EFFECTS OF VARIOUS NUTRIENTS ON ORGANOLEPTIC AND PHYSICOCHEMICAL PROPERTIES OF FLOUR AND CAKE

by

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INTRODUCTION

Since the current cereal grain enrichment standards were adopted in the early 1940's (1), a number of significant changes have occurred in the food consumption habits of the U.S. population. This is evidenced in many households by modification of traditional meal patterns and by the increased consumption of snack foods. In addition, total individual energy requirements have decreased as improved transportation systems and mechanization have reduced the physical labor required in many occupations. Large numbers of people have become weight-conscious and are dieting, thus further restricting energy intake and the consumption of specific foods that have traditionally been sources of many nutrients. Consequently, data used in developing the enrichment pattern established in the 1940's may no longer be valid.

Research over the past 30 years demonstrates the contribution enriched cereal grain products have made toward elimination of nutritional diseases such as beri-beri (2). Based on this and the important position cereal grain products hold in the American diet, a thorough review of the current enrichment programs is in order.

NRC/NAS, therefore, has recommended that the cereal-grain products be fortified with an expanded list enrichment formula. Because 26 % of the daily caloric intake in the U.S. comes from cereal-grain product, cereal-grain products appear best to meet many of the criteria for multiple nutrient fortification. In view of this study on technical feasibility of this expanded enrichment is conducted on the grain products to determine the stability, availability, uniformity of dispersion, freedom from segregation in commercial handling, and consummer acceptance of such characteristics as color, flavor and odor (3). The enrichment in bread has more or less been studied. Therefore, this study will emphasize the effects of enrichment on cakes.

The objectives of the study were to determine: (1). the effects of the new levels of enrichment on the flour physical and chemical properties, (2). the effects of the new levels of enrichment on product qualities, and (3). the effects of storage temperature and period on either the enriched flour or the cakes made by this kind of flour.

REVIEW OF LITERATURE

A major contribution to nutrition in the U.S. has been made by the cereal product enrichment program began in the 1940's. During the 1930's, public health authorities became greatly concerned by the alarming incidence of nutritional deficiency disease such as pellagra and beriberi; iron deficiency anemia was widespread. Then in 1938, riboflavin deficiency was identified as contributing to other deficiency diseases (4). At that time, more than one-third of the population had inadequate diets, and it was clear that the diseases were due largely to diets low in thiamin, niacin, riboflavin, and iron (5). A movement was soon underway to introduce the needed nutrients into the diet by adding them to white flour, and thus assure their presence in a food eaten by most people, namely bread. Then in 1941 (6), the United States Food and Drug Administration set legal standards to the levels of the B-vitamins and iron to be added to flour. Enrichment was made mandatory in 1943 (7). Finally in 1952, standards of identity for bakery products were issued. They specified that 1-lb enriched bread must contain: 1.1 to 1.8 mg thiamin, 0.7 to 1.6 mg riboflavin, 10.0 to 15.0 mg niacin, and 8.0 to 12.5 mg iron. Optional nutrients specified were calcium, 300 to 800 mg, and vitamin D, 150 to 750 USP units, per pound of bread (8).

A consequence of the enrichment program has been the virtual elimenation of deficiency disease due to inadequate uptake of the three B-vitamins (thiamin, niacin and riboflavin). In fact in just five years after introduction of enrichment, pellagra and beriberi were no longer evident among the alcoholics in large cities (4).

In 1971, the Food and Drug Administration proposed that the level of the three B-vitamins in bread and cereal products be increased by at least 50 % and the level of iron by a factor of three. An amendment was issued in 1973 to establish these higher levels (9).

According to a 1974 report of the Food and Nutrition Board of the National Academy of Science, significant segments of the U.S. population show evidence of potential deficiency of vitamin A, thiamin , iron, calcium, magnesium and zinc (2). Since 26% of the daily caloric intake in the U.S. comes from cereal-grain products, the board proposed increased fortification of cereal products with 10 nutrients (10). The proposal recommends increasing the iron level and adding vitamin A, vitamin B_6 (pyridoxine), folic acid, magnesium and zinc in addition to 1941 standards. Table 1 (9,10) shows the proposed levels compared to the levels in current use. The functions of these 10 nutrients are discussed in the appendix.

The point in the food process at which nutrients are added to insure uniformity is important. Ideally, they are introduced as close to the end of the processing stage as possible, particularly if they are heat labile, in order to decrease loss of uniform distribution or decomposition. Four basic methods have been developed for adding nutrients to foods. They are:

a. Pure compound or direct addition: Weight amounts of single or combination nutrients are added, with sufficient mixing to insure uniformity (12).

b. Tablet, wafers, or cubes: Compressed units containing the nutrients along with edible fibers can be prepared in the amount to nutrify a container or given weight or volume of food (12).

c. Solutions, emulsions or dispersions: Concentrated solutions or emulsions and micronized suspensions (dispersions)- or the nutrients

Table 1: Fortification Comparisions
(in mg/lb flour)

Nutrient	Presently used[a]	Proposed[b]
Thiamin	2.9	2.9
Riboflavin	1.8	1.8
Niacin	24.0	24.0
Iron	13.0-16.5	40.0
Calcium	-	900.0
Vitamin A	-	2.2 (7,300 IU)
Pyridoxine	-	2.0
Folic Acid	-	0.3
Magnesium	-	200.0
Zinc	-	10.0

[a]Levels listed in Code of Federal Regulations, Title 21 (1973) (9).

[b]NRC (1974) proposed fortification (3).

with edible suspendity and/or stabilizing agents in digestible carriers can be prepared (12).

d. Dry premixes: A premix blended into the food product can give greater assurance of final product uniformity since the weight of the pure individual nutrients added is quite tiny (13). Dry premix addition is the major method of nutrifying foods. The addition of premix is generally done at the mill by metering a vitamin-mineral premix into the stream of flour flowing to the packing bins or be incorporating a weighed quantity of the vitamin-mineral premix into flour in a batch mixing operation (14). Currently in the U.S., flour is fortified with premixes at the rate of 0.25-0.50 oz/100 lb. of flour. This level is delivered by feeders installed in the mills. If calcium is added, it is fed in separately at 120-600g/100 lb (10).

To assure the stability and availability of the fortified nutrients during storage and baking process is also an important requirement. Because one of the principal sources of loss of nutrients activity during processing occurs in the baking stage.

Vitamin A: Vitamin A is unstable in the presence of oxygen or air and when exposed to ultraviolet light (15). In flour with 11 % moisture, typical vitamin A retention was about 90 % after being stored six months at room temperature. The effects of temperature and time on vitamin A loss in fortified wheat flour and premix was significant (16). In fortified wheat flour, there was basically no change in vitamin A content at any storage temperature when stored less than two months. The baking loss of vitamin A at $395 \pm 0.5^{o}F$ for one hour and ten minutes was more than 50% (15).

Vitamin A palmitate, which is one of two commonly available types of vitamin A, was more stable in baked products than was vitamin A acetate (15). Vitamin A palmitate is stabilized with an antioxidant and by encapsulation, usually with gelatin or gum acacia as the encapsulating medium, and is furnished in beadlet form (17). This dry vitamin A palmitate product is very stable in baking wheat bread (15).

There is no natural vitamin A in flour. Any overage has to come from the amount added. Since previous work (10,18) has shown vitamin A to have a greater stability problem than other vitamins, an additional safety margin was allowed by the proposed level, as indicated.

Thiamin: Thiamin is probably the most heat sensitive of the B vitamins, especially in non-acid foods. Temperature, pH, heating time, thiamin molecule form, trace metals, oxygen, and processing or storage conditions are the most important factors contributing to the loss of thiamin in food product (19). Improved stability of thiamin in dry vitamin premixes and in dry foods and also at low pH was reported by Borenstein (20). Cakirer et al confirmed that the effect of temperature on the thiamin content in both fortified wheat flour and the original premix was significant, but the effect of time was not (21).

Obermeyer and Chen (22) reported the initial cleavage of thiamin to its pyrimidine and thiazole moieties, together with some unknown degradation products under the conditions encountered in bread making. Later, Dwivedi and Arnold studied the mechanism of the degradation of thiamin, and reported that thiamin, on heating in a glucose solution, produced a brown discoloration and fluorescence(23). This behavior is analogous to the maillard reactions of sugars and amino acids and could be important in the loss of thiamin during processing (19). A 20 % loss of thiamin in bread was found by Coppock et al and 29.5 % loss in European bread also was found by Bottomley and Nobile (21).

