Distribution of Vitamin A in Fortified Flours and Effect of Processing, Simulated Shipping, and Storage[1]

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ABSTRACT

Wheat and corn flours and corn meal were prepared containing nutrients according to the 1974 fortification proposal by the National Research Council, as modified by the Inter-Industy Committee of the American Bakers' Association, to take into account natural levels of nutrients in grains and commercial requirements and procedures. Tests were conducted on the distribution in flours of the stabilized vitamin A of the fortification mix, tendency of the vitamin A to segregate in pneumatic handling and shipping, and its stability during storage of flours for up to six months. The proposed fortification policy appeared to be technically feasible with respect to ability to distribute vitamin A satisfactorily in flours, stability of vitamin A at warehouse temperatures for up to six months, and resistance to segregation in handling and shipping. The recommended overage of 15% stabilized vitamin A for fortification of flours may be more than necessary under normal commercial conditions.

In 1974 the Food and Nutrition Board (NAS/NRC 1974a) proposed a new fortification policy for cereal grain products and suggested that before full implementation, products should be prepared and tested to answer questions on the technical feasibility. This article is a report on the effects of some milling and processing procedures on uniformity of distribution of a stabilized vitamin A added to flours, resistance of the vitamin A to segregation and handling losses, and stability of the vitamin A during storage. Studies on other fortification nutrients will be reported separately.

MATERIALS AND METHODS

Flours Used

White bread flours milled from mixed Kansas hard winter wheat in the Grain Science and Industry production mill were fortified with different iron sources (Table I) and labeled A and B. Soft wheat cake flour was a commercial product supplied by Acme-Evans Co., Indianapolis, IN. Corn meal and flour were degermed commercial products made from yellow corn by Lauhoff Grain Co., Crete, NE. Semolina was milled from durum wheat by the Peavey Co., Hastings, MN.

Fortification

Flours were fortified as proposed by the Food and Nutrition Board (NAS/NRS 1974a) with two exceptions. Iron fortification was not increased over present enrichment levels because of the opinion, later verified, that the proposed increase of iron in the enrichment standards would not be approved (Federal Register 1977). Vitamin A fortification was at the revised level (1.3 mg/lb of retinol equivalent) because the value in the original proposal was based on an erroneous calculation (Hepburn 1976). The calculated quantities of nutrients added were those suggested by the Inter-Industry Committee (Ranum 1976), intended to raise nutrient contents to the proposed fortification levels with overages of 5–20% to provide for normal product and manufacturing variations and storage losses. With respect to vitamin A, those values are: level to add, 5,000 IU/lb; NAS fortification level, 1.3 retinol equivalents/lb (4,333 IU); mean percent natural in fortification level, 0; mean overage, 15%.

The carrier for the vitamin-iron premix (Table I) was corn starch, about 40% of the premix weight. Premix formulated with iron as ferrous sulfate was added to white bread flour A at 15 g/cwt. The premix added to white bread flour B, corn flour, corn meal, and semolina (made from durum wheat) at 10 g/cwt contained iron in the electrolytically reduced form. Because of lower contents of certain nutrients in soft wheat flour and the fact that only two premix preparations were supplied, the latter premix was added to cake flour at 11 g/cwt. Calcium, as sulfate, and magnesium and zinc, as oxides, were added separately to the flours (Table I). Vitamin A in the enrichment premix was dry retinyl palmitate, type 250-SD (Hoffmann-LaRoche), a dry product containing preservatives with acacia, lactose, and fractionated coconut oil as principal ingredients of the matrix. Because of processing overages in supplement and premix, vitamin A added to flours was found by analysis to be about 25% higher than the proposed fortification level, except that in cake flour it was 34% higher, resulting from the greater amount of fortification mix used. We discovered after mixing that some semolina supplied as an unenriched product was in fact vitamin-iron enriched according to present commercial standards, causing levels of water-soluble vitamins and iron in the final flour product to be higher than intended, but vitamin A content was not affected.

TABLE I
Nutrient Fortification of Flours, g/100 kg

	Flour Type			
Ingredient[a]	White Bread A	White Bread B and Semolina	Cake[b]	Corn Flour and Meal
Thiamin mononitrate	0.565	0.565	0.621	0.565
Riboflavin	0.396	0.396	0.436	0.396
Niacinamide	4.62	4.62	5.08	4.62
Pyridoxine HCl	0.44	0.44	0.48	0.44
Folic acid	0.057	0.057	0.063	0.057
Vitamin A palmitate, 250-SD	4.4	4.4	4.8	4.4
Tricalcium phosphate	0.66	0.66	0.73	0.66
Reduced iron	···	2.52	2.77	2.52
Ferrous sulfate	9.64	···	···	···
Calcium sulfate	665.0	665.0	687.0[c]	694.0[c]
Magnesium oxide	44.0	44.0	66.0[d]	44.0
Zinc oxide	2.1	2.1	2.57[d]	2.1

[a]All except last three ingredients were added as a fortification premix in a corn starch base, approximately 40% of premix weight.
[b]Premix level adjusted because of lower B-vitamin content of soft wheat cake flour.
[c]Adjusted because of lower natural calcium content of these flours.
[d]Adjusted because of lower natural magnesium and calcium content of this flour (Ranum 1976).

