

# Calcium and Magnesium Absorption from the Colon and Rectum Are Increased in Rats Fed Fructooligosaccharides<sup>1</sup>

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**ABSTRACT** We investigated the effects of fructooligosaccharides on the absorption of calcium, magnesium and water from the colon and rectum of rats fed a control diet or the control diet containing 50 g fructooligosaccharides/kg. Chromium-mordanted cellulose was used as an unabsorbable marker to calculate apparent absorption of calcium and magnesium. There was a positive correlation ( $r = 0.982$ ,  $P < 0.001$  in rats fed the control diet and  $r = 0.975$ ,  $P < 0.001$  in rats fed the fructooligosaccharides-containing diet) between the amount of chromium and the dry weight of each fecal pellet in the colon and rectum. Ratios of calcium to chromium and magnesium to chromium in fecal pellets in the colon and rectum were calibrated from the Ca:Cr and Mg:Cr ratios of cecal contents. In rats fed the fructooligosaccharides-containing diet, but not in rats fed the control diet, these ratios were correlated with the fractional length of transit along the colon and rectum, indicating linear disappearance of calcium and magnesium during the colorectal passage. Total apparent absorption of calcium and magnesium, predicted from regression equations with the Ca:Cr and Mg:Cr ratios of cecal contents, agreed well with those calculated from the Ca:Cr and Mg:Cr ratios of feces. The consumption of fructooligosaccharides did not affect net water absorption from the colon and rectum. These results indicated that fructooligosaccharides significantly increased calcium and magnesium absorption and that indigestible and fermentable carbohydrate facilitates colorectal absorption of calcium and magnesium. *J. Nutr.* 125: 2417-2424, 1995.

### INDEXING KEY WORDS:

- calcium • magnesium • rats
- fructooligosaccharides • large intestine

calcium and magnesium absorption is unknown. Recent reports demonstrated that the cecum and colon have a large capacity for calcium and magnesium absorption (Karch 1989, Karch and Feldmeier 1993, Lutz and Scharrer 1991, Scharrer and Lutz 1990).

We found that fructooligosaccharides, a mixture of 1-kestose, nystose and 1F- $\beta$ -fructofuranosyl nystose, which are not digestible by human enzymes, increased the total calcium, magnesium and phosphorus absorption in rats (Ohta et al. 1993, 1994a and 1994b). Other indigestible carbohydrates such as resistant starch (Schulz et al. 1993) and inulin (Rémésy et al. 1993) have similar effects. However, the site of action (i.e., small intestine vs. hindgut) on these carbohydrates is unclear. Several authors have suggested that indigestible carbohydrates increase calcium and magnesium absorption in the small intestine (Brommage et al. 1993, Goda et al. 1992, Heijnen et al. 1993). On the other hand, the effects of feeding indigestible carbohydrates on mineral absorption from the hindgut can be attributed to the effect of short-chain fatty acids (SCFA) produced by the fermentation of indigestible carbohydrates in the large intestine. A mixture of acetate and propionate or acetate and butyrate stimulated cecocolonic calcium and magnesium absorption in humans and rats (Lutz and Scharrer 1991, Scharrer and Lutz 1990, Trinidad et al. 1993). Thus it appears that both the small and large intestine are involved, but the relative contributions of these two regions of gut to such increases in

Both the small and large intestine have the ability to absorb calcium and magnesium (Hardwick et al. 1991, Marcus and Lengmann 1962). However, the segment of the intestine that is the main site for net

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total calcium and magnesium absorption in response to indigestible carbohydrates remain unclear.

In this study we assessed the absorption of calcium, magnesium and water along the colon and rectum quantitatively using chromium-mordanted cellulose as an unabsorbable marker. We also expected to exclude the possible effect of coprophagy by using this technique (Cree et al. 1986).