Thiamin is stable in dry vitamin premixes and in dry foods. It is commercially available both as the mononitrate and hydrochloride salts. The mononitrate is less hygroscopic and, hence, preferred in dry mixes. The hydrochloride is more soluble in water (20).

Currently mills are adding to a level of 2.9 mg/lb thiamin to flour during the enrichment process. This level assumes based on a natural thiamin level of about 0.35 mg/lb. A recent base line study (24) shows the natural level to be higher than that (0.60 mg/lb). Analysis of enriched flour confirm higher than necessary thiamin levels (11).

Riboflavin: Riboflavin causes few problems in food fortification, but its poor solubility in all food-type solvents is an inconvenience. In some applications its intense yellow color is undesirable. A more soluble analogue, riboflavin 5' phosphate, is available (3). A rice enrichment process using 5'phosphate, to prevent the color spotting has been reported (20). Riboflavin is less sensitive to heat than thiamin and quite stable in dry products, but it is readily degraded by light and unstable at high pH level (3).

Mills currently add 1.75 mg/lb riboflavin to enriched flour. Assays on enriched flour show a number of enriched flours hovering around the 1.8 mg/lb standard. An addition of 1.8 mg/lb was chosen to provide an extra safety margin and to improve the vitamin B_1/B_2 ratio (11).

Niacin: One of the most stable vitamins, niacin is not affected by heat, light and other baking conditions and, therefore, poses few problems in food fortification. It also is relative stable during breadmaking (21). No niacin is lost during the baking of enriched bread at 425°F for 30 minutes (25).

Niacin is commercially avialable both as nicotinic acid and nicotinamide (niacinamide) (20). The amide has a bitter flavor but is preferred in many applications because of its superior solubility (3).

Because it is totally stable, niacin does not require as high a safety margin as the other vitamins. The addition rate of 21 mg/lb for the new expanded enrichment level is the same as currently used (11).

Vitamin B_6 (pyridoxine): The stability of this vitamin is influenced by oxygen, heat, pH and light. Vitamin B_6 occurs in natural products. It has three forms, pyridoxine, pyridoxal, and pyridoxamine (3). In pharmaceutical products, pyridoxine is usually very stable, although in mixtures with minerals, significant losses may be observed (26). Stability data for foods indicate that pyridoxal and pyridoxamine are significantly less stable than pyridoxine (27). Excellent recoveries were obtained when pyridoxine hydrochloride was added to flour and baked into bread. The published data on loss of vitamin B_6 during bread baking show losses in the range of 5 to 10 % (26). More research is definitely needed on the stability of this vitamin (21).

It has been recommended (11) that a 10 % overage for pyridoxine is adviseable, since previous work (10) has shown this to be an adequate safety margin.

Folic Acid: Folic acid is quite unstable; is readily destroyed by heat under acid conditions, and is subject to storage losses. Its solubility in the acid pH range is very limited (20). High losses of both free and total folic acid can occur in foods during short cooking periods. The natural folic acid in the flour is fairly stable at $84^{o}F$, with 86 % retention after 12 months, but it shows progressively increasing losses as the temperature increases. Almost one-third of the natural folic acid is lost in bread processing. Folic acid added to flour at levels of 1 or 5 ug/g showed excellent stability during 12 months of storage at 84^{o} and $100^{o}F$. Total folic acid in bread dough (65% of yeast origin) increased with fermentation time but decreased by an average of 31 % during baking (21). Oxidizing maturing agents had little or no effect on natural folic acid activities in bread made from baker's patent flour. Native flour folic acid decreased in activity at early during storage and stabilized at a level dependent on storage temperature. The added folic acid showed small losses at high temperature. These results suggest that flour and bread are practical vehicles for folic acid enrichment of the American diet (28).

The proposed levels of folic acid provide around a 10 % overage which is what previous work (10) has shown to be an adequate safety margin (11).

Iron: There is high incidence of iron deficiency anemia in the United States, especially among young, rapidly growing children and adult women during their fertile years . The one important factor which is susceptible to control is the selection of iron sources with superior bioavailability (3). Most cereal enrichment is accomplished with four iron sources: ferrous sulfate, reduced iron, ferric orthophosphate, and sodium iron pyrophosphate (21). Ferrous sulfate is the preferred iron source because of its bioavailability and is the usual standard against which other iron sources must be compared for bioavailability (29,30). Reduced iron and ferric orthophsophate are too variable to be given an average relative biological value. Sodium iron pyrophosphate is very insoluble and relatively unavailable, so it is significantly less well utilized than the other iron sources (31,32).

There are some real and imagined effects on the color, odor, and taste of some foods due to the added iron. One of these effects is rancidity, which may result from the chemical reaction of the iron with components of the food or as a physical effect, due to the use of a colored iron source (33).

Research on the storage stability of flour enriched to a high level of ferrous sulfate is clearly needed. Rancidity did not develop in any of the flour samples that were stored for two years at room temperature (34). Further evidence is seen in the successful use of ferrous sulfate by many millers. Discoloration may result from chemical action between the iron and various other food factors. Tannis and chocolate have caused some off-colors. Some insignificant grayness may occur in white flour when enriched with finely powdered reduced iron. This can be detected by the 'Pekar Slick Test', but not by looking at the flour under normal conditions (33,34,35).

Iron is the only nutrient with a minimum-maximum enrichment range. To obtain flour with 20% of the U.S. RDA, a minimum level of 14.4 mg/lb is required, leaving an allowable range of 2.1 mg/lb (11).

Calcium: The major problem in fortifying with large quantities of calcium is the possibility of producing chalky flavors, sandy mouth feel, opacity, sediment and color changes in foods (20). Calcium salts commonly used for enrichment are calcium phosphate dibasic, anhydrous ($CaHPO_4$), calcium phosphate tribasic ($Ca_3(PO_4)_2$), and calcium carbonate ($CaCO_3$) (21). A 5% loss of calcium carbonate during baking was reported (25). This was attributed to the reactivity of the calcium ions with proteins, gums, etc., during baking (21).

Because the natural level of calcium in flour is quite low compared to the fortification level, variation in natural levels will have little effect on the final level (11).

Magnesium: Magnesium salts suitable for the fortification of foods are magnesium acetate, magnesium carbonate, magnesium chloride, magnesium oxide, magnesium phosphate-dibasic, magnesium trisilicate (3,20). Ranhotra et al (36) suggested that magnesium oxide and probably magnesium hydroxide and carbonate, can be used in proposed fortification of flour without noticeable loss of bread quality if necessary modifications in the bkaing procedures are made. However, the interaction of bread ingredients with added magnesium and the resulting flavor profile must be examined in greater detail to fully assess the potential of magnesium rich sources in a fortification program (36). Ranhotra et al (37) also found out that the difference probably has little physiological significance, and concluded that magnesium was equally available from all magnesium sources.

Unlike calcium, the natural level of magnesium varies considerably, and it consistitutes a significant part of the fortification level. This makes it very difficult to arrive at a common addition rate. Flour with different ash levels require different amount of magnesium to reach the target level. It may be that each mill will have to determine magnesium addition rates for each type of flour they produce (11).

Zinc: Zinc salts suitable for the fortification of foods are zinc acetate, zinc chloride, zinc lactate, and zinc sulfate (20). The solubility of zinc was suspended in 0.85% NaCl solution, adjusted to pH values between 4.5 and 7.5, than when unleavened bread was so suspended. The solubility of zinc in suspensions of wholemeal bread either leavened or unleavened increased as pH decreased from 7.0 to 4.5. Fermentation with yeast markedly increases the physiological availability of zinc in wholemeal bread. This is attributed in part to the action of yeast in destroying phytate. The zinc of the wheat kernal is intimately associated with phytate and protein in several structures. Addition of zinc to a wholemeal for conversion into bread might not adequately simulate the behavior of naturally occuring zinc (38,39).

The situation with zinc is similar to magnesium. Addition rates vary with flour type. If zinc is included in a vitamin-iron premix it should be added at a rate of 8-9 mg/lb (11).

A study by Cort et al (10) of enrichment premixes containing the six NRC/NAS recommended vitamins plus iron and vitamin E showed good stability of all components. The further study showed excellent retention of these vitamins in flour, which was additionally fortified with calcium, magnesium and zinc (10). But Rubin et al (18) determined that calcium and magnesium may decrease vitamin A availability, and, to a lesser extent, increase folic acid deterioration at the temperature and moisture levels in the baking process. They compared two different kinds of premixes. Premix A provided all the NRC recommended vitamins and minerals; premix B contained all the proposed nutrients for the new fortification levels, except calcium and magnesium. The data shows very good stability for vitamin B_1, B_2, B_6 and niacin in both experiments. But vitamin A and folic acid showed a slight loss in bread fortifed with premix A, while almost no loss was observed in samples enriched with premix B (18).

