Certain B Vitamin and Phytic Acid Contents of Pearl Millet [*Pennisetum americanum* (L.) Leeke]

Chipo G. Simwemba, Russell C. Hoseney,* Elizabeth Varriano-Marston, and Kathleen Zeleznak

This study leads to the following conclusions on the B vitamin content of pearl millet [Pennisetum americanum (L.) Leeke]. Little variation occurs in riboflavin and thiamin contents among cultivars grown at the same location but niacin varies significantly among cultivars grown at the same location. Location has a strong effect on thiamin content, a moderate effect on niacin, and no significant effect on the riboflavin content. Significant losses of B vitamins occur as a result of milling. Riboflavin, niacin, and thiamin are high in the germ and bran of pearl millet but relatively low in the endosperm. Cooked pearl millet showed elevated B vitamin levels presumably by improving extractability. The level of phytic acid in pearl millet is slightly lower than that reported for wheat and appears to vary both with location and with cultivar. As expected, the level of phytic acid is high in the germ and low in the endosperm.

Pearl millet is a staple food for millions in the dry and semiarid regions of Asia, Africa, and the Middle East. The potential of pearl millet as a food grain is large and its genetic variability offers opportunity for breeding highyielding, disease-resistant cultivars (Rachie and Majmudar, 1980).

As a cereal for human food, it is often considered highly palatable and is among the most nutritious of grain crops (Rachie and Majmudar, 1980). Virtually all the pearl millet grain, except that saved for seed, is consumed as human food. The various food preparations in Asia have been described by Chavan (1958) and Vogel and Graham (1979). The most important use of millet flour is for baking flat rounded unleavened bread, called chapati, roti, or bhakri. The cracked grain can also be cooked like rice or the flour is cooked into a porridge of suitable consistency. Other preparations include frying in deep fat or mixing with pulses, other cereals, and spices.

In Africa, pearl millet is often consumed in the form of a cooked porridge, soup, or gruel. The grain is cleaned, ground (including wet milling), and sifted to obtain the flour. Occasionally, it is baked in the form of unleavened bread but more often it is boiled in earthenware pots and eaten with milk or melted butter (Rachie and Majmudar, 1980).

A wide range of results have been reported for the B vitamin content of pearl millet and the major concentration is found in the germ (Adrian and Sayerse, 1957; Chitre et al., 1955; Samson and Adrian, 1971; Rama Rao and Swaminathan, 1953; James et al., 1947; Hinton et al., 1953). Those workers, however, do not agree on whether the B vitamins, thiamin, niacin, and riboflavin, are influenced genetically or environmentally.

All cereal grains are rich in phytic acid. Phytic acid has been implicated in binding certain minerals, particularly divalent ions, thus, making these nonabsorbable. No reports were found in the literature on the level of phytic acid in pearl millet.

The purposes of this study were (a) to determine the niacin, riboflavin, and thiamin content of pearl millet samples from different populations grown in different locations, (b) to determine the content of different milling fractions, and (c) to determine the effect of food preparation including boiling, fermenting, and baking on vitamin retention in prepared foods. We also determined genetic and location effects on the phytate phosphorus content of pearl millets.

MATERIALS AND METHODS

Pearl millet samples grown at the following locations were used: Lubbock, TX; Minneola, KS; Garden City, KS; St. John, KS. From each location the following lines were analyzed: 2 (TIFT 2 23 DAE \times 656 + 653); 10 (79-2202 \times 79-4104); 12 (79-2017 \times 79-4104); 19 (79-2161 \times 78-7024); 28 (79-2055 \times 78-7024); 29 (79-2042 \times 79-4104). Three bulk samples grown at Manhattan, KS, HMP 561, HMP 1700, and G 2279, were used in the processing studies.