## MATERIALS AND METHODS

**Animals and diets.** Six-week-old male Sprague-Dawley rats (Clea Japan, Tokyo, Japan) were housed in individual stainless-steel wire-mesh cages in a room maintained at 25°C and 55% relative humidity. All rats were fed a pelleted diet (MF, Oriental Yeast, Tokyo, Japan) for 1 wk; they were then divided into two experimental subgroups of 14 rats each and fed one of two experimental diets (Table 1). Fructooligosaccharides were added to one diet at 50 g/kg diet, by replacing an equal amount of sucrose in the control diet with fructooligosaccharides. Both diets contained the same level of chromium-mordanted cellulose. Animals were allowed free access to their respective diets and to water throughout the experimental period.

Animals were maintained in accordance with the guidelines for animal experimentation of the Japanese Association for Laboratory Animals (1987).

**Preparation of chromium-mordanted cellulose (Cr-cellulose).** The method is based on that of Udén et al.

(1980). One part cellulose (AVICEL®, Asahi Chemical Industry, Tokyo, Japan) was mixed in a glass beaker with four parts (v/v) Na<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>·2H<sub>2</sub>O solution containing chromium equivalent to 13 g/100 g of the cellulose. After the beaker was covered with aluminum foil, the mixture was baked at 100°C for 24 h. The baked mixture was thoroughly washed with tap water and then suspended in tap water. Ascorbic acid (one half the weight of cellulose) was added to the suspension. After standing for at least 1 h, the mordanted cellulose was washed thoroughly with tap water until the effluent became free of green color. The material was dried at 65°C.

**Collection of samples.** All feces were collected between 0600 and 0800 h on d 7 of the experimental diet. At 0600 h on d 8, rats were killed by an overdose of diethyl ether inhalation, and the cecum, colon and rectum quickly removed. Distances between the cecocolonic junction and the anus and between the cecocolonic junction and each fecal pellet in the colon and rectum were measured. Cecal contents and all fecal pellets in the colon and rectum were collected separately for further analysis.

**Quantification of calcium, magnesium, chromium and water.** The amount of calcium, magnesium and chromium in the diets, cecal contents, fecal pellets in the colon and rectum and excreted feces was determined with an inductive coupled plasma emission spectrometer (ICPS-5000, Shimadzu, Kyoto, Japan). All samples were weighed, dried at 110°C for 6 h and reweighed. Water content was calculated by subtracting the dry weight from the fresh weight of the same sample. The dried samples were then ashed at 550°C for 24 h in a muffle furnace. The ashed samples were further processed in preparation for mineral analysis by the method of Williams et al. (1962).

**Calculations.** Fractional lengths of transit (*Lt*) of each fecal pellet in the colon and rectum were calculated as follows:

$$Lt (\%) = \frac{\text{distance from the cecocolonic junction to pellet (mm)}}{\text{distance from the cecocolonic junction to anus (mm)}} \times 100$$

The apparent absorption of calcium and magnesium from each sample (excreted feces or cecal contents) was calculated as the proportion of decreased calcium and magnesium according to Ali and Evans (1967).

$$\text{Sample apparent calcium absorption (\%)} =$$

$$\left[ 1 - \frac{\text{Ca:Cr (mol/mol) in sample}}{\text{Ca:Cr (mol/mol) in the diet}} \right] \times 100$$

$$\text{Sample apparent magnesium absorption (\%)} =$$

$$\left[ 1 - \frac{\text{Mg:Cr (mol/mol) in sample}}{\text{Mg:Cr (mol/mol) in the diet}} \right] \times 100$$

TABLE 1

Composition of experimental diets

Ingredient	Control diet	FO diet
	g/kg diet	
Casein	250	250
Cornstarch	495	495
Corn oil	60	60
AIN-76 vitamin mix	10	10
AIN-76 salt mix	35	35
Cellulose <sup>1</sup>	50	50
Sucrose	100	50
Fructooligosaccharides <sup>2</sup>	—	50
	mmol/kg diet <sup>3</sup>	
Calcium	103	103
Magnesium	16.9	16.8
Chromium	4.54	4.60
	mol/mol	
Ca:Cr	22.6	22.4
Mg:Cr	3.72	3.65

<sup>1</sup>AVICEL®, Asahi Chemical Industry.

<sup>2</sup>Meiologo-P® (concentration of oligosaccharides were >95% of total mixture).

<sup>3</sup>After chemical analysis described in the text.