Fortified breads, in the baking test made by Rubin et al (18), showed a slightly darker color than unfortified breads. The bread containing the full mineral supplement (premix A) gave a slightly lower grain score. The scoring data showed no change in the volume of the loaves fortified with the various enrichment premixes. Bread fortified with full mineral supplement (premix A), received lower scores than premix B, suggesting that addition of large amounts of calcium and magnesium affects color and grain.

In the tasting test made by Rubin et al (18), no off-flavor of the fortified bread was detected by the taster. But tasting tests carried out after a week's storage of the bread at room temperature indicated a very slight off-flavor in the bread fortified with NRC proposed vitamin-mineral supplement. The off-flavor was probably due to magnesium. Bread fortified with the vitamins, iron and zinc was judged not to have an off-flavor taste.

Stability of vitamins in flour containing approximately 12% moisture is excellent (18). Cakirer, Lachance (21) and Rubin et al (18) found no loss of vitamins, change of odor or appearance in the fortified flour which was stored for six months at room temperature.

Currently, soft wheat flour research and testing are still in the early stages of development (40). The relatively large quantities of shortening required in cake formulation may inhibit gluten development, and the use of sugar may bring about changes in water distribution among ingredients and flour components (41). Since so little is known about the mechanism of the cake-baking process and the factors affecting quality, the flour testing now being done is being conducted on an emperical basis (40).

Yamazaki (42) outlined the procedure for soft wheat and flour evaluation in the U.S., and Yamazaki and Lord (43), and Pratt (44) described the legal standard and the various quality components that are related to end-use applications (44). The procedure employs the use of both chemical and physicochemical tests in outling flour specifications, only some of these tests are used currently. They are moisture content, pH, alkaline water retention capacity, MacMichael viscosity, color and baking test.

Reducing moisture content to lower levels results in better batter and cake yields and an improvement in the shelf life (41).

The pH value of cake flour provides a partial index to the extent of chlorine treatment. It has been observed, however, that cake flour with high ash content require more chlorine treatment to mellow gluten and produce a given pH than do flours of lower ash content. Practical experience has shown that the best flour performance is obtained when it is carefully bleached to a pH value between 4.6 and 5.0 (45).

The alkaline water retention capacity (AWRC) is highly correlated with cookie diameter (40). Bode also confirmed this result (46). In 1953, Yamazaki reported that the tailing fraction of the starch gives a good indication of varietal hydration properties; those from varieties baking poorer cookies had the higher retention values (47). An advantage of this test over other physicochemical methods, in intervarietal comparison, is that the AWRC results may be compared directly with cookie diameter without adjustment for flour protein or ash content (40).

The method of determining the "apparent viscosity of acidulated flour-water suspensions", based on the work of Sharp and Gortner (48) and described in Cereal Laboratory Methods (49), is widely used by the soft wheat milling industry. Most of the previous investigators, such as Bayfield, Reiman, and Rich (50), observed that viscosity increased with the flour protein content and decreased with ash content. Higher viscosity results are associated with stronger gluten characteristics (41). Previous work has shown that cake volume or cookie spread ratio decreased with increased viscosity (41, 50,51). Although this test is used extensively for classifying soft wheat flours, Loving (51) commented that viscosity has never been accepted as an important quality factor in cake flours.

Flour color is determined by M-500-A agtron which is a reflectance spectrophotometer designed to measure relative reflectance of a sample at four monochromatic spectral frequencies (52). The green mode is used for determining color in flour (53). Previously published data (53-56) shows the reliability, precision, and consistency of the agtron method. The ash content of flour provides an index of the degree of refinement of the milling process, and is useful in comparing quality of flours milled from a given wheat mix. However, ash is less useful in comparing flours from different wheat mixes, and there is little relation between flour ash content and flour quality per se (57). Such a criterion would appear logical because the consumer is more concerned with product color than flour ash content. The degree of agtron color thus appears to be useful for quality control measurements: the whiter the flour, the higher the grade. Watson and Shuey (57) determined that there were no pronounced changes in the green or blue agtron values of the flour during the storage peirods, and they also confirmed that flour color as measured by the agtron is essentially stable.

A baking test requires considerable skill, experience and the ability to recognize and interprete the important baking qualities in a given situation. The white layer cake is said by many authorities to be the best testing medium for cake flours, because it offers the most favorable conditions for judging crumb color as well as the other characteristics that are normally scored. Others are of the opinion that the balancing of a yellow cake formula to a flour provides a more critical test and hence prefer that method (58).

The first clue to be judged to determine baking performance is the crust character. This should be smooth, of a desirable medium brown color, and free of a taffy-like appearance or other indications that the sugar is not being carried properly (41,42,51,58).

Volume in cake is an important measure of the baking performance of the flour because commercial baking is geared to a definite size, and if volume is variable the final package may be over or under-filled even though the scaling weight is the same (41,42,51,58).

Next the cut cake is critically examined for symmetry. A bold symmetry is indicative of a good rise in the oven and high sugar carrying ability. A flat or dipped cake reflects a formula that is unbalanced or a flour that is unable to carry the desired amount of sugar (41,42,51,58).

Grain and texture are examined next to see that the cells are fine, uniform and thin-walled. Coarseness, irregularity, thick cell wall, or excessively large cells are all indicative of inferior baking performance (41,42,51,58).

The color of the crumb is an important characteristic. Grain and testure exert an influence on color. A coarse grain and irregular texture make the color appear darker (41,42,51,58).

The final characteristics to be evaluated are flavor and eating quality. A properly baked cake from a well balanced formula will be free from doughiness, yet at the same time possesses a moist, tender character that produces a good "mouth feel" (58). Cake tasting panel is usually used for testing these qualities.

MATERIALS AND METHODS

MATERIALS

The flour used for the flour and cake fortification studies was a typical baker's cake flour obtained from International Multifoods, with an protein content of 8.1%, a moisture content of 9.89%, and an ash content of 0.36%.

The other ingredients of the cake formula were obtained from the Baking Laboratory in the K.S.U. Dept. of Grain Science and Industry.

Two types of vitamin-iron premix were utilized in this study. One premix (supplied by Pennwalt Comp.) was designed to enrich flour to regular or present FDA-approved levels, while the other premix (supplied by Hoffman-LaRoche Inc.) was designed to meet the NRC recommended enrichment levels. Two variations of each of the two premix types were used, providing for different forms of iron. The designations used in this report to describe the premixes are as follows:

Premixes to enrich to regular or current FDA levels.

- Reg A - Regular enrichment with reduced iron.
- Reg B - Regular enrichment with ferrous sulfate.

Premixes to enrich to NRC recommended levels.

- PA - NRC enrichment with reduced iron.
- PB - NRC enrichment with ferrous sulfate.

Compositions and the feed rates of these vitamin-iron premixes are shown in Tables 2-5.

Thiamin mononitrate was used as the thiamin source. It was preferred over the chloride form since a previous worker found it was more stable in flour (59).

Table 2: Regular Vitamin-Iron Premix Formulations

Ingredients	With reduced iron (Electrolytic)	With ferrous sulfate (Bakery grade)
Thiamin Mononitrate	3.83	1.92
Riboflavin	2.47	1.23
Niacin	29.60	14.81
Iron Source	16.92	9.87
Starch	Q.S.	Q.S.

Table 3: NAS Flour Fortification Premix Formulations (1)

Ingredients	With reduced iron (2) (Electrolytic)	With ferrous sulfate (3) (Bakery grade)
Thiamin Mononitrate	2.57	1.71
Riboflavin HCl, Type S	1.80	1.20
Niacinamide	21.00	14.00
Iron Source	11.46	29.17
Pyridoxine HCl	2.00	1.33
Folic Acid	0.26	0.17
Vitamin A Palmitate, 250 SD	20.00	13.33
Tricalcium Phosphate	3.00	3.00
Starch	O.S.	Q.S.

(1). This is not a manufacturing formula. No overages are included to insure premix label claims.

(2). Electrolytic reduced iron. Glidden-Durkee, A-131.

(3). Mallinckrodt's Bakery Grade ferrous sulfate, Code 5098.

Table 4: Feed Rates (in g/cwt) for Meeting Regular Fortification Standard.