For the vitamin content study, cleaned samples of pearl millet were milled with a Udy cyclone mill by using a 0.5-mm steel screen. To produce samples for analysis of the kernel components, three bulk pearl millet samples were cleaned and decorticated to 15% in a Satake grain testing mill, No. 55336 (Stake Engineering Co., Ltd., Tokyo, Japan). Whole grain was tempered to 20% for 6 h and then milled with a Ross laboratory mill. The stock was sifted by using Tyler Rotap sifters (Abdelrahman et al., 1983).

Vitamin Analysis. Niacin. AACC method 86-50 was used for niacin. Two-gram samples were analyzed and readings were taken on a Spectronic 21 (Bausch & Lomb) colorimeter.

Riboflavin. AACC method 86-70 (American Association of Cereal Chemists, 1969) was used to determine riboflavin in 5-g samples with slight modification of the oxidation step. After the sample solution, standard solution, and water were placed in the test tube and before the potassium permanganate was added, 1 mL of glacial acetic acid was added. Without the glacial acetic acid, a precipitate appeared after oxidation. Fluoresence was determined with an Electronic photofluorometer, Model 12C (Colemen Instruments Inc., Maywood, IL), by using filters PC2 and B-2.

Thiamin. AACC method 86-80 was used to determine thiamine in 3-g samples but 0.1 N hydrochloric acid was used instead of sulfuric acid in the extraction step. An insoluble complex formed when sulfuric acid was used. Fluoresence was determined with an Electronic photofluorometer, Model 12C (Coleman Instruments, Inc., IL,), using filters PC1 and B-1.

Food Preparation. Bulk samples were decorticated to 10% for food preparation studies. The decorticated grain was milled in a Udy mill and the meal was used to prepare the following foods.

Uji. Uji is a thin porridge consumed in many African

Department of Grain Science and Industry, Kansas State University, Manhattan, Kansas 66506.

Table I. Riboflavin Content $(\mu g/g)$ in Pearl Millet Samples^a

lines	Lubbock,	Minneola,	Garden	St. John,
	TX	KS	City, KS	KS
2	1.92 bx	2.08 axy	1.99 bxy	1.87 ax
10	2.26 ax	2.62 by	2.49 cxy	1.78 az
12	2.20 ax	2.00 ax	2.10 abx	1.95 ax
19	2.11 abx	2.13 ax	2.03 abx	1.97 abx
28	2.41 ax	2.17 ax	2.39 acx	2.14 bcx
29	2.24 ax	2.17 ax	2.21 ax	2.23 cx

^a Within each column means with different letters (a, b, c, d) are significantly different among cultivars and within each row means with different letters (x, y, z) are significantly different among location (P < 0.05). Values are on a dry weight basis.

countries and made from fermented or unfermented millet or sorghum flour. The meal (65 g) was mixed with 100 mL of water, placed in a covered container, and allowed to stand for 48 h in a warm place (30 °C). the remaining water (300 mL) was boiled and added to the fermented pearl millet meal (pH 4.6) and cooked until smooth and thick (~15 min). The cooked uji was cooled, freeze-dried, and then ground in the Udy mill.

 $T\hat{o}$. This is a thick unfermented porridge made from millet, sorghum, or cornmeal. It forms the basic food in many African countries. Half of the meal (100 g) was mixed with 100 mL of water to make a smooth slurry. The slurry was then added to 300 mL of boiling water while stirring to prevent lumps from forming. The porridge was stirred continuously until it started to boil. Then the remaining meal (100 g) was added and stirred in rapidly until the tô was very thick and smooth (about 20 min). The cooled, cooked tô was freeze-dried and ground in the Udy mill.

Roti. The roti is an unleavened, flat, round bread that is also called a chapatti. Meal (50 g) was mixed with 37 mL of water and allowed to stand for 45 min. The dough was then rolled out ($^{1/8}$ in.) and cut to a 6 in. diameter circle. Excess dough was discarded. The roti was loosened from the board carefully, placed on a 200 °C grill, and cooked for 1.5 min/side, flipping 3 times. The rotis were sprayed with water just before the first flip. The cooked roti was then placed in a tandoor-type cooker to allow it to puff. It was then immediately removed to a coolng rack. When cooled, the rotis were air-dried in a warm place (35 °C), broken into small pieces, and milled in a Udy cyclone mill.