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Mixing of Flours

To test the effects of processing procedures, simulated shipping and handling, and storage on segregation and stability of the added vitamin A, the six flours, in 100-lb or 200-lb lots, were mixed in a Wenger double-ribbon horizontal batch mixer. Vitamin and mineral enrichment materials were premixed with 5 lb of flour by "fluidizing" (shaking) in a closed plastic bag filled with air. Equally divided portions of the premix were distributed on top of each 50 lb of flour after it was spread out in the mixer. Flour and premix were blended for 15 min. Samples for the initial analysis (0-time) were taken at the beginning, middle, and end of discharge of flour from the bottom of the mixer.

Storage

The fortified flours were stored up to six months under refrigeration (4°C), under warehouse conditions (temperature range 20–32°C, mean 24°C), and at 40°C. Samples for vitamin A determinations were taken after mixing (0-time), and after one, three, and six months of storage. Determinations were in triplicate or more at 0-time, and usually in duplicate at later storage times. Data in Table II are averages of the replicated determinations.

Pneumatic Conveying

To determine whether pneumatic conveying of prepared flours might cause segregation or loss of vitamin A in the system, a section of the production mill was cleaned as completely as possible and flushed with fortified flour. In a continuous operation, about 40 lb of the same fortified flour was elevated pneumatically four floors, collected in a cyclone, passed through an air lock and allowed to flow by gravity two floors lower in the mill. Triplicate samples were taken for analysis from the first 3–4 lb of flour collected, from the middle, and from the end of the run.

Simulated Shipping and Handling

To test effects of shipping and handling on segregation of vitamin A in fortified flour, 20-lb paper bags of flour were stacked on end firmly together in a specially constructed shaker mounted on rollers that passed over small rods to simulate the jarring effect of rail and highway joints. The stroke of the shaker was about 4¾ in., and it was operated at a rate of about 158,000 complete cycles and 624,000 jolts per 30-hr test period. Each bag was handled four times before starting the shaking and four additional times during brief interruptions in the shaking period, during which each bag was laid flat.

At the end of the test period, bags were laid flat on a table and cut open; six approximately 1-lb samples were taken from right and left sides at the top, middle, and bottom sections of each bag.

A Mill Run

For a test of distribution of nutrients in flour under typical milling conditions, flour was made in the production mill, with the fortification materials added through a Sterwin enrichment feeder. Because of the bulk of materials added, the feeder had to be operated at an accelerated rate of 56–59 g/min. A premix of fortification materials was prepared by "fluidizing" a part of the calcium sulfate and the other mineral and vitamin additions and blending that with the rest of the calcium sulfate in a tumbler.

Fortified flour was produced in the mill for 1 hr to stabilize production. Then flour was made for an additional hour, with samples drawn for analysis at 10-min intervals just before the final sifting stage and from every second 50-lb bag of flour sacked from the final bagging scale. Results at the end of the test period showed that flour was produced at a rate 10% greater than expected, which decreased the intended fortification level in flour.

Analytical Methods

The method for determining vitamin A was that of the Association of Official Analytical Chemists (1975), modified slightly by addition of sodium sulfite and ascorbic acid before the alkaline hydrolysis, which appeared to increase some analytical values slightly and make extractions a little easier.

For the storage tests, carotene and monohydroxy carotenoids were determined on corn flour and corn meal samples by alumina column chromatography, eluting those pigments with 4 and 15% acetone-in-hexane, respectively (AOAC 1975). Those analytical values were converted to retinol equivalents (NAS/NRC 1974b). In other tests, carotenoids in corn flour and meal were not separated, but in calculating vitamin A values, corrections were made for color produced by carotenoids with $SbCl_3$ (Kimble 1939).

Moisture was determined on all flours analyzed for vitamin A in the storage tests, and results were corrected to the same moisture basis as that of the 0-time samples. The correction amounted to 5–7% in samples stored at 40°C.