The Ca:Cr and Mg:Cr ratios (mol/mol) in each pellet in the colon or rectum were calibrated from their ratios in cecal contents by the following formula:

Calibrated Ca:Cr (mol/100 mol) =

$$\frac{\text{Ca:Cr (mol/mol) of pellet in the colon and rectum}}{\text{Ca:Cr (mol/mol) of cecal contents}} \times 100$$

Calibrated Mg:Cr (mol/100 mol) =

$$\frac{\text{Mg:Cr (mol/mol) of pellet in the colon and rectum}}{\text{Mg:Cr (mol/mol) of cecal contents}} \times 100$$

Total apparent absorption of calcium and magnesium predicted from regression equations between the fractional length of transit and the calibrated Ca:Cr or Mg:Cr ratio of pellet in the colon and rectum with Ca:Cr or Mg:Cr ratio values of the cecum, was calculated using the following formula:

Calculated Ca:Cr of pellet in the colon and rectum (mol/100 mol) =  $A \times Lt + B$

Calculated Ca:Cr (mol/100 mol) of feces =  $A \times 100 + B$  (1)

Calculated Mg:Cr of pellet in the colon and rectum (mol/100 mol) =  $A \times Lt + B$

Calculated Mg:Cr (mol/100 mol) of feces =  $A \times 100 + B$  (1')

where A and B are the regression slope and intercept, respectively (Table 4).

Apparent calcium absorption (%) =

$$100 - \frac{\text{Cecal Ca:Cr (mol/mol)} \times \text{calculated Ca:Cr (mol/100 mol) of feces}}{\text{Ca:Cr (mol/mol) in the diet}} \quad (2)$$

Apparent magnesium absorption (%) =

$$100 - \frac{\text{Cecal Mg:Cr (mol/mol)} \times \text{calculated Mg:Cr (mol/100 mol) of feces}}{\text{Mg:Cr (mol/mol) in the diet}} \quad (2')$$

**Chemicals.** Fructooligosaccharides are a mixture of 42% 1-kestose, 46% nystose and 9% 1F- $\beta$ -fructofuranosyl nystose (Meiologo-P®, Meiji Seika Kaisha, Tokyo, Japan) (Hidaka et al. 1988). Other dietary components apart from the minerals were from Oriental

Yeast. All other reagents were of analytical grade from Wako Pure Chemical (Tokyo, Japan).

**Statistics.** The data are expressed as mean values with SD. The Mann-Whitney U test (Ichihara 1991) was used to compare means between groups. A simple linear regression equation was calculated by the least squares method using Microsoft Excel Version 4.0 (Microsoft, Tokyo, Japan). Student's *t* tests were used to compare regression slopes between groups (Ichihara 1991).

When at least one data set was distributed parallel to the x-axis (Fig. 2, 3), the entire data set was rotated counterclockwise by 45° using the following conversion:

$$Xi = xi + yi \times \cos [\pi/4 + \tan^{-1} (yi/xi)],$$

$$Yi = xi + yi \times \sin [\pi/4 + \tan^{-1} (yi/xi)],$$

where *xi* and *yi* denote original data and *Xi* and *Yi* denote converted data. Slopes of regression lines on converted data were then compared. Differences were considered significant at  $P < 0.05$ .

## RESULTS

**Body weight and food consumption.** Initial and final body weights did not differ between the two groups (Table 2). Total food intake of rats fed the fructooligosaccharides diet was 9.7% less ( $P < 0.05$ ) than that of rats fed the control diet. Therefore, the intake of calcium, magnesium and chromium by rats fed the fructooligosaccharides diet was 8.5–10.1% less ( $P < 0.05$ ) than that of rats fed the control diet.

