## The Effect of Cellulose Supplementation of Diets on Gastrointestinal Tract Function of Humans

# M. Szakács,<sup>a</sup> A. Halász,<sup>a</sup> I. Sawinsky,<sup>b</sup> Á. Kovács,<sup>b</sup> & L. Vámos-Vigyázó<sup>a</sup>

<sup>a</sup> Department of Biochemistry, Central Food Research Institute, 1022, Budapest Herman Ottó 15, Hungary. <sup>b</sup> 'Balassa János' Municipal Hospital, 1088, Budapest Vas 17, Hungary

(Received 3 April 1987, revised version received and accepted 21 January 1988)

## ABSTRACT

The objective was to determine the cellulase activity of the gastrointestinal tract and the effect of cellulose supplementation of a nutritionally complete diet on cellulose digestion in human adults. All the subjects were given ordinary, low fibre and high fibre  $(3 \times 12g$  wheat bran/subject/day) controlled diets. The cellulose digestion and the cellulase activity of the samples taken of the stomach and small intestine contents were found to be sugnificant. Blood sugar content also tended to increase when dietary fibre was administered to the patients. No correlations have been found between the age or sex of patients and cellulase activity; however, higher values were found for male patients. The quantities of sugars released from dietary fibre can be relevant in the case of diabetes mellitus.

## INTRODUCTION

According to latest data in the literature, dietary fibre supplementation of certain processed foods seems to be advisable (Kay & Strasberg, 1978; Spiller & Kay, 1980; Jenkins *et al.*, 1983; Kies *et al.*, 1984). With some exceptions (Wahlqvist *et al.*, 1979; Tredger *et al.*, 1981; Ullrich & Albrink, 1982), most of the studies have reported beneficial effects. A great variety of

137

Food Chemistry 0308-8146/88/\$03.50 © 1988 Elsevier Science Publishers Ltd, England. Printed in Great Britain commercial products of high fibre content is available on the market. The addition of fibre to the diet was found to lower postprandial glucose levels by adsorption in normal subjects (Goulder *et al.*, 1978) as well as in patients with type II diabetes (Anderson, 1980). In spite of these findings, an elevated fibre content in the diet might give rise to glucose formation, e.g. by the action of cellulases. No information is available on the cellulase activity of the gastrointestinal tract in human adults.

The objective of this work was to investigate whether there was any activity in the gastrointestinal tract or if such activity could be induced by cellulose supplementation of a nutritionally complete diet. A second point was to investigate the effect of such a diet on cellulose digestion and blood sugar content.

### METHODS

### Determination of cellulase activity of human digestive tract

Six adult men and seven women, aged 50-70, served as subjects. All the subjects were given ordinary, low fiber and high fiber  $(3 \times 12g \text{ of wheat} bran)$  controlled diets. The 12-day study was divided into a 3-day adaptation period and three randomly arranged 3-day experimental periods. The day after each period, samples were taken of the stomach and small intestine contents. The cellulose digestion and the cellulase activity of the samples were determined by the reductometric method of Mandels *et al.* (1976) (Table 1). The activity values are given in International Unit/cm<sup>3</sup> (IU). One IU = 1  $\mu$ mole glucose equivalent per minute. The C<sub>1</sub> activities are given in Units (U). 1 U = released glucose equivalent, as mg from 100 mg of cellulose in 24 h at pH 4-8 and 50°C.

Method	Substrate	Time (min)	Reaction product
FPA	Filter paper (Whatman No. 1)	60	Glucose
C <sub>1</sub>	Cotton 100 mg	1440	Cellobiose
C <sub>x</sub>	Carboxymethylcellulose-Na salt (CMC-Na) 1%, 9.5 cm <sup>3</sup>		
	(Sigma, St. Louis)	30	Cellobiose

 TABLE 1

 Cellulase Activity Assays

## **Blood sugar determination**

Blood sugar content was determined according to Sós (1974). 1.5 ml of diluted (1:200) blood sample was boiled with 2 ml of o-toluidine reagent (1.5 g thiocarbamide and 60 ml of o-toluidine made up to 1000 ml with acetic acid). After cooling, the glucose was determined photometrically at 610-650 (635) nm. As standard glucose (1, 2, ... 6 g w/v % in 0.2% cenzoic acid) was used.

