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The Amount and Distribution of Water, Dry Matter, and Sugars in the Digestive Tract of Rats Fed Xanthan Gum (41567)

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Abstract. Diets containing a nutritionally adequate, high-maltose nutrient mixture and either 4% xanthan gum or 4% cellulose were fed *ad libitum* to rats. The feeding of this gum increased the combined weight of the small intestine and its contents by 110%. This effect was partially due to an enlarged intestinal cell mass and to extra dry matter in the contents but chiefly to a 400% increase in intraluminal water. Xanthan feeding enhanced greatly the persistence of sugars beyond the proximal quarter of the small intestine and increased their total recovery in the first three quarters of that organ by 150%. The xanthan-induced increase in intraluminal water in the small intestine was partially due to a slowed absorption of osmotically active substances from the gut.

It has been proposed that the level of intake of dietary fiber among groups of people influences the incidence of diabetes, obesity and atherosclerotic heart disease (1, 2). Technical problems arise in testing these concepts. Hemicelluloses and lignin are much altered in extraction from food. Cellulose appears to have little metabolic effect. Edible gums have been widely used as models for exploring the metabolic effects of fiber. Xanthan gum appears to be a suitable example. This gum, which is produced by a specific bacteria, has a well-defined structure (3, 4) and can be produced with reasonable purity and with reproducible physical properties from batch to batch (5, 6). Further, it is relatively resistant to bacterial breakdown in the digestive tract (6).

A primary approach to exploring the metabolic effects of fiber is to observe how it alters the gastrointestinal processing of food. In a recent, unpublished study, we observed that the feeding of xanthan gum caused the combined weight of the small intestine and its contents to be doubled. In the present study, we sought to determine (a) what regions of the digestive tract are enlarged by the feeding of xanthan gum; (b) to what extent is the enlargement due to extra contents versus an increased cell mass; (c) to what degree are extra contents due to extra water versus extra dry matter; and (d) how fully may retarded absorption of simple sugars account for the extra water in the small intestine.

Materials and Methods. Male Wistar rats, weighing 175 to 250 g, were purchased from Hilltop Laboratory Animals, Inc., Scottdale, Pennsylvania. They were then housed in individual cages at 21° to 23°C and 40 to 60% relative humidity and provided continuous access to distilled water. For the first 1 to 2 weeks, they were fed a commercial pelleted diet (Rodent Laboratory Chow 5001 from Ralston Purina Company), then one of two test diets.

Test diets were nutritionally adequate and contained either 4% xanthan gum or cellulose.[2] Maltose was provided in place of glucose in order to reduce hypertonicity of the ingesta and the possibility of osmotic insult in the proximal digestive tract. Groups of six and eight rats, respectively, were *ad libitum*

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[2] Diets contained the following (g/100 g): maltose monohydrate, 63.91; casein, vitamin-free, 19.86; *l*-methionine, 0.14; corn oil, 4.00; partially hydrogenated vegetable oil (Crisco, Procter and Gamble, Cincinati, Ohio), 4.00; AIN-76 mineral mixture (8), without carbohydrate, 3.09; AOAC vitamin mixture (9), 1.00; and xanthan gum (Kelco Company, San Diego, California) or cellulose (Alphacel, ICN Nutritional Biochemicals, Cleveland, Ohio), 4.00. Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture, and does not imply its approval to the exclusion of other products that may also be suitable.

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TABLE I. DIETARY CONDITION OF RATS

	Experiment 1		Experiment 2	
	Xanthan-fed	Cellulose-fed	Xanthan-fed	Cellulose-fed
Food intake (g/day)[a]	19.9 ± 0.7[b]	23.0 ± 1.6	23.1 ± 0.7	25.3 ± 0.8
Weight gains (g/day)[a]	6.9 ± 0.5	8.2 ± 0.7	4.6 ± 0.3	4.4 ± 0.3
Final body weight (g)[a]	296 ± 7	299 ± 8	339 ± 6	337 ± 6
Final eating period (h)[c]	9.5 ± 0.34 (8.3–10.4)	9.5 ± 0.62 (8.1–11.1)	9.7 ± 0.22 (8.9–10.6)	9.7 ± 0.33 (8.4–10.9)[b]
Diet consumed (g)[c]	17.8 ± 1.5	18.6 ± 1.5	17.5 ± 0.6	18.9 ± 1.3

[a] Average values during 7 days of *ad libitum* feeding of diets containing 4% xanthan gum or 4% cellulose, prior to final day.
[b] Mean ± SEM for groups of six (Experiment 1) or eight (Experiment 2) rats. Parentheses indicate range.
[c] Assumed to begin with start of dark period on final day.

fed the xanthan and cellulose diets for 7+ days in Experiments 1 and 2. Animals were decapitated 8 to 11 hr into the dark period of Day 8, when the digestive tract was expected to be relatively full of ingesta (7).