Ingredients	Cake Flour
Regular Vitamin-Iron Premix	
-with reduced iron (Type 41 N-RICHMENT-A)	7.09
-with ferrous sulfate (Type 540 N-RICHMENT-A)	14.18

Table 5: Feed Rates (in g/cwt) for Meeting NAS Fortification Standards.

Ingredients	Cake Flour
Vitamin-Iron Premix (Table 4)	
-with reduced iron	15
-with ferrous sulfate	16
Calcium Salt	
-Sulfate, anhy. (29% Ca)	310
-Carbonate (40% Ca)	225
Magnesium Oxide (60% Mg)	27
Zinc Oxide (80% Zn)	1.1

Niacinamide and niacin are equivalent in their functionality and stability. But niacinamide was included in the premix because it does not have the vasodilation properties of niacin.

Two types of iron, reduced iron and ferrous sulfate, were involved in this study.

The reduced iron source we used was electrolytic iron. This product is manufactured by Glidden-Durkee under their code A-131. It is assumed that other forms of reduced iron (hudrogen reduced and carbonyl) will be similar to electrolytic iron in their functionality, stability and interaction effects. Their main differences are in relative bioavailability (11).

The ferrous sulfate product specified for the vitamin-iron premix was Mallinckrodt's bakery grade ferrous sulfate, code No. 5098. This product has a very fine particle size. It has been found to be the only form available which will not produce the so called "black spot" problem which ocassionally crops up in rolls and buns made from ferrous sulfate enriched flour (11).

The mineral of the NAS fortification formula are calcium, magnesium and zinc. Feed rates of these minerals are shown on Table 5 (11).

Two food grade calcium sources are used in this enrichment: calcium sulfate and calcium carbonate. Calcium sulfate is manufactured by U.S. Gypsum. The anhydrous form which was used in this experiment is sold under the name "Snow White Filler, F & P Grade". It is 29.4% calcium. Calcium carbonate (ground limestone) is produced by Georgia Marble Co.

Magnesium oxide was used as the source of magnesium. It is also one of the cheaper sources. Magnesium hydroxide and carbonate may also be acceptable sources. Food grade forms of these three salts are all from Mallinckrodt.

The only zinc product currently approved for use in foods is zinc oxide. This was also provided by Mallinckrodt.

METHODS

Flour Physical and Chemical Tests

Flour moisture content, pH value, MacMichael viscosity and color were tested according to AACC methods 44-19, 02-52, 56-80 and 14-30 (49). The method developed by Yamazaki in 1953 (40) was used for the determination alkaline water retention capacity.

Preparation of Cake

In addition to the flour physicochemical tests, baking performance tests were used to provide direct information on cake flour functionality.

Hobert mixer, model C-100, with 3.5 Quart Hobart mixing bowl, made by the Hobart MFG. Co., Troy, Ohio, were used for mixing batter.

Typical Yellow Layer Cake Formula

Ingredients	Baker's %	Total Grams
Flour	100	500
Sugar	120	600
Salt	3	15
High ratio shortening	20	100
NFDM	8	40
Whole egg solid	10	50
Food color (Goldex)	3	15
Emulsifier (Panalite 40SA)	1.3	6.5
Vanilla	1.07	5.35
Baking powder	6	30
Water	130	650

Panalite 40SA composes of mono- and di- glyceride.
40: 40% monoglyceride. S: soft. A: animal

For the baking of yellow layer cakes, the oven was brought to a baking temperature at 375°F and it was conditioned by baking cake using scrap batter and the above formula. All the dry ingredients were combined and sifted well. They were then transfered to the mixing bowl. Shortening, vanilla, panalite 40SA and 65% of water were introduced to these dry ingredients and mixed at low speed for 0.5 minute by paddle. Then the slurry was scraped down. The rest water was divided to two portion for addition. After each addition, the slurry was mixed for 3 minutes at low speed and was scraped. After following these procedures, the batter specific gravity should be about 0.9. The bottom of the pans, which were greased lightly with commercial pan grease, was lined with parchment paper. The 425 gm of batter was scaled into each of four pans and was baked at 375°F. After 25 minutes, cakes were cooled in pans for about 30 minutes. Then the cakes were removed from pans and cooled further. The cakes were then stored in polyethylene bags before presenting for evaluation.

Typical Chocolate Cup Cake Formula

Ingredients	Baker's %	Total grams
First Stage:		
Flour	100	500
Sugar	120	600
Salt	3	15
High ratio shortening	20	100
NFDM	6	30
Whole egg solid	5	25
Natural cocoa	15	75
Vanilla	1.07	5.35
Water	70	350
Second Stage:		
Baking powder	1.8	9
Soda	2.4	12
Water	70	350

For the baking of chocolate cup cakes, the oven was brought to the baking temperature at 400°F and it was conditioned by baking cake using scrap batter and above formula. The dry ingredients were combined in the first stage and sifted well. Then they were transferred to a mixing bowl. Shortening, vanilla and water were introduced to these dry ingredients and mixed for 1 minute at low speed by wire beater. The slurry was then scraped down and was mixed for another 2 minutes at medium speed. After the mixing, the slurry was also scraped down. Then, baking powder, soda and water in the second stage were added to the slurry. These were mixed at low speed for 1 minute, scraped down and mixed at low speed for another 1 minute. After all of these procedures, the batter temperature should be 75°F and specific gravity should be 0.95. Then the pans were lightly greased with commercial cake grease and 36 gm of batter were scaled into each cavity of the pans. The batter was then baked at 400°F for 14 minutes. After baking, the cakes were cooled in pans for about 30 minutes. Then the cakes were removed from pans and cooled for 15 minutes at room temperature.

Physicochemical Determinations of Cake Characteristics

The following determinations were conducted:

1. Specific gravity of batter.
2. Batter pH.
3. Batter color.
4. Cake weight.
5. Cake volume.
6. Crumb pH.
7. Crumb firmness.
8. Crumb color.

Specific Gravity of Batter. This was obtained by dividing the weight of a precise volume of batter by the weight of an equal volume of water.

Batter pH. This determination was made by using a Waring blender and a Beckman Expanomatic pH meter. The cake batter was blenderized for 1 minute at high speed and a pH determination was made.

Batter Color. The agtron M-300-A at yellow mode was used for measuring crumb color. The disk was selected to furnish the widest range of readings in order to distinguish small differences in color. The instrument was standarized at 0 reflectance with disk 75 and at 100 % reflectance with disk 100. Disk 0 and 24 were selected for measuring the batter color of chocolate cup cake.

Cake Weight. Cake weight was determined by weighing the finished cake in grams.

Cake Volume. Cake volume was determined by rapeseed displacement in cc.

Crumb pH. According to AACC method 02-52 (49), the determination was made by using a Waring blender and a Beckman Expanomatic pH meter. Ten grams of cake crumb were placed in the blender with 100 cc of distilled water. The blender was operated for 1 minute at high speed and a pH determiantion was made on the homogenized solution.

Crumb Firmness. Firmness was measured by a Bloom Gelometer equipped with a plunger of one-inch diameter. Five slices from each yellow layer cake and one slice from each chocolate cup cake were taken for firmness measurements. Each slice of cake, one inch thick, was placed on the platform of the gelometer; the platform was vertically adjusted so that the slice just touched the bottom surface of the plunger. Lead shot was then released such that the shot compressed the slice by 4 mm (48). The samples were compressed in the direction toward the central slice. After the firmness measurements were completed, the cake samples were saved for the crumb color measurement.

Crumb Color. The agtron M-300-A at yellow mode was used for measuring crumb color. Five slices of yellow layer cake and one slice of chocolate cup cake were taken for color measurements from each sliced cake. The selection of the disks was done the same way as the disk selection for the color of cake batter. A mask of black paper 1" x 3" was used. The sliced cake was placed on the mask to obtain the reading.

All the cake qualities were measured 1, 3 and 6 days after yellow layer cakes were baked and 1 day after chocolate cup cakes were baked, so the effects of storage at room temperature on cake qualites could be detected.

Taste Test

A taste panel was held after the cakes were baked. An untrained panel of 40 volunteers was asked to examined the cakes made with variously enriched flours and to judge each sample by tasting. Non-parametrics, statistical methods based on ranks, was utilized to examine the data.

Storage Test

The effects of storage temperature and time on the qualities of NRC-level enriched cake flour and the cakes made from this enriched flour were investigated. The flours fortified with NRC proposed enrichment were stored at three different temperatures, $0^{o}C$, $25^{o}C$ and $40^{o}C$, for tow different times, 3 months and 6 months, separately. After the storage, flour physicochemical test, baking tests on one day old yellow layer cake and taste tests on one day old yellow layer cakes were conducted. In addition, flour odor was tested. The odor test was conducted by the same method that was used for the taste tests except the samples were replaced by the flours and were judged by smell rather than taste.