Statistical Analysis. Data from all measurements was subjected to analysis of variance. The means were compared by using Duncan's multiple range test (P < 0.05).

Phytate Phosphorus. The level of phytate phosphorus was determined as described by Tangkongchitr et al. (1981).

RESULTS AND DISCUSSION

Vitamin Content. Effect of Cultivar. Riboflavin, niacin, and thiamin contents of pearl millet samples grown at the four locations are given in Tables I–III. As shown in Table I, there was little difference in riboflavin content among cultivars grown at the same location. The differences that occur among some lines, though significantly different (P < 0.05), are relatively small and appear to occur randomly.

The niacin content of different cultivars grown at each location is given in Table II. At Lubbock, TX, lines 2, 10, 12, and 28 were significantly higher than lines 19 and 29. At Minneola, KS, lines 10, 19, and 29 were significantly higher than lines 2, 12, and 28. At Garden City, KS, lines 12, 19, 28, and 29 were significantly higher than lines 2 and

Table II. Niacin Content $(\mu g/g)$ in Pearl Millet Samples^a

lines	Lu bboc k,	Minneola,	Garden	St. John,
	TX	KS	City, KS	KS
2	50.15 ax	24.86 by	24.83 cy	23.19 by
10	42.99 abx	37.32 ay	26.69 cz	26.05 bz
12	41.05 abx	23.24 by	40.11 bx	41.81 ax
19	32.05 bz	36.96 az	55.33 ax	36.11 abyz
28	50.38 ax	25.66 bz	40.57 by	41.47 ay
29	38.84 aby	40.51 ay	57.40 ax	22.70 bz

^a Within each column means with different letters (a, b, c, d) are significantly different among cultivars and within each row means with different letters (x, y, z) are significantly different among locations (P < 0.05). Values are on a dry weight basis.

Table III. Thiamin Content $(\mu g/g)$ in Pearl Millet Samples^a

lines	Lubbock, TX	Minneola, KS	Garden City, KS	St. John, KS
2	8.36 bx	6.16 cy	4.41 ak	5.38 cz
10	8.37 bx	6.47 by	4.33 az	4.60 bz
12	8.63 ax	6.43 by	4.44 az	4.03 zk
19	8.55 ax	6.51 aby	4.51 az	4.22 az
28	8.76 az	6.52 aby	3.78 bz	4.13 az
29	7.94 cx	6.72 ay	4.49 az	4.29 abz

^a Within each column means with different letters (a, b, c, d) are significantly different among cultivars and within each row means with different letters (x, y, z, k) are significantly different among locations (P < 0.05). Values are on a dry weight basis.

10. At St. John, KS, high values were observed for lines 2, 10, and 29. There does not seem to be a strong genetic effect on niacin values. However, lines 12, 19, and 28 were in the higher value group at three of the four locations and lines 10 and 29 were in the higher group at two of the four locations.

The thiamin content of the pearl millet lines studied for each location are given in Table III. At Lubbock, TX, line 12, 19, and 28 have the highest values and line 29 has the lowest thiamin content. At Minneola, KS, little variability occurs among the cultivars. However, line 29 is significantly higher than lines 2, 10, and 12 and line 2 is lower than the other lines. No significant differences occur in the lines from Garden City, KS, except line 28, which is lower. At St. John, KS, line 2 is significantly higher than the other lines, which show little variability. There appears to be no consistent effect of genetics on the thiamin content of pearl millet.

Effect of Location. Lines 2, 12, 19, 28, and 29 grown at the four locations show no significant differences in riboflavin content attributable to location. The riboflavin content of line 10 varied at the four locations.

With all the lines studied, there was significant variation in niacin content for samples grown at the four locations. In general, samples from Lubbock, TX, and Garden City, KS, were higher than those from Minneola and St. John, KS. There appears to be a line by location interaction for niacin content in pearl millet.