**Correlation between dry weight and amount of chromium in fecal pellets in the colon and rectum.** A total of 65 fecal pellets from the colon and rectum of

TABLE 2

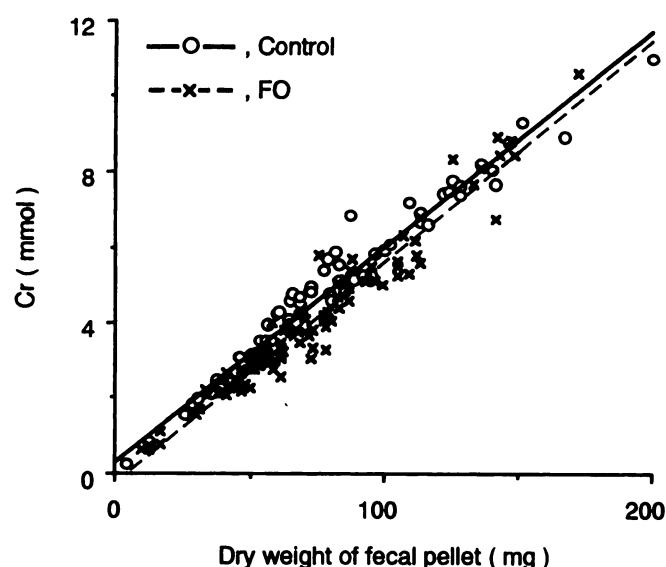
**Body mass and intake of food, calcium, magnesium and chromium in rats fed control or fructooligosaccharides (FO) diet<sup>1</sup>**

	Control diet		FO diet <sup>2</sup>	
Body mass				
Initial, g	234	± 13	232	± 14
Final, g	285	± 16	280	± 15
Food intake, g/d	22.1	± 2.3	19.9	± 1.9*
Mineral intake				
Calcium, $\mu\text{mol/d}$	2262	± 232	2052	± 191*
Magnesium, $\mu\text{mol/d}$	372	± 38	334	± 31*
Chromium, $\mu\text{mol/d}$	100	± 10	92	± 9*

<sup>1</sup>Values are means  $\pm$  SD,  $n = 14$ .

<sup>2</sup>One half the sucrose (50 g/kg diet) of the control diet was replaced with the same mass of fructooligosaccharides.

\*Significantly different from control group ( $P < 0.05$ ).



**FIGURE 1** Relationship between the amount of chromium and the dry weight of each fecal pellet in the colon and rectum of rats fed the control or fructooligosaccharides (FO) diet. Control diet group. Regression equation:  $y = 15.268 + 2.971x$ ,  $r = 0.982$ ,  $P < 0.001$ , ( $n = 79$ ). FO diet group. Regression equation:  $y = -12.722 + 3.041x$ ,  $r = 0.975$ ,  $P < 0.001$ , ( $n = 96$ ).

14 control rats and 82 fecal pellets from the colon and rectum of 14 fructooligosaccharides-fed rats were collected at killing. Dry weight and the amount of chromium in each pellet were linearly correlated ( $r = 0.982$ ,  $P < 0.001$  in control rats and  $r = 0.975$ ,  $P < 0.001$  in fructooligosaccharides diet-fed rats) in both groups (Fig. 1).

**Analysis of cecal contents and excreted feces.** Forty-four fecal pellets from 10 rats in the control group (four rats excreted no feces during the sampling period) and 57 fecal pellets from 12 rats in the fructooligosaccharides group (two rats excreted no feces during the sampling period) were collected. The wet weight of cecal contents from fructooligosaccharides diet-fed rats was ~three times ( $P < 0.001$ ) that of control rats (Table 3). Concentrations of calcium, magnesium and chromium as well as the Ca:Cr and Mg:Cr ratios were lower in both cecal contents and feces in fructooligosaccharides rats than in control rats.