RESULTS AND DISCUSSIONS

The Effects of Enrichment on Flour Physicochemical Properties

A study of the effects of NRC-level enrichment on flour physicochemical properties was necessary to determine whether the new enrichment levels are acceptable. In current commercial products, the regular enrichment is added to the flour as a vitamin and iron premix. In this study, the six vitamins and iron specified by the NRC/NAS were also combined as a premix.

Currently, two iron sources, reduced iron and ferrous sulfate, are used for the regular vitamin-iron premix. These two kinds of iron sources, therefore, also were applied to the NRC proposed vitamin-iorn premix.

The effects of premixes with two different forms of iron, either in regular-premix or NRC-premix, on the flour physicochemical properties were compared. Table 7 presents the results. The data shows no significant difference in flour physicochemical properties between NRC-premix and regular-premix, if reduced iron was the iron source for both premixes. When ferrous sulfate was the iron source, no significant difference was found between the regular-premix and the NRC-premix, except that the NRC premix gave a lower viscosity than the regular-premix. The two different iron sources produced the same effects in the regular premixes, except for a higher viscosity found in the premix made with reduced iron. Reduced iron seems to be the better source for fortifing the NRC enriched flour than the ferrous sulfate, since it increased the alkaline water retention capacity less and reduced the flour viscosity less. These results suggest that both kinds of NRC-premixes are very acceptable, but reduced iron seems to be the better iron source for fortification.

Table 7: The Effects of Enrichment on Flour Physicochemical Properties

Samples	pH[a]	Agtron[b] Rd.	AWRC[c]	MacMichael[d] viscosity
F	4.75	65.23	50.63	62.75
F+RA	4.66	64.18	49.88	67.38
F+PA	4.70	65.15	50.90	65.50
F+PA+CaA	4.90	64.13	50.98	8.00
F+PA+CaB	6.12	63.30	48.08	22.50
F+PA+Mg	5.58	63.00	51.23	38.25
F+PA+Zn	4.77	64.08	54.05	66.00
F+PA+CaA+Mg+Zn	5.48	63.43	50.75	6.13
F+PA+CaB+Mg+Zn	6.49	63.00	51.45	16.75
F+RB	4.66	64.80	50.73	57.25
F+PB	4.70	65.45	51.58	52.00
F+PB+CaA	4.87	64.88	50.95	7.75
F+PB+CaB	6.12	62.63	51.83	20.13
F+PB+Mg	5.55	62.13	52.38	35.25
F+PB+Zn	4.73	65.20	51.83	49.25
F+PB+CaA+Mg+Zn	5.45	63.40	50.18	6.50
F+PB+CaB+Mg+Zn	6.52	61.13	53.95	15.25

F - Unenriched flour.
RA - Regular vitamins-reduced iron premix.
RB - Regular vitamins-ferrous sulfate premix.
PA - NRC-level vitamins-reduced iron premix.
PB - NRC-level vitamins-ferrous sulfate premix.
CaA- $CaSO_4$ Mg - MgO
CaB- $CaCO_3$ Zn - ZnO

[a]LSD = 0.087 [c]LSD = 1.943
[b]LSD = 0.780 [d]LSD = 3.609

(Calculation was based on 5 % level)

Beside the regular vitamin-iron premix, the flour also was fortified with calcium, magneasium and zinc, according to the NRC proposal.

Generally, there are two forms of calcium used in enrichment, $CaSO_4$ and $CaCO_3$. These two calcium sources, and also magnesium and zinc were added to both NRC premixes separately, because of the large bulk of the Ca and the unknown effect of the minerals on flour. Adding the minerals separately would help determine what changes, if any, were produced by each addition.

The data indicates that $CaSO_4$ was the better calcium source for fortification, because $CaCO_3$ increased the pH notably, darkened the flour color and lowered the viscosity. However, $CaSO_4$ affected only viscosity.

pH value increased after MgO or either calcium sources was added. Thus, when the full NRC proposed enrichment were added to the flour the flour pH was higher than that of the unenriched one. The data also indicates that the flour color was darkened by the adding of MgO or $CaCO_3$, alkaline water retention capacity was increased by the addition of either $CaCO_3$ or ZnO, and the viscosity was decreased by the addition of $CaCO_3$, $CaSO_4$ or MgO. Thus when the full NRC-level enrichments were added to the flour, the flour color was darkened, alkaline water retention was increased and viscosity was decreased.

The Comparison of Regular and NRC Proposed Enrichment

The physicochemical and baking tests were made on flours fortified with regular enrichemnt (with reduced iron) and the NRC proposed enrichment (with reduced iron and $CaSO_4$) separately. Reduced iron and $CaSO_4$ were found to be the better iron and calcium sources for fortification as noted in the previous results. The results of this comparison are presented in Table 7-16.

Flour Physicochemical Test: The results of the tests on physicochemical properties are presented in Table 7. The results indicate that the NRC proposed enrichment gave the flour a higher pH and lower viscosity than that of the regular enrichment.

Cake qualities are best if the pH of cake flour is in the range of 4.6-5.0 (44). The data indicates, however, that the NRC-level enrichment increased the pH to 5.48. This increase may be due to the addition of the basic minerals, $CaSO_4$ and MgO, and or to the interaction of minerals with protein (21). Ranhotra et al (37) also reported that the addition of MgO was found to raise bread pH.

The optimum range for flour MacMichael viscosity is 35-60 (44), but NRC enrichment decreased the viscosity to 6.13. Previous work by Bresson et.al (50) show that viscosity increases with the flour protein content and decreases with ash content. Some workers have noted an interaction between calcium, magnesium and zinc with protein (21,36,61,62) and, hence, the protein structure is degraded. This may explain why viscosity decreased when the NRC-level enrichment was added to the flour.

Baking Test: Baking tests were made to investigate the effects of regular enrichment and NRC-level recommended enrichment on the qualities of yellow layer cakes and chocolate cup cakes.

Table 8 and 9 show the effects of regular and NRC proposed enrichment on cake batter properties. These two enrichments did not cause a significant changes in the batter specific gravity, pH or color.

Table 8: The Effects of Regular and NRC-Level Enrichment on the Batter of Yellow Layer Cake

Samples	Specific Gravity[a]	pH[b]	Agtron Rd.[c]
F	0.908	7.145	35.75
F+REG Enrichment	0.888	7.190	38.38
F+NRC Enrichment	0.908	7.238	36.75

[a] LSD = 0.0762

[b] LSD = 0.0543 (Calculation was based on 5 % level)

[c] LSD = 4.9064

Table 9: The Effects of Regular and NRC-Level Enrichment on the Batter of Chocolate Cup Cake

Samples	Specific[a] Gravity	pH[b]	Agtron[d] Rd.
F	0.856	6.85	62.51
F+REG Enrichment	0.868	6.85	59.50
F+NRC Enrichment	0.885	6.90	57.52

[a]LSD = 0.062

[b]LSD = 0.130 (Calculation was based on 5 % level)

[c]LSD = 5.663

Regular or NRC-level enrichments had the same general effects on yellow layer cake qualities when judged by optical evaluation. With these two enrichments, the cakes had some tunnels, very good golden brown color and structural symmetry. The unenriched cakes had a little bit finer, firmer and more uniform grain than both the regular and NRC-level enriched cakes. Cakes with the NRC-level enrichments tended to be a darker yellow than the cake with the regular enrichment, and both of these enrichments made the cakes a darker yellow than the unenriched cakes. No difference in taste was noted by an informal bench panel.

Regular and NRC-level enrichment also had the same general effects on chocolate cup cake qualities judged by optical evaluation. Both enrichments gave the cakes some holes, very good dark brown color, structural symmetry, and a little peak on the top. However, the NRC-level enriched cakes had smaller peaks than the others. No difference in taste was noted by an informal bench panel.

Table 10 and 11 show data on yellow layer cakes and chocolate cup cakes, respectively. The data indicates that these two enrichments did not significantly change such cake qualities as cake weight, volume, pH, firmness and color. The higher pH in the NRC-level enriched flour may be masked or buffered by the other ingredients in the cakes. In Yamazaki's previous study (41), cake volume was found to decreased when MacMichael viscosity increased. In this study, the low flour viscosity did not significantly affect the cake volume. This suggests that the criterion ranges of cake flour pH and viscosity may not need to be strictly limited.

