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Dietary fiber analysis of cassava using gravimetric methods

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SUMMARY. We report the application of a method which combines digestion with pancreatin and neutral detergent treatment in the analytical study of dietary fiber from cassava. The use of pancreatin previous to the detergent extraction enabled rapid filtration, thus giving more reproducible results for neutral detergent fiber (NDF). Acid detergent fiber (ADF), hemicellulose, lignin and pectin were also determined. The values obtained for NDF (4.65%) and pectin (1.17%) are very important, considering their role in the digestive process.

RESUMEN. Análisis de fibra dietética en casabe utilizando métodos gravimétricos. Se reportan la aplicación de métodos que combinan la digestión con pancreatina y la extracción con detergente neutro en el estudio del contenido de fibra dietética en casabe. A las muestras se les determinó FDN, FDA, hemicelulosa y lignina. Los valores de 4,65 para FDN y 1,17 para pectina son de suma importancia considerando el papel de ambos en el sistema digestivo humano. El empleo de pancreatina antes de la extracción con detergente promueve una filtración rápida con el beneficio de resultados más reproducibles.

INTRODUCTION

Cassava is a typical home food made from *Manihot esculenta crantz,* that is produced and consumed in several country areas of Venezuela. In particular the autochthonous Indian groups have a high consumption of this food. The cholesterol levels of these populations with a high cassava intake have been found to be lower than those from urban areas, and one reason for this could be the high content of dietary fiber (DF) in cassava (3). The DF from other foods is associated with the control of digestion and absorption of different components from foods, including lipids, proteins etc. Also, DF is associated with a reduction in some human diseases such as: bowel cancer, diabetes, hypercholesteremia, amongst others (4,6). For the above reason, it is important to determine the precise content of DF in order to design effective and convenient diets for humans. Although, there are methods which combine enzymatic and gravimetric procedures to determine DF of food samples, most of these give useful results with material that is relatively poor in starch (2,15,16), as they are unsatisfactory for the quantitative determination of DF in starch-rich food (20). These methods are in addition, relatively expensive and not accessible to small laboratories. We tested, however, procedures which combine amylase and detergent treatment to overcome the problem presented by starch (18). We report the analysis of the following DF components of cassava: hemicellulose, cellulose, lignine and pectin. The total starch content was also included in these analysis.

MATERIALS AND METHODS

Cassava was purchased four times per year at different dates in San José de Río Chico, Miranda State, Venezuela. Each stock sample (n=4) was dried between 24 and 48 hrs until reaching constant weight at 50±5°C. All samples were milled

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to a particle size less than 0.4 mm in a Cyclone mill (Tecator, Sweden) and sifted in two groups: (i) 20-30 mesh and (ii) 60-80 mesh. For each stock sample, three assays by octuplicates were carried out.

Analysis of dietary fiber.

0.5g dried samples of cassava were incubated at 40°C for 6 hrs in the presence of 0.033 mg/ml pig-pancreatin (Merck, West Germany) in 0.1M phosphate buffer, 0.056 M NaCl, pH 7.0 (Merck, INC) following the method of Hart and Fisher (7), and slight modification by Asp et al (2). Each sample was washed by filtration with hot water, then with 50 ml acetone and dried to constant weight at 100°C.

The dietary fiber determination was made using the neutral detergent method modified by Holechek and Vavra (8) in order to reduce the time of the assay, as well as the amounts of sample and reactants used. Thus permitting the processing of a greater number of samples. This method was applied to the casava sample previously digested by pancreatin. The acid detergent method was applied to 0.5 g of the first sample group (20-30 mesh) according to Van Soest (22) and modification by Holechek and Vavra (8). Starch was determined in 2,463 g of the second sample group (60-80 mesh) by the method of Hart and Fisher (7).

Pectin was measured starting from the second sample group (60-80 mesh). Briefly, 1.0 gr of this sample was washed with diethyl-ether and dried (1). Pectin was then extracted from the sample according to he method of Sabir (19) and quantitated by the method of Dische (5), modified by McCready (13) and Knutson and Jeanes (12).