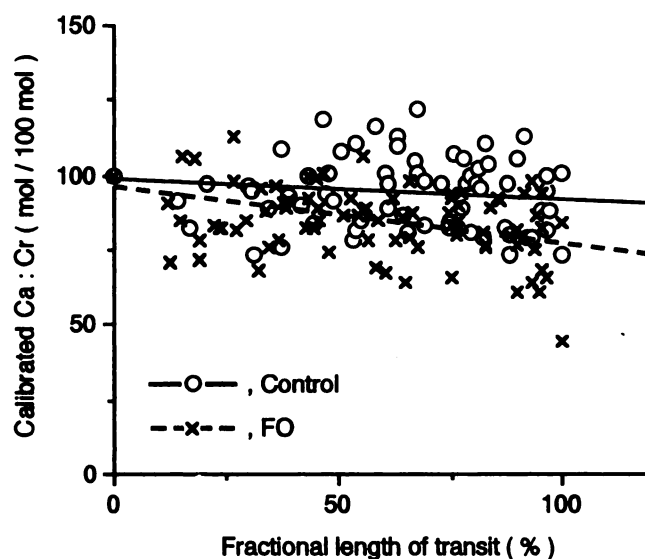
**Fractional length of transit from cecum to anus and calibrated Ca:Cr or Mg:Cr ratio of fecal pellets in the colon and rectum.** Both the calibrated Ca:Cr ratio ( $r = -0.513$ ,  $P < 0.001$ ) and the calibrated Mg:Cr ratio ( $r = -0.745$ ,  $P < 0.001$ ) correlated negatively and linearly to the fractional length of transit in fructooligosaccharides-fed rats, but not in control rats (Fig. 2, 3).

**Correlation between fractional length of transit from cecum to anus and water content of the contents in the colon and rectum.** The fractional length

of transit and the water content of the colonic and rectal contents correlated negatively and linearly in both the control ( $r = -0.903$ ,  $P < 0.001$ ) and fructooligosaccharides ( $r = -0.876$ ,  $P < 0.001$ ) rats (Fig. 4).

**Intergroup comparison of regression slopes.** The calibrated Ca:Cr and Mg:Cr ratios remained constant over a wide range of values for the fractional length of transit in the control group. Therefore, all data were rotated counterclockwise by  $45^\circ$  to compare the slopes of the regression lines for both groups. The slopes of the rotated regression lines of the fractional length of transit and the calibrated Ca:Cr and Mg:Cr ratios calibrated differed between the control and fructooligosaccharides groups (calcium:  $P < 0.02$ , magnesium:  $P < 0.001$ ). However, the slopes of the regression lines between the fractional length of transit and the water content of the colonic and rectal contents did not differ between the two groups (Table 4).

**Apparent absorption of calcium and magnesium.** The apparent absorption of calcium and magnesium was significantly higher in the fructooligosaccharides group than in the control group (Table 5). The apparent absorption of calcium and magnesium calculated from the regression equation agreed well with that calculated from the Ca:Cr and Mg:Cr ratios in feces (Table 5). The absolute amounts of calcium and magnesium that were absorbed were also larger in fructooligosaccharides diet-fed rats than in control rats ( $P < 0.01$ ).



**FIGURE 2** Relationship between the fractional length of transit and Ca:Cr ratio of each pellet in the colon and rectum calibrated from cecal Ca:Cr ratio of rats fed the control or fructooligosaccharides (FO) diet. Control diet group. Regression equation:  $y = 98.568 - 0.0635x$ ,  $r = 0.184$ ,  $P = 0.1025$ , ( $n = 79$ ). FO diet group. Regression equation:  $y = 96.457 - 0.197x$ ,  $r = 0.513$ ,  $P < 0.001$ , ( $n = 96$ ).



TABLE 3

*Analysis of cecal contents and feces of rats fed control or fructooligosaccharides (FO) diet<sup>1</sup>*

	Control diet	FO diet <sup>2</sup>
<b>Cecal contents</b>		
Wet weight, g	1.78 ± 0.38	6.13 ± 2.71*
Water, g/100 g	70.5 ± 1.3	73.0 ± 5.8
Calcium, μmol/g dry contents	849 ± 80	662 ± 123*
Magnesium, μmol/g dry contents	99.6 ± 12.0	62.9 ± 21.1*
Chromium, μmol/g dry contents	58.8 ± 3.6	52.7 ± 3.8*
Ca:Cr, mol/mol	14.4 ± 1.2	12.6 ± 2.3
Mg:Cr, mol/mol	1.70 ± 0.19	1.20 ± 0.40
<b>Feces</b>		
Calcium, μmol/g dry contents	839 ± 70	471 ± 119*
Magnesium, μmol/g dry contents	105.3 ± 3.2	30.5 ± 13.0*
Chromium, μmol/g dry contents	59.6 ± 2.3	50.4 ± 5.9*
Ca:Cr, mol/mol	14.09 ± 0.39	9.42 ± 2.60*
Mg:Cr, mol/mol	1.77 ± 0.21	0.62 ± 0.29*