Location had a significant effect on the thiamin content in pearl millet cultivars. The samples grown at Lubbock, TX, were significantly higher than those from the other three locations. Samples harvested at Minneola were significantly higher than those at Garden City and St. John, KS. The location average thiamin value ranged from 8.44 μ g/g for pearl millet samples grown at Lubbock, TX, to 4.33 μ g/g for those grown at Garden City, KS.

The differences indicated could be a result of a number of factors, including seed size (Hinton et al., 1953), envi-

Table IV. Riboflavin, Thiamin, and Niacin Contents $(\mu g/g)$ of Pearl Millet Kernel Components^a

lines	germ	bran	endosperm	decorticated	whole meal
riboflavin	······	·····			
HMP 561	9,99 a	9.30 b	2.06 d	3.23 c	3.39 c
HMP 1700	8.34 a	7.83 b	1.37 e	2.80 d	3.18 c
G 2279	7.08 a	6.62 a	1.71 c	2.22 bc	2.45 b
thiamin					
HMP 561	8.88 a	9.01 a	3.21 c	4,95 b	5.26 b
HMP 1700	7.38 a	7.66 a	3.82 c	5.53 b	5.84 b
G 2279	6.80 a	6.61 a	3.65 d	4.74 c	5.26 b
niacin					
HMP 561	80.06 a	78.78 a	23.03 d	44.48 c	46.37 b
HMP 1700	75.93 a	62.04 b	18.73 d	37.65 c	42.06 c
G 2279	85.01 a	71.35 b	17.54 d	41.45 c	44.03 c

^a Within each row means with the same letter are not significantly different (P < 0.05). Values are on a dry weight basis.

Table V. Riboflavin, Thiamin, and Niacin Contents $(\mu g/g)$ of Pearl Millet Food Products^a

Table VI.	Phytate Phosphorus in Two Lines of Pearl
Millet Gro	wn at Four Locations

lines	meal	tô	uji	roti
riboflavin				
HMP 561	3.39 c	3.55 b	3.63 ab	3.69 a
HMP 1700	3.38 d	3.49 c	3.57 b	3.67 a
G 2279	2.58 c	2.79 b	2.87 ab	2.94 a
thiamin				
HMP 561	4.95 d	5.88 c	6.04 b	6.18 a
HMP 1700	5.58 c	5.99 b	6.34 a	6.46 a
G 2279	4.92 d	5.46 c	5.97 b	6.17 a
niacin				
HMP 561	46.92 c	49.31 bc	53.39 a	51.78 ab
HMP 1700	48.13 d	49.23 d	53.71 a	51.36 b
G 2279	47.70 a	48.52 a	51.77 a	50.06 a

^a Within each row means with the same letter are not significantly different (P < 0.05). Values are on a dry weight basis.

ronmental and genetic factors (Samson and Adrian, 1971), and fertilizers in the soil (Pai et al., 1958).

Our results lead to the following conclusions on the B vitamin content of pearl millet. Little variation occurs in riboflavin and thiamin contents among cultivars grown at the same location but niacin content varies significantly among cultivars grown in the same location. Location has a strong effect on thiamin content, a moderate effect on niacin, and no significant effect on the riboflavin content of the cultivars.

B Vitamin Content in Pearl Millet Kernel Components. It is unusual in any human society for cereals to be eaten as uncooked whole seeds. Even when used for animal feed, cereals are often ground, flaked, or cooked to increase their digestibility. For human consumption, the seeds are customarily milled before being cooked.

It is the purpose of most milling processes, even simple traditional household methods, to bring about some separation of the three cereal seed components: germ, endosperm, and bran (pericarp). The three B vitamins studied are known to be reduced by removal of the pericarp layers, and significant losses of all three have been reported following traditional milling (Hulse et al., 1980).

Table IV gives the riboflavin, niacin, and thiamin contents of pearl millet kernel components. Riboflavin, thiamin, and niacin are high in the germ and bran and relatively low in the endosperm of all three bulk samples studied. This causes a loss of B vitamins during milling processes because a marked preference for cereal flours composed largely of endosperm is not unusual among people who subsist upon the millets.