Hemicellulose, cellulose and lignin was determined according to Van Soest (22).

The statistical analysis employed was Analysis of Variance (non parametric) by Shefler (21).

RESULTS

When the Van Soest method (22) was applied to a cassava sample, the NDF yielded values about 85% of the dry weight, and with the iodine test this NDF sample was strongly positive. This suggests the presence of starch (data not shown). In addition, the slow rate of filtration during this procedure was a serious problem.

We decided, therefore, to apply the modification to this procedure suggested by Robertson and Van Soest (18). Who proposed the use of amylase and neutral detergent to overcome the problem of high levels of starch. We used first the amylase followed by the detergent because if this order is inverted, the residual starch content increased (unpublished results). This has also been found in other assays with bran, using neutral detergent and amylase (18).

This modified method yielded values of 4.65±0.45% for DF, which is 2.73 times higher than that reported by the National Nutrition Institute (INN) of Venezuela (10). Also the sample was negative to the iodine test, indicating that most of the starch was solubilized.

This difference is probably because the INN determined crude fiber, which is in lower concentration than dietary fiber, and the latter corresponds better to the natural fiber processed during food digestion. On the other hand it is important to note that the first step, in which pancreatin is used, allowed a rapid filtration with an enhancement in reproducibility and precision, previously described by McQueen and Nicolson (14). This is particulary true for vegetable foods with a high content of starch, such as cassava. Values for acid fiber (2.79±0.43%) are shown in Table 1. This value probably corresponds to cellulose (1.76±0.29%) and lignin (0.54±0.04%), the latter being one of the dietary fiber components which, together with pectin, are reported to affect cholesterol levels (9).

TABLE 1
DIETARY FIBER AND STARCH FROM CASSAVA (%)

	Mean	±	SD	C.V
NDF	4.65		0.45	9.84
ADF	2.79		0.43	15.48
Hemicellulose	1.96		0.25	13.07
Cellulose	1.76		0.29	17.37
Lignin	0.56		0.04	7.34
Pectin	1.17		0.10	8.74
Starch	83.02		5.26	6.34

Values are in relative percentage for dry samples.
NDF estimates cellulose, lignin and hemicellulose.
ADF estimates cellulose and lignin.
Abreviations:
NDF: neutral detergent fiber; ADF: acid detergent fiber; C.V.: coefficient of variation.

If the values in Table 1 (NDF, hemicellulose, pectin, starch) are added to the protein content (1.3%), fat content (0.6%) and ash (0,9%) published by the I.N.N. (10), we obtain 93.6% of the total dried cassava composition. The coefficients of variation (C.V.) showed a minumun value for starch of 6.34 and a maximun value of 17.37 for cellulose (Table 1) indicating homogeneity in the estimations.

DISCUSSION

We could not determine quantitatively DF in cassava without applying pancreatin, due to the difficulty in filtering the viscous solution formed by starch. Althought the NDF shown in Table 1 resulted negative to the iodine test, it is not possible to eliminate the possible presence of retrograde starch (20).

From the data obtained we confirm the convenience of applying methods which combine the use of pancreatin and detergent in the determination of DF. Most of the methods

available for the determination of DF give useful results with samples which are relatively poor in starch. Our result of a 1.17% pectin content in DF may be very important, because it retains significant amounts of water in the digestive tract, due to its gelifying property. This, combined with its reported effect in lowering human serum cholesterol levels (11,15), supports the hypothesis of cassava dietary fiber as being a factor in decreasing the cholesterol levels in our Indian population. Experimental studies have frequently, but not invariably, supported this observation. Nevertheless, many factors which may be unrelated to dietary fiber, such as food forms, processing, low intake of fats, intake of unknown vegetable foods, may also affect the rate of digestion. We are interested in measuring the content of pectin in several vegetable foods and to correlate its consumption with other factors in urban area. The above method allow more precise determination of the content of dietary fiber, abolishing the artifacts produced by other components. Further work will be required to determine the effect of such components in lowering the cholesterol levels, and the possible protective role of dietary fiber.