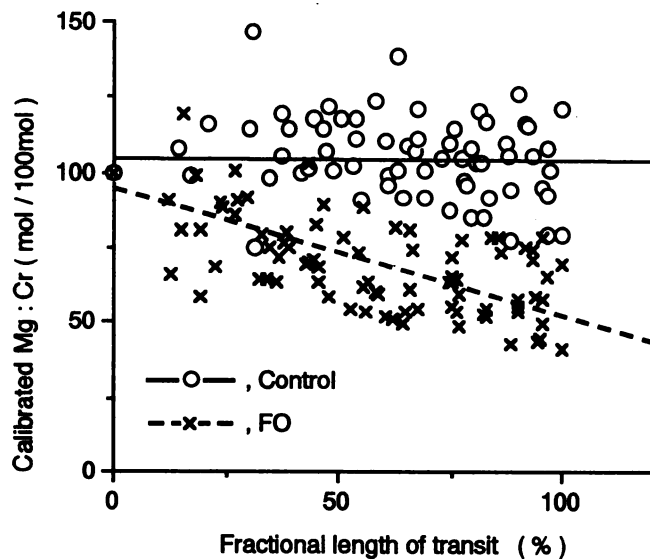
<sup>1</sup>Values are means ± SD, n = 10–14.<sup>2</sup>One half the sucrose (50 g/kg diet) of the control diet was replaced with the same mass of fructooligosaccharides.\*Significantly different from control group ( $P < 0.05$ ).

## DISCUSSION

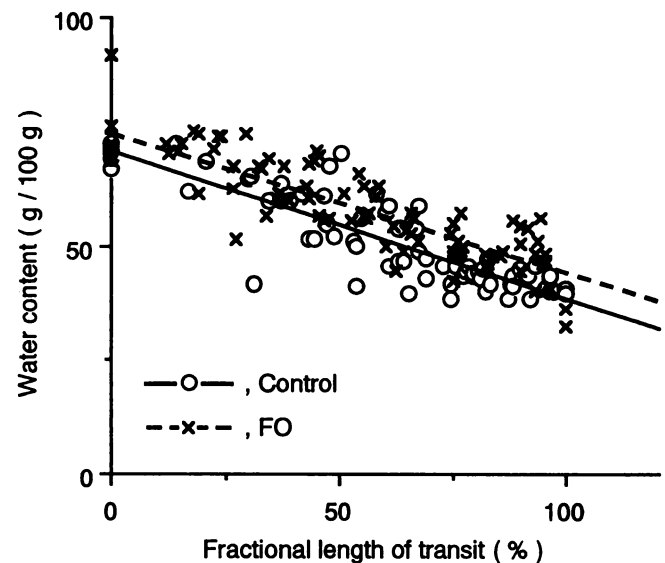
The linear correlation between the amount of chromium and the dry weight of each fecal pellet in the colon and rectum of the control and fructooligosaccharides groups (Fig. 1) indicated a homogeneous distribution of this marker throughout the

hindgut contents and supported the usefulness of Cr-mordanted cellulose as an unabsorbable marker.

The significant linear decline in the calibrated Ca:Cr and Mg:Cr ratios in pellets along the colon and rectum in rats fed the fructooligosaccharides diet, but not in the control rats (Fig. 2 and 3), indicated that fructooligosaccharides feeding increased net



**FIGURE 3** Relationship between the fractional length of transit and Mg:Cr ratio of each pellet in the colon and rectum calibrated from cecal Mg:Cr ratio of rats fed the control or fructooligosaccharides (FO) diet. Control diet group. Regression equation:  $y = 104.850 - 0.00711x$ ,  $r < 0.001$ ,  $P = 0.8732$ , ( $n = 79$ ). FO diet group. Regression equation:  $y = 94.502 - 0.420x$ ,  $r = 0.745$ ,  $P < 0.001$ , ( $n = 96$ ).



