of the easily hydrolyzed portion of the cell wall fiber to its rate and percentage digestibility in ruminant animals.

LITERATURE CITED

Albersheim, P.; Nevins, D. J.; English, P. D.; Karr, A. *Carbohydr. Res.* **1967**, *5*, 340.
Barton, F. E., II; Amos, H. E.; Burdick, D.; Wilson, R. L. *J. Anim. Sci.* **1976**, *43*, 504.
Collings, G. F.; Yokoyama, M. T. *J. Agric. Food Chem.* **1979**, *27*, 373.
Goering, H. K.; Van Soest, P. J. *U.S., Dep. Agric., Agric. Handb.* **1970**, *No. 379*.
Gum, E. K.; Brown, R. D. *Anal. Biochem.* **1977**, *82*, 372.
Himmelsbach, D. S.; Barton, F. E., II *J. Agric. Food Chem.* **1980**, *28*, 1203.
Kleinbaum, D. G.; Kupper, L. L. "Applied Regression Analysis and Other Multivariable Methods"; Duxburg Press: North Scituate, MA, 1978.
Linden, J. C.; Lawhead, C. L. *J. Chromatogr.* **1975**, *105*, 125.
Palmer, J. K. "Applied Polymer Symposium"; Wiley: New York, 1975a.
Palmer, J. K. *Anal. Lett.* **1975b**, *8*, 215.
Palmer, J. K.; Brandes, W. B. *J. Agric. Food Chem.* **1974**, *22*, 709.
Steel, R. G. D.; Torrie, J. H. "Principles and Procedures of Statistics"; McGraw-Hill: New York, 1960.

Received for review August 25, 1981. Revised manuscript received March 22, 1982. Accepted June 17, 1982. Reference to a company or product name does not imply approval or recommendation of the product by the U.S. Department of Agriculture to the exclusion of others that may be suitable.

Dietary Fiber Content of Some Tropical Fruits and Vegetables

Eric D. Lund* and John M. Smoot

The dietary fiber composition of some common tropical fruits and vegetables was studied. Samples analyzed were pineapple, carambola, sapodilla, papaya, mango, grapefruit, sweet potato, and yam. Values for cellulose, hemicellulose, lignin, cutin, ash, neutral detergent residue (NDR), and enzymatic (insoluble and soluble) fractions were obtained (percent of fresh weight). NDR for most samples was 0.9–1.2%. Cellulose was 0.1–0.9%, except sapodilla (2.4%) and sweet potato peel (1.4 and 1.9%). Lignin was around 0.025–0.17%; carambola (0.3%), sapodilla (2.3%), and sweet potato peel (0.4%) were exceptions. Hemicellulose was about 0.04–0.4%; exceptions were pineapple (0.5%), sapodilla (0.6%), and tropical (Puerto Rican) sweet potato peel (0.9%). In some samples nutritionally valuable fiber components were relatively concentrated, and they may be potential sources of concentrated fiber fractions with useful physiological properties. Sapodilla, in particular, seems to be an abundant source of lignin and cellulose.

The composition of fiber fractions in forages and cereals has been thoroughly investigated, but fiber in fruits and vegetables has not been as well studied. Previous investigations (Kamath and Belvady, 1980; Shipley, 1978) showed some differences between cereal products, fruits, and vegetables. Grain products have relatively high hemicellulose concentrations, vegetables have been found somewhat low in lignin, and the composition of fruits is usually somewhere between. Although some tropical fruits and vegetables have been analyzed, fiber in most of the common examples has not been reported. Tropical fruits and vegetables may contain unusual fiber components, some of which may have unique physiological properties, such as specific lipid binding capacities.

Previous studies have shown that in a few tropical fruits and vegetables fiber components, in general, did not differ significantly from those of other fruits and vegetables. Among the more common tropical species, these authors (Kamath and Belvady, 1980; Shipley, 1978) reported values for sweet potato, yellow pumpkin (*Curcurbita maxima*), plantain, banana, mango, guava, grapefruit, and mandarin oranges. A report on olive fiber has also been published (Moreno and Diez, 1979). The Van Soest detergent procedure or the Southgate procedure was used in the studies cited above. The Van Soest procedure yields values for cellulose, lignin, hemicellulose, and neutral detergent residue (NDR). The Southgate method gives cellulose and lignin values, but the hemicellulose and pectin are usually reported together as "noncellulosic polysaccharides". Most samples had 0.2–6% cellulose, 0.04–2.4% hemicellulose (or noncellulosic polysaccharides), and 0–0.8% lignin. A few samples deviated significantly from the usual composition range. Thus, plantain and guava contained relatively large lignin fractions (1.23 and 0.80%, respectively), and sweet potato had a large amount of noncellulosic polysaccharides (5.24%).