In Experiment 1, the wet and dry weights of contents from the stomach, large intestine, cecum, and segments of the small intestine were determined, as well as the wet and dry weight of tissue in these various portions of the digestive tract. The abdominal wall was opened and clamps were placed on either side of the pyloric sphincter. The small intestine was pulled free of the mesentery, care being taken to keep all portions at approximately the same level in order to minimize postmortem movements of the ingesta. The small intestine was excised and placed on a water-repellant surface (Parafilm by American Can Company), measured, marked off, and cut into six equal lengths. Each length of small intestine was slit open; and the wet contents were transferred into weighed beakers by repeated, gentle scraping with a small spoon-like stainless steel spatula (No. #21-401-15, Fisher Scientific Company). Contents of the stomach, cecum, and large intestine were similarly removed, and the remaining portions of digestive tissues were placed in weighed beakers. A prompt weighing indicated wet weights of these samples. Samples were then freeze-dried and reweighed for calculation of dry weight.

The above procedure was practiced in preliminary studies. We learned first to avoid gouging the intestinal mucosa, as observed with 3× magnification. We scraped each section of the digestive tract repeatedly, until one extra scrape yielded less than 5 mg contents. Wet-weight measurements on the small intestine were completed within 5 min of killing the animal. In test trials, we were able to recover 95 to 98% of the wet weight of the intact intestine in the six fractions of contents and of tissues.

The amounts of simple sugars in successive portions of the small intestine were examined in Experiment 2, under the same dietary conditions as in Experiment 1. Rats were *ad libitum* fed the same xanthan and cellulose diets and were killed 8 to 11 hr into the dark period of the eighth day. The small intestine was then removed with extra concern for speed because of the known rapidity of intestinal absorption of sugars. The organ was quickly measured, marked, and cut into four equal lengths, and the proximal three of these quarters were promptly placed in chilled 1 mM sodium citrate buffer, pH 4.2 (containing 0.25 g sodium fluoride/100 ml) (10). These intestinal segments were then homogenized, heat-treated to inactivate enzymes (11), and incubated with amyloglucosidase to insure the hydrolysis of any remaining maltose (10) and analyzed enzymatically for "total" glucose (12). The low level of glycogen in the intestinal wall introduced little error in the procedure (11). Total glucose was not measured in the distal quarter of the small intestine because of its low concentration in samples from control animals and because of high turbidity in samples from gum-fed rats.

Data were evaluated statistically with Student's t test.

TABLE II. EFFECT OF FEEDING 4% XANTHAN GUM VERSUS 4% CELLULOSE DIETS ON CONTENTS OF WATER AND DRY MATTER IN PORTIONS OF THE DIGESTIVE TRACT[a]

	Water[b]		Dry matter	
	Xanthan-fed (g)	Cellulose-fed (g)	Xanthan-fed (g)	Cellulose-fed (g)
Stomach	2.91 ± 0.29	3.26 ± 0.74	3.26 ± 0.50	3.24 ± 0.67
Small intestine:				
Portion a (proximal)	0.74 ± 0.24[c]	0.12 ± 0.02	0.062 ± 0.013[c]	0.026 ± 0.004
Portion b	1.05 ± 0.09[c]	0.25 ± 0.03	0.139 ± 0.017[c]	0.056 ± 0.009
Portion c	1.25 ± 0.09[c]	0.32 ± 0.07	0.133 ± 0.012[c]	0.077 ± 0.017
Portion d	2.01 ± 0.22[c]	0.39 ± 0.05	0.187 ± 0.018[c]	0.116 ± 0.019
Portion e	2.15 ± 0.14[c]	0.37 ± 0.04	0.197 ± 0.021[c]	0.146 ± 0.022
Portion f (distal)	1.72 ± 0.09[c]	0.33 ± 0.04	0.238 ± 0.009[c]	0.129 ± 0.016
Cecum	3.26 ± 0.28[c]	1.15 ± 0.13	0.66 ± 0.03[c]	0.41 ± 0.04
Large intestine	1.05 ± 0.13[c]	0.40 ± 0.06	0.44 ± 0.07[c]	0.24 ± 0.03

[a] Measured 8 to 11 hr after the start of the dark period, when the digestive tract was estimated to be relatively full.
[b] Weight loss during freeze-drying.
[c] Indicates that values for xanthan-fed rats are different ($P < 0.05$) from those of cellulose-fed controls.