For statistical comparisons of data among flours on the same basis, vitamin A values for cake flour were reduced by 10% to correct for the 10% extra addition. Standard methods (Snedecor and Cochran 1967) were used to test significance and calculate coefficients of variation, which are used as measures of uniformity and/or precision.

RESULTS AND DISCUSSION

Stability of Vitamin A in Stored Flour

The coefficient of variation of replicated determinations of vitamin A in the six batch-mixed fortified flours at 0-time was 4.70%, indicating satisfactory distribution of vitamin A. Changes in vitamin A content during storage of flours are shown in Table II. Differences due to time and temperature are highly significant. The few aberrations in the three-month and six-month data (up to about 10%), possibly due to sampling, are not infrequently encountered in work of this type. No nutritionally meaningful changes occurred in vitamin A content of the wheat flours stored at 4°C, but vitamin A tended to decrease somewhat in corn flour and meal. Losses of vitamin A tended to be greater during storage at warehouse conditions (21–32°C) than at 4°C. The loss was 11% or less at three or six months in wheat flour, but in corn flour and meal it was more than 20%. At 40°C, losses of vitamin A were definite

TABLE II
Changes in Vitamin A Contents[a] of Flours During Storage

Flour	Temperature (C°)	Storage Time 0	Months 1	3	6
White bread A[b]	4	5,330	5,325	5,163	5,586
	21–32 (24)[c]		5,000	4,581	4,899
	40		4,630	3,481	2,918
White bread B[d]	4	5,370	5,120	5,052	5,481
	21–32 (24)		4,890	4,798	5,010
	40		4,640	3,594	2,830
Cake[d,e]	4	5,815	5,204	5,243	5,874
	21–32 (24)		4,942	4,799	5,291
	40		4,710	3,662	3,059
Semolina[d,f]	4	5,080	5,155	5,011	5,327
	21–32 (24)		4,380	4,077	4,488
	40		4,170	3,174	2,789
Corn[d,g]	4	5,624	5,250	5,097	4,725
	21–32 (24)		4,852	4,303	4,310
	40		4,425	3,493	2,392
Corn meal[d,g]	4	5,430	4,600	4,820	4,757
	21–32 (24)		4,508	4,333	4,112
	40		4,540	3,345	2,290

[a] IU/lb.
[b] Iron in form of ferrous sulfate.
[c] Range and mean temperature in warehouse.
[d] Iron in form of electrolytic reduced iron.
[e] Made of soft wheat. Data adjusted in statistical analysis for extra vitamin A added.
[f] Made of durum wheat.
[g] Degermed.

even at one month, and at six months, 50% was lost in all flours.

In corn grits containing spray-dried vitamin A palmitate stored at 23°C and 50% relative humidity, Carroll et al (1977) noted that vitamin A dropped rapidly during the first month but stabilized after three months at 40–50% of the original value. Cort et al (1976) and Rubin et al (1977) reported essentially no losses of stabilized vitamin A (type 250-SD) in flour during six months of storage at room temperature, but 20% loss in three months at 45°C. In corn meal, 90% of the vitamin A was retained in six months at room temperature, but only 67% in three months at 45°C. For corn grits, retention of vitamin A was about 82% in six-month storage at room temperature.

Our data, supported by those of Cort et al (1976) and Rubin et al (1977), indicate that the Inter-Industry Committee's recommendation of a 15% mean overage of vitamin A in wheat flour fortification would allow the declared vitamin A content to be delivered in flours stored for at least six months under warehouse conditions. Corn flour and meal would have a vitamin A value less than 10% below the declared value under those conditions. But use of stabilized vitamin A in flours is of little value when storage is three or six months at 40°C.

Carotenoids

The vitamin A contribution from carotenoids receives little overall attention in the fortification program because wheat and many other grains contain only traces at most. But in yellow corn products it could affect the amounts of vitamin A fortification used. We used yellow corn flour and meal, which contain measurable quantities of natural carotenoids with provitamin A activity. Changes in carotene and monohydroxy carotenoid (cryptoxanthin) were followed during the storage tests. Some of the carotene isolated was not *trans-β*-carotene, which has the highest provitamin A activity, and some monohydroxy carotenoids did not have provitamin A activity; nevertheless, the data reflect overall changes in vitamin A activity from provitamin A sources during storage.

Results are in Table III. Carotene contributed 50–70% of the calculated vitamin A value from carotenoids in most samples, but the carotenoids with provitamin A activity contributed only 14–20% of the total vitamin A value in the 0-time samples. In these storage tests, percentage loss of vitamin A value from carotenoids was approximately twice that from the stabilized vitamin A.