In this report, the dietary fiber content of 15 samples from 8 different fruits and vegetables was determined. Modified forms of the Van Soest detergent and Hellendoorn enzymatic methods were used (see Experimental Section). Differences between varieties, the effects of processing, and variations between different parts of the plant were studied for some samples.

EXPERIMENTAL SECTION

Materials. Pineapple and papaya samples and one sample of sweet potato were purchased at local markets. The yams (five types, listed in Table I) and the tropical sweet potato were obtained from the Mayaguez Institute of Tropical Agriculture, Mayaguez, PR. Carambolas, sapodillas, and mangos were obtained from the USDA Subtropical Horticultural Research Station at Miami, FL. The grapefruit were obtained from USDA Horticultural Field Station, Orlando, FL.

General Procedure. Unless otherwise noted, the edible, fleshy portion of the fruit or vegetable was analyzed. Since the peels of sweet potato are also edible, they were analyzed separately from the flesh. The peel of carambola is normally consumed with the fruit flesh, thus the ana-

U.S. Citrus and Subtropical Products Laboratory, Southern Region, U.S. Department of Agriculture, Agricultural Research Service, Winter Haven, Florida 33880.

Table I. Fiber Content As Determined by Nonsequential (N) and Sequential (S) Detergent Methods

	fiber content, % of fresh wt			
	NDR (N)	NDR (S)	ADR (N)	ADR (S)
carambola				
Tean Ma	0.89 ± 0.01	1.10 ± 0.003	0.66 ± 0.02	0.78 ± 0.02
No. 44	1.04 ± 0.08	1.21 ± 0.02	0.65 ± 0.09	0.85 ± 0.03
sapodilla	2.46 ± 0.11	5.30 ± 0.18	1.98 ± 0.06	4.67 ± 0.18
sweet potato				
supermarket[a]				
flesh	1.14 ± 0.02	1.20 ± 0.09	0.95 ± 0.04	0.83 ± 0.03
peel	1.99 ± 0.11	2.07 ± 0.04	2.10 ± 0.08	1.70 ± 0.05
tropical[b]				
flesh	1.13 ± 0.03	1.14 ± 0.03	1.31 ± 0.04	1.05 ± 0.001
peel	3.15 ± 0.01	3.27 ± 0.07	2.90 ± 0.04	2.33 ± 0.04
yam (*Dioscorea*)				
alata				
Forestero	0.89 ± 0.04	0.90 ± 0.03	0.89 ± 0.02	0.72 ± 0.03
Gunung	1.28 ± 0.06	1.37 ± 0.02	1.13 ± 0.04	0.98 ± 0.02
unspecified	0.80 ± 0.03	0.79 ± 0.02	0.79 ± 0.01	0.63 ± 0.02

[a] Obtained from a local market. [b] Obtained from Puerto Rico.

lytical sample included peel. For all samples (800–900 g), the fresh fruit was cut into small pieces, then frozen in liquid nitrogen, and homogenized in a blendor prior to analysis.

Dry weight was determined by holding a 5-g sample of the fresh blended material in a vacuum oven at 70 °C and 1 torr for 6 h. The dried sample was allowed to cool to room temperature in a desiccator and then weighed.

Detergent Analysis. Samples were analyzed for fiber by a modified form of the Van Soest detergent procedure (Robertson, 1978). The sample was extracted with neutral and acid detergent solutions. The residues were designated respectively the neutral detergent residue (NDR) and acid detergent residue (ADR). The NDR contained the plant cell wall without pectins (cellulose, hemicellulose, lignin, cutin, and insoluble minerals), and the ADR contained lignin, cellulose, cutin, and insoluble minerals. The amount of hemicellulose was determined by subtracting the two values. Lignin was determined by the permanganate method. The amount of cellulose was determined as the difference between the permanganate-insoluble residue and ash. Cutin was determined by treatment of the cellulose residue with 72% H_2SO_4. When the ADR did not contain dark particles, a cutin analysis was not carried out.

