Packaging Effects on Riboflavin Content of Pasta Products in Retail Markets¹

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ABSTRACT

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Twenty enriched pasta products purchased at the retail level were analyzed for riboflavin content and were found to have a range of 1.50–7.35 μ g of riboflavin per gram of pasta. Lumichrome, a degradation product of riboflavin, was detected in eight of these products. Under controlled conditions of light, pasta packaged in paperboard cartons with or without

transparent windows had approximately 100% riboflavin retention after one week. Pasta packaged in transparent bags showed 70% retention, with light intensities of 50, 150, or 250 ft-c having the same relative impact on riboflavin degradation.

Because most of the pasta sold in the United States is enriched, the stability of the added nutrients is an important consideration for pasta manufacturers and consumers. Whereas niacin and iron are considered stable in pasta products, thiamin stability is dependent on temperature and water activity (Kamman et al 1981), and riboflavin stability is primarily influenced by light exposure (Woodcock et al 1982).

Riboflavin instability in foods was noted as a problem by Peterson et al in 1944, when they found that riboflavin in milk rapidly decomposed when exposed to light. Woodcock et al (1982) reported that riboflavin in pasta was extremely sensitive to light. When a single layer of elbow macaroni was exposed to moderate intensities of light, more than half of the riboflavin was lost the first day, and approximately 10% more was lost after 35 days of exposure. Riboflavin in pasta remained stable when protected from light.

Lumichrome (7,8-dimethylalloxzaine) has been identified as a photodegradation product of riboflavin in neutral and acidic solution (Karrer et al 1934), in milk (Parks and Allen 1977), and in enriched elbow macaroni (Woodcock et al 1982). In elbow macaroni, riboflavin was only partially converted to lumichrome, and with continued light exposure, lumichrome appeared to degrade. Thus, lumichrome may not be useful as a quantitative measure of riboflavin photodegradation in pasta, but may be used as a qualitative indicator that riboflavin has been degraded by light.

Because riboflavin is sensitive to light, the packaging material used for pasta may be a key factor in protecting riboflavin from degradation. Three types of packaging material commonly used for pasta are the transparent bag, the paperboard carton, and the paperboard carton with a transparent window. These types of packaging provide different degrees of light protection and, therefore, may play a role in maintaining riboflavin at a level within the Standards of Identity for enriched pasta (Code of Federal Regulations 1979).

The intensity of light that reaches a food is one of the factors that determines the penetration of light into a food product and the rate of photodegradation of its nutrients (Karel 1975). In milk, an increase in light intensity increased the rate of riboflavin loss (Singh et al 1975); however, when a single layer of elbow macaroni was exposed to light at intensities of 100, 200, or 300 foot-candles (ft-c), no differences in the rates of loss were observed (Woodcock et al 1982). Although exposing a single layer of macaroni to light is useful for determining the effect of light on individual pieces of pasta, the effect of various light intensities on actual products may differ because of packaging considerations.

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This study was undertaken to analyze pasta at the retail level to determine the amount of riboflavin and lumichrome in products available to the consumer. Transparent bags, paperboard cartons, and paperboard cartons with a window were evaluated for their ability to protect riboflavin from photodegradation. Retention of riboflavin in pasta held in three types of packaging exposed to various controlled intensities of light was also determined.

MATERIALS AND METHODS

Product Survey

Many pasta shapes are available, but spaghetti and short-cut products such as elbow macaroni make up the majority of the retail market (Winston 1971). Therefore, spaghetti and macaroni were used as representative pasta products in this study. Enriched elbow macaroni and enriched spaghetti samples were purchased from seven retail outlets throughout the Minneapolis/St. Paul area.

Pasta products were selected from the front of the shelf and, after purchase, were stored in the dark until analyzed. The light intensity in each of the retail outlets was tested using a General Electric type 214 light meter approximately one m from the floor as well as on the front of the display shelf. Light intensities ranged from 40 to 120 ft-c in the aisle and from 25 to 100 ft-c on the front of the shelf.

Eight brands of spaghetti or macaroni were purchased, and at least three different brands of pasta were analyzed within each of three packaging classifications. Analysis was done on 10 products packaged in transparent bags, five products in paperboard cartons, and five products in paperboard cartons with transparent windows. The contents of each package were mixed before sampling to obtain a representative sample. Triplicate analyses were done on each product.

