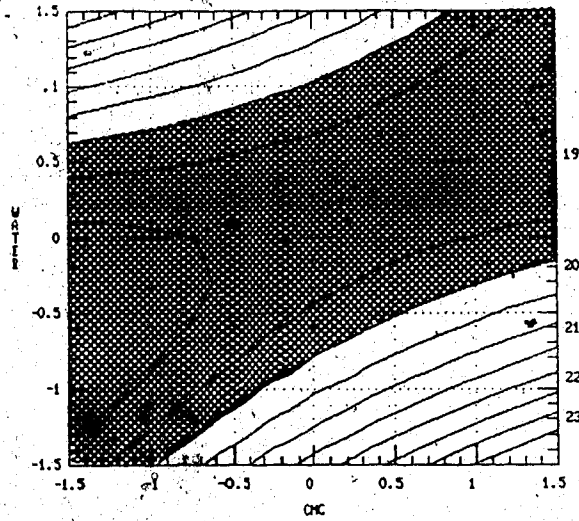
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HPMC



CRUST COLOR "b"

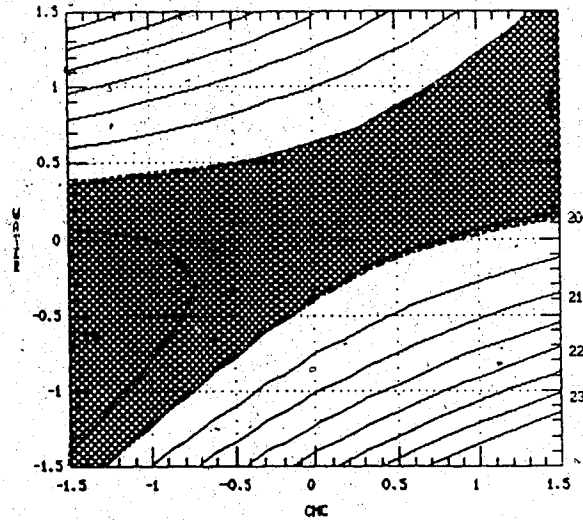
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(HPMC held at Level -1.633)

-1.633
HPMC



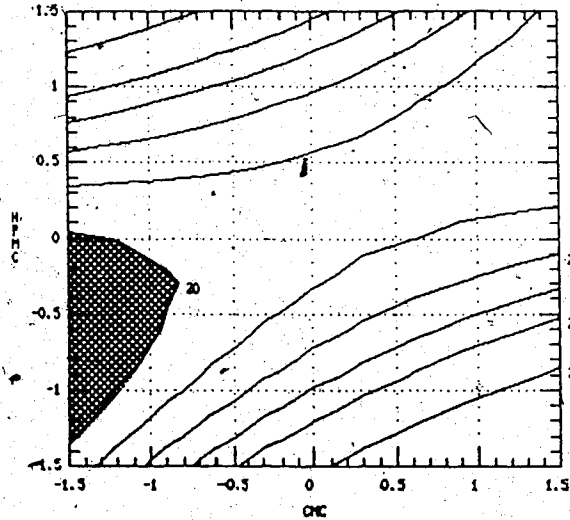
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(HPMC held at Level 0)

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HPMC



CRUST COLOR "b" (Dainty)
(HPMC held at Level 1.633)

1.633
HPMC

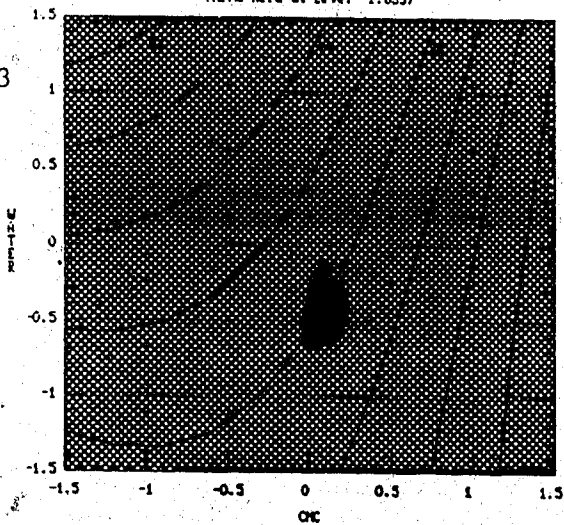


CRUMB COLOR "L"

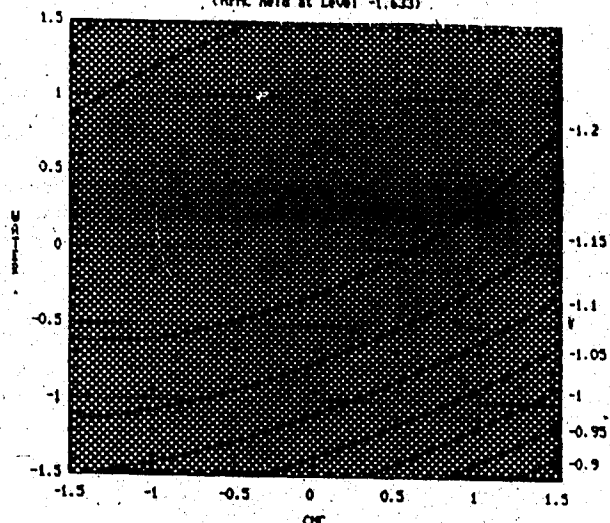
CRUMB COLOR "a"

-1.633
HPMC

CRUMB COLOR 'L' (Dainty)
(HPMC held at Level -1.633)

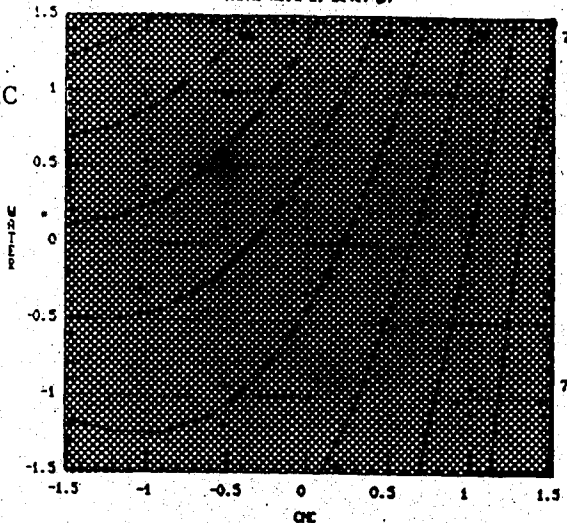


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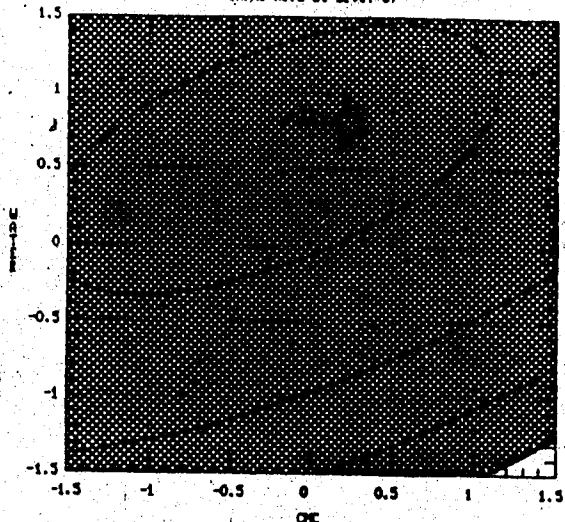


0
HPMC

CRUMB COLOR 'L' (Dainty)
(HPMC held at Level 0)

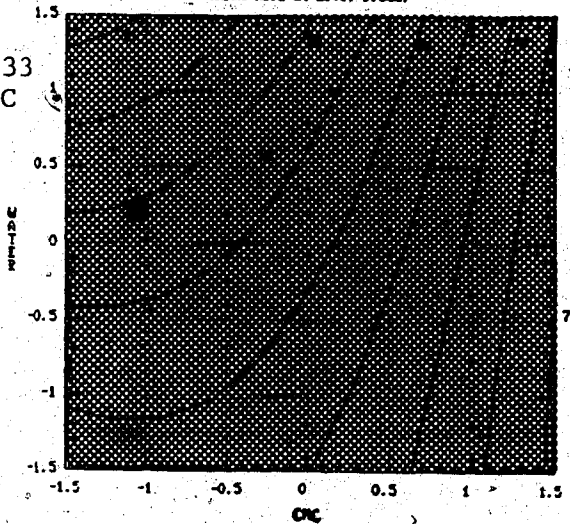


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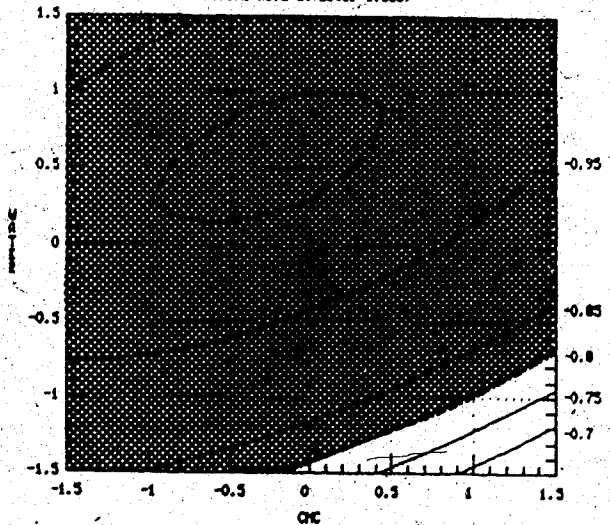


1.633
HPMC

CRUMB COLOR 'L' (Dainty)
(HPMC held at Level 1.633)

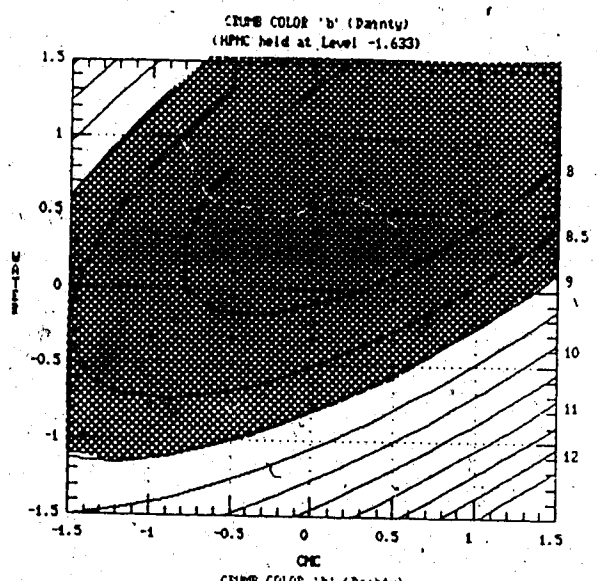


CRUMB COLOR 'a' (Dainty)
(HPMC held at Level 1.633)

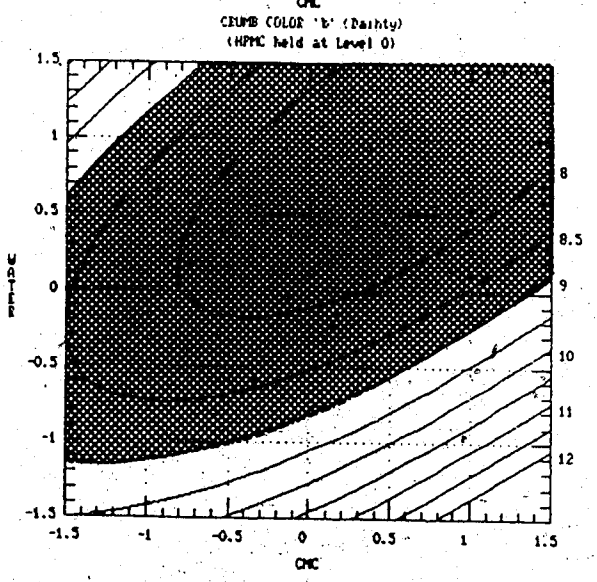


CRUMB COLOR "b"

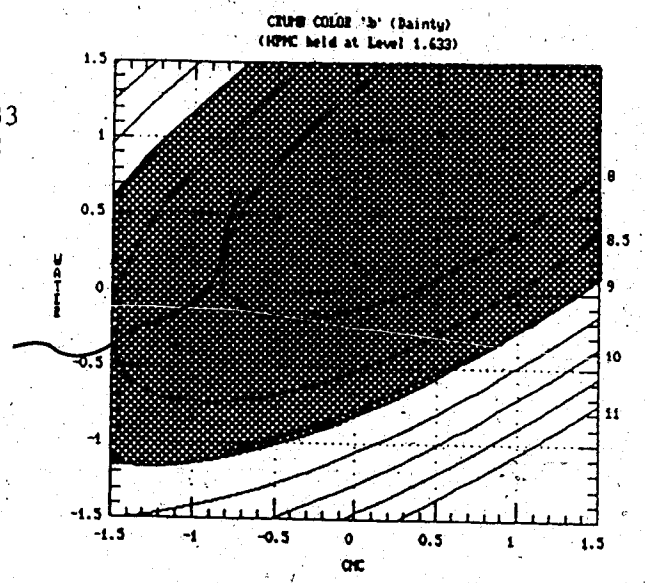
-1.633
HPMC



0
HPMC



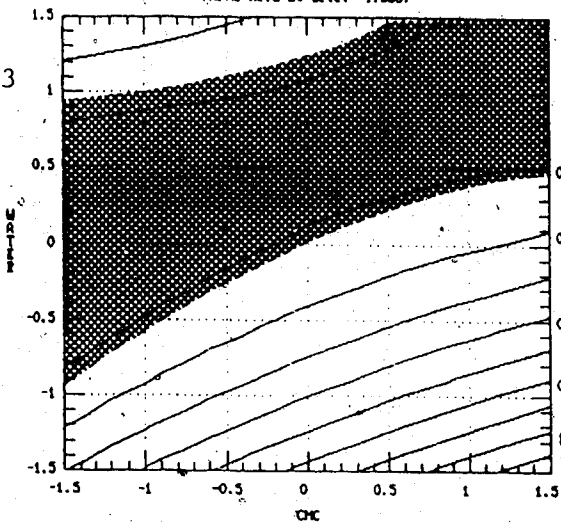
1.633
HPMC



INSTRON FIRMNESS

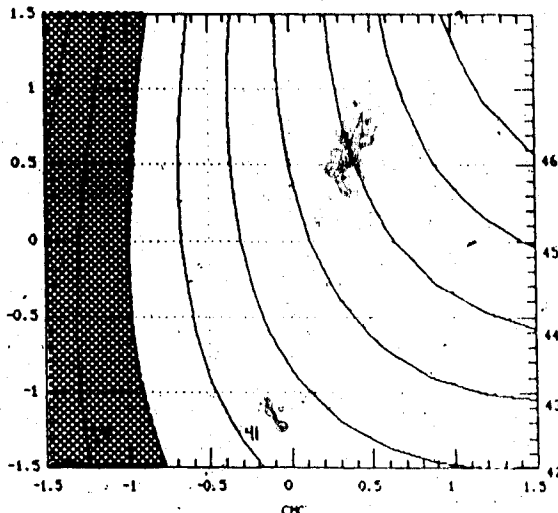
INSTRON (Dainty)
(HPMC held at Level -1.633)

-1.633
HPMC



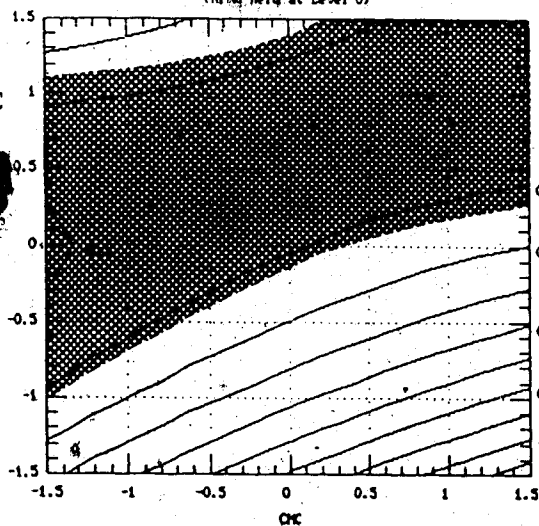
PERCENT MOISTURE

% MOISTURE (Dainty)
(HPMC held at Level -1.633)

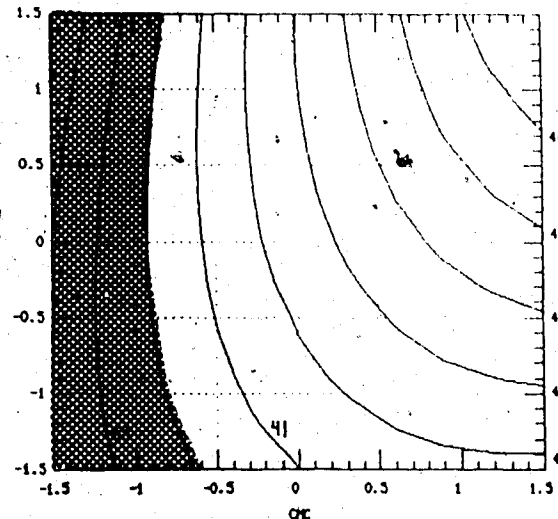


INSTRON (Dainty)
(HPMC held at Level 0)