The investigations of the properties of yellow layer cake also carried our after three and six days's storage of the cake at room temperature.

Table 10: The Effects of Regular and NRC-Level Enrichment on One Day Old Yellow Layer Cakes

Samples	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d] (gm)	Agtron[e] Rd.
F	384.75	1352.50	7.48	161.25	32.88
F+REG Enrichment	385.50	1358.75	7.47	168.08	32.08
F+NRC Enrichment	385.25	1377.50	7.48	167.85	32.05

[a]LSD = 5.128

[b]LSD = 74.105

[c]LSD = 0.076

[d]LSD = 20.562

[e]LSD = 8.909

(Calculation was based on 5 % level)

Table 11: The Effects of Regular and NRC-Level Enrichment on One Day Old Chocolate Cup Cakes

Samples	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d]	Agtron[e] Rd.
F	30.3	106.0	8.93	103.21	16.5
F+REG Enrichment	30.3	106.5	8.85	116.47	16.5
F+NRC Enrichment	30.7	109.5	9.15	115.42	16.5

[a]LSD = 0.860
[b]LSD = 3.182
[c]LSD = 0.310
[d]LSD = 20.875
[e]LSD = 2.250

(Calculation was based on 5 % level)

Tables 12-16 show the results. The data indicates that the weight, volume, pH, firmness and color of the 3-day old or 6-day old yellow layer cakes fortified with NRC-level enrichment were indistinguishable from those of the regular fortified cakes. The storage time of the cake at room temperature was considered to be very related to cake staling. After 3 days' storage, the firmness and pH of the NRC-level enriched cakes increased while the unenriched or regular enriched cakes did not show any change. After 6 days' storage time, not only the NRC-level enriched cakes, but also the unenriched and regualr enriched cakes showed an increase in firmness.

The taste of the yellow layer cakes fortified with either kind of enrichments also was indistinguishable from the other. An untrained panel of 40 volunteers failed to detect any off-flavor taste in the one-day old yellow layer cake fortified with NRC-level enrichment. The NRC-level enriched chocolate cup cake was even preferred to either unenriched or regular enriched one by the panel. After the yellow layer cakes were stored for three and six days, a taste test also was done. The taste test panel detection of off-flavor in the cakes fortified with NRC-level enrichment was statistically non-significant at the 5 % level. NRC-level enrichment was, therefore, found to be very acceptable according to these results.

The Effects of Storage on the Flour Fortified with NRC-Level Enrichment

Varying storage conditions, either length of storage or temperature may adversly affect flour qualities. In order to investigate the effects of various storage conditions on the qualities of the NRC-level enriched flour, the fortified flours were stored at three different temperature (0^oC, 25^oC and 40^oC) for three months and six months, separately.

Table 12: The Effects of Regular and NRC-Level Enrichment on 3-Day Old Cakes

Samples	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d] (gm)	Agtron[e] Rd.
F	383.75	1357.50	7.51	175.55	33.88
F+REG Enrichment	385.00	1341.25	7.53	184.80	34.70
F+NRC Enrichment	383.25	1375.00	7.59	192.53	30.88

[a]LSD = 4.094

[b]LSD = 54.746

[c]LSD = 0.064

[d]LSD = 15.456

[e]LSD = 19.059

(Calculation was based on 5 % level)

Table 13: The Effects of Regular and NRC-Level Enrichment on 6-Day Old Cakes

Samples	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d] (gm)	Agtron Rd.
F	381.50	1333.75	7.49	205.00	29.73
F+REG Enrichment	384.50	1307.50	7.48	205.68	26.10
F+NRC Enrichment	384.25	1358.75	7.53	224.70	28.05

[a]LSD = 4.485

[b]LSD = 52.680

[c]LSD = 0.055

[d]LSD = 35.875

[e]LSD = 18.286

(Calculation was based on 5 % level)

Table 14: Staling of the Unenriched Cakes

Days	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d] (gm)	Agtron[e] Rd.
1	384.75	1352.50	7.48	161.25	32.88
3	383.75	1357.48	7.51	175.55	33.88
6	381.50	1333.82	7.49	205.00	29.73

[a]LSD = 3.936

[b]LSD = 54.421

[c]LSD = 0.033

[d]LSD = 33.975

[e]LSD = 10.637

(Calculation was based on 5 % level)

Table 15: Staling of the REG Enriched Cakes

Days	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d] (gm)	Agtron Rd.
1	385.48	1358.75	7.46	168.08	32.08
3	385.02	1341.25	7.53	184.80	34.70
6	384.53	1307.50	7.48	205.68	26.10

[a]LSD = 5.592 [d]LSD = 21.457

[b]LSD = 71.456 [e]LSD = 18.719

[c]LSD = 0.098

(Calculation was based on 5 % level)

Table 16: Staling of the NRC Enriched Cakes

Days	Weight[a] (gm)	Volume[b] (ml)	pH[c]	Firmness[d] (gm)	Agtron[e] Rd.
1	385.25	1377.50	7.50	167.85	32.05
3	383.25	1375.00	7.59	192.53	30.88
6	384.25	1358.75	7.53	224.70	28.05

[a]LSD = 4.656 [d]LSD = 21.807

[b]LSD = 56.534 [e]LSD = 17.703

[c]LSD = 0.0497

(Calculation was based on 5 % level)

Tables 17-19 compare the results of the stored flour to the control, which was also fortified with NRC-level enrichment but without storage.

Flour Physicochemical Test: The results of the flour physicochemical properties determination after the various storage are presented in Table 17.

Storage temperature caused many more variations in moisture content than storage time. Moisture content increased as the storage temperature decreased. Flour stored at both 0°C and 25°C had a higher moisture content than the control. Flour stored at 40°C had the lowest moisture content, which was a little bit lower than the control, when stored for three months. When the flour was stored for 6 months at 40°C, however, the moisture content was significantly much lower.

Flour pH decreased as the storage temperature increased. The pH of the control flour was 5.20. When the flour was stored for 3 months, the pH decreased from 5.18 at 0°C to 4.98 at 40°C. The pH of the flours stored at the same temperature, but for different lengths time, was indistinguishable. This result suggests that storage time does not affect flour pH as much as storage temperature.

Flour color was not affected by being stored for three months, at 0°C and 25°C. However it changed when stored for three months at 40°C. When the storage time was increased to 6 months, the color became darker. The color also became darker as the storage temperature was increased. Both storage temperature and time were found to affect flour color.

Storing for three months at 40°C caused no notable effect on alkaline water retention capacity. If, however, the flour was stored for six months at 40°C, alkaline water retention capacity became higher than the control,

Table 17: The Effects of Storage Temperature and Time on the Flour Physicochemical Properties

Samples	Moisture[a] Content	Grouping*	pH[b]	Grouping*	Agtron[c] Rd.	Grouping*	AWRC[d]	Grouping*	MacMichael[e] Viscosity	Grouping*
Control	9.89	C	5.20	A	66.13	A	58.45	B	6.75	A
300	16.57	A	5.18	A	66.20	A	45.70	D	7.63	A
325	12.27	B	5.09	B	65.75	A	52.20	C	6.63	A
340	9.43	D	4.98	C	62.25	C	57.08	B	6.50	A
600	16.79	A	5.05	B	62.93	BC	44.48	D	7.00	A
625	12.58	B	5.10	B	63.80	B	51.90	C	6.75	A
640	6.33	E	4.99	C	59.18	D	66.05	A	6.50	A

* Values with the same letter are not significantly different at 5% level.

300, 325, 340 is the flour stored for 3 months under 0^oC, 25^oC, 40^oC separately.

600, 625, 640 is the flour stored for 6 months under 0^oC, 25^oC, 40^oC separately.

$^{a}LSD = 0.371$

$^{b}LSD = 0.0562$

$^{c}LSD = 1.470$

$^{d}LSD = 1.564$

$^{e}LSD = 1.798$

As the storage temperature decreased, the alkaline water retention capacity was decreased. Three months' storage had a greater effect on decreasing the alkaline water retention capacity than six months.

The effects of storage temperature and time on flour odor was detected by an panel of 40 untrained volunteers. All the stored flour, according to the panel, except for the flour stored at $0^{o}C$ for six months, had no significant off-flavor, compared to the control. High moisture levels may lead to release of free fatty acids in flour. In Cuendet's previous work (63), high percentages of free fatty acids were produced at 14% moisture content, and this led to off-odors in the flour. Because the moisture content of the flour stored at $0^{o}C$ for six months increased to 16.69% in this study, the off-flavor may have been caused by the degradation of fats at such high moisture levles.