Absorbance of sample Absorbance of standard concentration of standard

= blood sugar content (w/v %)

## Administered dietary fiber

The composition of wheat bran was determined according to Hellendoorn *et al.* (1975) (Tables 2 and 3).

## **RESULTS AND DISCUSSION**

The results of the cellulolytic activities of the samples are shown in Figs 1 and 2. The reducing sugar content of the samples was found to be considerable. Higher values were detected after administration of wheat

The Con	position of Wh	eat Bran (Ho	orváth, E., 1987	, unpublished resul	its)
<u> </u>	Water	Fat	Protein	Carbohydrate	Starch <sup>a</sup>
Wheat bran	13.0	4.6	14.5	53	20

 TABLE 2

 The Composition of Wheat Bran (Horváth, E., 1987, unpublished results)

The values are given in g/100 g wheat bran.

" Data according to Schall (1962).

TABLE 3
Dietary Fiber Content of Wheat Bran (Horváth, E., 1987, unpublished
results)

	Water soluble	Water insoluble	Total dietary fiber
Wheat bran	5·6 ± 0·4	53·2 ± 0·8	58·8 ± 0·7

The values are given in g/100 g wheat bran.



**Fig. 1.** Reducing sugar content and cellulase activity of female digestive tract (A) before, and (B) after administration of high fiber diet. Activity (IU/cm<sup>3</sup>): FPA **(B)**; C<sub>1</sub> **(B)**; C<sub>2</sub> **(C)**; reducing sugar content **(C)**. Pairs of columns represent data from (1) stomach and (2) small intestine.



Fig. 2. Reducing sugar content and cellulase activity of male digestive tract (A) before and (B) after administration of high fiber diet. Symbols as in Fig. 1.

	(Fer	I (Female)		II ale)		II. (Induc	l tion)		Ratio inte	o of acti stine an	ivity in s ad stoma	mall chª
					Fen	nale	М	ale	Fem	ale	Ma	le
	S	i	\$	i	5	i	s	i	Before administ	After ration o	Before of high fil	After bre diet
Reducing sugar	2	2	2	2	4	4	4	2	5	3	6	2
FPA-activity	2	1	2	4	4	3	4	5	1	2	1	4
C <sub>1</sub> -activity	3	3	3	5	3	4	5	6	3	2	0	4
C <sub>x</sub> -activity	1	1	5	5	5	2	6	6	0	1	3	6

 
 TABLE 4

 Number of Subjects with Elevated Reducing Sugar Contents in the Stomach and Small Intestine and Induced Cellulolytic Activities after Consumption of High Fibre Diet

In columns I, II and III, the figures in the body of the table mean:  $>0.5 \text{ mg/cm}^3$  reducing sugar;  $>0.10 \text{ IU/cm}^3$  FPA-activity;  $>0.10 \text{ U/cm}^3$  C<sub>1</sub>-activity;  $>0.10 \text{ IU/cm}^3$  C<sub>x</sub>-activity. s = Stomach sample; i = small intestine sample.

<sup>a</sup> In many cases cannot be evaluated, because the initial value was 0.

bran. For the small intestine the increase was significant for all the males and, with one exception, for the females, too (Table 4). For the stomach, the increase was significant for the great majority of the subjects. (Tables 5 and 6). Since the free reducing sugar content of the wheat bran was insignificant, the high sugar content of the samples can be explained by the enzymic degradation of the starch content of the wheat bran. Acidic breakdown in the stomach can also lead to the release of reducing sugars. No digestive disorders were observed when dietary fiber was administered to the patients. No correlations have been found between the age of subjects and cellulase activity. In some instances, however, higher values were found for male subjects (Figs 1 and 2).