0
HPMC

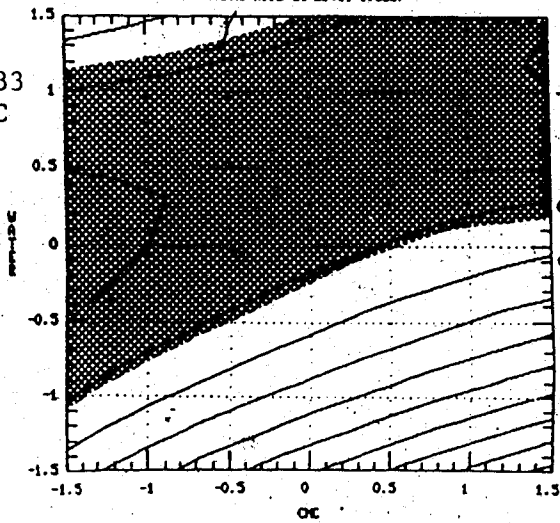


% MOISTURE (Dainty)
(HPMC held at Level 0)

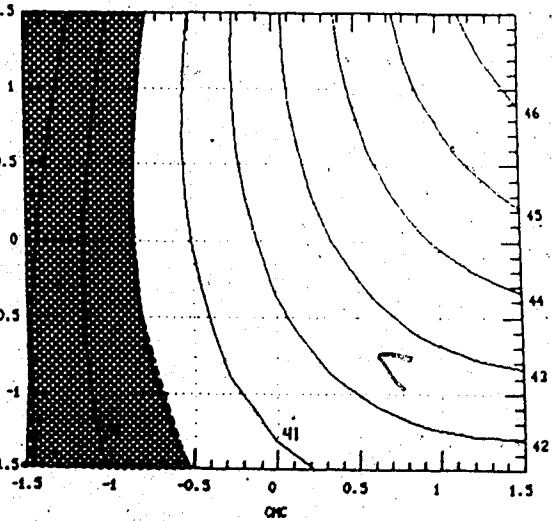


INSTRON (Dainty)
(HPMC held at Level 1.633)

1.633
HPMC



% MOISTURE (Dainty)
(HPMC held at Level 1.633)

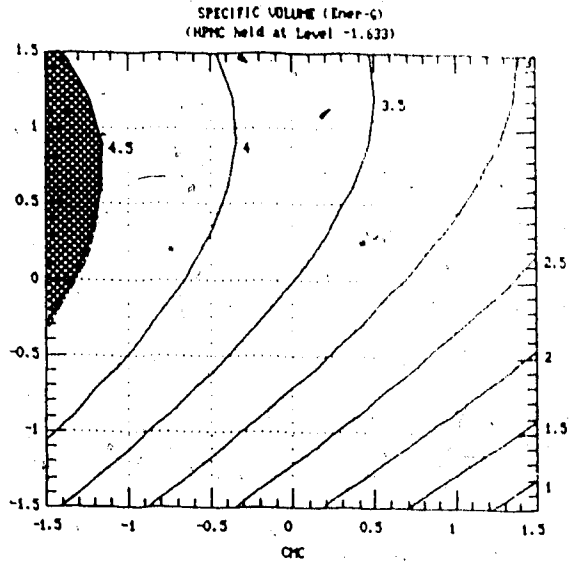
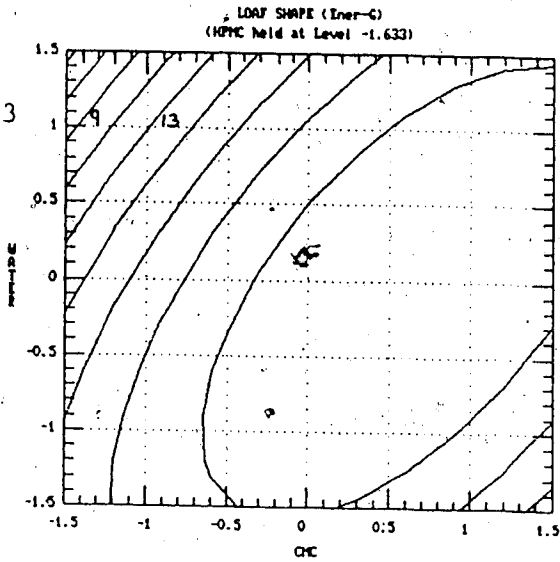


Appendix 19 - Contour plots for Flour B rice bread objective responses. Levels of HPMC, water and CMC are coded values. The shaded regions met reference standards.

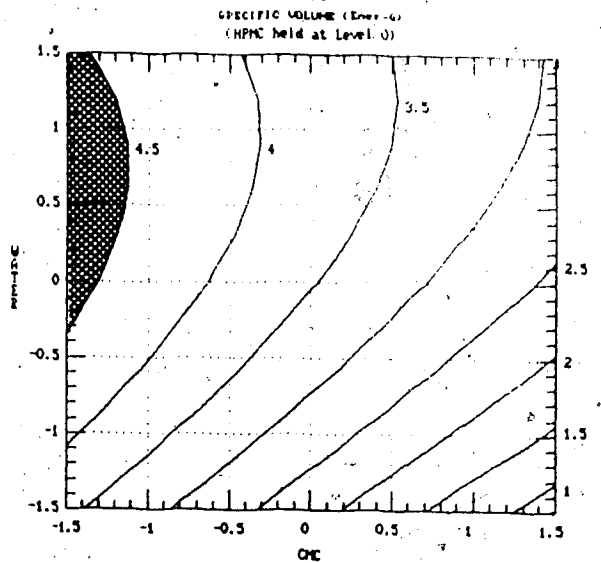
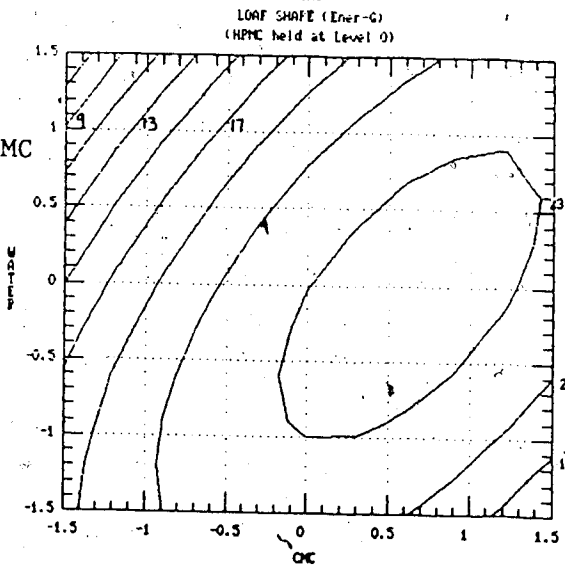
LOAF SHAPE

SPECIFIC VOLUME

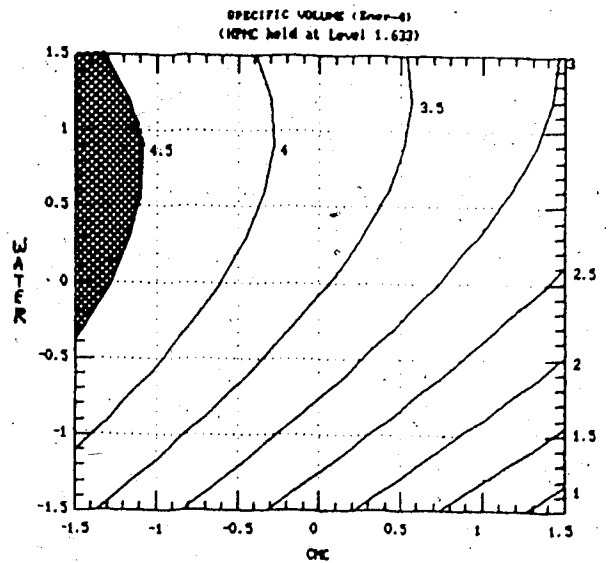
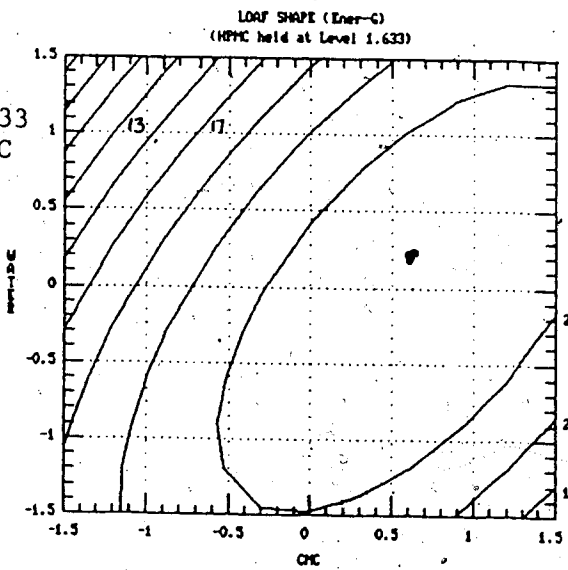
-1.633
HPMC



0
HPMC

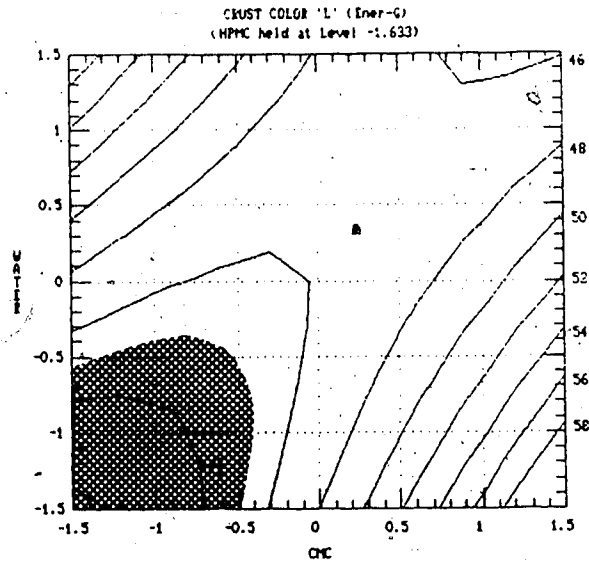


1.633
HPMC

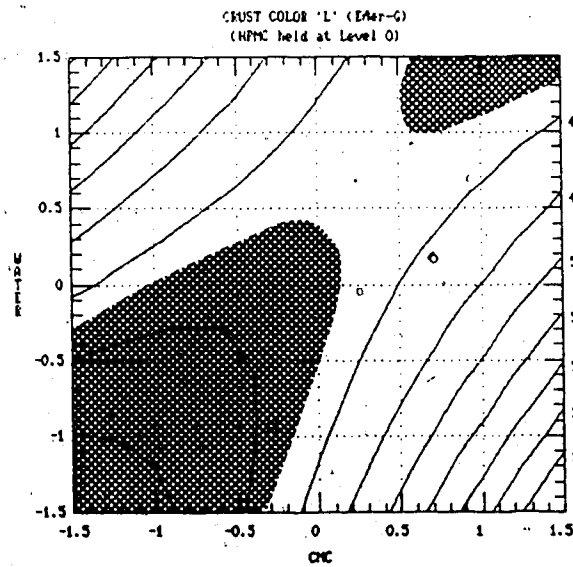


CRUST COLOR "L"

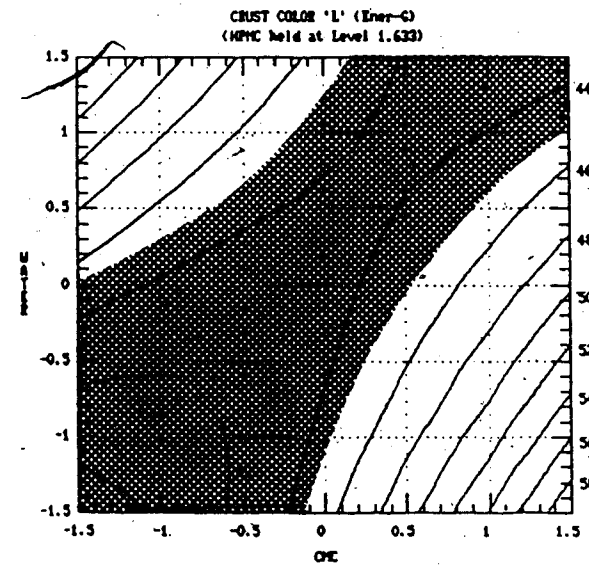
-1.633
HPMC



0
HPMC



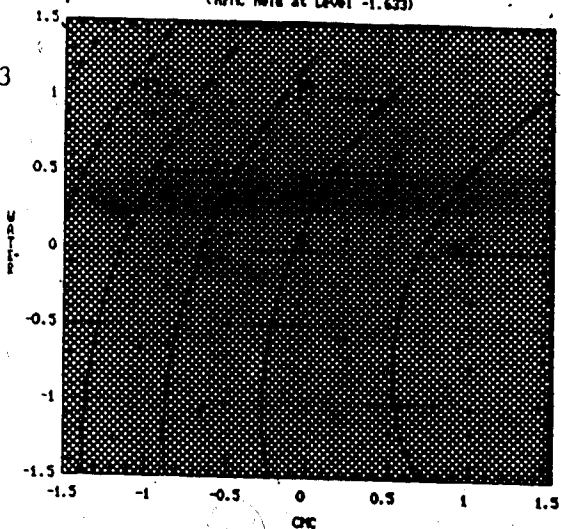
1.633
HPMC



CRUMB COLOR "L"

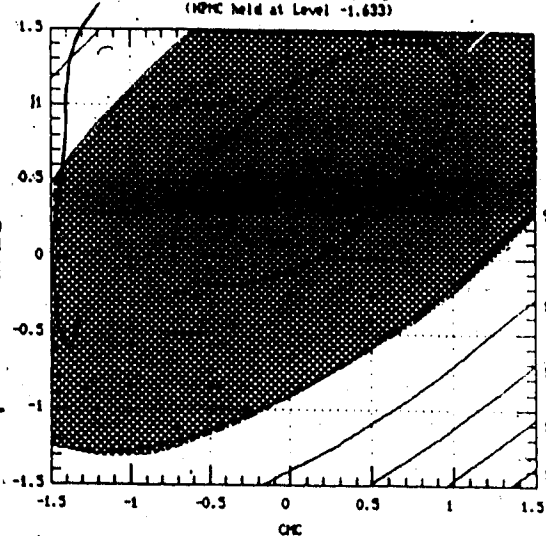
CRUMB COLOR 'L' (Ener-G)
(HPMC held at Level -1.633)

-1.633
HPMC



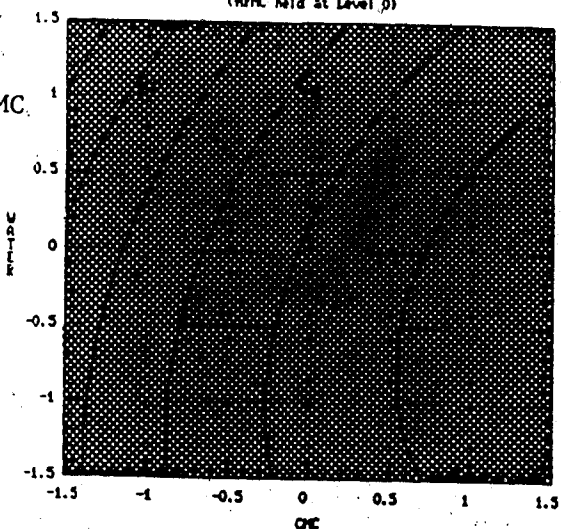
CRUMB COLOR "b"

CRUMB COLOR 'b' (Ener-G)
(HPMC held at Level -1.633)

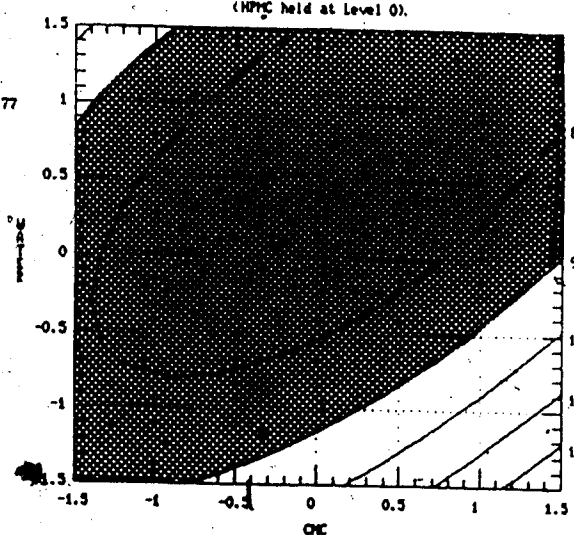


CRUMB COLOR 'L' (Ener-G)
(HPMC held at Level 0)

0
HPMC

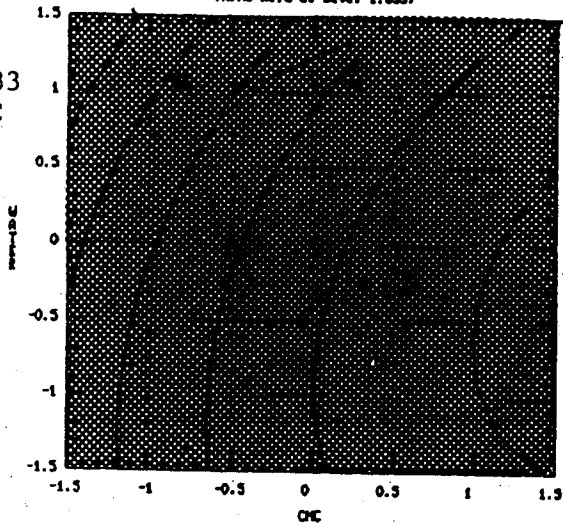


CRUMB COLOR 'b' (Ener-G)
(HPMC held at Level 0)