Baking Test: Baking tests were conducted with the stored flour because although the results of physicochemical tests provided some information about the flour qualities, the results still cannot be used to predict the cake qualities.

The optical evaluation was carried out on the cakes made from each of the flours stored at $0^{o}C$, $25^{o}C$ and $40^{o}C$ for three months. No notable difference was found in symmetry and color, compared to the control. But, the cakes made from stored flour had a finer, more uniform grain with more tunnels than the control. Cake made from flours stored for six months at the different temperatures showed no notable difference in cake color. Grain was reasonably uniform except in the cakes made from flour stored at $40^{o}C$ storage temperature. Cakes made from the flour stored at $40^{o}C$ were a bit more dense, coarse, and more open. No off-flavor was detected in any of the cakes by an informal bench panel.

Table 18 shows the effects of flour storage on the qualities of cake batter. When the flour was stored for three months, the different storage temperatures caused no significant effects on batter specific gravity, pH or color. When the flour was stored for six months, the flour stored at 40°C caused an increase in batter specific gravity and batter acidity. The color of batter made from flour stored at 25°C was not significantly different from the control. But, if the flour had been stored at temperatures lower than 25°C, the color was lighter; if the temperature was higher than 25°C, the color was darker.

Table 19 shows the effects of different storage temperature and time on cake qualities. The data indicates that the different storage temperatures caused no significant effects on all the cake qualities, including cake weight, volume, pH, firmness or color, if the flour was stored for three months.

When the storage time increased to six months, effects on cake volume and firmness became significant. The flour stored for six months, regardless of the storage temperature, caused a significantly lower cake volume than the control one. But flour storage temperature of 25°C decreased cake volume less than either 0°C or 40°C. Cake firmness increased as the flour storage temperature increased. The flour stored at 25°C caused no significant change in firmness. However, when temperature was lower than 25°C, the firmness decreased; if the temperature was higher, the firmness increased.

Viscosity has a relationship with cake volume according to Yamazaki (41).

Table 18: The Effects of Flour Storage
on the Cake Batter

Samples	Specific Gravity[a]	Grouping*	pH[b]	Grouping*	Color[c]	Grouping*
Control	0.925	B	7.22	A	41.00	BC
300	0.910	B	7.26	A	40.25	C
325	0.908	B	7.20	A	38.85	C
340	0.923	B	7.21	A	38.03	C
600	0.900	B	7.21	A	49.45	A
625	0.925	B	7.27	A	45.00	AB
640	0.965	A	7.04	B	29.13	D

*Values with the same letter are not significant different at 5 % level.

300,325,340 is the flour stored for 3 months at $0^{o}C$,$25^{o}C$,$40^{o}C$ separately.

600,625,640 is the flour stored for 6 months at $0^{o}C$,$25^{o}C$,$40^{o}C$ separately.

[a]LSD = 0.00040 [b]LSD = 0.00148 [c]LSD = 4.48100

Table 19: The Effects of Flour Storage on the Cake Qualities

Samples	Weight[a]	Grouping*	Volume[b]	Grouping*	pH[c]	Grouping*	Firmness[d]	Grouping*	Agtron[e] Rd.	Grouping*
Control	385.00	A	1425.00	A	7.55	A B	176.00	A B	33.58	A
300	384.74	A	1408.75	A B	7.53	B	181.38	A B	32.80	A
325	386.25	A	1410.00	A B	7.53	B	184.53	A	31.58	A
340	387.50	A	1437.50	A	7.52	B	188.85	A	32.60	A
600	384.75	A	1326.25	C	7.55	A B	147.93	C	31.75	A
625	387.25	A	1376.25	A B C	7.61	A	162.83	B C	27.85	A
640	388.00	A	1352.50	B C	7.51	B	188.85	A	29.35	A

* Values with the same letter are not significantly different at 5 % level.
300, 325, 340 is the flour stored for 3 months at 0°C, 25°C, 40°C separately.
600, 625, 640 is the flour stored for 6 months at 0°C, 25°C, 40°C separately.

$^{a}LSD = 4.110$

$^{b}LSD = 65.309$

$^{c}LSD = 0.054$

$^{d}LSD = 18.913$

$^{e}LSD = 6.001$

Since storage did not affect flour viscosity, the reduction in cake volume in this storage study may not related to flour viscosity. Although alkaline water retention capacity was found to be highly correlated with the spread ratio of cookie, lower flour alkaline water retention capacity gave the cookie a higher spread ratio (40,41,43). This relationship does not appear to apply to cakes, since the cake volume decreased while the alkaline water retention capacity of the flour decreased. The relationship between alkaline water retention capacity and cake qualities needs further study. Reduction of cake volume probably can be explained by the increased specific gravity of the batter made from flour stored for six months. The higher specific gravity could result in a reduced ability to trap air, and thus, cake volume could decrease.

Storage temperature had more effect on acceptability than the storage period according to the taste panel results. Cakes made from flour stored at 40°C for both three and six months were judged unacceptable by the taste panel.

The Relationship of Slice Position to the Color and Firmness

The relationship of the slice position to the cake color and firmness was investigated. Table 20 presents the results.

Position 3, which is the central portion of the cake, had a significantly lighter color than the other portions. Firmness also was significantly related to the slice position. The central portion was most more firm than the others. Figure I shows the relation of slice position to color and firmness.

Table 20: The Effects of Slice Position on Cake Color and Firmness

Position	Agtron[a] Rd.	Firmness[b] (gm)
1	30.54	182.33
2	30.10	176.55
3	33.21	203.09

[a]LSD = 2.497

[b]LSD = 5.387

(Calculation was based on 5 % level)

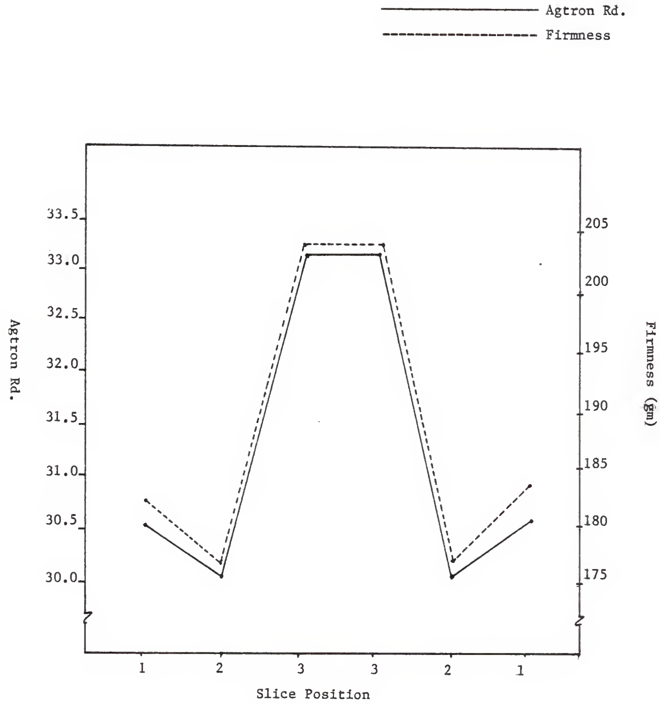

Fig I: The Relationship of Slice Position to the Cake Color and Firmness

SUMMARY

The effects of regular enrichment and NRC/NAS recommended vitamins and minerals on cake flour and cake qualities by physicochemical, baking, storage and tasting tests were compared. The feasibility of NRC/NAS recommended enrichment was thus determined.

From the data obtained, NRC recommended vitamins and iron prmix, with either reduced iron or ferrous sulfate as the iron source, was very acceptable. However, reduced iron seems to be the better iron source for fortification because it increased the alkaline water retention capacity less and reduced the flour viscosity less. Two forms of calcium sources are $CaSO_4$ and $CaCO_3$. $CaCO_3$ was found to increase the flour pH, darkened the flour color and lowered the flour viscosity while $CaSO_4$ affected only viscosity. These results indicated that $CaSO_4$ was the better calcium source than $CaCO_3$.

When the flour was fortified with NRC-level enrichment, a significant increase in flour pH and decrease in flour MacMichael viscosity was observed. However, these changes on flour by the new level enrichment did not affect the cake qualities. By the baking test and tasting test, no difference was detected on qualities of cakes enriched with NRC-level enrichment.

Storage of the NRC/NAS enriched baked producted at room temperature for few days, the time usually required to distribute the products form the producer to the consumer, had little adverse effect on the acceptability. However, after six days' storage, the cake volume decreased and the firmness increased, but these differences in cake qualities after storage were not detected by the taste panel.