Results. Table I summarizes information on the dietary condition of rats. In both experiments, animals ate and gained weight at similarly good rates. When killed 8 to 11 hr into the dark period, animals had consumed, on the average, about 80% as much food during that interval as in a typical 24-hr period.

Table II summarizes data on digestive contents of rats in Experiment 1. Xanthan feeding did not affect the weight of either water or dry matter in the stomachs but increased both components in all portions of the digestive tract beyond the pyloric sphincter. The effect was particularly large for water contents.

Figure 1 indicates that gum feeding increased the dry weight of tissue mass of all six portions of the small intestine but not that of the stomach, cecum, or large intestine.

Table III summarizes information from Experiment 1 relating to the 110% increase by xanthan feeding in the combined weight of the small intestine and its contents. Contents accounted for 53% of the total intestinal

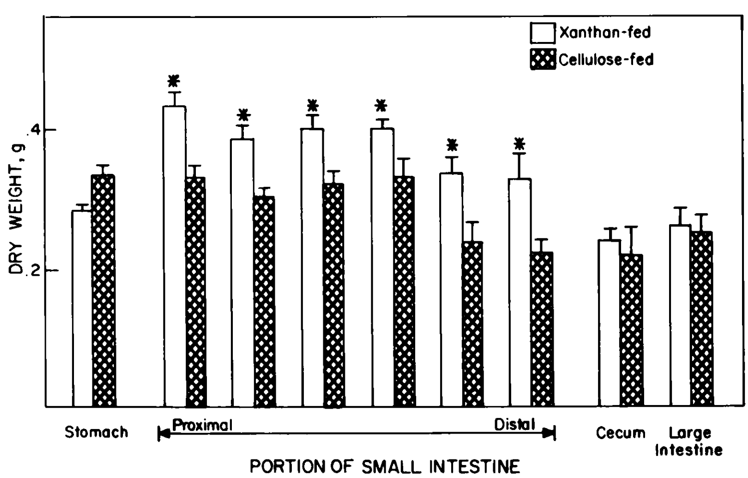

FIG. 1. Effect of feeding 4% xanthan gum versus 4% cellulose diets on dried tissue weights in various portions of the digestive tract in Experiment 1. Means + SEM are shown. *, $P < 0.05$.

TABLE III. COMPONENTS IN THE XANTHAN-INDUCED WEIGHT INCREASE OF THE SMALL INTESTINE AND ITS CONTENTS[a]

	Xanthan-fed (g)	Cellulose-fed (g)	Difference (g)	Difference % total
Intestine + contents	18.66 ± 0.43[b]	8.80 ± 0.45	9.86	100.0
Water of contents	8.93 ± 0.35[b]	1.79 ± 0.17	7.14	72.4
Dried contents	0.96 ± 0.05[b]	0.55 ± 0.05	0.41	4.2
Tissue water	6.49 ± 0.35[b]	4.72 ± 0.27	1.77	17.9
Dried tissue	2.28 ± 0.07[b]	1.74 ± 0.08	0.54	5.5

[a] Measured 8 to 11 hr after the start of the dark period, when the digestive tract was estimated to be relatively full.
[b] Indicates that values for xanthan-fed rats are different ($P < 0.05$) from those of cellulose-fed controls.

weight in gum-fed animals and for 27% of the total in controls. The 75% increase in dry matter contents in gum-fed animals accounted for less than 5% of the difference in total intestinal weight. Xanthan-fed animals contained 36% more wet cell mass than did controls; and this component accounted for 23% of the total difference in intestinal weights. However, the 400% increase in the water in intestinal contents accounted for more than 70% of the gross difference.

Figure 2 records the combined amounts of glucose and maltose monohydrate in the first three quarters of the small intestine in rats fed the two test diets in Experiment 2. Portions of several intestinal extracts were examined for both free and total glucose and were found to contain mostly the free form in the middle two quarters. Xanthan feeding did not significantly increase the sugars found in the proximal quarter of the small intestine, but largely prevented the sharp decline in sugar contents which was observed in the second and third quarters of the small gut among cellulose-fed controls. Xanthan gum increased the sugar recovered in the total of these three segments by 150%.

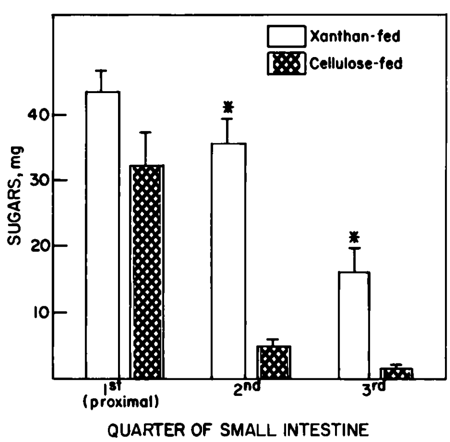

FIG. 2. Amount of sugars recovered from first three quarters of the small intestine in rats fed xanthan and cellulose diets in Experiment 2. Bars represent mean + SEM for amounts of glucose recovered after hydrolysis of any maltose present. *, $P < 0.05$.