As a check for anomalous ADR values, nonsequential and sequential methods were employed (three samples by each method). Use of the sequential method eliminated any possible error caused by the presence of tannins and acid-insoluble pectins, which can appear in the ADR when the sample is extracted directly with acid detergent solution. In the nonsequential method, the neutral and acid detergent extractions were carried out separately on different portions of the same sample. For the sequential analysis, the sample was first extracted with the neutral detergent solution, and then the residue was extracted with the acid detergent solution. Sodium sulfite was added to the nonsequential neutral detergent solution as an antioxidant for improved extraction of protein. It was not used in the sequential procedure.

High starch samples (yam and sweet potato) were degassed by evacuation at 1 torr for 1 h, heated in an autoclave for 1 h at 130 °C, and then treated with α-amylase for starch removal as described by McQueen and Nicholson (1979). However, we had to use twice the concentration of enzyme that they recommended.

Interference from pectin was estimated by analysis of a sample of grapefruit albedo. The albedo was analyzed by the modified detergent method described above (nonsequential analysis). Pectin was determined in the initial albedo sample, the NDR, and the ADR by the modified uronic acid carbazole reaction (Bitter and Muir, 1962).

Enzymatic Analysis. Fiber composition was also determined by a modified version of Hellendoorn's enzymatic method (Schweizer and Wursch, 1979). Analyses were carried out in triplicate. The yam and sweet potato samples were degassed as described above before the autoclaving step. For starchy vegetables (yam and sweet potato) we used 3 times the amount of amyloglucosidase recommended by Schweizer and Wursch.

RESULTS AND DISCUSSION

Dietary fiber composition was determined for a number of relatively common tropical fruits and starchy vegetables (sweet potatoes and yams). The interference of large amounts of starch and pectin required modification of the analytical procedure for many of the samples.

Application of the reported procedures to starchy vegetables resulted in abnormally large and variable NDR and enzymatic insoluble values. Therefore, both methods were modified until reasonably low and consistent values were obtained. Starch is usually removed by pretreatment with amylase. However, several modifications of the pretreatment procedure were found necessary. Twice the amount of amylase suggested by McQueen and Nicholson (1979) was necessary for complete removal of starch prior to detergent analysis. Three times the amount of amylase recommended by Schweizer and Wursch (1979) was necessary for complete removal of starch prior to enzymatic analysis. In addition, in the procedure for detergent analysis we included the autoclaving step prior to the amylase pretreatment [as recommended by Schweizer and Wursch (1979) for the enzymatic analysis]. This step was necessary for complete removal of starch probably because the swelling of the starch granules made them more susceptible to enzymatic degradation. Some of the yams appeared to contain a natural foam stabilizer that resulted in a significant loss of sample during the autoclaving step. Vacuum degassing prior to autoclaving eliminated this problem.

The maximum error that might be caused by incomplete removal of pectin was estimated from the analysis of a high pectin sample (grapefruit albedo, 10.1% AGA) by the detergent method. Approximately 67.4% of the pectin was solubilized during the NDF procedure and 90% during the ADF procedure. Our results indicated, for most fruits and vegetables, which contain much less pectin, the error in the detergent values resulting from incomplete removal