The flours fortified to the NRC/NAS enrichment were stored at 0°C, 25°C and 40°C for 3 and 6 months separately to determine if this enrichment will affect the flour properties when stored under various storage conditions.

When the flour was stored at temperature below room temperature (25°C), the moisture content increased as the temperature decreased. But the increase became smaller as the storage time was increased. When the flour was stored at 40°C, higher than room temperature, the moisture content was decreased. As the storage time was increased the decrease in moisture content became greater.

Storage, regardless of the temperature or length of time, gave the NRC enriched flour a lower pH and darker color than the unstored flour. The effect became greater as the storage temperature and time were increased.

Although storage also decreased the alkaline water retention capacity of flour, the higher storage temperature and longer storage time lessened the effect.

From the data obtained, no significant effect on MacMichael viscosity of flour by the various storage conditions was found.

Off-flavor was detected by the odor test if the flour was stored for 6 months at 0°C. Off-flavor also was detected by the taste panel on cakes made from the flour stored at 40°C for either 3 or 6 months.

The effects of three months' flour storage, regardless of the storage temperature, on the cake qualities, including cake weight, volume, pH, firmness and color, were not significant. But when the storage time increased to six months, some effects on decreasing cake volume and increasing cake firmness were detected. 25°C had less effect on decreasing cake volume than either 0°C or 40°C. 25°C also showed no significant change in firmness. But, when temperature was lower than 25°C, the firmness decreased; if the temperature was higher, the firmness increased.

In summary, NRC/NAS recommended enrichment, which would provide essential nutrients for humans, appears to be technically feasible as applied to cake flour and cake making. The baking industry sould consider implementing the NRC/NAS recommendations because of their importance to public health.

APPENDIX

The Function of These Ten Nutrients (64-67)

1. Vitamin A:

   a. Maintenance of visual purple for vision in dim light.

   b. Stimulation of growth.

   c. Promotion of fertility (reproduction).

   d. Health of epithelial cells.

   e. Other functions: release of proteolytic or protein-splitting enzymes; effective in the synthesis of the hormone corticosterone from cholesterol in the adrenal cortex.

   Vitamin A deficiency symptoms include night blindness, changes in the eye, skin, gastrointestinal tract and nervous tissue, respiratory infections and failure of tooth enamel (64).

2 Thiamin:

   a. As part of the enzyme thiamin pyrophosphate or cocarboxylase, which is required in metabolism of carbohydrate, leads eventally to the release of energy and excretion of carbon dioxide and water.

   b. Activates transketolase, an enzyme involved in the direct oxidative pathway for metabolism of glucose.

   Thiamin deficiency may result in loss of appetite, failure of absorption, decreased muscle tone, mental depression and confusion, neurological changes and beriberi.

3. Riboflavin:

   a. A necessary reaction in the liberation of energy from glucose, fatty acid and protein within the cell mitochondrion.

   b. An essential part of the enzymes usually referred to as FMN (Flavin MonoNucleotide) and FAD (Flavine Adenine Dinucleotide).

c. Integral part of other enzymes specifically involved in the transfer of hydrogen atoms within the protein metabolism.

d. A source of the vitamin niacin in the body.

The lack of riboflavin will result ariboflavinosis, a condition known as cheilosis, in which the tongue becomes smooth and takes on a characteristic purplish red in condition of described as glossitis, growth retardation, reduced reproductive capacity, loss of hair (alopecia) and infiltration of blood vessels into the cornea of the eye.

4. Niacin:

a. A vital role in the release of energy from all three energy-building nutrients-carbohydrates, fats and proteins.

b. Part of the coenzymes, nicotinamide adenine dinucleotide phosphate (NASP), both of which can accept or release hydrogen atoms readily.

c. Involved in the synthesis of protein, fat and pentoses.

Niacin deficiency: In pellagra, the skin, the gastrointestinal tract, and the central nervous system are affected; dermatitis, diarrhea, and depression preceding death, irritability, headaches, and sleeplessness, loss of memory, hallucinations, delusions of persecution.

5. Vitamin B_6 (pyridoxine):

a. Functions as a coenzyme in the form of pyridoxal phosphate.

b. Necessary for the process of transamination and deamination.

c. Forms a precursor of porphyrin.

d. As part of the enzyme phosphorylase, facilitates the release of hydrogen from the liver and muscles as glucose phosphate.

e. Plays a role in the metabolism of the central nervous system.

f. Regulations the formation of the enzyme in the brain.

Deficiency symptoms were result of a low dietary intakes; microcytic hypochromic anemia in association with high serum iron; nervousness, irritability, insomnia and difficulty in walking.

6. Folic acid:

a. The transfer of single-carbon units.

b. Synthesis of the purines adenine and guanine, and the pyrimidine, thyamine, all part of the nucleic acids.

c. The conversion or oxidation of part essential amino acid.

A deficiency of folic acid results in various disorders including thalassemia to toxemia of pregnancy and rheumatoid arthritis, megaloblastic anemia and reduction of the production of leukocytes or white blood cells and hence the ability of the body to produce antibodies.

7. Iron:

a. Carrier of oxygen and carbon dioxide.

b. Blood cell formation and metabolism.

Anemia which is a deficiency of iron in quantity and/or quality of the red blood cells.

8. Calcium:

a. Bone and tooth formation.

b. Growth.

c. Blood clotting.

d. Catalyst for biological reactions.

e. Regulation of permeability of cell membrane and strontium uptake.

Deficiency of calcium will result in tetany and rigor.

9. Magnesium:

   a. Catalyst to several hundred biological reactions.

   b. Activates the production of ATP by oxidative phosphorylation.

   c. Crucial in cellular respiration.

   d. Influences protein synthesis through an influences on the arrangement of the protein-synthesizing organelles of the cell.

   e. One of the minerals involved in providing the proper enviroment in the extra cellular fluid of nerve cells to normal muscular contraction.

   f. Increase the stability of calcium in tooth enamel.

   g. Influence the secretion of thyroxine and the maintenance of normal basal metabolic role and facilitates adaption to cold.

   Deficiency symptoms: Diagnosed uncontrolled neuromuscular activity, cardiovascular and rectal system also are affected, with symptoms such as vasodilation and skin changes.

10. Zinc:

   a. Constituent of many enzymes involved in digestion and metabolism.

   b. Associated with the hormone insulin secreted by the pencrease and necessary for carbohydrate metabolism.

   c. Plays a role in the contraction and relaxation of muscle.

   d. Related to protein and nucleic acid synthesis.

   Zinc deficiency symptoms include poor growth, loss of appetite, inpaired testicular maturation, and parakertotic skin lesions and distal esophagusi irregularities in sulfur amino acid metabolism and diminished taste and smell.

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EFFECTS OF VARIOUS NUTRIENTS ON ORGANOLEPTIC AND PHYSICOCHEMICAL PROPERTIES OF FLOUR AND CAKE

by

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In line with the fortification policy for cereal-grain products proposed by the Food and Nutrition Board of the National Academy of Science-National Research Council (NRC/NAS), vitamin A, vitamin B_6 and folic acid plus calcium, magnesium and zinc have been added to cake flour and the most popular cakes in U.S., yellow layer cakes and chocolate cup cakes, in addition to the normal fortification with thiamin, riboflavin, niacin and iron. In view of this expanded list of nutrients, studies of technical feasibility were conducted on the cake flour and cakes to determine the uniformity, freedom from segregation and such influences on consumer acceptance as color, flavor and odor by flour physicochemical tests, baking tests, taste panel tests and storage tests.

Studies showed that the recommended NRC fortification levels caused an increase in flour pH and decrease in flour MacMichael viscosity, compared to normal cake flour. However, baking tests and taste panel tests found the cake qualities not to be affected. In fact, in some cases, 6-days old cakes enriched to NRC recommended levels had no adverse flavor as determined by a taste panel. These cakes, however, had less volume and were firmer, compared to the control.

The effects of various storage conditions ($0^{\circ}C$, $25^{\circ}C$ and $40^{\circ}C$ for three and six months respectively) on the NRC-level enriched flour was also investigated in this study. Storage temperature and storage time were shown to have some effects on flour physicochemical and cake qualities. Storage temperature is much more important than storage period as a factor causing variation in qualities. When the storage temperature was higher than room temperature, off-flavor was developed in the cake which could be detected by a taste panel, the acceptability was then decreased. Under normal storage

conditions, at room temperature and for less than six months, flour enriched to NRC recommended levels can be successfully utilized in cake-making.