Discussion. During times of day when stomachs were relatively full, rats receiving a 4% xanthan gum diet had five times as much intraluminal water in their small intestine as did their cellulose-fed controls. In a preliminary, unpublished study of ours, small numbers of rats were *ad libitum* fed 4% guar gum, 4% xanthan, or 4% cellulose in otherwise identical diets. Guar appeared to increase intraluminal water in the gut but to a lesser degree than did xanthan gum.

Guar and xanthan gums form highly viscous suspensions in water. Guar suspensions, however, tend to be slow to thicken fully (13); and guar is less resistant to bacterial breakdown in the gut than is xanthan (14, 6). It seems likely that xanthan affected the rheology within the rat gut more that did guar. It also seems likely that extra water in gut contents is a general effect of feeding viscous gums.

The feeding of xanthan gum increased the wet and dry weight of the small intestine per se. This was not surprising, for the feeding of 15.2% pectin (15) or of 20 or 40% of each of five other edible gums (14) increased the length and/or wet weight of the small intestine in rats.

Xanthan feeding did not affect the amount of diet eaten during the 8 to 11 hr of the final dark period or the amount of dry matter finally recovered in the stomach. If one assumes stomachs to have been essentially empty at the start of the dark period (7), the average rate of gastric emptying of ingested food during the final 8 to 11 hr was similar in both groups. However, edible gums do slow gastric emptying when fed acutely (16–19). It remains to be determined whether chronic feeding of gums may restrain gastric emptying during and shortly after meals and may thereby influence metabolic regulation.

Water constituted only about 50% of stomach contents in both the xanthan-fed and control rats. The dry matter of ingesta became much diluted in the proximal portion of the small intestine in both groups and especially in the xanthan-fed animals.

The feeding of gums has been reported to slow the distal progression of ingesta (18–20). This appears to have occurred in the present study, for xanthan-fed rats retained 1.75 times as much dry matter in the small intestine as did the cellulose-fed animals.

A greater amount and a more distal distribution of sugars was found within the small intestine of xanthan-fed than of cellulose-fed rats. Apparently, the presence of a viscous gum slowed intestinal absorption, as others have postulated (18–20). Mechanisms for a slowed absorption can readily be suggested. The greater dilution of nutrients in the small intestine in gum- versus cellulose-fed animals reduced the concentration difference between lumen and blood and presumably also the concentration gradient for the passive portion of intestinal absorption. Presumably, the viscosity produced by xanthan gum in intestinal contents reduced mixing and the frequency of contact between nutrient molecules and mucosal cells. An altered histology of the small intestine may also have been involved (21, 15).

A major question is how to account for the greatly increased amount of intraintestinal water in gum- versus cellulose-fed animals. Theoretically, this effect might involve the movements of water both into and out of the small intestine. In the absence of hydrophilic colloids, intraluminal water leaves the small intestine mainly through osmosis or diffusion. This occurs when contents become hypotonic, due ultimately to an active transport of some sugars, amino acids, and inorganic ions out of the tract. Xanthan gum promoted the retention of intraluminal water in the small intestine by increasing the retention of sugars and perhaps of other osmotically active solutes. Sugar retention, however, was far too small to account fully for the extra 7 g intraintestinal water in xanthan-fed rats, for the 57-mg increase in intraintestinal sugars was enough to render only 1 g water isotonic to blood serum.

Xanthan gum also has a water-holding capacity which reduced the migration of water from the small intestine to an unknown degree. It appears likely from the magnitude of the xanthan effect on intraintestinal water that this gum also speeded certain aqueous secretions into the small intestine.

Many nutritionists are particularly interested in reports that the feeding of guar gum and other forms of fiber may be beneficial to maturity-onset diabetics [see review (19)]. Gums are thought to affect gastric emptying and the secretion of enteric hormones in ways that restrain insulin release after a meal (19). The sensitivity of cells to insulin is usually subject to down regulation by circulating insulin (22). It is tempting to attribute the enhanced insulin sensitivity after the chronic feeding of gums to a reduced degree of down regulation.

Xanthan gum appears to affect the endocrine activity of the gut in at least two ways. Intestinal distention, as produced by xanthan feeding, is known to influence the secretion of some enteric hormones. Xanthan gum appears to move distally the region of principal contact between nutrients and the intestinal lining. The latter effect should be important since secretory cells for various enteric hormones are somewhat concentrated within different portions of the gut.

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