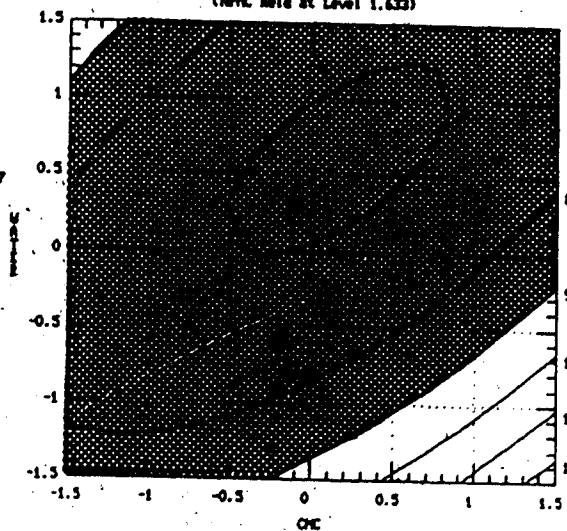


CRUMB COLOR 'L' (Ener-G)
(HPMC held at Level 1.633)

1.633
HPMC



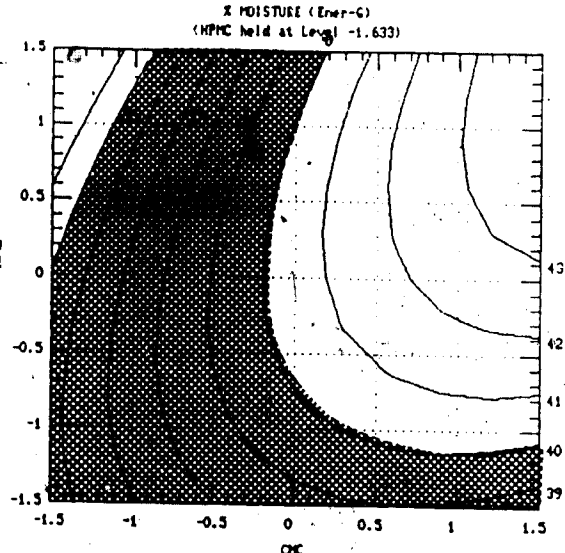
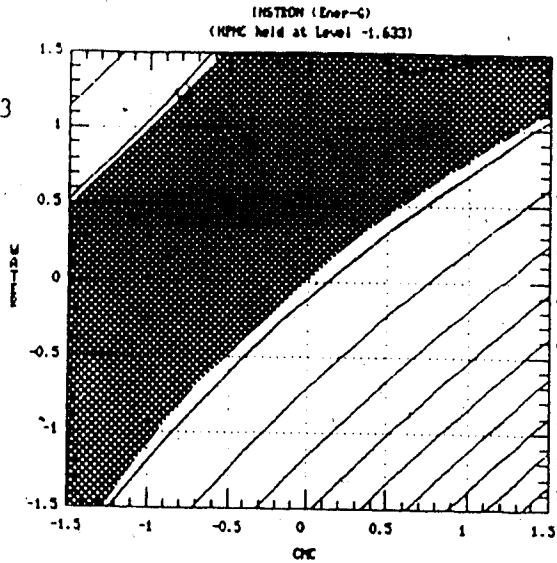
CRUMB COLOR 'b' (Ener-G)
(HPMC held at Level 1.633)



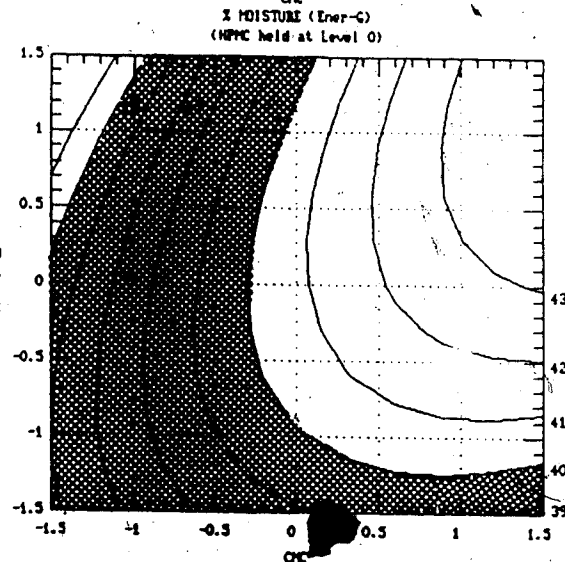
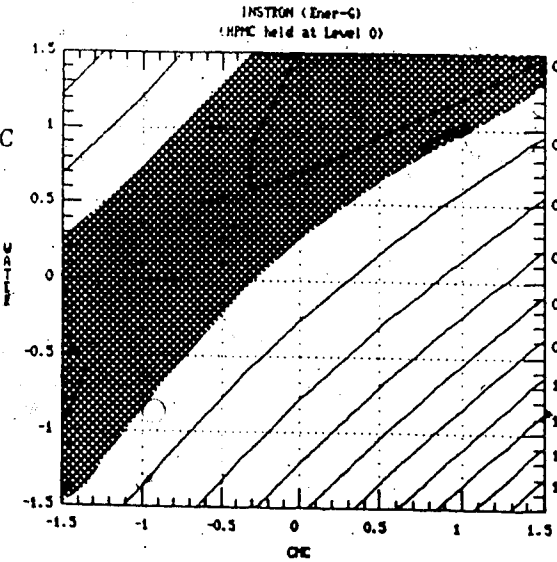
INSTRON FIRMNESS

PERCENT MOISTURE

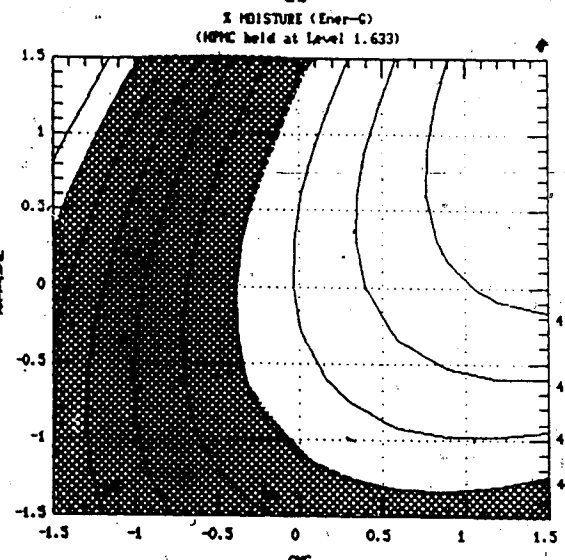
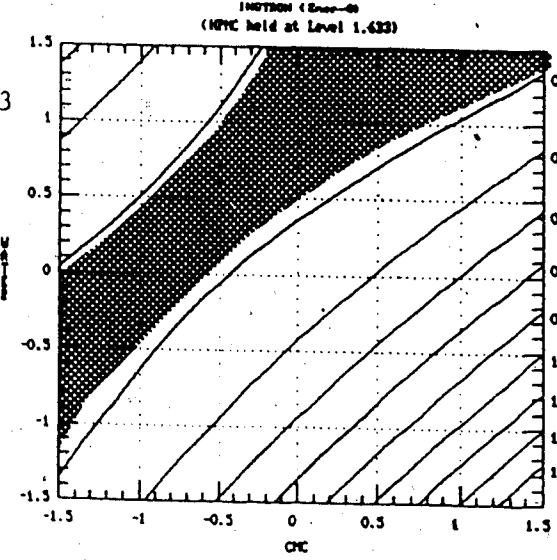
-1.633
HPMC



0
HPMC

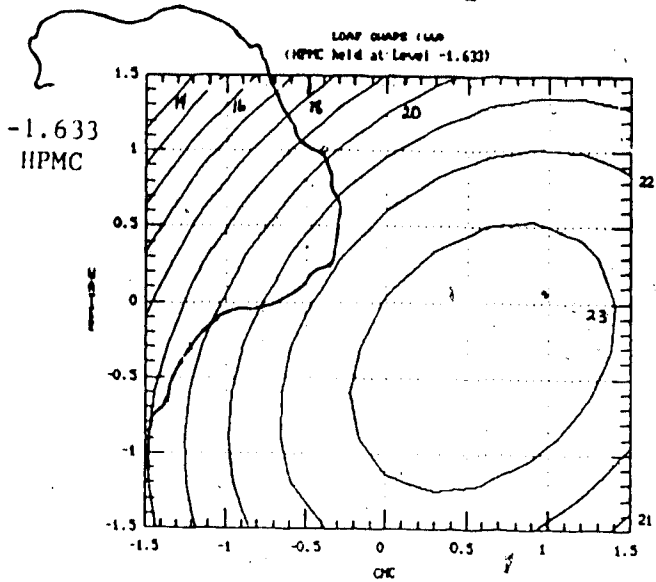


1.633
HPMC

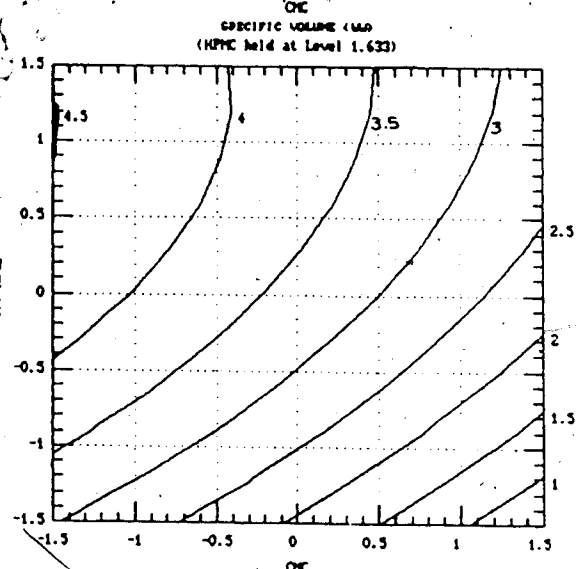
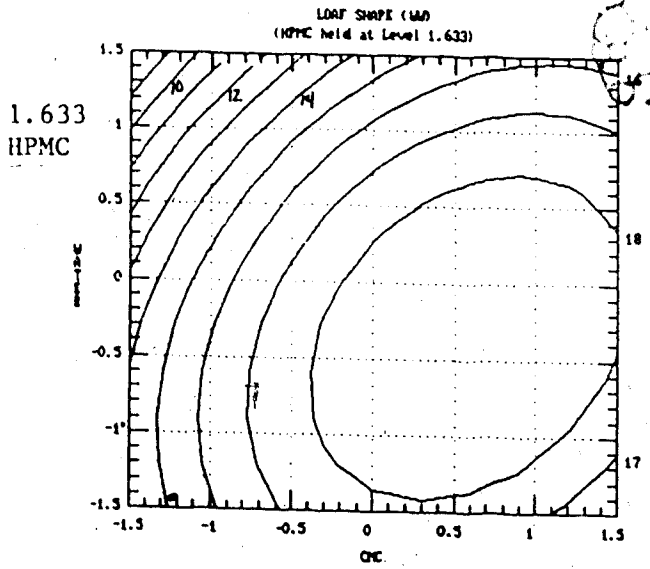
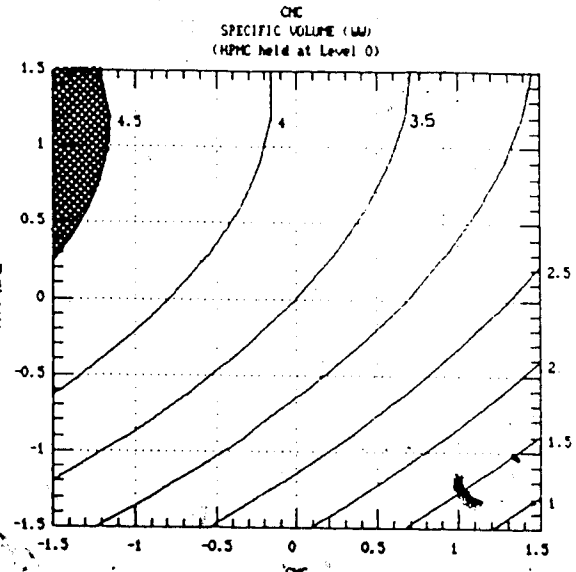
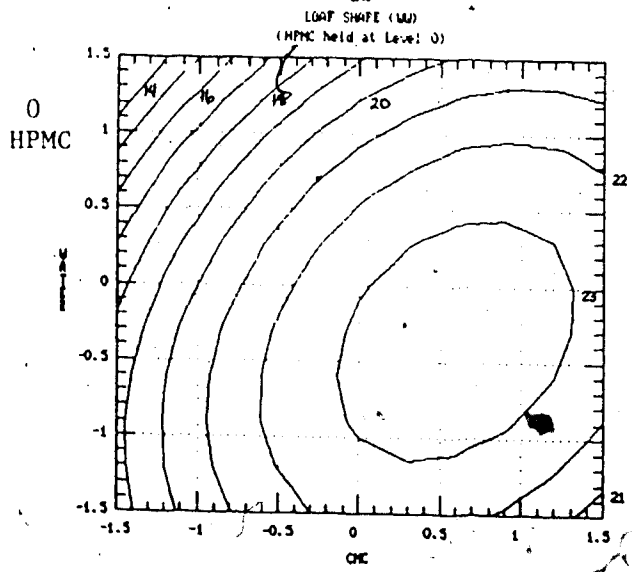
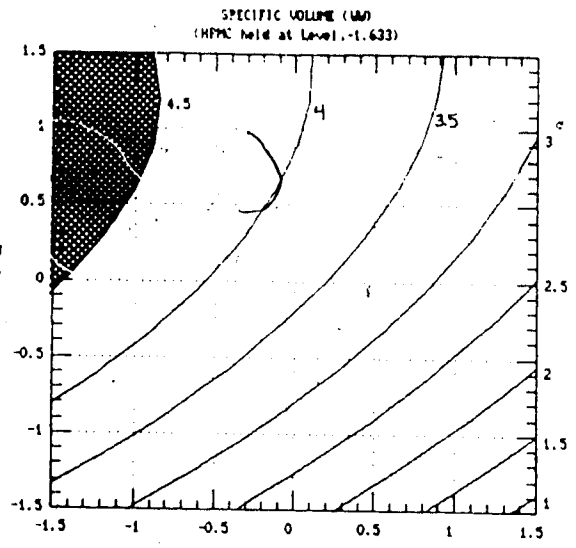


Appendix 20 - Contour plots for Flour C rice bread objective responses. Levels of HPMC, water and CMC are coded values. The shaded regions met reference standards.

LOAF SHAPE



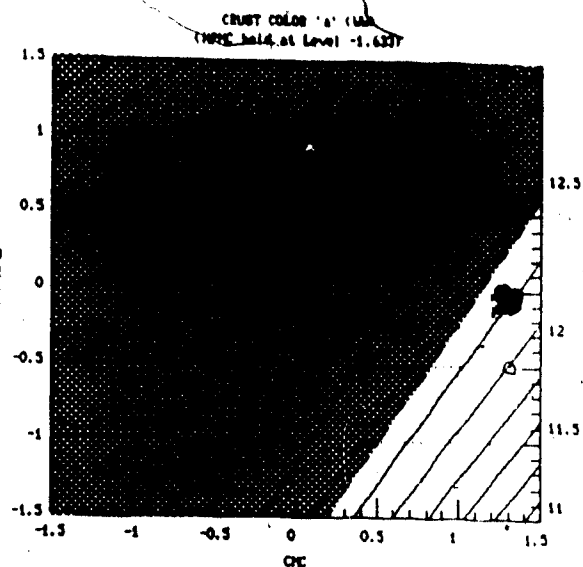
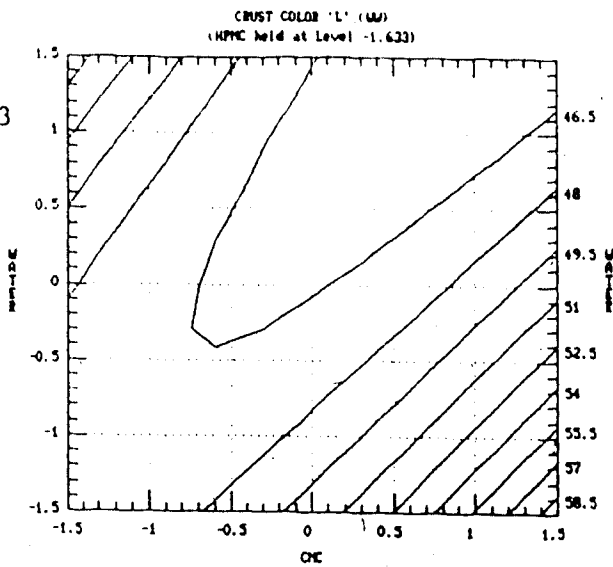
SPECIFIC VOLUME



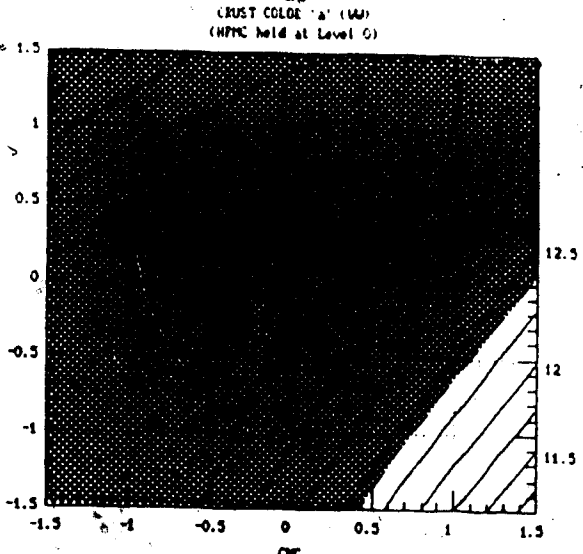
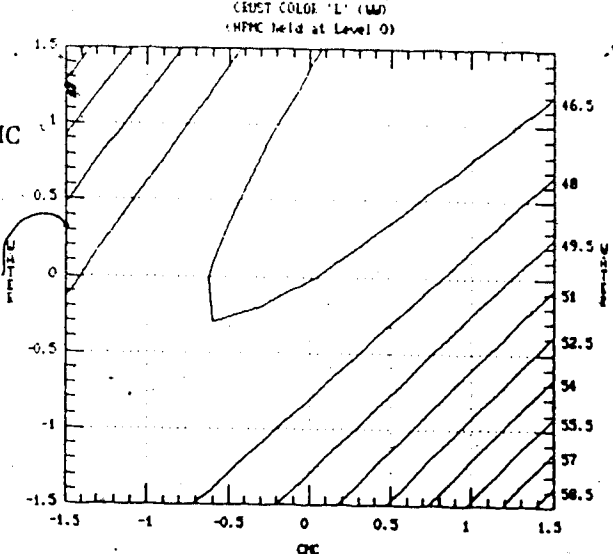
CRUST COLOR "L"