Table II. Dietary Fiber Content of Tropical Fruits and Vegetables

	% of fresh wt ± SD							
	detergent[a,b]				enzymatic fiber			
	cellulose	hemicellulose	lignin	ash	NDR	insoluble	soluble	dry wt
fruits								
pineapple								
fresh: A	0.37 ± 0.02	0.51 ± 0.02	0.050 ± 0.005	0.028 ± 0.019[c]	0.93 ± 0.01			16.7 ± 0.1
B	0.37 ± 0.00	0.46 ± 0.01	0.040 ± 0.003		0.87 ± 0.01	0.80 ± 0.00	0.160 ± 0.004	13.1 ± 0.0
canned: A	0.44 ± 0.03	0.47 ± 0.01	0.021 ± 0.003	0.024 ± 0.010[c]	0.92 ± 0.02			16.3 ± 0.4
B	0.44 ± 0.02	0.49 ± 0.02	0.042 ± 0.006		0.97 ± 0.05			16.8 ± 0.1
carambola[b]								
Tean Ma	0.46 ± 0.01	0.32 ± 0.01	0.31 ± 0.01	0.014 ± 0.002	1.10 ± 0.003	1.30 ± 0.02	0.44 ± 0.01	9.9 ± 0.0
No. 44[d]	0.44 ± 0.02	0.35 ± 0.01	0.33 ± 0.01	0.012 ± 0.003	1.20 ± 0.02			9.8 ± 0.2
sapodilla	2.4 ± 0.1	0.63 ± 0.02	2.28 ± 0.09	0.038 ± 0.029	5.30 ± 0.18	4.16 ± 0.02	0.93 ± 0.01	22.1 ± 0.3
papaya	0.72 ± 0.05	0.103 ± 0.028	0.086 ± 0.022	0.016 ± 0.002	0.91 ± 0.01	1.11 ± 0.04	0.79 ± 0.04	13.3 ± 0.1
mango								
Tommy Atkins	0.67 ± 0.01	0.34 ± 0.04	0.053 ± 0.012	0.011 ± 0.001	1.06 ± 0.05	1.07 ± 0.01	0.61 ± 0.02	14.1 ± 0.0
Keitt	0.66 ± 0.01	0.40 ± 0.04	0.033 ± 0.010	0.011 ± 0.004	1.10 ± 0.04	1.06 ± 0.03	0.64 ± 0.03	15.0 ± 0.1
grapefruit[e]	0.104 ± 0.005	0.036 ± 0.004	0.025 ± 0.002	0.004 ± 0.003	0.165 ± 0.010	0.21 ± 0.01	0.37 ± 0.01	9.8 ± 0.1
vegetables								
sweet potato[f]								
flesh	0.76 ± 0.02	0.37 ± 0.06	0.068 ± 0.007	0.015 ± 0.001	1.20 ± 0.09	1.47 ± 0.12	1.02 ± 0.01	21.0 ± 0.2
peel[g]	1.36 ± 0.02	0.35 ± 0.02	0.36 ± 0.04		2.07 ± 0.04	3.26 ± 0.12	1.56 ± 0.02	14.0 ± 0.3
sweet potato[h]								
flesh	0.94 ± 0.01	0.115 ± 0.001	0.112 ± 0.012	0.029 ± 0.011	1.14 ± 0.03	1.67 ± 0.05	1.30 ± 0.08	22.4 ± 0.1
peel[b]	1.91 ± 0.03	0.94 ± 0.04	0.41 ± 0.02		3.27 ± 0.07			16.5 ± 0.1
yam								
D. esculenta								
Muni	0.66 ± 0.03	0.202 ± 0.031	0.056 ± 0.002	0.243 ± 0.086	0.91 ± 0.02	1.26 ± 0.18	0.23 ± 0.01	28.2 ± 0.1
Beti	0.56 ± 0.04	0.29 ± 0.05	0.089 ± 0.007	0.093 ± 0.033	0.93 ± 0.00	3.18 ± 0.10	0.77 ± 0.08	38.4 ± 0.2
D. alata								
Forastero	0.59 ± 0.01	0.180 ± 0.025	0.130 ± 0.017	0.033 ± 0.001	0.90 ± 0.03	1.96 ± 0.08	0.62 ± 0.05	26.4 ± 0.2
Gunung	0.81 ± 0.01	0.38 ± 0.02	0.170 ± 0.008	0.042 ± 0.013	1.37 ± 0.02	3.27 ± 0.09	0.60 ± 0.03	18.7 ± 0.2
undetermined[i]	0.57 ± 0.02	0.181 ± 0.008	0.061 ± 0.003	0.027 ± 0.009	0.79 ± 0.02	1.90 ± 0.04	0.90 ± 0.02	21.4 ± 0.6

[a] Values derived from sequential method. [b] Cutin values: for carambola, Tean Ma = 0.011 ± 0.005 and No. 44 = 0.085 ± 0.013; for sweet potato peel (Puerto Rico) = 0.006 ± 0.001. [c] Combined average for samples A plus B. [d] Numerical designation of variety. [e] Marsh (sections). [f] Obtained from a local market. [g] The peel fraction was 7.44% of total fresh weight. [h] Obtained from Puerto Rico. [i] Unknown variety, obtained from Puerto Rico.

of pectin is negligible [see Belo and deLumen (1981)].