**FIGURE 4** Relationship between the fractional length of transit and water concentration of each pellet in the colon and rectum of rats fed the control or fructooligosaccharides (FO) diet. Control diet group. Regression equation:  $y = 70.929 - 0.323x$ ,  $r = -0.903$ ,  $P < 0.001$ , ( $n = 79$ ). FO diet group. Regression equation:  $y = 74.551 - 0.304x$ ,  $r = -0.876$ ,  $P < 0.001$ , ( $n = 96$ ).

TABLE 4

*Slopes and intercepts of regression equations between fractional length of transit (Lt) and Ca:Cr, Mg:Cr or water in colorectal contents of rats fed control or fructooligosaccharides (FO) diet with or without data rotation*

Variables		Rotation	Experimental group	Slope	Intercept	$r^1$
X (Independent)	Y (Dependent)					
Lt	Ca:Cr	None	Control	-0.06	98.6	-0.184
			FO diet <sup>2</sup>	-0.20	96.5	-0.513**
Lt	Ca:Cr	45° counterclockwise	Control	0.71	126.1	0.794**
			FO diet	0.55*	110.9	0.790**
Lt	Mg:Cr	None	Control	-0.01	104.9	-0.0001
			FO diet	-0.42	94.5	-0.745**
Lt	Mg:Cr	45° counterclockwise	Control	0.73	138.1	0.736**
			FO diet	0.32*	92.6	0.672**
Lt	Water content	None	Control	-0.32	70.9	-0.903**
			FO diet	-0.30	74.6	-0.876**

<sup>1</sup>Correlation coefficient.

<sup>2</sup>One half the sucrose (50 g/kg diet) of the control diet was replaced with the same mass of fructooligosaccharides.

\*Significantly different from control group ( $P < 0.05$ ).

\*\*Linear relationship between X (independent) and Y (dependent) ( $P < 0.001$ ).

absorption of calcium and magnesium from the colon and rectum. Conversely, there was no net absorption of calcium and magnesium from the colon and rectum of the control rats. Schulz et al. (1993) demonstrated that resistant starch increases the absorptions of Ca and Mg. The cornstarch used in both diets in the present study should have contained a certain amount of resistant starch although we did not estimate its concentration. However, the lack of colorectal calcium and magnesium absorption in the control rats (Fig. 2 and 3) indicated that the amount of resistant starch contained in the present diets was not so large as to increase the absorption of calcium and magnesium in the colon and rectum.

Apparent absorption of calcium and magnesium predicted from the regression equation based on the

Ca:Cr and Mg:Cr ratios in cecal contents agreed well with the apparent absorption of calcium and magnesium calculated from Ca:Cr and Mg:Cr ratios in excreted feces. This good agreement supports the reliability of the regression equation between the fractional length of transit and the calibrated Ca:Cr and Mg:Cr ratios in colorectal pellets. The apparent absorption of calcium and magnesium from mouth to cecum calculated from cecal Ca:Cr and Mg:Cr ratios (Table 5) was approximately 26% greater in fructooligosaccharides diet-fed rats than in control rats. The apparent absorptions of calcium and magnesium from mouth to anus calculated from fecal analysis or from regression equations were about 40–50% and 60% greater in fructooligosaccharides-fed rats than in

TABLE 5

*Segmental apparent absorption of calcium and magnesium in rats fed control or fructooligosaccharides (FO) diet<sup>1</sup>*

	Calcium		Magnesium	
	Control diet	FO diet <sup>2</sup>	Control diet	FO diet <sup>2</sup>
Mouth to cecum, %	35.9 ± 5.3	45.5 ± 12.7*	54.5 ± 5.2	68.6 ± 11.6*
Mouth to anus, %				
(from fecal analysis)	37.5 ± 5.5	57.7 ± 11.7*	52.4 ± 5.7	83.1 ± 7.9*
(from regression equation) <sup>3</sup>	40.9 ± 4.9	58.2 ± 9.7*	52.6 ± 5.4	83.8 ± 6.5*
Apparent absorption, <sup>4</sup> μmol/d	924 ± 142	1185 ± 157*	197 ± 32	278 ± 24*

<sup>1</sup>Values are means ± SD,  $n = 10$ –14.