## Female subjects

In the case of females, no or only small quantities (subjects 2, 4, 5, 7) of FPA activity could be detected in the subjects before administration of wheat bran. The FPA activity was found to be significantly inducible in the stomach and small intestine of subjects 6 and 5, 6, 7, respectively. In 50% of the cases, higher FPA activity was found in the stomach than in the small intestine, which might be due to additional reducing sugar formation caused by the acidic breakdown of the filter paper in the extremely acidic medium in the stomach. In subjects 3 and 4 the C<sub>1</sub> activity was found to be lost after

iect	nean (me eli	ucing sugar cu ucose equinale	ontent vnt/cm <sup>3</sup> )		FPA			ت ا			പ്	
ļ	0				ť			ſ	,	•	ſ	•
	Y	R.	1	V	R	1	V	R	7	¥	Ð	-
						Female stu	omach					
	$0.34 \pm 0.01$	$0.08 \pm 0.01$	31-84***	Q	0	0	0	0	0	0	ĉ	0
	$0.48 \pm 0.02$	$0.46 \pm 0.02$	1-66	$0.03 \pm 0.01$	$0-01 \pm 0-0$	3-46	0	0	0	0	00 <del>+</del> 001	10-39**
	$0.20 \pm 0.01$	$0.42 \pm 0.01$	26.94**	0	$0.02 \pm 0.0$	0	$0.04 \pm 0.01$	0	<del>\$</del> -93	0	$0.03 \pm 0.02$	260
_	$0.10 \pm 0.01$	$0.10 \pm 0.01$	0	0	0	0	0	0	0	0	$0.02 \pm 0.01$	3.46
	$0.21 \pm 0.01$	$0.31 \pm 0.01$	12-25**	$0.01 \pm 0.0$	$0.02 \pm 0.01$	1-73	$0.04 \pm 0.02$	0-11 ± 0-01	5.42*	0	0	0
	$0.42 \pm 0.01$	$0.60 \pm 0.01$	22-05**	0	$0.12 \pm 0.03$	6.93*	$0.22 \pm 0.01$	$0.31 \pm 0.01$	11-02**	0	$0.02 \pm 0.01$	38-11==
	$0.50 \pm 0.01$	$0.90 \pm 0.01$	48.99***	$0.09 \pm 0.01$	$0.10 \pm 0.01$	1·22	$0.72 \pm 0.06$	$0.78 \pm 0.02$	1-64	0	$0.22 \pm 0.01$	38-11**
						<sup>-</sup> emale small	l intestine					
	$0.46 \pm 0.02$	$0.04 \pm 0.01$	32-53***	0	$0.01 \pm 0.0$	0	0	0	0	0	0	•
	$0.49 \pm 0.01$	$0.42 \pm 0.01$	8-57*	$0.03 \pm 0.02$	0	2.60	0	$0.60 \pm 0.01$	10-39**	0	0	0
	$0.20 \pm 0.01$	$0.39 \pm 0.01$	23-27**	0	0	0	$0.12 \pm 0.01$	$0.02 \pm 0.01$	12-25**	0	0	0
	$0.02 \pm 0.01$	$0.12 \pm 0.01$	12-25**	$0.02 \pm 0.0$	0	0	$0.14 \pm 0.01$	0	23.5**	0	0	0
	$0.26 \pm 0.01$	$0.26 \pm 0.01$	•	0	$0.06 \pm 0.01$	10-39**	$0.06 \pm 0.02$	$0.46 \pm 0.02$	30-2**	$0.02 \pm 0.01$	$0.02 \pm 0.01$	0
_	$0.46 \pm 0.01$	$0.82 \pm 0.01$	44.09***	0	$0.08 \pm 0.01$	13-86**	$0.30 \pm 0.01$	$0.37 \pm 0.02$	5.6*	0	$0.01 \pm 0.0$	0
	$0.40 \pm 0.01$	$0-99 \pm 0-01$	72.20***	$0.05 \pm 0.01$	$0.21 \pm 0.01$	19-60**	$0.10 \pm 0.01$	$0.15 \pm 0.01$	8-5*	0-09 ± 0-02	$0.30 \pm 0.02$	12-86**

and Callulace Activity of Famala Directive Tract TABLE S Deducing Sugar Content

compared by the *t*-test. t: critical value of *t*-test. Values with asterisk superscripts indicate significant differences between the respective A and B values: \* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001.