A comparison of NDR and ADR data from analyses by the nonsequential and sequential methods for five samples is shown in Table I. These samples (carambola, sapodilla, sweet potato, and the Forastero, Gunung, and unspecified yams) were the only samples in which the two methods differed significantly (greater than 0.02–0.04% of fresh weight). For carambola and sapodilla, the sequential ADR and NDR values were greater than the nonsequential, and for ADR in sweet potatoes and the three yams, the nonsequential values were greater. The sequential method has been reported to reduce interference from tannins or acid-insoluble pectins. The lower values from the sequential analyses of sweet potato and the three yams probably indicates that one (or both) was (were) present. Although the pectin content of yams has not been reported, sweet potato had a relatively high total pectin level (Reddy and Sistrunk, 1980). The higher values from sequential analyses of carambola and sapodilla are probably due to other factors (see below).

As the data in Table II show, NDR and enzymatic insoluble fractions were comparable for most of the fruits. In contrast, values for some varieties of starchy vegetables varied considerably. Neutral detergent residue values varied from 0.16 to 5.2%. For most samples, the NDR was between 0.9 and 1.2%. Sapodilla contained much more cellulose, hemicellulose, lignin, and NDR than the other samples.

NDR for starchy vegetables was usually lower than the enzymatic insoluble fraction. The difference between these two is probably due to a combination of insoluble pectin and other insoluble noncellulosic polysaccharides (Schweizer and Wursch, 1979). Apparently the fruits we analyzed did not contain significant amounts of these two fractions. Previous studies (Reddy and Sistrunk, 1980; Campbell and Palmer, 1978) reported 0.8–1.3% total pectin in most commercial sweet potatos (0.2–0.3% water insoluble). The difference between the NDR and the enzymatic insoluble fraction for sweet potato obtained from the supermarket (1.5–1.2 = 0.3%) could therefore be entirely water-insoluble pectin. Pectin in tropical (Puerto Rican) sweet potato and yams has not been reported. Since there are probably large amounts of mannans in yams (Sefa-Dedeh and Rasper, 1977), it seems likely that, in addition to pectin, mucilages of the mannan type may account for the large difference between the NDR and the enzymatic insoluble fraction for certain yams (*Dioscorea esculenta* var. Beti and *Dioscorea alata* var. Gunung).

Since phenolics such as anthocyanins and tannins are abundant in some yams (Martin, 1979), polyphenolic oxidation products can form during the analysis. These products could form insoluble complexes with the proteins or enzymes that could contribute to the abnormally high insoluble fraction found in yams by the enzymatic method.

Pineapple contained somewhat more hemicellulose than most other fruits and vegetables. One of the fresh samples (A) contained more hemicellulose, NDR, and lignin than the other sample (B). Sample A also had a higher dry weight, 16.5 vs. 13.1%. The cellulose content of canned pineapple was slightly greater than that of the fresh fruit. Lignin values of the two canned samples were both low but differed considerably (0.042 vs. 0.021%).

Lignin and cutin were both highly concentrated in carambola. The high cutin value (see footnote *b*, Table II) was apparently derived from the peel, since cutin particles were not observed in the flesh. The No. 44 variety contained larger amounts of cutin and NDR than Tean Ma. The cutin value by the nonsequential method (not reported in Table II) was about half the cutin value reported in Table II for the sequential analyses. The lower NDR value for the nonsequential method (Table I) could have been partly due to reaction of cutin with sulfite (used in this method as an antioxidant) and subsequent formation of a water-soluble product. The difference could also possibly be due to formation of an insoluble protein fraction during the sequential procedure by reaction of protein with oxidized phenolics. These possible sources of error are discussed in detail by Robertson (1978).