Controlled Light Exposure

In studies in which light intensity was controlled, chambers maintained at a constant light intensity and relative humidity were used. The custom-built chambers were mounted with variable-intensity standard fluorescent bulbs (Cool White, General Electric no. F15T8-CW). Light intensity was adjusted with a rheostat and monitored with a General Electric type 214 light meter. The relative humidity in the chambers was maintained at approximately 44% with saturated K_2CO_3 . Studies were conducted at room temperature.

Two packages of elbow macaroni in each of the three types of packaging materials were exposed to 50 ft-c of light for one week. In addition, macaroni packaged in transparent bags was also exposed to light intensities of 150 and 250 ft-c to determine whether increased light intensity would affect the riboflavin retention in packaged pasta. Control samples were taken before the packages were exposed to light. The packaged products used in this controlled-lighting study were purchased in retail outlets and had no detectable lumichrome before light exposure. Triplicate analyses were done for each packaged product at each light intensity.

To determine the photodegradative behavior of riboflavin and the concurrent formation of lumichrome in enriched pasta, single layers of spaghetti and macaroni were placed in petri dishes (6.7 cm in diameter) and were exposed to a light intensity of 50 ft-c. Duplicate samples were removed over a two-week period, and the amounts of riboflavin and lumichrome were determined in both the spaghetti and elbow macaroni.

Determination of Riboflavin and Lumichrome

The AOAC (1980) riboflavin procedure with slight modifications was used for the extraction of riboflavin and lumichrome from pasta. A 1.50-g ground pasta sample (which passed through a No. 20 sieve) was placed in a 50-ml Pyrex test tube with a Teflon-lined screw cap. These Pyrex test tubes were wrapped with aluminum foil to prevent light exposure. Approximately 15 ml of 0.1N HCl was added, and the samples were mixed and then autoclaved for 30 min at 15 psi. After autoclaving, the samples were mixed for 20 sec while still hot, centrifuged for 30 min at $270 \times g$, and then the supernatants were decanted into 25-ml volumetric flasks. The remaining sediment was rinsed with 10 ml of 0.1N HCl, centrifuged again, and decanted. Combined supernatants were brought up to volume with 0.1N HCl and then were filtered through 0.45- μ m membrane filters. All extractions were conducted under subdued lighting, using light-protective glassware.

High-performance liquid chromatography (HPLC) with fluorescence detection was used for the separation and quantification of riboflavin and lumichrome. The HPLC system consisted of a model 6000A pump (Waters Associates, Inc.), a mode 7120 Rheodyne injector with a 10- μ l sample loop, a model FS 950 Fluoromat fluorometer (Kratos, Inc.), and a Hewlett Packard model 3380A recorder integrator. The separation of riboflavin was accomplished using a μ Bondapak C₁₈ column (30 cm \times 3.9 mm, i.d., 10- μ m particles, Waters Associates, Inc.) with a mobile phase of 43% methanol, 56% distilled water, and 1% glacial acetic acid at a flow rate of 1.0 ml/min. A 7-59 excitation filter and a 3-70 emission filter were used for the detection of riboflavin. The limit of detection for riboflavin was 1 pg injected.

In a separate analysis of the same extract, lumichrome was determined using a Z Module Radial Compression Separation System with a μ Bondapak C₁₈ radial compression cartridge (10 cm \times 8 mm, i.d., 10- μ m particles, Waters Associates, Inc.). A mobile phase of 49% methanol, 50% distilled water, and 1% glacial acetic acid was used. Fluorescence detection of lumichrome was optimized using an FSA 403 excitation filter and an FSA 426 emission filter. The limit of detection for lumichrome was 30 pg.

Riboflavin (Sigma Chemical Co.) and lumichrome (Aldrich Chemical Co.) were dissolved in 0.1N HCl and methanol, respectively, using heat and stirring. The standard solutions were stored at 4°C in amber flasks and were serially diluted with 0.1N HCl to the desired concentrations of 0.15 μ g of riboflavin per milliliter and 0.30 μ g of lumichrome per milliliter. Determination of riboflavin and lumichrome was completed by comparing the peak heights of the samples to the peak heights of the standards.

When a known amount of riboflavin or lumichrome was added to pasta samples before analysis, the recovery of added riboflavin was 100%, and the recovery of lumichrome was 85%.

Moisture Determinations

Moisture content was determined for each pasta product purchased. Duplicate samples were analyzed by the AACC (1976) vacuum oven method.