142

TABLE 6 Reducing Sugar Content and Cellulase Activity of Male Digestive 1		Irac
TABLE 6           Reducing Sugar Content and Cellulase Activity of Male D		igestive <b>T</b>
<b>TABLE 6</b> Reducing Sugar Content and Cellulase Activity o		f Male D
TABLE Reducing Sugar Content and Cellulase	9	Activity o
Reducing Sugar Content and (	TABLE	Cellulase
Reducing Sugar Content		and (
Reducing Sugar		Content
Reducing		Sugar
		Reducing

Effect of cellulose in diets on human gastrointestinal tract

wheat bran administration. This may be due to the adsorption of the enzyme on the cotton surface.

In subjects 5, 6, 7 considerable  $C_1$ -activity was induced by wheat bran. With the exception of subject 7, higher activity values were found in the small intestine. Very low quantities of  $C_x$  activity could be detected in the subjects. However, this activity was also found to be inducible, mainly in the stomach (Fig. 1). (The increase was significant feel stomachs of subjects 2, 6 and 7.)

#### Male subjects

In the majority of cases higher activity values were found for the male subjects (Fig. 2). In subjects 1 and 2 the FPA activity of the stomach was induced to a considerable extent after wheat bran administration. However, the increases were significant for subjects 3, 5 and 6 as well (Tables 5 and 6). The highest  $C_1$  and  $C_x$  activities were detected also in subjects 1 and 2, although the induced  $C_x$  activities were significantly higher for all the subjects.



**Fig. 3.** Blood sugar content before and after administration of high fiber diet. Empty stomach (------); 0.5 h sample (-----); 1 h sample (----).

Sign of subject	Blood sug samj	gar content of ples (mol/dm <sup>3</sup> )	0•5 <i>h</i>	Sign of subject	Blood sugar content o samples (mol/dm <sup>3</sup>		gn of Blood sugar content of 1 h bject samples (mol/dm <sup>3</sup> )	f 1 h
	A	B	t	-	A	B	t	
1	4·37 ± 0·15	$5.23 \pm 0.06$	9.22*	1	5·40 ± 0·10	$6.63 \pm 0.15$	11.82**	
2	$102 \pm 0.10$	$10.33 \pm 0.06$	1.93	2	$4.50 \pm 0.10$	$5.97 \pm 0.06$	21.83**	
3	5·8 ± 0·10	5·87 ± 0·06	1.18	3	$6.17 \pm 0.06$	$6.50 \pm 0.2$	2.74	
4	5·3 ± 0·20	5·33 ± 0·06	0.25	4	5·77 ± 0·06	$5.03 \pm 0.55$	2.32	
5	6·03 ± 0·15	$6.33 \pm 0.06$	3.22	5	$5.17 \pm 0.06$	$5.27 \pm 0.06$	2.04	
				6	$6.27 \pm 0.06$	6·83 ± 0·06	11-43**	
				7	10·63 ± 0·12	$10.93 \pm 0.06$	3.87	
				8	$6.27 \pm 0.06$	6·8 ± 0·10	<b>?·</b> 87*	

 TABLE 7

 Blood Sugar Content before and after Administration of High Fiber Diet

A: Blood sugar content before administration of high fiber diet. Mean values of three replicates.

B: Blood sugar content after administration of high fiber diet. Mean values of three replicates.

A and B values pertaining to a given subject and parameter were compared by the *t*-test. t: critical value of *t*-test.

Values with asterisk superscripts indicate significant differences between the respective A and B values: \* P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001.

Induced activities were found in the other male subjects as well. With the exception of subjects 1 and 2 the small intestine activities were found to be always higher than those found in the stomach fluid (Table 4). All the induced activities were significantly higher than the control values, except for the  $C_1$  activity in subject 6.

No or only slight increase was detected in the blood sugar content 30 min after wheat bran administration (Fig. 3). In the 1 h samples the increase was significant in 50% of the samples. (Table 7).