CRUST COLOR "a"

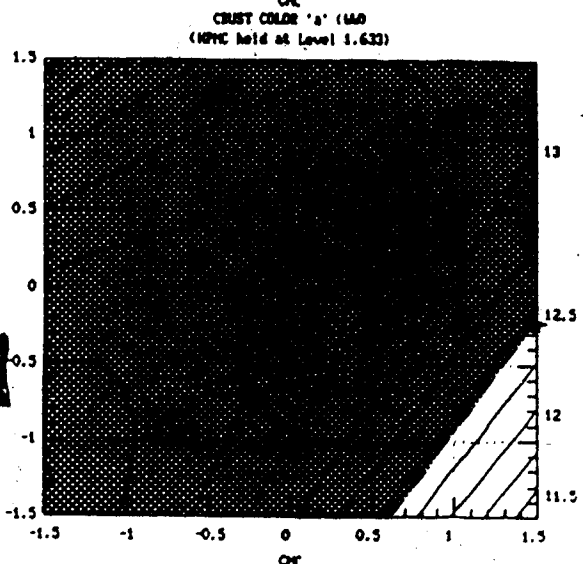
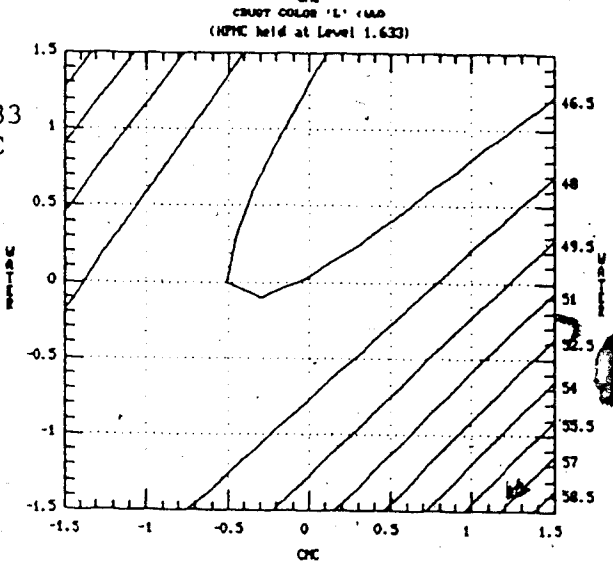
-1.633
HPMC



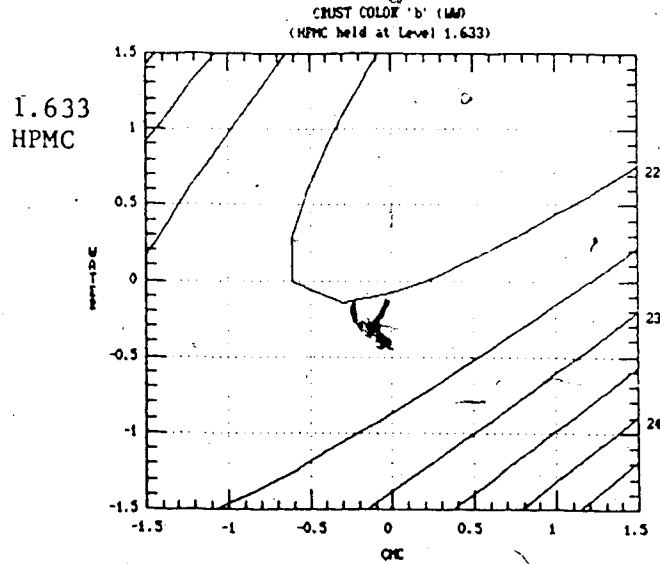
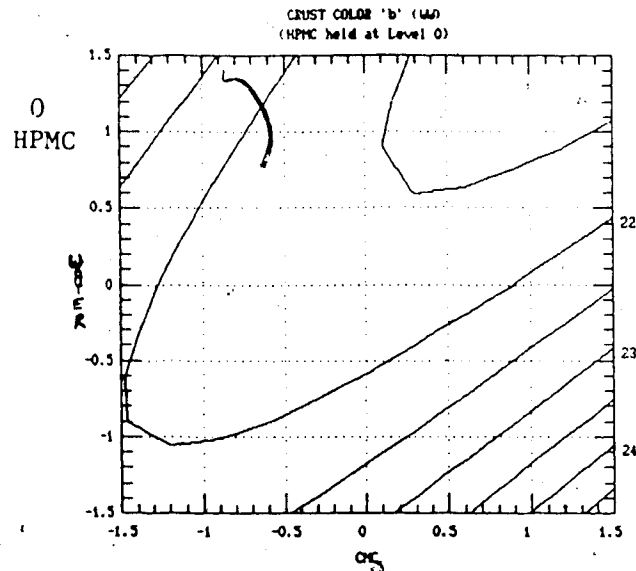
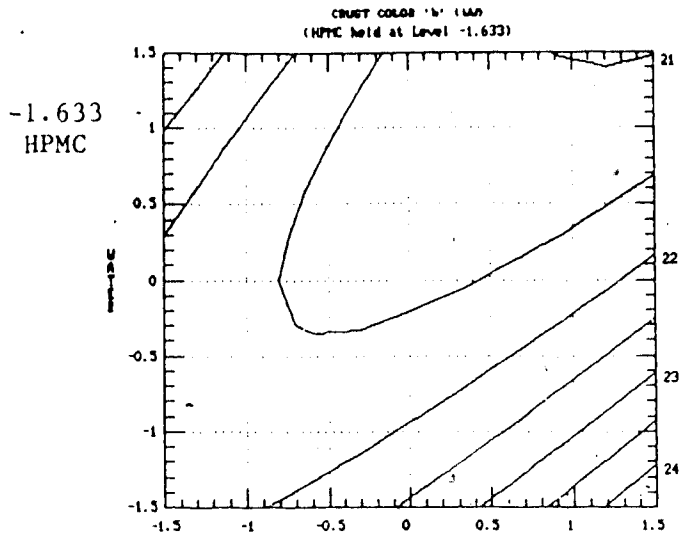
0
HPMC



1.633
HPMC

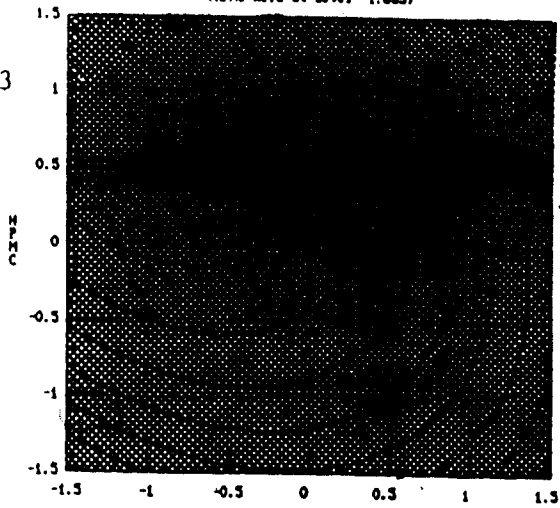


CRUST COLOR "b"



CRUMB COLOR "L"

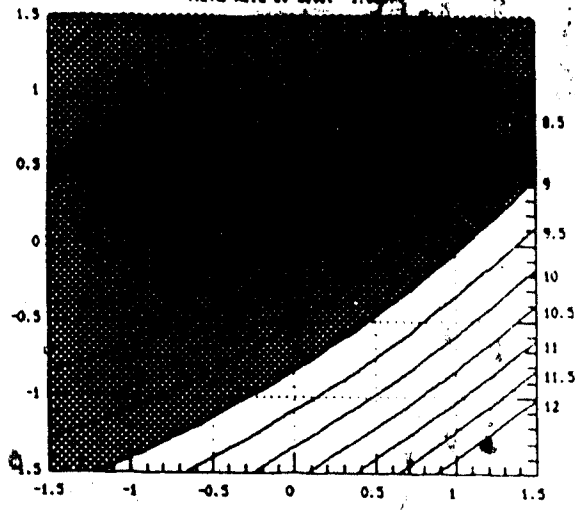
CRUMB COLOR 'L' (M) (HPMC held at Level -1.633)



-1.633
HPMC

CRUMB COLOR "b"

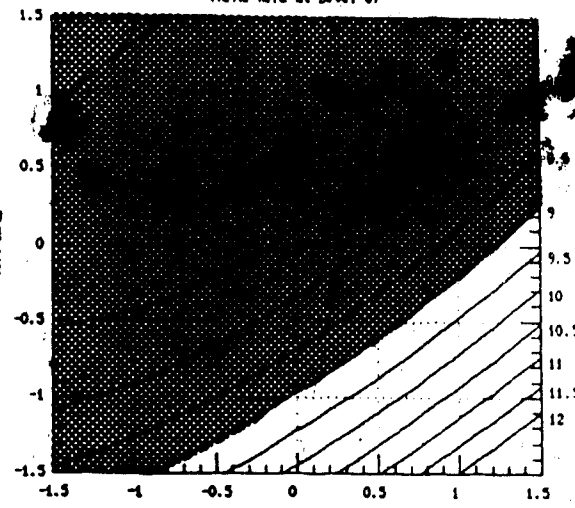
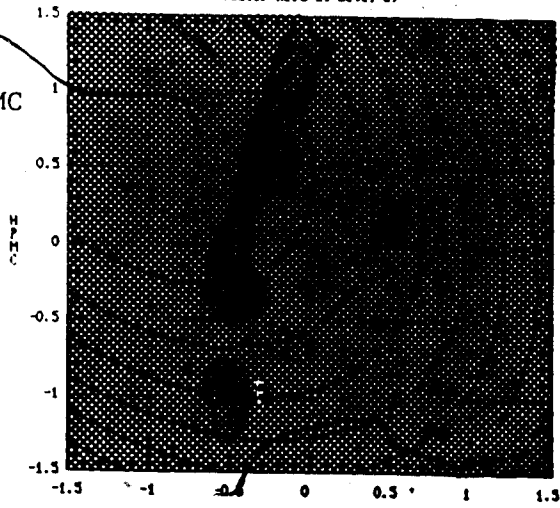
CRUMB COLOR 'b' (M) (HPMC held at Level -1.633)



CVC
CRUMB COLOR 'L' (M)
(Water held at Level 0)

CVC
CRUMB COLOR 'b' (M)
(HPMC held at Level 0)

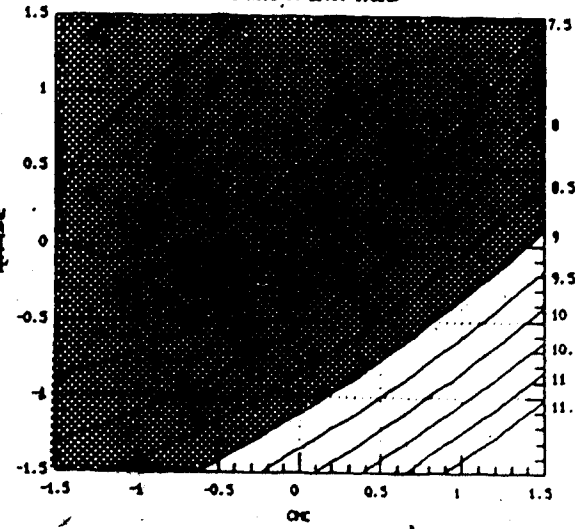
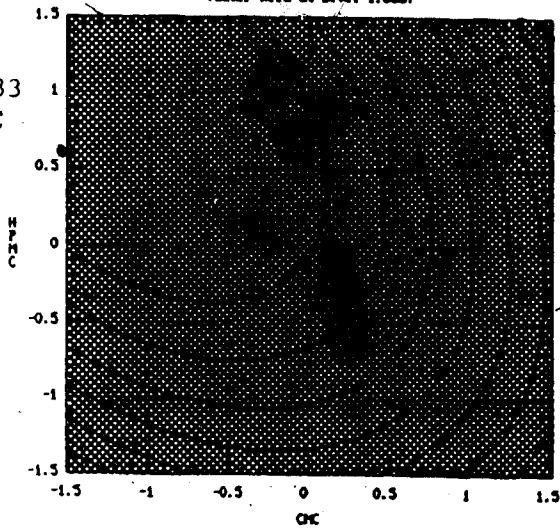
0
HPMC



CVC
CRUMB COLOR 'L' (M)
(Water held at Level 1.633)

CVC
CRUMB COLOR 'b' (M)
(HPMC held at Level 1.633)

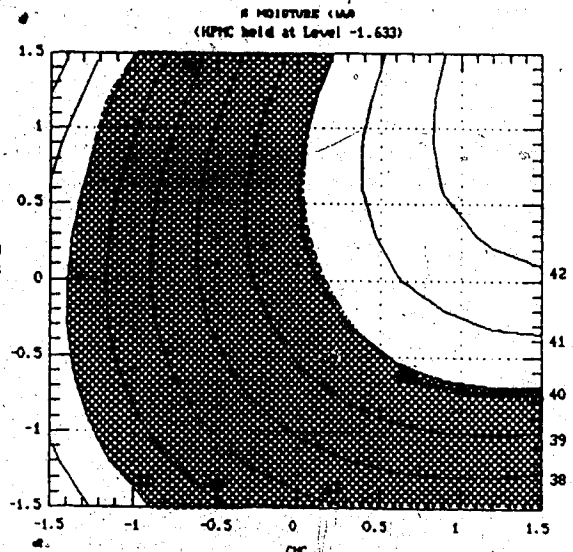
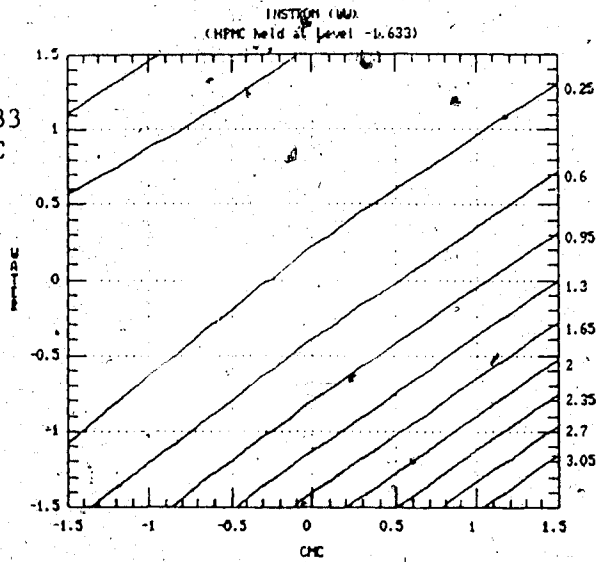
1.633
HPMC



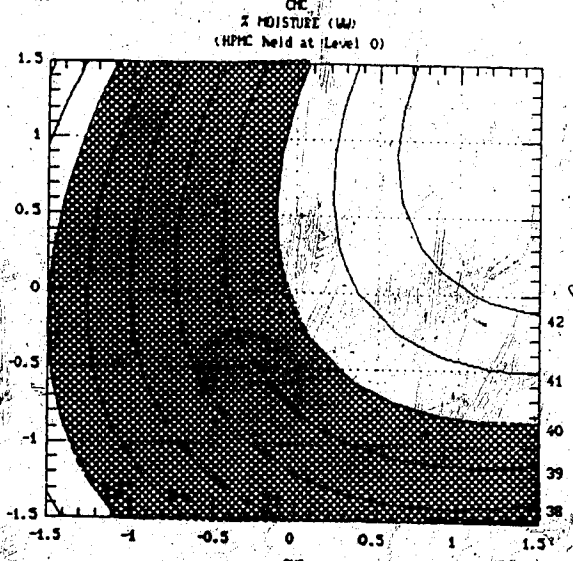
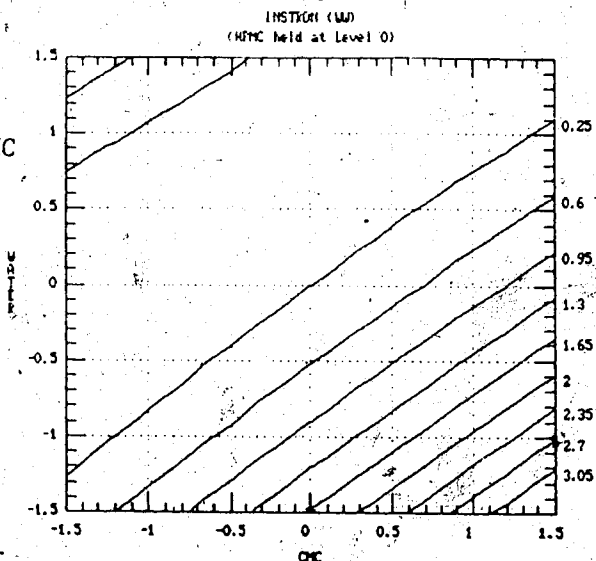
INSTRON FIRMNESS