RESULTS AND DISCUSSION

Product Survey

The Federal Standard of Identity (Code of Federal Regulations 1979) for enriched macaroni and spaghetti products specifies riboflavin content as not less than 1.7 mg nor more than 2.2 mg in each pound of pasta (3.75–4.85 μ g of riboflavin per gram of pasta). Analysis of 20 commercially available pasta products (Table I) showed that the riboflavin content was distributed above, within, and below the range specified by the Standard of Identity. The riboflavin content of the products surveyed ranged from 1.50 to 7.35 μ g of riboflavin per gram of pasta. This range is similar to that in the USDA's Nutrient Data Bank (Douglass and Matthews 1982). For enriched macaroni products analyzed from 1963 to 1982, the riboflavin content was 3.6–7.8 μ g of riboflavin per gram of pasta.

Moisture content in the samples surveyed in Table I ranged from 6.5 to 12.3% when calculated on a wet basis, which agrees with the range of 5.2–12.0% in the USDA's Nutrient Data Bank (Douglass and Matthews 1982). All experimental results are reported as riboflavin content in the product as purchased and are not adjusted for moisture content.

In Table I, the lumichrome concentration of each of the products surveyed is indicated qualitatively as greater than 0.50 μ g per gram of pasta, detected but less than 0.50 µg per gram of pasta, or not detected. With the limit of lumichrome detection at 30 pg per injection, a sample with no detectable lumichrome would have less than 0.05 μ g per gram of pasta. Of the six samples that had riboflavin contents below the Standard of Identity, four had greater than 0.50 μ g of lumichrome per gram of pasta, one sample had detectable lumichrome but less than 0.50 μ g per gram of pasta, and one sample had no detectable lumichrome. Since lumichrome has not been shown to occur without the photodegradation of riboflavin, these results indicate that riboflavin had photodegraded in all but one of these samples. The product with the lowest riboflavin content—1.5 μg per gram of pasta—had no detectable lumichrome. This product may have been underfortified with riboflavin when manufactured. However, because lumichrome is somewhat unstable (Woodcock et al 1982), the lumichrome produced as a riboflavin breakdown product may have been degraded with extended storage.

Seven out of the 20 products surveyed had riboflavin contents above the level set by the Standard of Identity and had no detectable lumichrome. Seven other samples had riboflavin

TABLE I
Riboflavin and Lumichrome Content in 20 Commercially Available Pasta Products
Relative to the Range Specified by the Standard of Identity^a

Relative to the Range Specifica by the Standard of Identity						
Above Range (μg/g of Pasta)		Within Range (µg/g of Pasta)		Below Range (μg/g of Pasta)		
Riboflavin ^b	Lumichrome ^c	Riboflavin	Lumichrome	Riboflavin	Lumichrome	
7.35 ± 0.11	0	4.79 ± 0.30	0	3.71 ± 0.17	++ ^d	
5.81 ± 0.15	0	4.60 ± 0.10	0	3.54 ± 0.12	$++^{d}$	
5.80 ± 0.26	0	4.56 ± 0.16	0	3.49 ± 0.21	++ ^d	
5.60 ± 0.18	0	4.55 ± 0.34	0	3.18 ± 0.16	+ ^d	
5.31 ± 0.28	0	4.37 ± 0.02	$++^{d}$	2.27 ± 0.23	++ ^d	
5.20 ± 0.28	0	4.09 ± 0.20	+ ^d	1.50 ± 0.02	0	
4.90 ± 0.45	0	3.85 ± 0.29	+ ^d	•••	•••	

^a Range = $3.75-4.85 \mu g$ of riboflavin per gram of pasta.

^b Mean riboflavin content ± standard deviation of three analyses on each product.

 $^{^{}c}0 = No$ lumichrome detected (less than 0.05 μ g per gram of pasta).

 $[^]d++=$ Greater than 0.50 μ g of lumichrome per gram of pasta detected. += Lumichrome detected, but less than 0.50 μ g per gram of pasta.

contents within the Standard of Identity, and one of these had a lumichrome concentration of greater than 0.50 μ g per gram of pasta. At this concentration of lumichrome, riboflavin would have undergone considerable degradation and, therefore, this product probably would have had an initial riboflavin content above the Standard of Identity. Overages of the nutrient above the label claim are frequently required in fortification of foods because of imperfect nutrient distribution, analytical error, and degradation during processing, handling, and storage (Watson 1981, Borenstein 1971).