## CONCLUSION

The results of this study indicated that dietary fiber was enzymatically digested in the human gastrointestinal tract. Of the substrates applied for the determination of the cellulolytic activity only glucose and/or cellobiose can be generated by the enzyme. The wheat bran contains hemicelluloses which are preferentially hydrolysed already at the beginning of the process. However, the blood sugar (glucose) content increases after administration of wheat bran in 50% of the cases. To our knowledge no evidence can be found

in the literature for the conversion of pentoses (e.g. xylose) into glucose by microorganisms; we must therefore assume that the  $\beta$ -bonds of the oligomers are split into glucose by cellulases.

The digestion of the starch content of the wheat bran starts already in the mouth tract due to  $\alpha$ -amylase. The glucose liberated from the starch is resorbed in the mouth. No differences would be found in the glucose production if it were due exclusively to the hydrolysis of starch.

In the case of microbial contamination there would be no difference in glucose production before and after administration of wheat bran.

The cellulolytic activities were found to be inducible by the addition of dietary fiber. This is particularly well supported by the results obtained with male subjects (for the stomach and the small intestine, respectively, out of 18 data each only 4 and 1 were non-significant). The quantity of the sugars thus released might be relevant for sufferers with diabetes mellitus. In the majority of the subjects investigated, considerable cellulolytic activities were found even without administration of wheat bran. This might be related to eating habits.

## ACKNOWLEDGEMENT

The skilful assistance of Tünde Csikhelyi is gratefully acknowledged.

#### REFERENCES

- Anderson, J. W. (1980). The role of dietary carbohydrate and fiber in the control of diabetes. In *Advances in Internal Medicine*, Vol. 26, ed G. H. Stollernan, Yearbook Medical Publishers, Chicago.
- Goulder, T. J., Alberti, K. G. M. M. & Jenkins, D. A. (1978). Effects of added fiber on the glucose and metabolic response to a mixed meal in normal and diabetic subjects. *Diabetes Care*, 1, 351-5.
- Hellendoorn, E. W., Noordhoff, M. G. & Slogman, J. (1975). Enzymatic determination of the indigestible residue (dietary fiber) content of human food. J. Sci. Food Agric., 26, 1461–8.
- Jenkins, D. J. A., Wolever, T. M. S., Jenkins, A. L., Lee, R. Wong, G. S. & Josse, R. (1983). Glycemic response to wheat products: Reduced response to pasta but no effect to fiber. *Diabetes Care*, 6, 155-9.
- Kay, R. N. & Strasberg, S. N. (1978). Origin, chemistry, physiological effects and clinical importance of dietary fiber. *Clin. Invest. Med.*, 1, 9-24.
- Kies, C., Sanchez, V. E. & Fox, H. M. (1984). Cellulose supplementation of a nutritionally complete, liquid formula diet: Effect on gastrointestinal tract function of humans and fecal fiber recovery. J. Food Sci., 49, (3), 815-16.

146

- Mandels, M., Andreotti, R. & Roche, C. (1976). Measurement of saccharifying cellulase. In *Enzymatic Conversion of Cellulosic Materials: Technology and Applications*, ed by E. L. Gaden, M. H. Mandels, E. T. Reese & L. A. Spano, J. Wiley, New York, pp. 21-33.
- Sós, J. (ed.) (1974). Determination of blood sugar content. In: Laboratory Diagnostics. Medicina, Budapest, p. 176.
- Spiller, C. A. & Kay, R. M. (Ed.) (1980). *Medical Aspects of Dietary Fiber*, Plenum Press, New York.
- Schall, H. (1962). Nahrungsmitteltabelle, Johann Ambrosius Barth Verlag, Leipzig.
- Tredger, J., Sheard, C., & Marks, V. (1981). Blood glucose and insulin levels in normal subjects following a meal with and without added sugar beet pulp. *Diabetes Metab.*, 7, 169–72.
- Ullrich, I. H. & Albrink, M. J. (1982). Lack of effect of dietary fiber on serum lipids, glucose, and insulin in healthy young men fed high starch diets. Am. J. Clin. Nutr., 36, 1-9.
- Wahlqvist, M. L., Morris, M. J., Littlejohn, G. O., Bond, A. & Jakson, R. V. J. (1979). The effect of dietary fiber on glucose tolerance in healthy males. Aust. NZ J. Med., 9, 154–8.