PERCENT MOISTURE

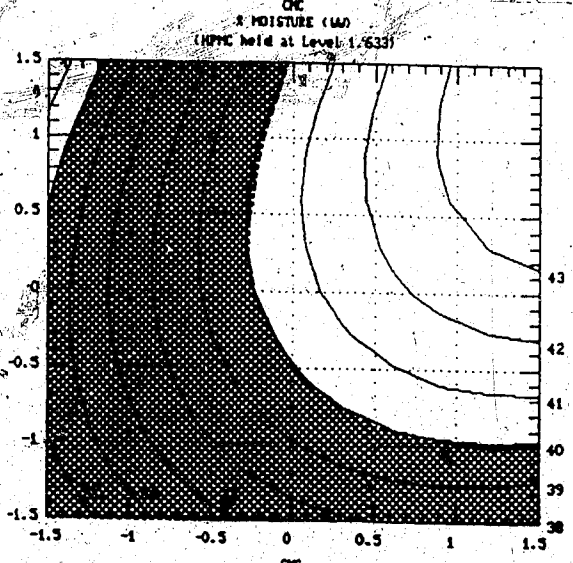
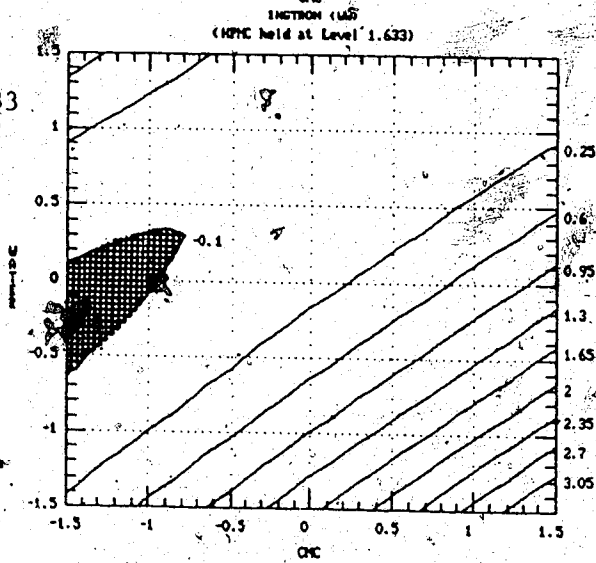
-1.633
HPMC



0
HPMC



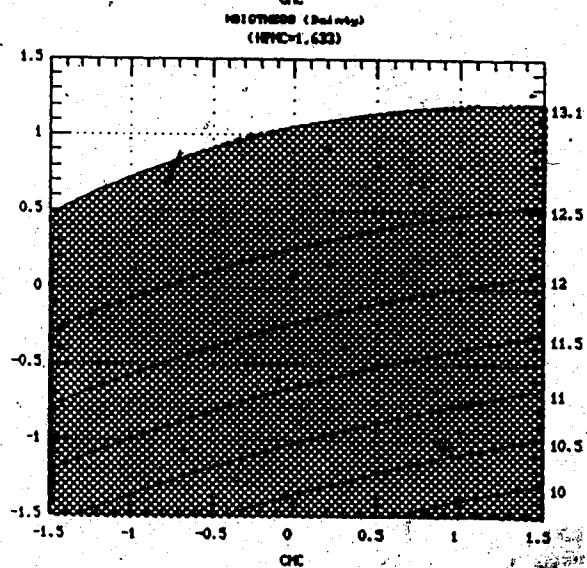
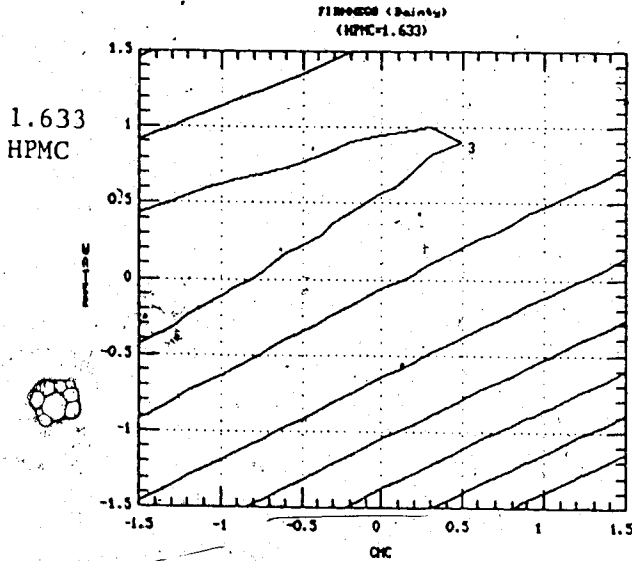
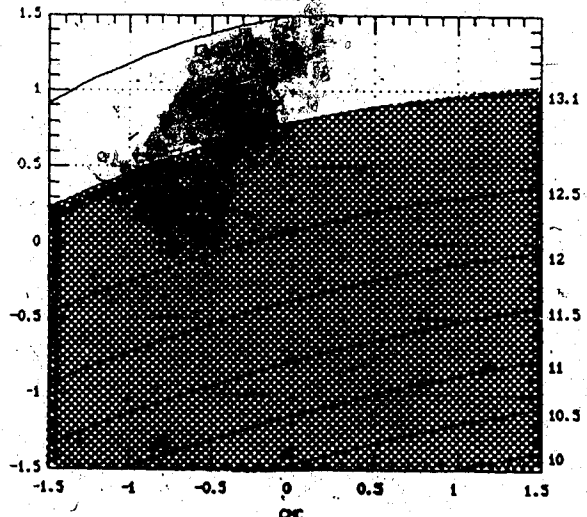
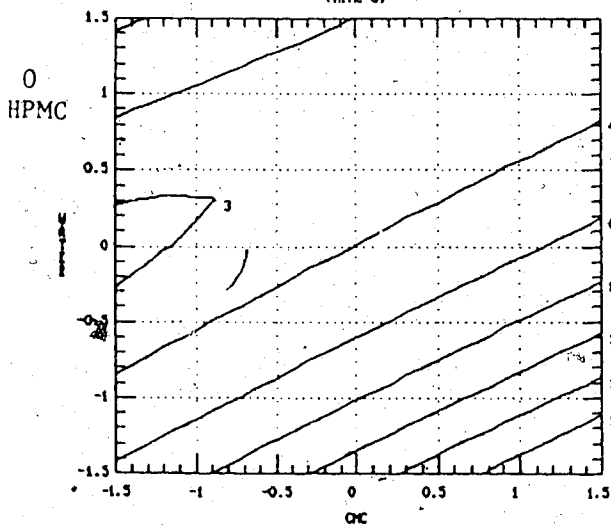
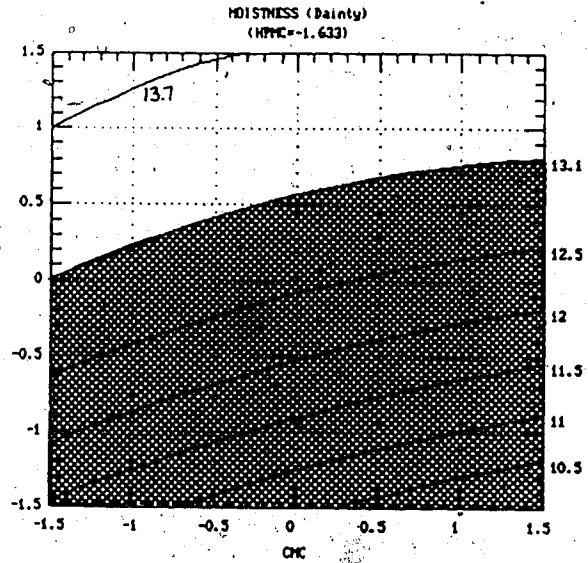
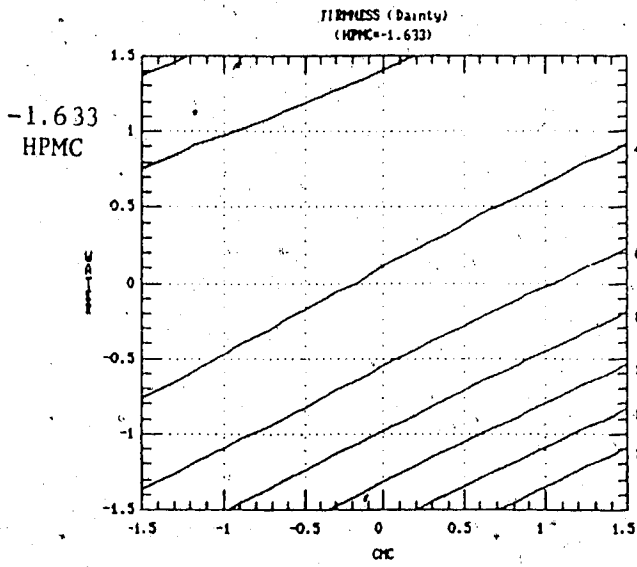
1.633
HPMC



Appendix 21 - Contour plots for Flour A rice bread sensory responses.
Levels of HPMC, water and CMC are coded values. The shaded regions met reference standards.

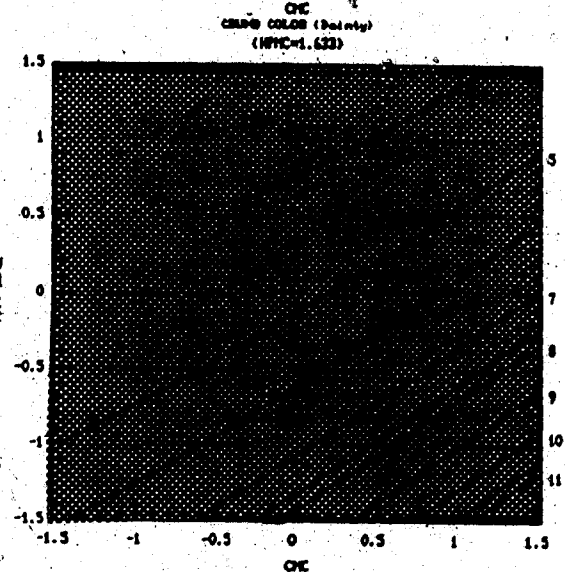
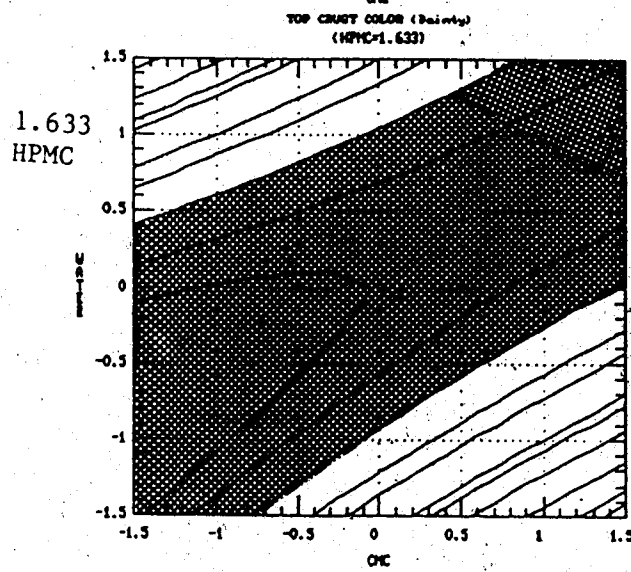
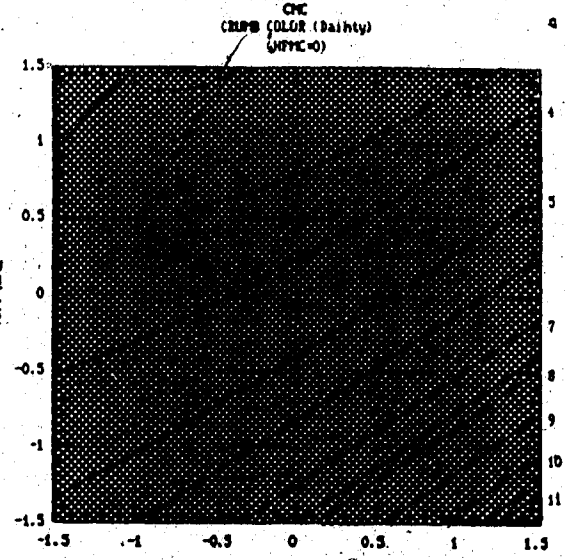
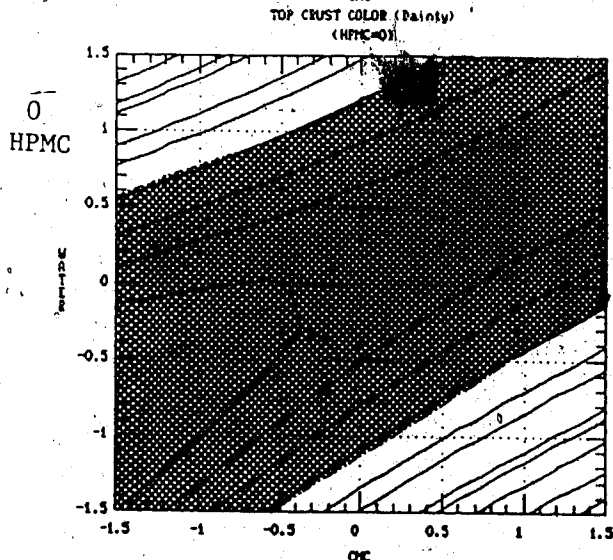
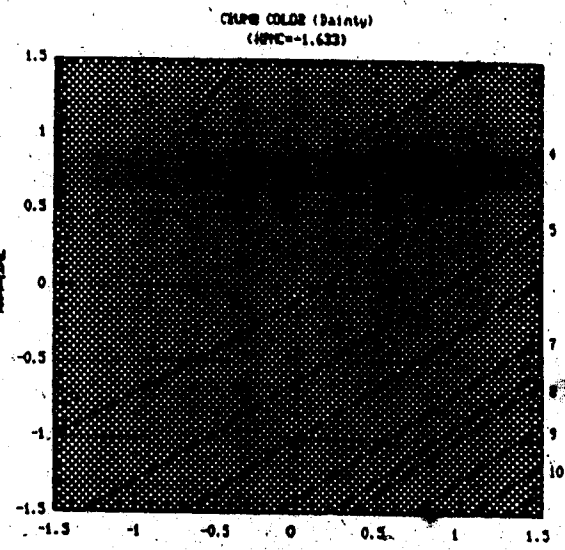
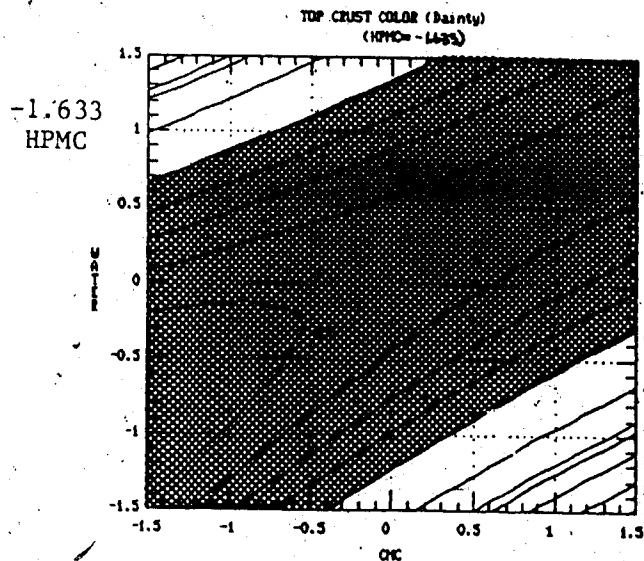
FIRMNESS