In Fig. 1, the riboflavin content in the pasta products is graphed versus the type of packaging material. All but one of the products packaged in paperboard cartons and paperboard cartons with windows had riboflavin contents within or above the 3.75-4.85 μ g per gram of pasta required by the Standard of Identity. The 10 products packaged in the transparent bags showed a wide distribution of riboflavin content. Unlike the products packaged in paperboard cartons with or without a window, half of the products packaged in transparent bags had riboflavin contents below the Standard of Identity. These samples also showed higher amounts of lumichrome, indicating that the low levels of riboflavin were likely the result of photodegradation rather than under fortification. The paperboard carton and the paperboard carton with a window seem to provide sufficient light protection to retain riboflavin content in commercial pasta products. The transparent bag appears to provide less protection, and products with this packaging may warrant larger overages.

Controlled Light Exposure

An additional measurement that can be used in the evaluation of riboflavin content and stability in pasta is the level of lumichrome present. To evaluate the formation of lumichrome during riboflavin photodegradation, single layers of spaghetti and elbow macaroni were exposed to light with an intensity of 50 ft-c. Only pasta samples with no detectable lumichrome were used for the study, and precautions were taken to avoid light before the exposure period. Changes in the concentration of riboflavin and lumichrome in spaghetti are illustrated in Fig. 2. Initially, riboflavin photodegraded rapidly and then did so more slowly with continued exposure. In contrast, lumichrome concentration

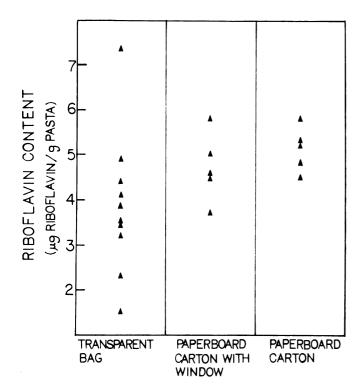


Fig. 1. Riboflavin content in commercial pasta products sold in different types of packages.

initially increased rapidly but remained fairly constant through two weeks of exposure. A similar pattern of riboflavin loss and lumichrome formation was observed for elbow macaroni. Even though the shape and physical structure for spaghetti and macaroni are different, the influences of light on riboflavin stability appear to be very similar for both products.

While lumichrome is a product of riboflavin photodegradation, Fig. 2 illustrates that it accounts for less than one-third of the degraded riboflavin. This low conversion ratio, along with the reported instability of lumichrome (Woodcock et al 1982), limits the use of lumichrome to a qualitative indicator of riboflavin loss.

When packages of elbow macaroni were exposed to light under controlled conditions for one week, the riboflavin retention was approximately 100% in the paperboard carton and in the paperboard carton with a window (Table II). Retention in a transparent bag was approximately 70%, with no significant trend of decreasing retention observed when the light intensity was increased from 50 to 150 or 250 ft-c. Riboflavin photodegradation in pasta packaged in a transparent bag is not significantly increased by light intensity above 50 ft-c.

CONCLUSIONS

Riboflavin contents in commerically available pasta products vary considerably, and some of the variation may be accounted for by photodegradation of riboflavin. The presence of lumichrome in a number of products showed that light-induced degradation of riboflavin had occurred. Paperboard cartons, with or without windows, provide excellent protective packaging for the photolabile nutrient. Transparent bags offer less protection and allow approximately one-third of the riboflavin to photodegrade in one week of light exposure. Light intensity above 50 ft-c does not increase the rate of riboflavin degradation in packaged pasta products.

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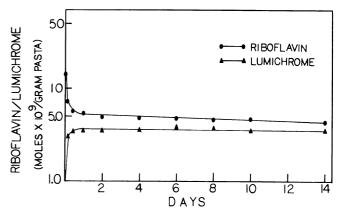


Fig. 2. Riboflavin and lumichrome content in a single layer of spaghetti exposed to a light of 50 ft-c.

TABLE II
Retention of Riboflavin in Macaroni in the Three Types of Packaging
After Seven Days of Light Exposure

Packaging Material	Light Intensity (ft-c)	Percent Retention (Mean ± Standard Deviation)
Transparent bag	50	71.1 ± 8.1
	150	65.6 ± 1.8
	250	68.5 ± 5.2
Paperboard carton		
with window	50	95.1 ± 2.4
Paperboard carton		
without window	50	102.1 ± 1.7

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