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REFERENCES

1. Asociation of Official Agricultural Chemistry. Officia l Methods of Analysis of the AOAC. 14th ed. Washington, D.C., 1984, p 95.
2. Asp, N.G.; Johansson, C.G.; Hollmer, H. and Siljeström, M. Rapid enzimatic assay of insoluble and soluble dietary fiber. J. Agric food Che, 31, 476-4822, 19833.
3. Bosch, V. and Camejo, G. Serum lipoprotein of Amazonian indians and inhabitants of an urban area of Venezuela fractionated by preparative ultracentrifugation. Metabolism 133, 11456-61, 1964.
4. Cummings, J.H. and Bingham. S.A. Dietary fibre, fermentation and large bowel cancer. Cancer Surv 6, 601-21, 1987.
5. Dische, Z. New color reactions for determinations of sugars in polysaccharides. In Methods of Biochemical Analysis, vol II, (D. Glick, Ed.), Interscience, New York, 1955, p.313
6. Ebeling, P.; Yki-Jarvinen, H,; Aro, A.; Helve, E.; Sinisalo, M. and Kolvisto, V.a. Glucose and lipid metabolism and insulin insensitivity in type 1 diabetes: The effect of guar gum. Am J Clin Nutr 48, 98-11033, 1988.
7. Hart, F.L. and Fisher, H.J. Modern food analysis. Springer-Verlag. New York, U.S.A. 1971. p.70.
8. Holechek, J.L. and Vavra, M. Comparison of micro and macro digestion methods for fiber analysis, J Rug Mgmt 35, 799-801. 1982.
9. Holloway, W.D.; Tasman-Jones, C. and Maher, K. Pectin digestion in humans. Am J Clin Nut 37, 22533-55, 1983.
10. I.N.N. Tabla de composición de alimentos para uso práctico M.S.A.S. Publication Nº 40, Caracas-Venezuela. 1983.
11. Keys, A.; Grande, F. and Anderson, J.T. Fiber and pectin in the diet and serum cholesterol concentration in man. Proc Soc Exp Biol Med. 106-555-58, 1961.
12. Knutson, C.a. and Jeanes, A. New modification of carbazole analysis: Aplication to heteropolysaccharides. Anal Biochem. 224, 470-811, 1968.
13. McCready, R.M. Pectin and pectin acid methods. Carboh Chem 5, 1167-170. 1963.
14. McQueen, R.E. and Nicholson, J.F.W. Fiber analysis. J. Assoc Offic Anal Chem 621, 676-680, 1979.
15. Olson, A.; Gray M.G. and Chiu M-C Chemistry and analysis of soluble dietary fiber. Food Technol 411, 71-80, 1987.
16. Prosky L.; Asp. N-G; Furda I.; Devries, J.G.; Schweizer, T.P.F. and Harland, B.F. Determination of total dietary fiber in food products: Collaborative study. J. Assoc Offic Anal Chem 68, 677-679.
17. Reiser, S. Metabollic effects of dietary pectins related to human health. Food Technology 41, 91-99, 1987.
18. Robertson, J.B. and Van Soest, P.J. The detergent system of analysis and its application to human foods. In basic and clinical nutrition. Eds. W.P.T. James and O. Theander. Marcel Dekker Inc. 1981. vol 3, pp 12233-1156.
19. Sabir, M.A.; Sosulski, F.W. and Stewart, J.C. Polymetaphosohate and oxalate extraction of sunflower pectins. J. Agric Food Chem 24, 348-350, 1976.
20. Selvendran, R.R.; ring, S.G. and Du Pont, S. Determination of the dietary fiber content of the EEC samples and a discussion of the various methods of analysis. In basic and clinical nutrition. Eds. W.P.T. James and O. Theander. Marcel and Dekker Inc. 1981. vol 3, pp 95-119.
21. Shefler, Bioestadística. Edit. Fondo Educativo Interamericano. México. 1979.
22. Van Soest, P.J. Collaborative study on acid-detergent fiber and lignin. J. Assoc Offic Agric Chem 56, 781-784, 1973.