Sapodilla had relatively higher amounts of the three major constituents, cellulose, hemicellulose, and lignin. The enzymatic insoluble fraction was also high. The dry weight was relatively high for a fruit (22 vs. 10–17%). Sapodilla contains large quantities of tannins and probably a large amount of latex. Many hard white particles that could be latex particles were observed in the flesh near the seeds. The lower NDR values found in the nonsequential method (Table I) could be partly the result of solubilization of latex by sulfite. Formation of insoluble protein complexes during the sequential procedure could also produce this difference. The role of tannins is more difficult to evaluate since they can affect the results in various ways.

The two mango varieties had very similar composition. Both results agreed closely with the data reported by Shipley (1978). The data for the two Indian varieties (Kamath and Belavady, 1980) differed considerably, and this probably reflects regional or varietal differences.

For fresh grapefruit sections, lignin was lower and cellulose higher than that reported by Shipley (1978) in canned sections. Processed pineapple also contained increased cellulose and slightly decreased lignin as compared with that of fresh; however, the difference was not as pronounced.

Increased lignin content of the peel accounted for most of the difference between the flesh and peel of the sweet potato sample obtained from the supermarket. Thus, 30% of the total lignin was in the peel, even though the peel fraction was only 7% of the total weight. In addition to increased lignin, the peel of the tropical (Puerto Rican) sweet potato contained more hemicellulose than the flesh.

A unique feature of the yam fiber was the very high amount of ash observed in one variety (Muni). Since high ash values are usually associated with silica (Cummings, 1976), it is possible that the Muni variety contains a high concentration of this inorganic material.

In general, these analyses show that some tropical fruits and vegetables contain significant amounts of certain fiber components. For example, lignin was relatively high in carambola, sapodilla, and sweet potato peel, hemicellulose was high in pineapple, sapodilla, and tropical (Puerto Rican) sweet potato peel, and cellulose was relatively high in sapodilla and sweet potato peel. Since these fiber components could have useful physiological properties, some of these tropical fruits and vegetables have potential as sources of dietary fiber.

ACKNOWLEDGMENT

We thank the following individuals for contributions of fruit and vegetables samples: Jose Vincente-Chandler of the USDA Agricultural Experiment Station, Rio Piedras, PR; Antonio Sotomayor-Rios and J. A. Santiago of the Mayaguez Institute of Tropical Agriculture, Mayaguez, PR; Robert J. Knight, Jr., Paul K. Soderholm, and Irene de B. Galvin of the USDA Horticultural Research Station, Miami, FL. We also appreciate the technical assistance offered by the following individuals: Joseph H. Bruemmer and Robert A. Baker of the USDA Citrus and Subtropical Products Laboratory, Winter Haven, Florida; Franklin E.

Barton, II, of the USDA Richard B. Russell Research Center, Athens, GA.

LITERATURE CITED

Belo, P. S.; de Lumen, B. O. *J. Agric. Food Chem.* **1981**, *29*, 370–373.
Bitter, T.; Muir, H. M. *Anal. Biochem.* **1962**, *4*, 330–334.
Campbell, L. A.; Palmer, G. H. In "Topics in Dietary Fiber Research"; Spiller, G. A.; Amen, R. J., Eds.; Plenum Press: New York, 1978; Chapter 4.
Cummings, J. H. In "Fiber in Human Nutrition"; Spiller, G. A.; Amen, R. J., Eds.; Plenum Press: New York, 1976; p 20.
Kamath, M. V.; Belavady, B. *J. Sci. Food Agric.* **1980**, *31*, 194–202.
Martin, F. W. In "Tropical Foods: Chemistry and Nutrition"; Ingeltt, G. E.; Charalambous, G., Eds.; Academic Press: New York, 1979; Vol. I, pp 249–263.
McQueen, R. E.; Nicholson, J. W. G. *J. Assoc. Off. Anal. Chem.* **1979**, *62*, 676–680.
Moreno, A. H.; Diez, M. J. F. *Grasas Aceites (Seville)* **1979**, *30*, 141–143.
Reddy, N. N.; Sistrunk, W. A. *J. Food Sci.* **1980**, *45*, 682–684.
Robertson, J. B. In "Topics in Dietary Fiber Research"; Spiller, G. A.; Amen, R. J., Eds.; Plenum Press: New York, 1978; Chapter 1.
Schweizer, T. F.; Wursch, P. *J. Sci. Food Agric.* **1979**, *30*, 613–619.
Sefa-Dedeh, S.; Rasper, V. F. *Cereal Chem.* **1977**, *54*, 746–759.
Shipley, E. A. In "Topics in Dietary Fiber Research"; Spiller, G. A.; Amen, R. J., Eds.; Plenum Press: New York, 1978; Appendix.

