Nutrient Composition of Selected Wheats and Wheat Products. VII. Total and Free Niacin[1]

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ABSTRACT

A total of 156 samples were analyzed for their content of total and free (unbound) niacin. The samples included 10 types of wheat foods obtained from each of 2 cities in each of 5 geographical areas of the country (100 samples) and an additional 56 samples comprised of 11 wheats, flour milled from those wheats, and products prepared from the flours. Total niacin was measured according to a conventional procedure. Free niacin was determined by *Lactobacillus plantarum* assay of aqueous extracts. Most niacin of enriched products was found in the free form (85 to 90%), whereas that of unenriched products was approximately one-half free niacin. The distribution of niacin was consistent for commercial products, and showed no variation with geographical source. Although both total and free niacin of wheat was reduced by milling, a greater proportion of free niacin was retained in each flour. Moreover, the proportion of niacin in the free form was higher in bread, cakes, and crackers than in the flours from which they were prepared.

This work is part of a study to determine the nutrient composition of typical wheat foods in geographical regions of the U.S. and to measure the effect of normal processing of grain to flour and of flour to product on the distribution of nutrients. Nutrients studied include: total ash and 15 mineral elements; protein and 18 commonly occurring amino acids; 5 carbohydrate fractions; total lipids and 15 individual fatty acids; and vitamins B-6, E, thiamine, riboflavin, and niacin. The purpose of this paper is to present results of analyses for total and free niacin.

The presence of a bound form of niacin in cereal grains was demonstrated by several groups of workers in the early 1940's (1,2,3). That of wheat was shown to be soluble and resistant to hydrolysis in water and dilute (1/10 N) acid, but was attacked by treatment with stronger acid and was extremely labile to alkali, under even mild conditions. Unless hydrolyzed, the bound form was found to be unavailable to microorganisms, rats and pigs, and possibly to man (4,5,6). Das and Guha (7) found as further evidence of its limited biological usefulness that the bound form could not be released by pepsin. Ghosh et al. (8) suggested that the ability of a foodstuff to improve a nicotinic acid deficiency would be determined by its content of free nicotinic acid and nicotinamide, not by the content of bound niacin.

The seriousness of the question prompted the attempt in this present study not only to determine niacin by the conventional procedure but also to estimate the degree to which the niacin may exist in free and bound forms.

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MATERIALS AND METHODS

The samples, and details of their collecting and processing, have been described previously (9). Briefly, 100 of the samples represented commercial wheat-food products. Composite samples of 10 types of food products were collected at the retail level in each of 2 cities in each of 5 geographical regions of the U.S. Products selected were: all-purpose flour, biscuit mix, breakfast cereals (whole-grain for cooking, wheat flakes, and shredded wheat), enriched white breads (conventional, prepared by the sponge-and-dough process, and continuous-mix), whole-wheat bread, hamburger rolls, and cake doughnuts.

The remaining 56 samples were selected to study changes in composition from grain to food product. These included: bread flours milled from 5 hard wheat blends; semolinas from 2 durum wheat mixes; and one or more cake flours plus one or more flours suitable for cracker manufacture from each of 4 soft wheat blends. Bread was prepared from the hard wheat flours by both sponge-and-dough and continuous-mix procedures; macaroni, from the durum semolinas; and either cake or crackers, from the appropriate soft wheat flours.

Microbiological determination of niacin was considered to be preferable to the chemical procedure because of the complications introduced in the latter by chromogenic substances. *Lactobacillus plantarum* was selected as the test organism because it has been shown to respond equally to both the free acid and amide forms of the vitamin.

Total niacin was estimated following AACC Method 86-51 (10). Preliminary tests with selected wheat-product types showed that the conditions employed for extraction of niacin (1N H_2SO_4, for 30 min. at 15 p.s.i.) were sufficient for complete release. Alkali extraction or treatment of the acid extract with sodium hydroxide gave identical or, with some samples, only slightly greater niacin values. Use of the official method had the obvious advantage for practical application that results could be compared to existing reference data.

The choice of methods for extracting free niacin was complicated by the observation that apparent results increased with severity of treatment. Trials with water and dilute acid as extractants showed that values tended to increase with both time and temperature of treatment. Although bound niacin is reported to be resistant to hydrolysis under such mild conditions, it was concluded that gradual breakdown does occur during extraction. It was decided to use the least stringent conditions possible for estimating free niacin. Samples were suspended in distilled water and were stirred continuously for 30 min. with a magnetic stirrer. Values so obtained tended to be lower than those obtained after autoclaving with N/10 acid, but results were found to be consistent in replication. Furthermore, niacin added in enrichment mixtures appeared to be fully recovered by the method.