Effect of Method of Food Preparation of B Vitamin Content. The riboflavin, niacin, and thiamin contents of foods prepared from three pearl millet bulk samples are shown in Table V. In general, the vitamin levels found were slightly higher in all cooked samples when compared

location	phytate phosphorus, line mg/100 g		
Garden City, KS	2	262	
Garden City, KS	12	284	
Minneola, KS	2	266	
Minneola, KS	12	306	
Lubbock, TX	2	179	
Lubbock, TX	12	214	
St. John, KS	2	264	
St. John, KS	12	279	

Table VII. Phytate Phosphorus in Millet Fractions from HMP 1700 Pearl Millet Bulk

fraction	phytate phosphorus, mg/100 g	
germ	752	
endosperm	86	
bran	278	
whole meal	245	
decorticated grain	312	

to the vitamin content of raw meal.

Niacin content of the cooked samples was higher than that of raw meal. In general, the uji and roti had niacin levels that were significantly higher than those found in tô. The high value of uji agrees with Vogel and Graham (1979), who state that fermentation elevates the B vitamin levels in comparison to the whole-grain starting values. Thiamin content was also significantly increased in the cooked samples. As with niacin, roti and uji had values higher than those of tô.

Three methods of cooking were used, boiling, fermenting followed by boiling, and baking. It was found that fermenting and baking had slightly higher values than did boiling. Baking was for a very short time period but gave values similar to those of fermenting and boiling. The increase in the B vitamin content of the cooked samples is bothersome. It would appear to indicate that all the vitamins are not being extracted from the raw meal or bound forms are being released as a result of thermal processing. In either case, additional work will be required to understand the phenomena.

Phytic Acid. The phytate phosphorus level of two lines of pearl millet grown at four locations is given in Table VI. The values found for pearl millet are slightly lower than those reported for wheat (Tangkongchitr et al., 1981). Line 12 had higher phytate phosphorus value than did line 2 at all locations. The samples grown in Lubbock, TX, were lower in phytate phosphorus than those grown at the three locations in Kansas. Thus, both genetic and environmental affects appear to be important in determining the phytate level. The level of phytate phosphorus in the mill fractions from the HMP 1700 bulk are given in Table VII. The level in the germ was quite high and the level in the endosperm was low as might be expected.

- **Registry No.** Riboflavin, 83-88-5; thiamin, 59-43-8; niacin, 59-67-6; phytic acid, 83-86-3.
- LITERATURE CITED
- Abdelrahman, A.; Hoseney, R. C.; Varriano-Marston, E. Cereal Chem. 1983, 60, 189.
- Adrian, J.; Sayerse, C. Br. J. Nutr. 1957, 11, 99.
- American Association of Cerial Chemists "Approved Methods of the AACC"; AACC: St. Paul, MN, 1969; Methods 86-50 Niacin, approved April 1966, 86-70 Riboflavin, approved April 1968, and 86-80 Thiamin, approved May 1960.
- Chavan, V. M. Indian Counc. Agric. Res. Farm Bull. 1958, 48.
- Chitre, R. G.; Desai, D. B.; Raut, V. S. Indian J. Med. Res. 1955, 43, 575.
- Hinton, J. J. C.; Peers, F. G.; Shaw, B. Nature (London) 1953, 172, 993.
- Hulse, J. H.; Laing, E. M.; Pearson, D. E. "Sorghum and the millets"; Academic Press: New York, 1980.