TABLE III
Changes in Vitamin A Activity[a] from Carotenoids of Corn Flour and Corn Meal During Storage

Product	Temperature (C°)	Storage Time 0	Months 1	3	6
Corn flour	4	255	228	260	256
	21–32 (24)[b]		213	189	197
	40		196	119	60
Corn meal	4	425	300	296	307
	21–32 (24)[b]		268	253	225
	40		229	139	77

[a] Expressed as retinol equivalents per pound.
[b] Range and mean of temperature in warehouse.

Pneumatic Conveying

Vitamin A contents of samples collected after pneumatic handling of flours are shown in Table IV. Coefficients of variation of vitamin A content of the different flours, based on averages of duplicates at each collection stage corrected for differences in moisture content, ranged from 0.5 to 6%. These are acceptable limits considering that the variations could result from original mixing, conveying, sampling, analysis, or combinations of those factors. Possibly the increased concentrations of vitamin A resulted from loss of some flour dust that did not contain the heavier or differently charged particles of the vitamin A product. Based on these data, an overage of vitamin A would not be required to guard against losses in pneumatic handling.

TABLE IV
Effect of Pneumatic Conveying on Segregation or Loss[a] of Vitamin A in Flours

Flour	Before Conveying	Collection Stage: First	Middle	Last	Coefficient of Variation[b] (%)
White bread A	5,126	5,638	5,460	5,437	0.5
White bread B	5,156	5,865	5,913	5,531	0.5
Cake	5,755	6,008	6,280	5,814	6.1
Semolina	5,516	6,038	6,257	6,618	1.5
Corn	5,014	5,426	5,180	5,385	0.5
Corn meal	5,692	5,539	5,526	6,214	6.0

[a] IU/lb.
[b] Based on duplicates for each collection.

Simulated Shipping and Handling

Under the handling conditions imposed in this study, no significant tendency for segregation of vitamin A in the bags of fortified flours was apparent (Table V). Coefficients of variation of averages of duplicate determinations of vitamin A at three levels on the two bags of each flour were from 2 to 4.4%, with an overall average of 3%. Flours became compacted rapidly, so little separation was expected. Although semolina and corn meal did not compact as tightly as other flours, vitamin A showed no greater tendency to segregate.

In tests using a laboratory vibrating table to simulate shipping conditions, Carroll et al (1977) found that spray-dried vitamin A palmitate applied to corn grits packaged in round commercial cylinders tended to segregate toward the bottom; the range of values at three levels in the package was 40% of the total vitamin A found.

Distribution of Vitamin A in Flour in a Typical Milling Operation

Vitamin A contents of flour to which the fortification mix was added during a flour production run are shown in Table VI. The coefficient of variation of samples drawn at 10-min intervals was 3.8% and on samples from 10 bags 1.8%. The average content of vitamin A in the flour was 103% of that calculated from the fortification mix placed in the Sterwin feeder. Thus no overall loss of vitamin A was found, and distribution was satisfactory in the milling-fortification operation.

Our studies indicate that the proposed fortification policy for

TABLE V
Effect of Simulated Shipping and Handling on Segregation of Vitamin A[a]

Flour	Bag	Samples[b]: Top	Middle	Bottom
White bread A	1	5,040	4,264	4,887
	2	5,044	5,044	5,269
White bread B	1	5,066	4,954	5,066
	2	5,475	5,291	5,336
Cake	1	5,061	5,156	5,246
	2	5,901	5,709	5,615
Semolina	1	5,566	5,397	5,486
	2	5,230	5,123	5,262
Corn	1	5,837	6,039	5,956
	2	6,065	6,136	6,448
Corn meal	1	7,060	7,006	6,953
	2	6,488	6,674	6,674

[a] IU/lb.
[b] Average of samples from left and right halves at each level.

TABLE VI
Uniformity of Additions of Vitamin A in Fortification Mix to Flour in Typical Mill Operation

Sample	Vitamin A (IU/lb)[a]
At 10-min intervals	
1	4,975
2	4,954
3	5,066
4	4,539
5	4,843
6	4,887
From bag No.	
1	4,843
2	4,777
3	4,712
4	4,887
5	4,777
6	4,843
7	4,975
8	4,712
9	4,787
10	4,887

[a]Average of duplicate determinations.

cereal grain products is technically feasible in that vitamin A fortification can be distributed satisfactorily in flours; vitamin A is sufficiently stable in flour at warehouse temperatures for up to six months; and it has no apparent tendency to segregate during pneumatic handling or shipping in bags. The 15% overage on vitamin A suggested by the Inter-Industry Committee (Ranum 1976) may be more than is needed in wheat flour under normal commercial conditions.

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