MOISTNESS



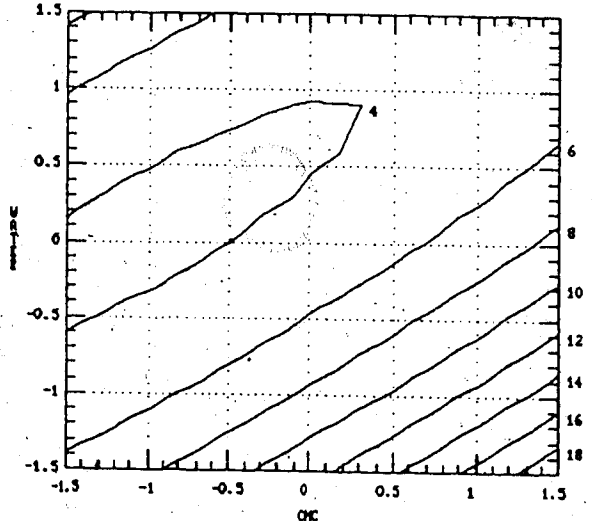
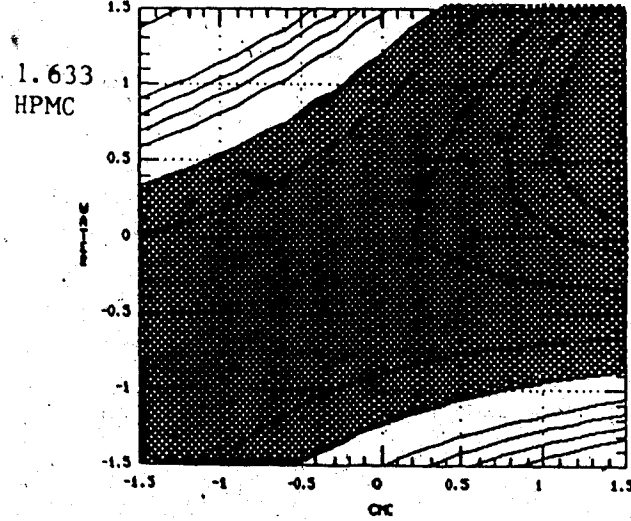
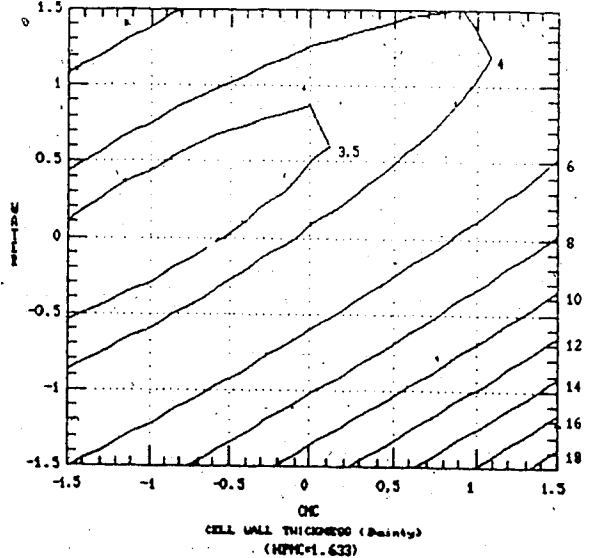
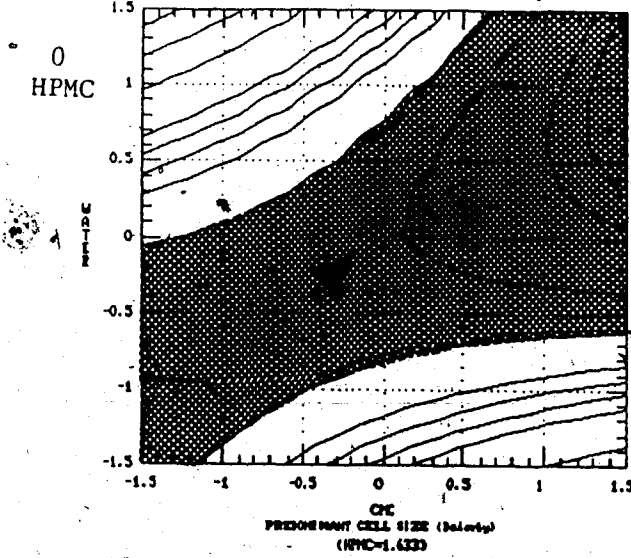
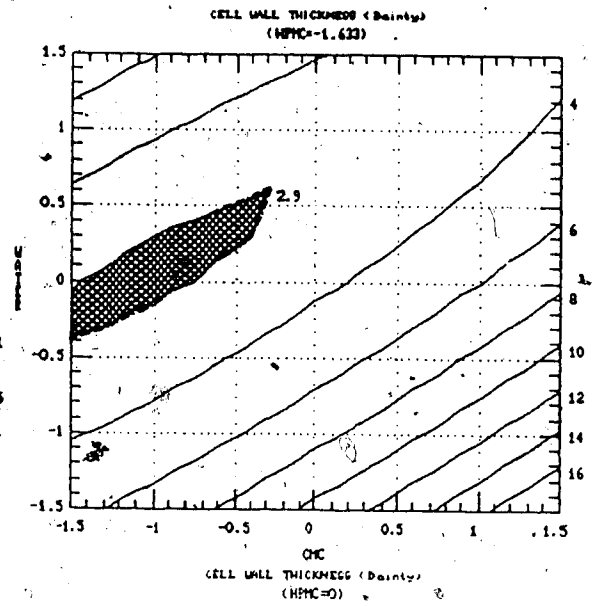
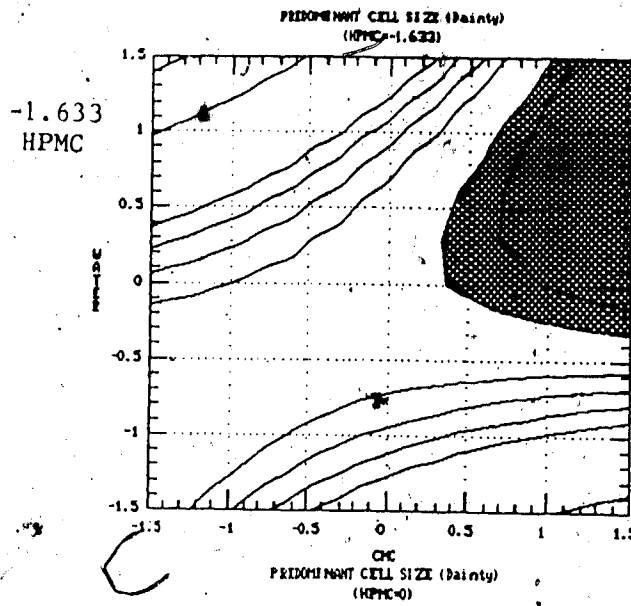
TOP CRUST COLOR

CRUMB COLOR



PREDOMINANT CELL SIZE

CELL WALL THICKNESS

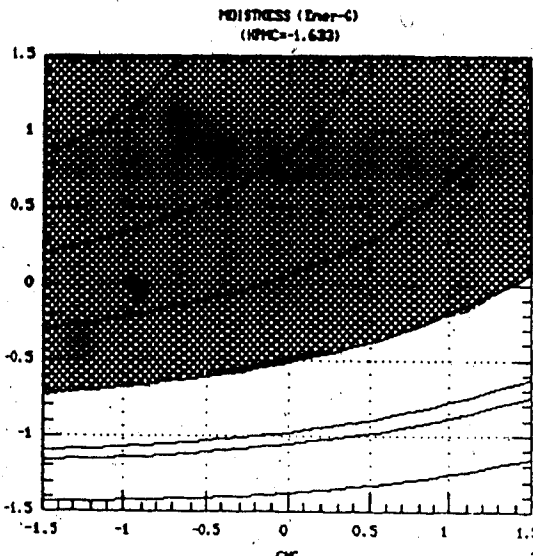
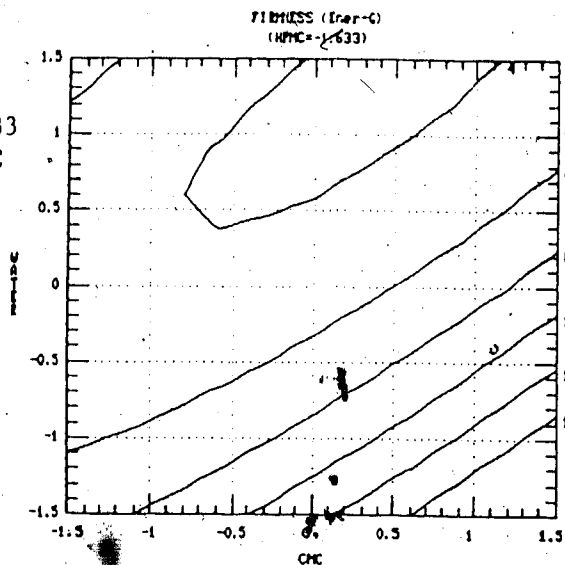


Appendix 22 - Contour plots for Flour B rice bread sensory responses.
Levels of HPMC, water and CMC are coded values. The shaded regions met reference standards.

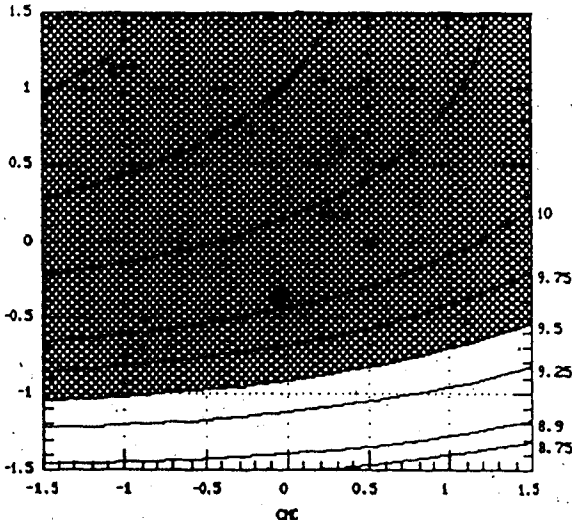
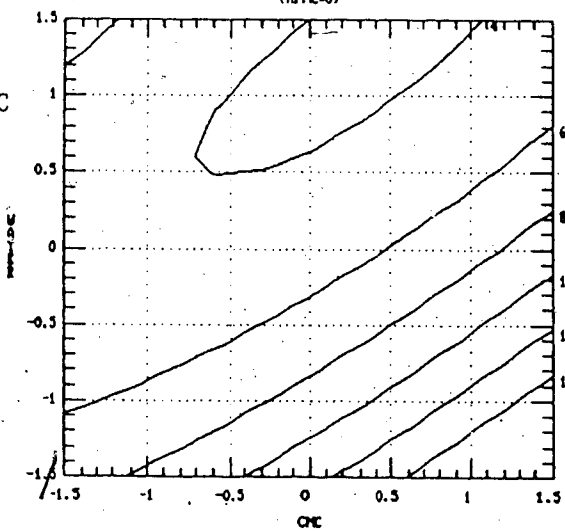
FIRMNESS

MOISTNESS

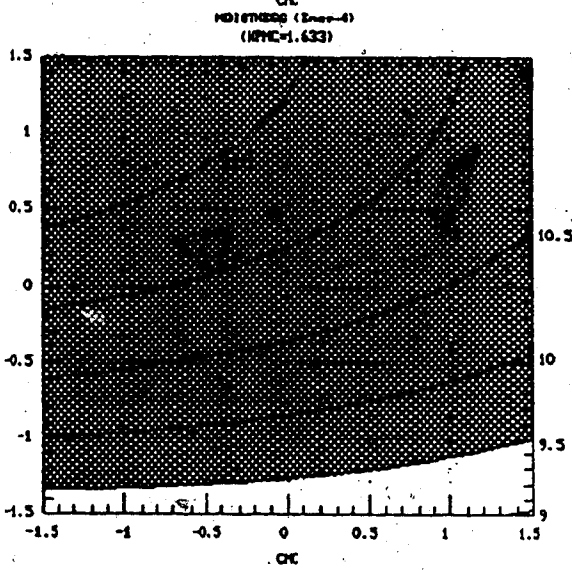
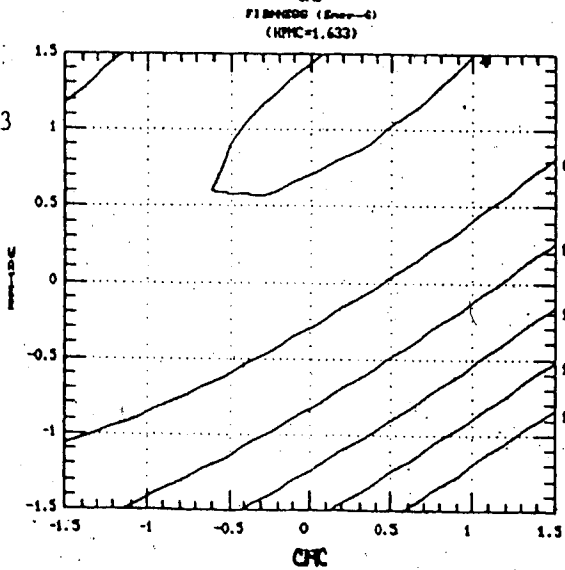
-1.633
HPMC



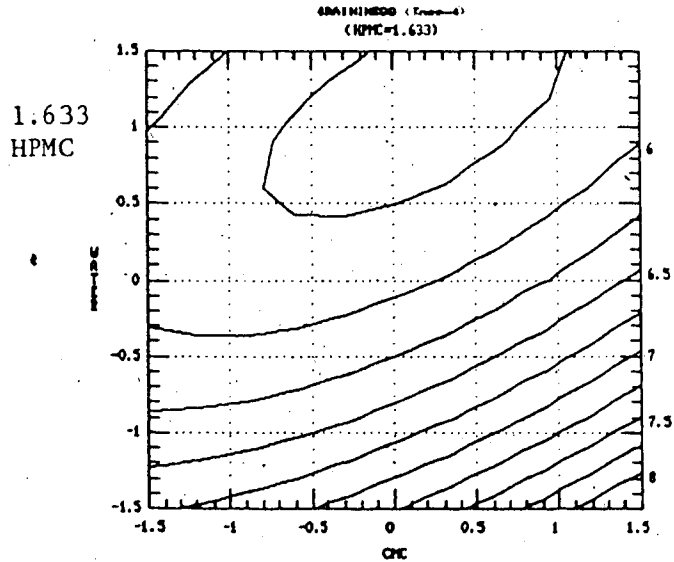
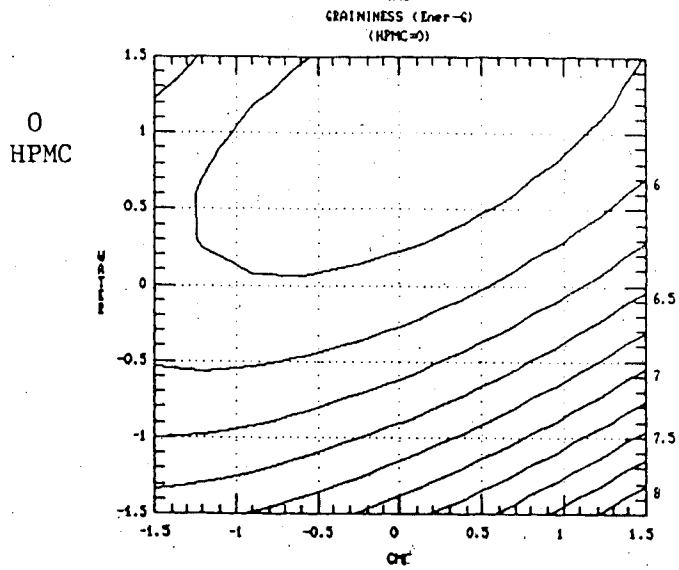
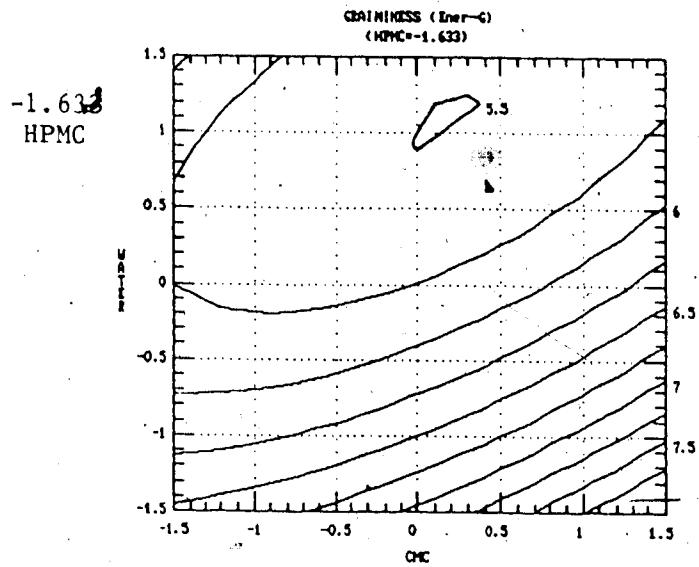
0
HPMC