RESULTS AND DISCUSSION

Results for the 100 consumer products are summarized in Table I. In general, most niacin of enriched products was found in the free form (85 to 90%), whereas that of unenriched products was divided approximately equally between free and bound forms. The relatively small standard deviations illustrate the consistency of results for each type of product. No pattern was exhibited to suggest a correlation

TABLE I. TOTAL AND FREE NIACIN CONTENT OF SELECTED WHEAT FOODS
(Averages of ten samples per product and standard deviation)

Product	Total Niacin	Free Niacin	
	mg./100 g.[a]	mg./100 g.[a]	% of total
All-purpose flour	4.72 (±0.68)	4.22 (±0.48)	89.7 (±3.8)
Biscuit mix	3.49 (±0.25)	2.93 (±0.31)	83.9 (±4.8)
Cereal to be cooked	5.27 (±0.68)	2.68 (±0.26)	51.2 (±4.4)
Shredded wheat	5.38 (±0.23)	2.55 (±0.11)	47.5 (±2.3)
Wheat flakes	5.55 (±0.48)	3.35 (±0.21)	60.7 (±5.2)
White bread, conventional	3.98 (±0.36)	3.32 (±0.28)	83.4 (±2.7)
White bread, continuous-mix	4.17 (±0.34)	3.68 (±0.28)	88.5 (±5.4)
Whole-wheat bread	5.84 (±0.75)	3.20 (±0.52)	54.7 (±3.8)
Hamburger rolls	3.98 (±0.24)	3.40 (±0.40)	85.2 (±6.6)
Cake doughnuts	0.66 (±0.03)	0.41 (±0.04)	62.5 (±6.9)

[a]Dry basis.

between niacin content and geographical source. Average results for the wheat-flour-product samples are arranged in Table II to facilitate comparison of processing steps. In general, about one-half of the niacin of wheats was present as free niacin. Although milling greatly reduced both niacin values, a greater proportion of the free form was retained in each flour than was present in the original wheat. Data for air-classified cake flours were combined with those of conventionally milled short patent flours because of their similarity in niacin distribution. Small differences were evident in the remaining soft-wheat flours, and

TABLE II. TOTAL AND FREE NIACIN CONTENT OF WHEAT-FLOUR-PRODUCT SAMPLES
(Averages per sample type and standard deviations)

Type and Number of Samples	Total Niacin	Free Niacin	
	mg./100 g.[a]	mg./100 g.[a]	% of total
HRW and HRS wheats (5)	7.44 (±1.67)	3.55 (±0.86)	47.6 (±2.6)
Bread flours (4)[b]	1.55 (±0.29)	1.02 (±0.20)	65.6 (±3.4)
Bread, sponge dough (4)	2.09 (±0.41)	1.58 (±0.34)	75.5 (±2.4)
Bread, continuous-mix (4)	2.08 (±0.39)	1.55 (±0.33)	74.3 (±3.8)
Durum wheats (2)	11.14 (±1.11)	4.67 (±0.17)	42.0 (±2.3)
Semolina (2)	3.88 (±0.30)	2.16 (±0.56)	55.2 (±10.2)
Macaroni (2)	4.42 (±0.42)	2.14 (±0.42)	48.2 (±5.0)
SRW and SWW wheats (4)	7.22 (±0.26)	3.77 (±0.10)	52.2 (±2.8)
Cake flours (6)[c]	1.01 (±0.10)	0.56 (±0.14)	55.4 (±13.7)
Cakes (6)	0.39 (±0.03)	0.26 (±0.03)	67.9 (±4.0)
Straight-grade flours (3)	1.28 (±0.11)	0.79 (±0.08)	61.2 (±2.4)
Crackers from straight flour (3)	0.83 (±0.13)	0.66 (±0.05)	73.5 (±10.5)
Cut-off flours (2)	1.64 (±0.26)	0.88 (±0.27)	53.2 (±7.9)
Crackers from cut-off flour (2)	1.20 (±0.28)	0.78 (±0.19)	65.4 (±0.05)
Flour, air-classified, for cracker (2)	1.12 (±0.08)	0.70 (±0.01)	63.1 (±4.1)
Cracker from air-classified flour (2)	0.88 (±0.21)	0.68 (±0.01)	78.2 (±17.3)

[a]Dry basis.

[b]Excluding one sample of enriched flour and breads prepared therefrom.

[c]Includes two flours produced by air classification.

these were grouped accordingly. Cutoff flours were highest in both free and total niacin but lowest in the free percentage. This is probably explained by the greater amounts of bran contained in this type of flour. No difference was observed between sponge-and-dough bread and that made by the continuous-mix process. The increase in bread niacin over the flour from which it was made appears to be entirely free niacin. This would be expected if the addition represented contributions from milk and yeast, whose niacin is reported to be completely available. Correspondingly, the higher proportion of free niacin in cake and crackers than in their respective flours may reflect additions from nonflour ingredients. The possibility is also considered that the processing step itself may release a portion of the bound niacin.

It should be pointed out that "free niacin," as determined by *in vitro* analytical methods, is defined by the methods employed. It would be expected that the mild extraction used in this study provides a minimal estimate of the amount which is biologically available. Further study incorporating an animal assay procedure is needed to establish the validity of the free-niacin estimate.

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