- James, E. M.; Norris, F. M.; Wokes, F. Analyst (London) 1947, 72, 327.
- Pai, M. L.; Mehta, B. V.; Shah, H. C. J. Maharaja Sayajirao Univ. Baroda 1958, 7, 41–48; Chem. Abstr. 1959, 53, 22657i.
- Rachie, K. O.; Majmudar, J. V. "Pearl Millet"; The Pennsylvania State University Press: University Park, PA, and London, 1980.
- Rama Rao, G.; Swaminathan, M. Bull. Cent. Food Technol. Res. Inst., Mysore 1953, 3, 68.
- Samson, M. F.; Adrian, J. Agron. Trop. (Paris) 1971, 26, 1090. Tangkongchitr, U.; Seib, P. A.; Hoseney, R. C. Cereal Chem. 1981, 58, 226.
- Vogel, S.; Graham, M. "Sorghum and Millet: Food Production and Use"; International Development Research Center, Ottawa, Canada, 1979.

Received for review May 16, 1983. Revised manuscript received September 19, 1983. Accepted October 16, 1983. This work was supported by Grant AID/DSAN/XII/G-1049 from the Agency for Internatonal Development, Washington, DC. Contribution No. 83-13J, Department of Grain Science and Industry, Kansas Agricultural Experiment Station, Manhattan, KS 66506

Determination of Total Available Glucose in Corn Base Materials

Clifford C. Budke

A method is described for the determination of total available glucose in corn base materials. The procedure involves gelatinization with water at 122 °C, conversion of starch and glucose polymers to free glucose with α -amylase and amyloglucosidase at 55 °C, and determination of the glucose content by using a Yellow Springs Instrument Co. Model 27 analyzer. This instrument uses an immobilized film of glucose oxidase to convert β -D-glucose to hydrogen peroxide, which is measured electrochemically to provide an absolute measurement of the glucose content. The method eliminates the inaccuracy and uncertainty associated with many of the chemical methods for determining glucose and is applicable to a wide variety of corn base materials. The procedure may be applicable to other agricultural products.

Interest in obtaining fuel and energy from nonpetroleum sources has increased in the past few years. Conversion of biomass materials to alternative fuels has received particular attention. It is estimated (*Chem. Week*, 1983) that consumption of corn for this purpose during the current year will increase about 3 times over the 1980 level. The use of corn base materials for the production of fermentation ethanol requires an accurate determination of total available glucose for process optimization and to monitor the quality of the feed materials.

Methods traditionally used for the determination of total available glucose or fermentable carbohydrates involve the hydrolysis of starch and lower α -D-glucose polymers with acid or amylolytic enzymes followed by the determination of glucose by chemical or enzymatic procedures. Of the chemical methods, modifications of those employing reduction by copper (Munson and Walker, 1906; Lane and Eynon, 1923; Zerban and Sattler, 1938) or alkaline ferricyanide (Friedemann et al., 1967) are more commonly used whereas glucose oxidase techniques are generally considered the most useful of the enzymatic methods. AOAC procedures have been published that use these methods for the determination of starch (AOAC, 1980, methods 7.808 and 14.075).

The copper reduction procedures for glucose determination are subject to interference from other reducing substances, require stringent analytical conditions, and are time consuming. On the other hand, enzymatic methods using glucose oxidase are relatively simple, are rapid, and have the distinct advantage of being specific for glucose. Numerous modifications of the glucose oxidase method have been reported including those using colorimetric (Banks and Greenwood, 1971; Norton and Smith, 1967) and electrometric measurement (Trop and Grossman, 1975; Enfors, 1981).

During the course of recent work, we used the Yellow Springs Instrument Co. (YSI) Model 27 analyzer for the routine determination of free glucose at various stages of the saccharification-fermentation process. This instrument (Taylor et al., 1977; Mason, 1983) uses a film of glucose oxidase immobilized on a membrane to convert β -D-glucose to hydrogen peroxide, which is measured electrochemically and related to the total glucose content. The instrument is calibrated with an equilibrated dextrose solution. Conversion of polymerized glucose to the free monomer results in an equilibrium mixture of the anomers that can then be directly analyzed. The method is simple, fast, and accurate and gives a digital readout of the glucose content of the sample solution.

U.S. Industrial Chemicals Co., Division of National Distillers and Chemical Corp., Research Department, Cincinnati, Ohio 45237.