1.633
HPMC

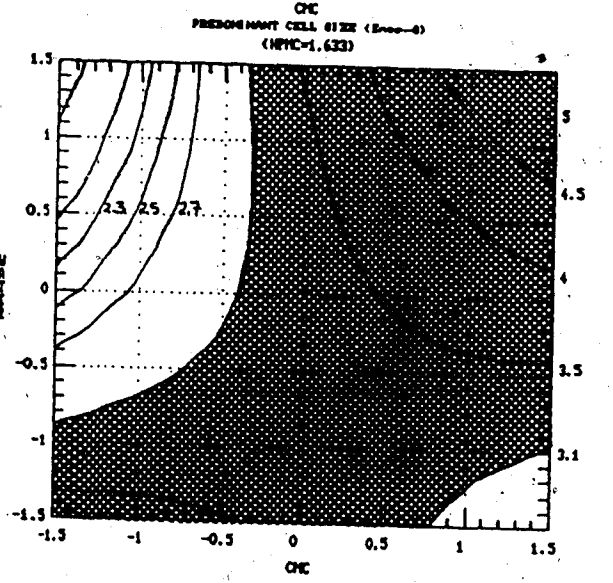
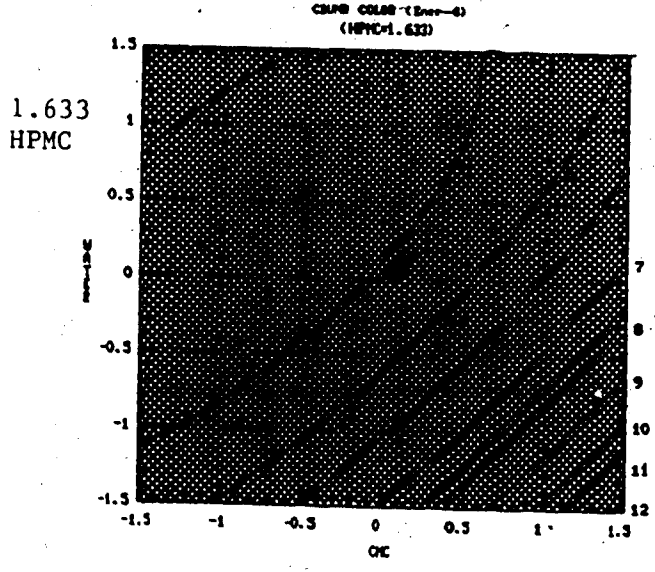
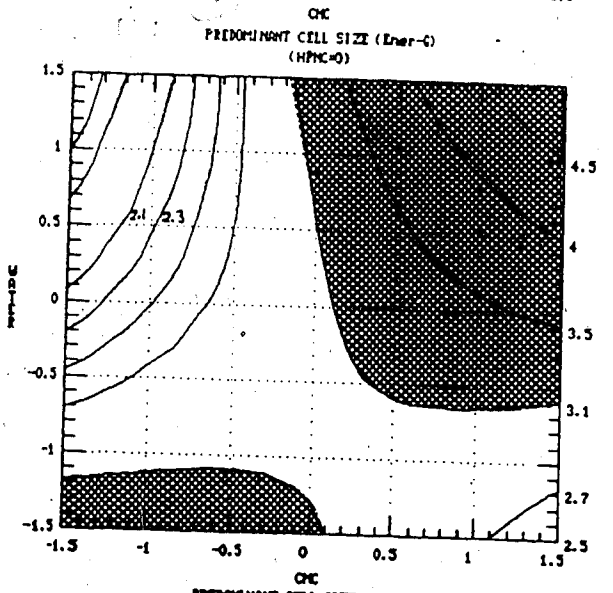
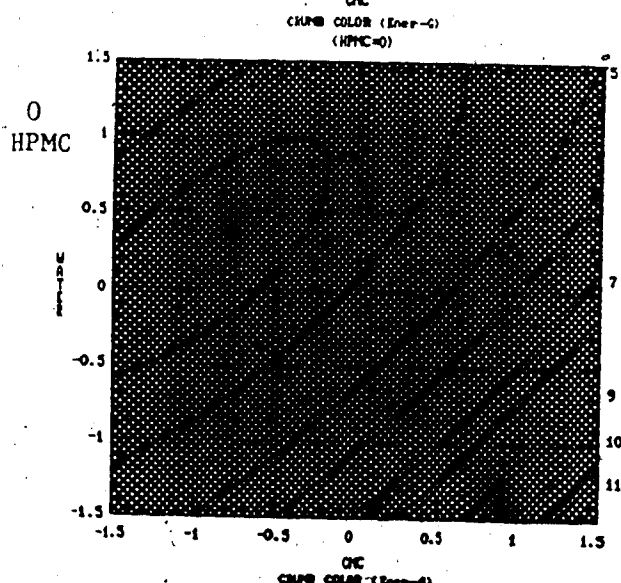
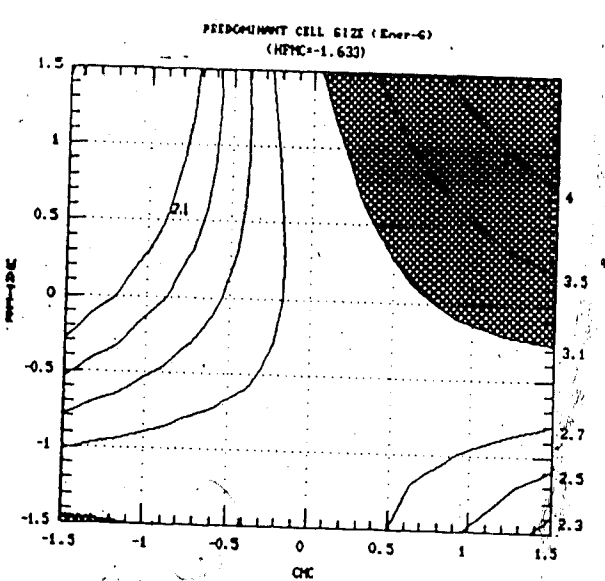
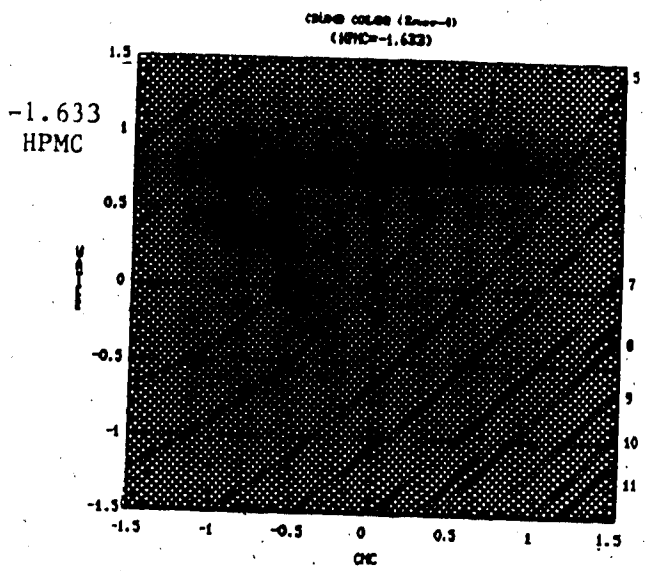


GRAININESS



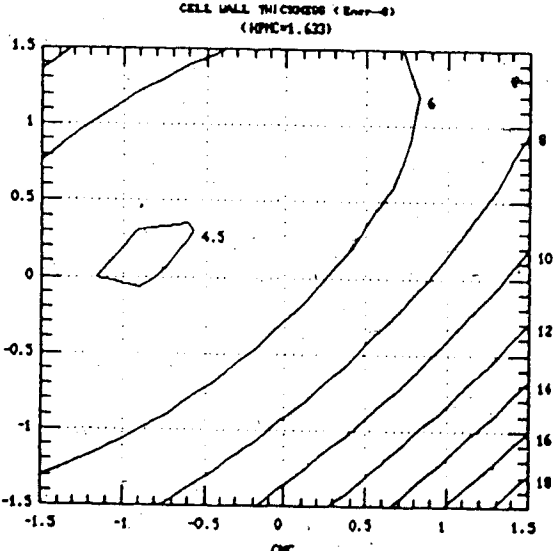
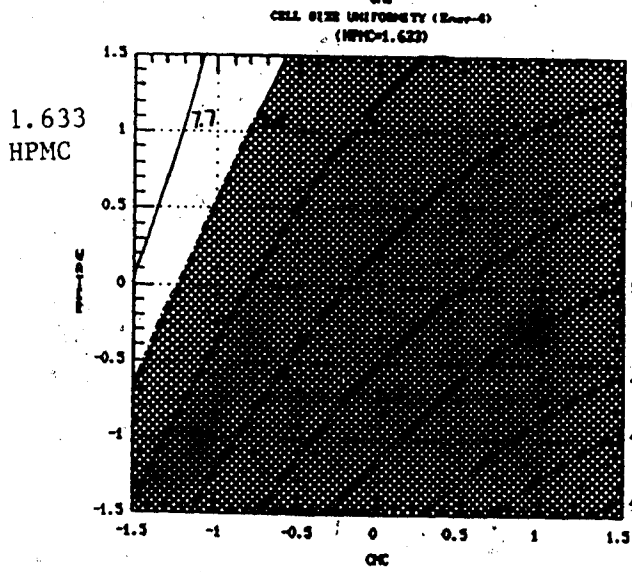
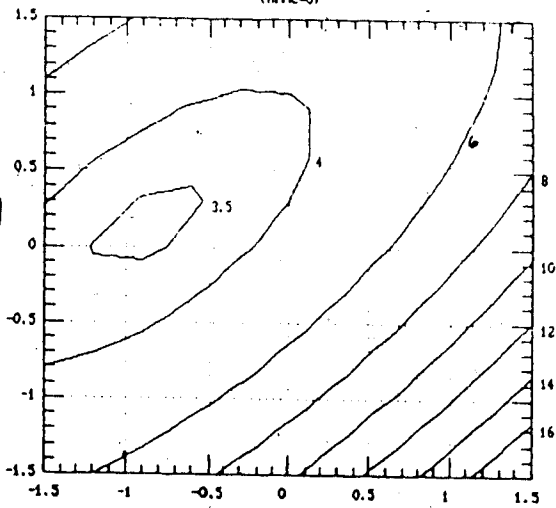
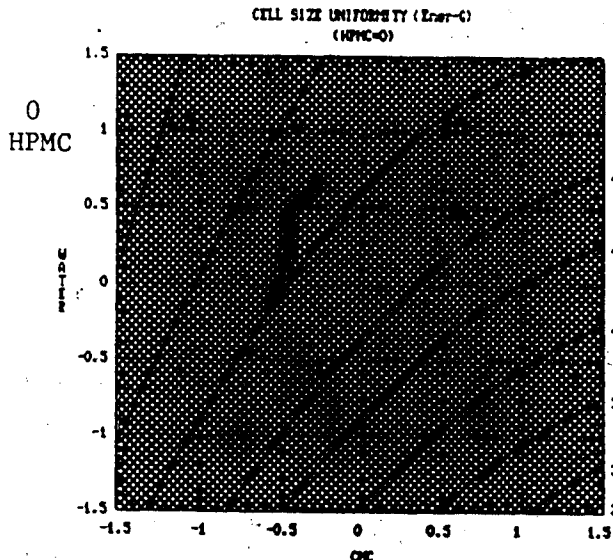
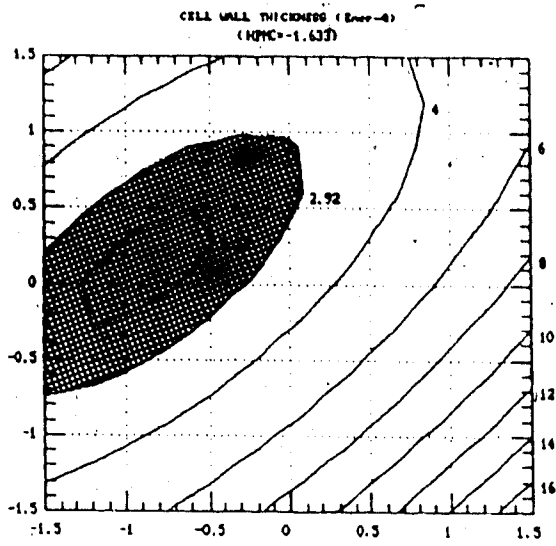
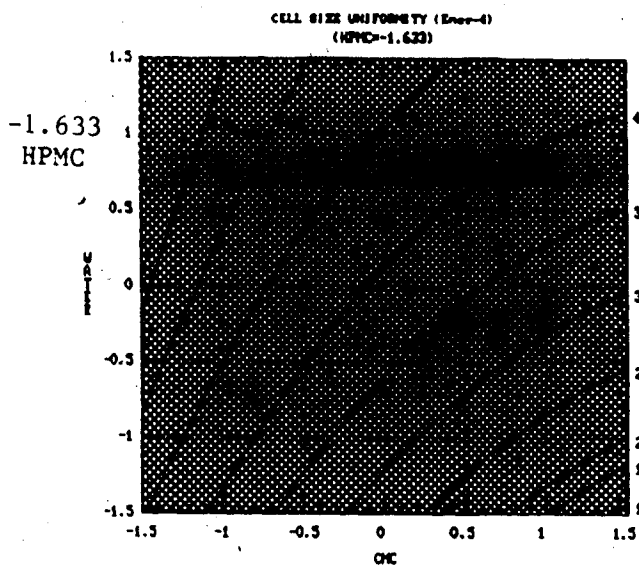
CRUMB COLOR

PREDOMINANT CELL SIZE



CELL SIZE UNIFORMITY

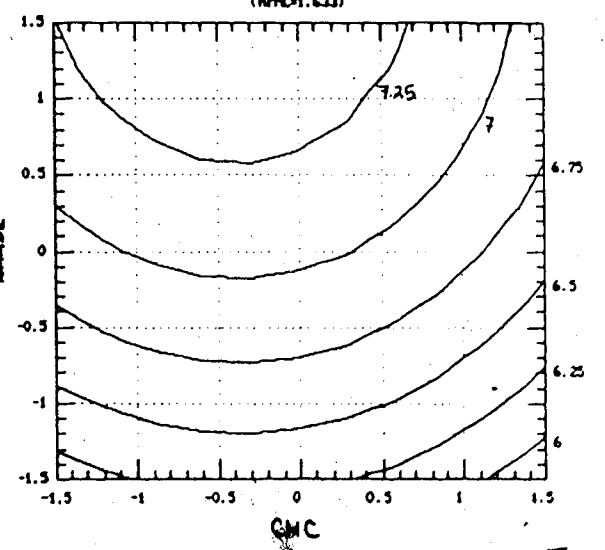
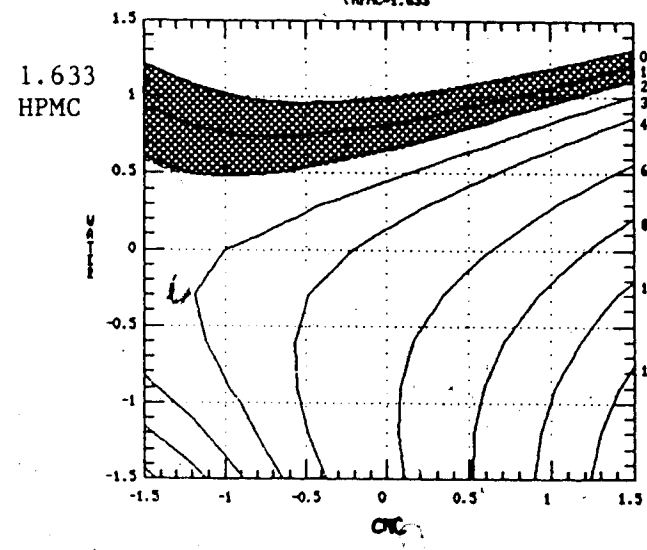
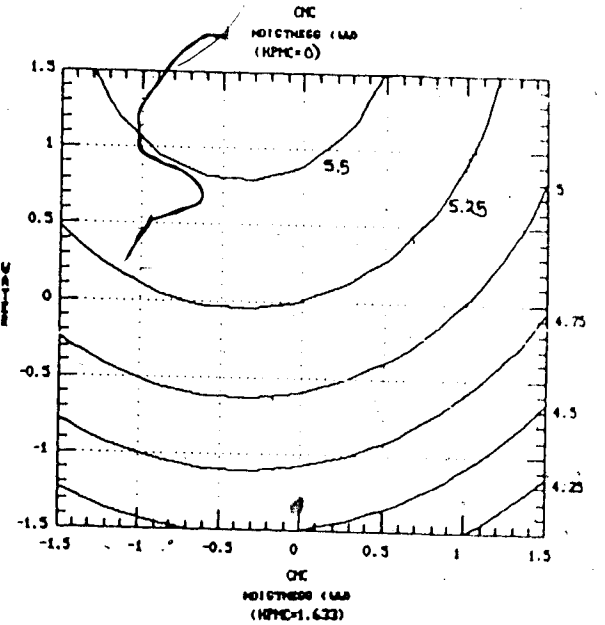
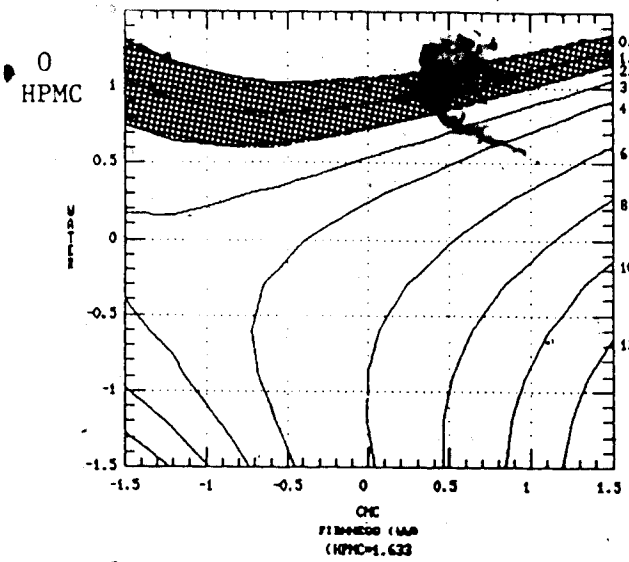
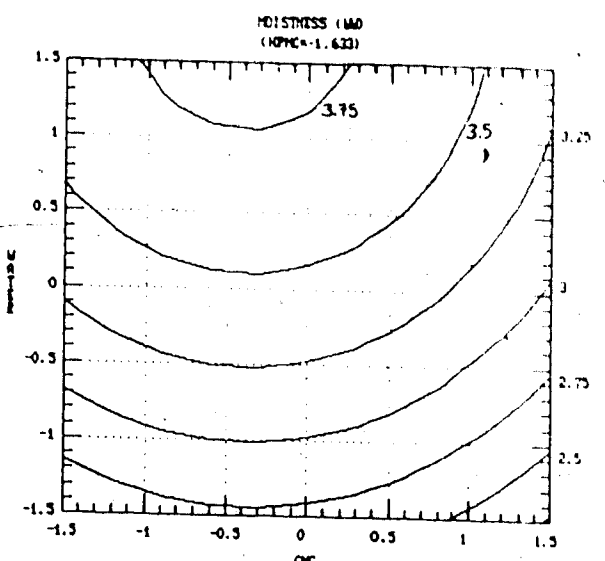
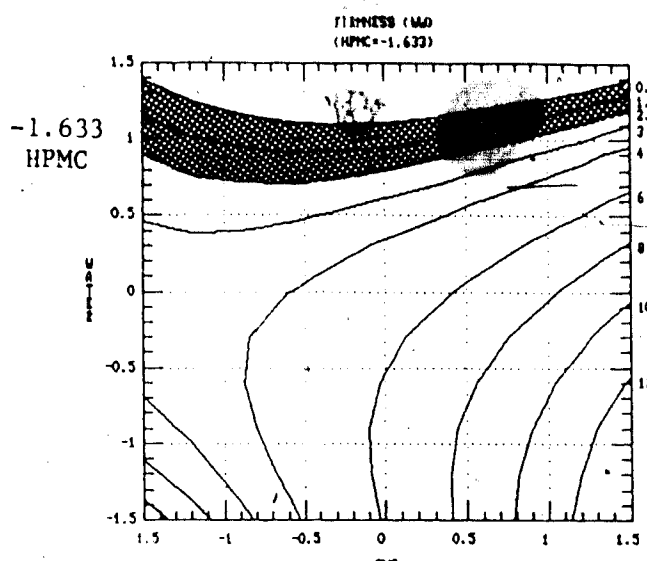
CELL WALL THICKNESS



Appendix 23 - Contour plots for Flour C rice bread sensory responses. Levels of HPMC, water and CMC are coded values. The shaded regions met reference standards.