<sup>2</sup>One half the sucrose (50 g/kg diet) of the control diet was replaced with the same mass of fructooligosaccharides.

<sup>3</sup>Values were predicted by calculation from regression equation with Ca:Cr or Mg:Cr of cecal contents.

<sup>4</sup>Average of mineral intake (μmol/d) × Apparent absorption ratio (%) from regression equation/100.

\*Significantly different from control group ( $P < 0.05$ ).

control rats, respectively (Table 5). Therefore, approximately one half of the additional absorption of calcium and magnesium from consuming fructooligosaccharides should have occurred between the mouth and cecum, and the other half in the colon and rectum. However, we cannot separate the effects of fructooligosaccharides on the cecum and the upper digestive tract, mainly due to insufficient contents in the ileum of rats in the present work.

The stimulative effects of fructooligosaccharides on the absorption of calcium and magnesium were about five times stronger than those of lactose (Ohta et al. 1993). The increased apparent absorption of calcium (by 28%) and magnesium (by 41%) from fructooligosaccharides consumption in this study were higher than those observed in our previous studies (15–30% for calcium and 20–40% for magnesium) when fructooligosaccharides were consumed at the same level (5%) (Ohta et al. 1993, 1994a and 1994b). Lower food intake by the rats fed the fructooligosaccharides diet in the present study (Table 2) may be the reason for this difference. However, it should be noted that the absolute amounts of calcium and magnesium absorbed were greater in fructooligosaccharides rats than in the control rats even though intakes of calcium and magnesium were lower (Table 5).

Goda et al. (1993) speculated that indigestible carbohydrates (maltitol) stimulated calcium absorption in the ileum based on studies of luminal  $^{45}\text{Ca}$  levels after an oral  $^{45}\text{Ca}$  dose in rats. Heijnen et al. (1993) speculated that indigestible carbohydrates (lactulose) stimulated magnesium absorption in the ileum of rats fed indigestible carbohydrates. They speculated that indigestible carbohydrates increased the soluble fraction of magnesium in the ileum, without mentioning the effects on hindgut magnesium absorption. However, these studies did not provide quantitative information on the proportional effects of indigestible carbohydrates on different gut segments.

Trinidad et al. (1993) reported that SCFA stimulated calcium absorption in the human colon. They also showed that calcium absorption from the colon did not occur when calcium was given without SCFA as well, agreeing with our present results in rats fed the fructooligosaccharides-free diet. Considering that fructooligosaccharides is indigestible and readily fermentable, and thus a good source of SCFA in rats and humans (Tokunaga et al. 1989), at least a part of the stimulative effect of fructooligosaccharides on calcium and magnesium absorption might be attributed to the SCFA produced from fructooligosaccharides.

The slopes of the regression lines between the water content of each pellet and the fractional length of transit in the control and fructooligosaccharides rats did not differ significantly, indicating that fructooligosaccharides feeding did not affect net water

absorption from the colon and rectum. Therefore, the increase in the absorption of calcium and magnesium was not related to water absorption in the colon and rectum.

Several authors reported that dietary indigestible carbohydrates lowered luminal pH and thereby increased the solubility of calcium and magnesium (Rémésy et al. 1993, Schulz et al. 1993). We also observed that luminal pH was lowered by fructooligosaccharides feeding (Ohta et al. 1994b). Calcium and magnesium seem to be absorbed via active transcellular and passive paracellular routes (Bronner 1987, Hardwick et al. 1991). However, the solubilization of calcium and magnesium should enhance their absorption via both routes. Thus, we could not determine from the present results whether fructooligosaccharides stimulated active or passive transport.

In conclusion, the present results (Figs. 2 and 3) indicated that calcium and magnesium are not substantially absorbed from the colon and rectum of rats fed a fructooligosaccharides-free diet. The addition of fructooligosaccharides to the diet significantly increased the absorption of both calcium and magnesium. Moreover, about half of the increase from fructooligosaccharides consumption takes place in the colon and rectum.

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