Received for review November 5, 1981. Accepted July 26, 1982. Mention of a trademark or proprietary product is for identification only and does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture, nor does it imply approval to the exclusion of other products that may also be suitable.

Sedimentation Equilibrium Study of the Interaction between Egg White Lysozyme and Ovomucin

Steven M. Miller,[1] Akio Kato,[2] and Shuryo Nakai*

Molecular weight distribution determination using sedimentation equilibrium ultracentrifugation was applied for investigating the interaction of lysozyme with native and reduced ovomucins. This method was found to be more sensitive and reliable than the earlier methods measuring precipitates or turbidity. The interaction seemed to be typically electrostatic: favored by lower temperatures and ionic strengths but decreased by acetylation of lysozyme. Removal of sialic acid residues from ovomucin had little effect. The extent of interaction of lysozyme with native ovomucin in 1:1 (w/w) mixtures was 20–30%, considerably lower than 70–80% with reduced, alkylated ovomucin at an ionic strength of 0.07. However, at an ionic strength of 0.13 the extent of interaction was about 6% with no difference between native and reduced, alkylated ovomucins. On the basis of these results using experimental conditions closer to those of natural egg white than have been previously reported, the ovomucin–lysozyme interaction is unlikely to be a cause of egg white thinning.

A number of workers have suggested that protein–protein interactions between ovomuucin and lysozyme are intimately involved in the maintenance of the gel structure of thick egg white and the process of egg white thinning (Kato and Sato, 1972; Kato et al., 1970a,b, 1971, 1975, 1976; Robinson, 1972; Robinson and Monsey, 1972). Attempts in the past to quantitate the extent of this interaction under varying conditions of pH and ionic strength have been based upon the observation that lysozyme and ovomucin will interact to form an insoluble complex (Dam, 1971; Dam and Bennett, 1963; Kato and Sato, 1972; Kato et al., 1971, 1975, 1976; Robinson, 1972; Robinson and Monsey, 1969). Detailed measurements of the extent of interaction have been obtained by using turbidimetric measurements at 450 nm (Robinson and Monsey, 1969, 1972), 550 nm (Kato et al., 1975, 1976), and 600 nm (Kato and Sato, 1972; Kato et al., 1971) of mixtures of lysozyme and reduced ovomucin. However, since not all interaction products could cause turbidity, these data may not be reliable.

Sedimentation equilibrium ultracentrifugation is more suitable for the study of macromolecule–macromolecule interactions (Howlett and Nichol, 1973). Molecular weight distributions of interacting proteins calculated by multiple regression analysis of sedimentation equilibrium data has recently been used to study the interaction of α_{s1}- and κ-caseins (van de Voort et al., 1979). An advantage of this technique over turbidimetric measurements is that soluble protein–protein interaction products can be detected. Moreover, the use of a UV scanning system provides a direct measure of protein concentration. Therefore, this method may be able to provide direct evidence of a lysozyme–ovomucin interaction.

Ovomucin is a complex of at least two distinct glycoproteins, α- and β-ovomucins (Hayakawa and Sato, 1976; Kato and Sato, 1971, 1972; Kato et al., 1971; Robinson and Monsey, 1971, 1975), and the interaction of lysozyme with β-ovomucin has been reported to be much stronger than that with α-ovomucin. Furthermore, it has been reported that the interaction is electrostatic in nature, involving the negative charges of the terminal sialic acid residues in ovomucin and the positive charges of the lysyl ϵ-amino groups in lysozyme (Kato et al., 1975, 1976). Since these determinations were made by using turbidimetric methods,

Department of Food Science, University of British Columbia, Vancouver, British Columbia, Canada V6T 2A2.
[1]Present address: Department of Neurosciences, McMaster University, Hamilton, Ontario, Canada L8N 3Z5.
[2]Present address: Department of Agricultural Chemistry, Yamaguchi University, Yamaguchi, Japan.

0021-8561/82/1430-1127$01.25/0