FIRMNESS

MOISTNESS

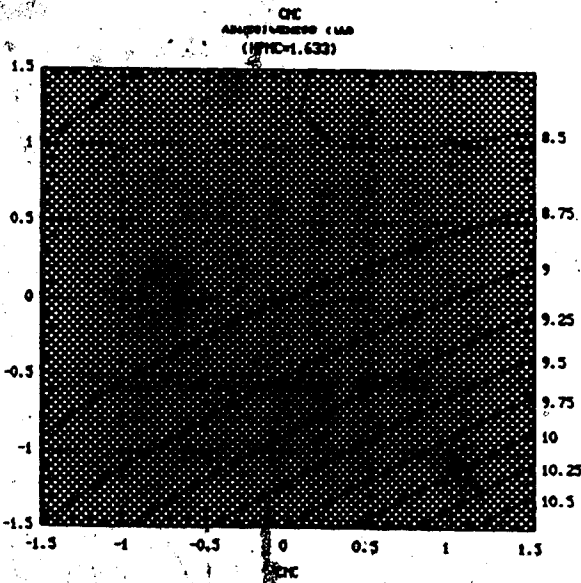
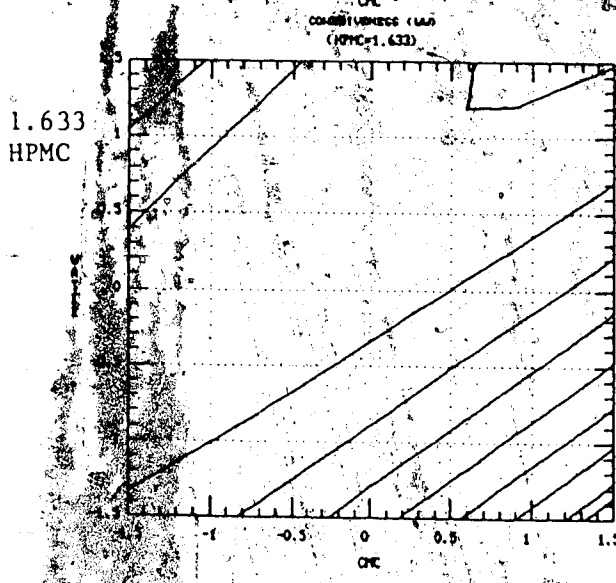
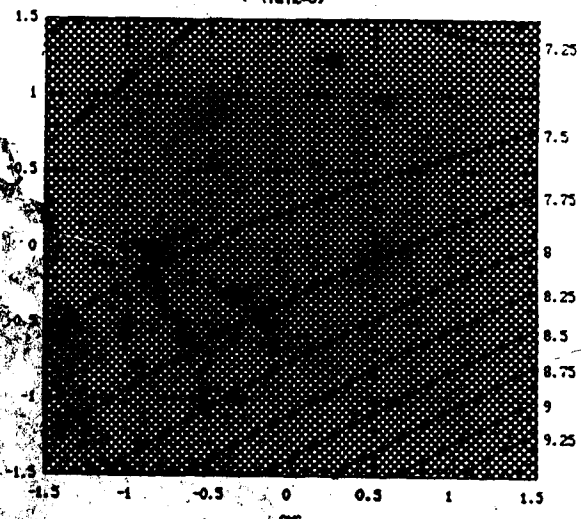
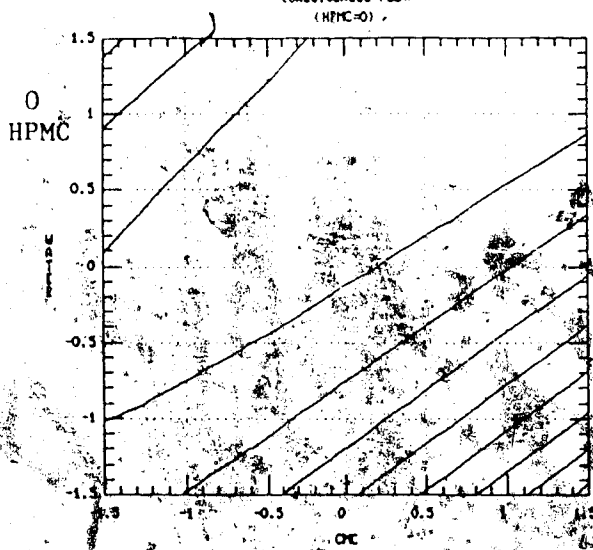
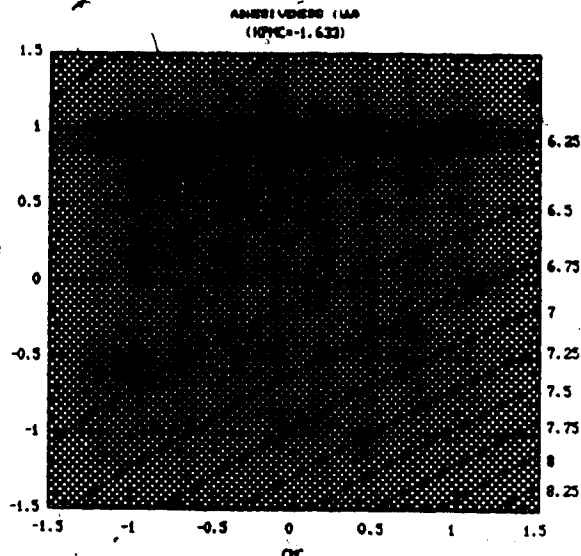
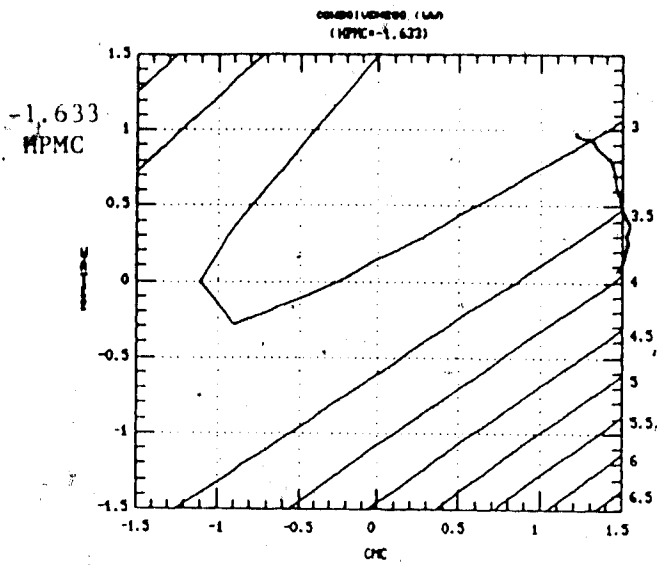


OC

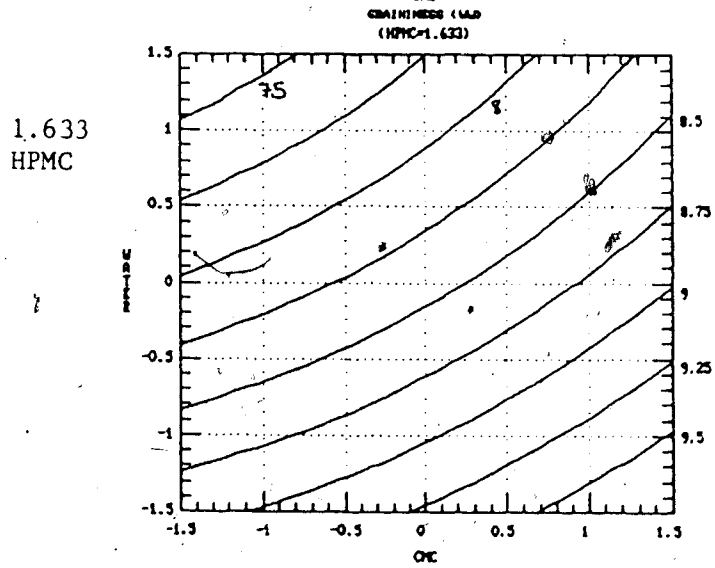
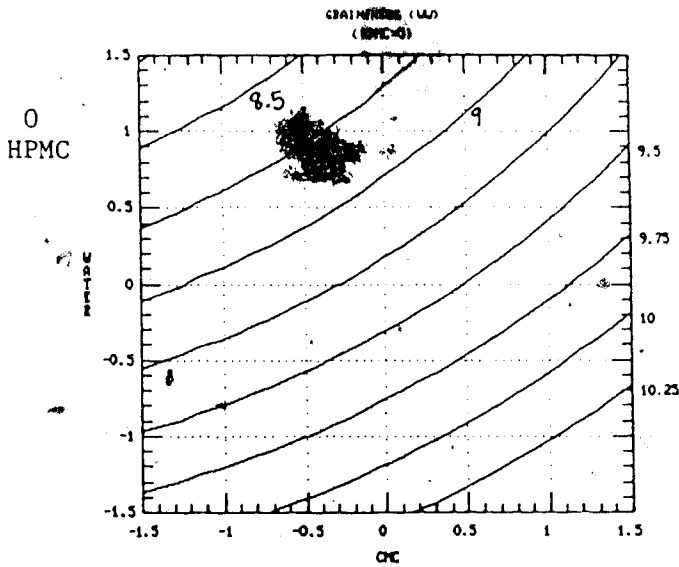
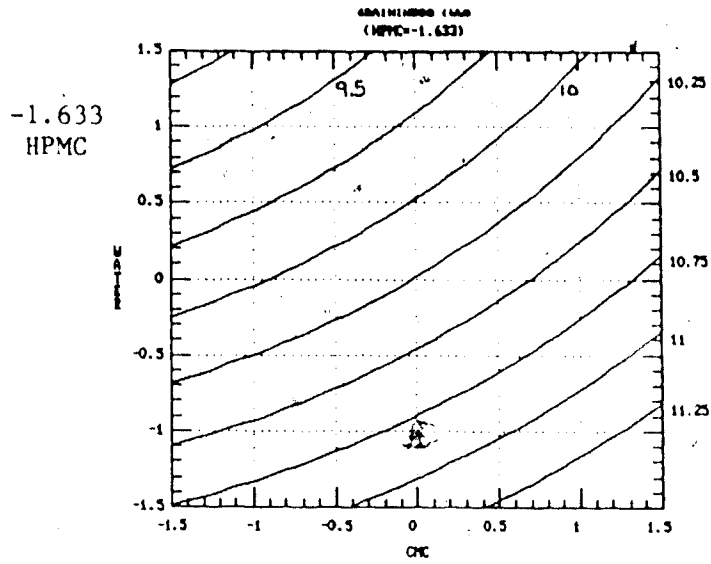
OC

COHESIVENESS

ADHESIVENESS

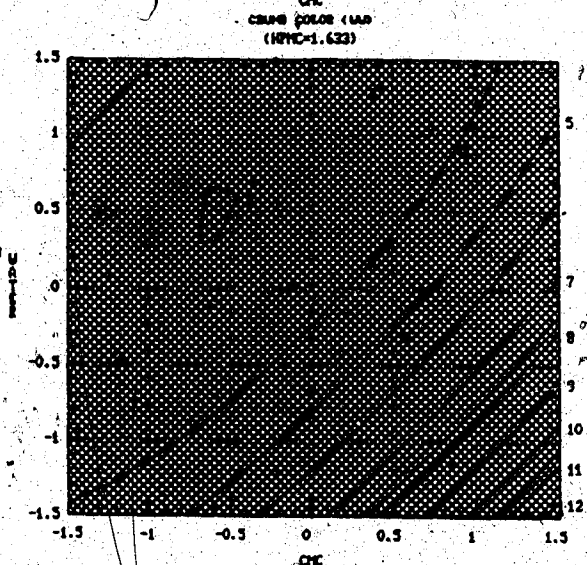
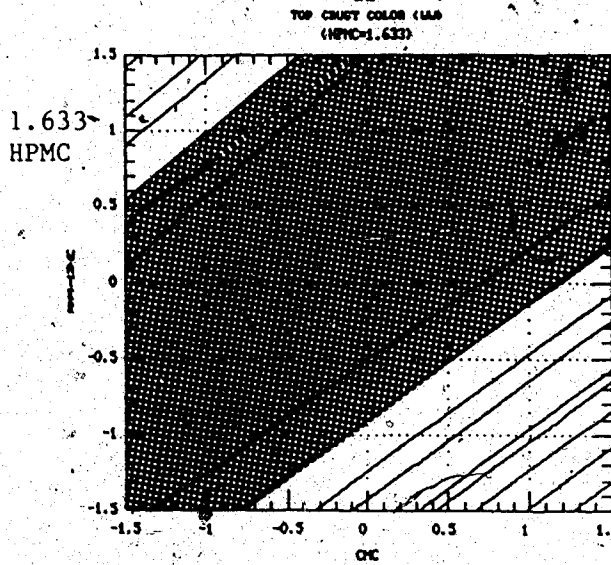
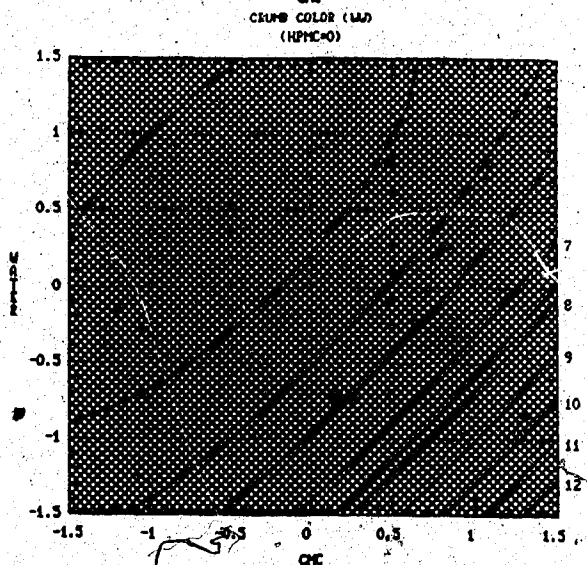
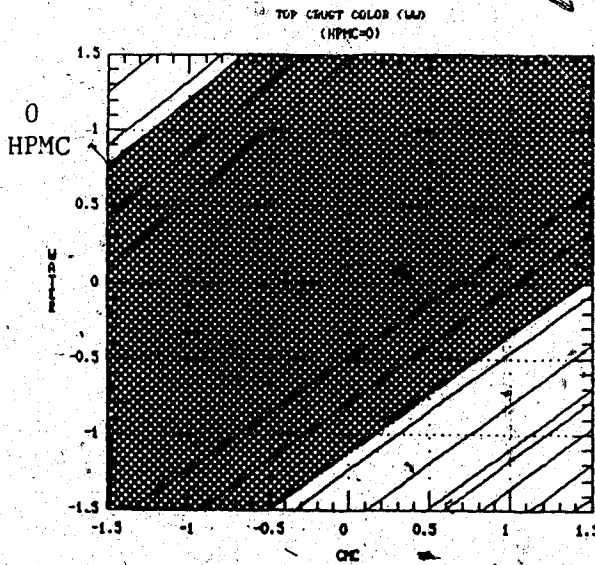
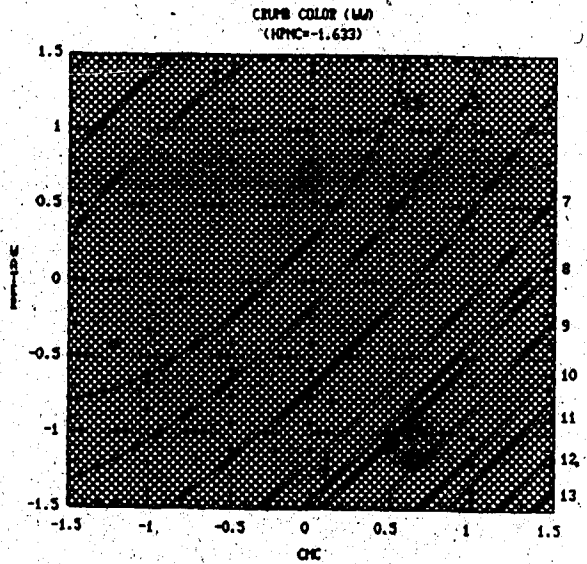
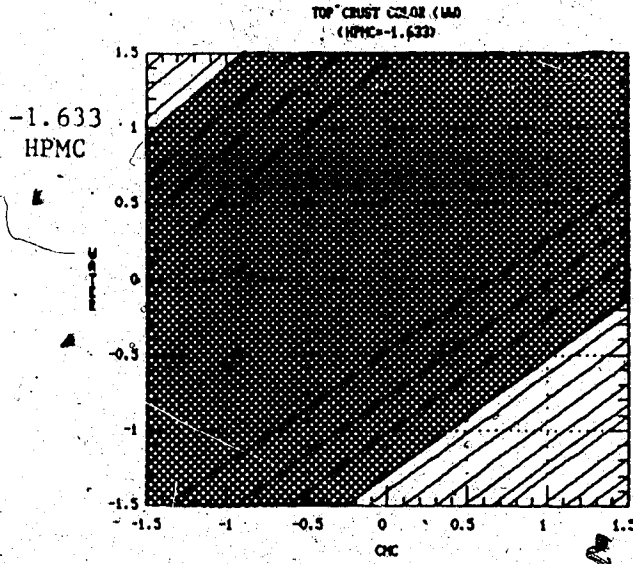


GRAININESS



TOP CRUST COLOR

CRUMB COLOR



CELL WALL THICKNESS

