Molecular weight of guar gum affects short-chain fatty acid profile in model intestinal fermentation

Maria L. Stewart and Joanne L. Slavin

University of Minnesota, St. Paul, MN, USA

Dietary fiber exerts many beneficial physiological effects; however, not all types of dietary fiber display the same effects. Partially hydrolyzed guar gum (PHGG), a lower molecular weight form of guar gum, is more easily incorporated into food, but may have less pronounced physiological effects than the native form. The aim of this study was to identify differences in intestinal fermentability based on the molecular weight of guar gum. Guar gum of four molecular masses (15, 20, 400, and 1100 kDa) was fermented using a batch *in vitro* fermentation system. Human fecal inoculum was the source of microbes. The 400-kDa fraction produced the greatest concentrations of total short-chain fatty acid (SCFA) at 8 h and the highest amounts of butyrate at 24 h. At 24 h, the 400-kDa fraction produced more total SCFA and propionate than the 15 kDa, but was not different than 20 kDa or 1100 kDa fractions. The molecular weight of guar gum was positively correlated with acetate production and negatively correlated with propionate production. This study concludes that 400-kDa guar gum may be optimal for intestinal fermentability. In conclusion, the molecular weight of guar gum affects *in vitro* fermentability and should be considered when adding to a food or beverage.

Keywords: Dietary fiber / Fermentation / Guar gum / Molecular weight / Short-chain fatty acids

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1 Introduction

Dietary fiber is a nutrient found in plant-based foods. Some types of fiber undergo bacterial fermentation in the colon to produce short-chain fatty acids (SCFA). Acetate, propionate, and butyrate are the SCFA produced in the highest concentrations. Acetate is a fuel for skeletal and cardiac muscle, kidney, and the brain, while butyrate is the preferred fuel of the colonic epithelium, in particular, the distal colon and rectum [1, 2]. Bacterial fermentation produces different proportions of each individual SCFA and concentrations of total SCFA. Physiological status may be improved by consuming fermentable fiber, so it is important to understand the fermentability of each type of fiber.

Studying fiber fermentability in a closed laboratory system provides an estimate of fiber fermentability without losing SCFA to colonic absorption. *In vitro* fiber fermentation mimics the colonic system using representative colonic bacteria under anaerobic conditions to ferment isolated fiber.

Correspondence: Dr. Joanne L. Slavin, 166 Food Science and Nutrition, 1334 Eckles Ave, St. Paul, MN 55108, USA

E-mail: jslavin@umn.edu **Fax**: +1-612-625-5272

Abbreviations: **PHGG**, partially-hydrolyzed guar gum ; **SCFA**, short-chain fatty acid

Guar gum is a naturally occurring fiber found in the seeds of the guar plant, *Cyamoposis tetragonolobus* [3]. The guar gum fraction of the seeds is the major component of the endosperm, and serves as the main carbohydrate storage for seed germination. Guar gum is composed of galactose and mannose residues in a ratio of 1:1.5-1.8 [4]. Mannose units linearly linked by $\beta(1-4)$ bonds form the backbone with galactose units branching from the O-6 position, joined by $\alpha(1-6)$ bonds. These bonds cannot be cleaved by human alimentary enzymes, rendering guar gum indigestible in the upper gastrointestinal tract.

Partially hydrolyzed guar gum, a lower molecular weight form of guar gum, is produced from guar gum by enzymatic hydrolysis with endo-β-D-mannanase [5]. Approximately 5% of the 1,4-β-D-mannosepyranose bonds are susceptible to enzymatic hydrolysis, resulting in an average molecular mass of 20 kDa. Partially hydrolyzed guar gum's main advantage over guar gum is its increased solubility, which makes it easily incorporated into food. Doses up to 20 g/day of PHGG have been established as safe, showing no adverse physiological effects (as cited in [3]). Partially hydrolyzed guar gum has many positive physiological effects such as increased laxation, decreased diarrhea, reduced blood lipids, increased beneficial colonic microorganisms, altered mineral absorption, and improved glucose tolerance [3].



Most *in vitro* fermentations of guar gum have shown that acetate is the major SCFA produced by fecal bacteria [6–11]. Propionate is often the next most abundant SCFA produced, with butyrate being produced in the lowest concentrations. Two studies have reported butyrate as being the second most abundant SCFA produced with propionate being the least abundant SCFA [12, 13]. One study, investigating the fermentation of two molecular masses of guar gum (10 and 15 kDa), reported butyrate as the greatest proportion of SCFA with acetate being second and propionate being last [14].

This study investigated the *in vitro* fermentabilities of guar gum of four different molecular weights by measuring the SCFA production over the course of 24 h.

2 Materials and methods

Four samples of guar gum (Taiyo Kagaku Co., Mie, Japan) were fermented, each having different molecular mass (15, 20, 400, and 1100 kDa). Molecular mass was determined via HPLC using water as the mobile phase (TSK-gel Super AW2500 and TSK-gel Super AW4000 column, TOSO Co Ltd, Japan; column temperature 40°C; flow rate 0.6 mL/min). The chosen molecular weights of guar gum are currently manufactured for the food and supplement industry. Chemical reagents were obtained from Fisher Scientific (New Hampton, NH, USA), Sigma Aldrich (St. Louis, MO, USA), and VWR Scientific (West Chester, PA, USA).

Fecal inoculum was prepared as described by McBurney and Thompson [6]. Briefly, fecal samples from three human subjects consuming a nonspecified Western diet were pooled (125 g total) and diluted with 400 mL distilled water. The solution was homogenized in a blender. Reducing solution (950 mL distilled water, 6.25 g cysteine hydrochloride, 40 mL 1 N NaOH, 6.25 g sodium sulfide nonahydrate) was added to the fecal inoculum to obtain a ratio of 15 parts fecal inoculum to 2 parts reducing solution [15]. Five 100-mL serum bottles were prepared for each guar gum sample (0.5 g), one for each of the five time points: 0, 4, 8, 12, and 24 h. Control bottles containing either 0.5 g glucose or no added carbohydrate were prepared in the same manner. Glucose was chosen as a positive control because the monomers are completely available to the bacteria, i. e., no glycosidic bonds must be broken for metabolism of the carbohydrate. No carbohydrate was added as a negative control to quantify the SCFAs produced by substrate originating from the fecal sample. Sterile trypticase peptone fermentation media (40 mL) plus reducing solution (2 mL) was added to each bottle [6]. The fermentation media included resazurin as a marker color-indicator for anaerobic conditions. Resazurin turns pink or blue under

aerobic conditions and appears colorless under anaerobic conditions. The bottles were sealed with a rubber stopper and crimped metal seal for storage at 4° C for 12 h to hydrate samples. At 2 h prior to inoculation with fecal solution, sample bottles were warmed to 37° C.

Ten milliliters of fecal inoculum was added into each serum bottle along with 0.8 mL Oxyrase1 oxygen-reducing enzyme (Oxyrase Inc., Mansfield, OH, USA). The bottles were immediately flushed with carbon dioxide gas to eliminate oxygen and generate anaerobic conditions. The bottles were gently shaken in a 37°C water bath. One sample bottle for each fiber was removed at 0, 4, 8, 12, and 24 h. Immediately upon removal, 1 mL copper sulfate (200 g/L) was added to each bottle to kill the bacteria and cease fermentation. Two 2-mL aliquots were removed for SCFA analysis.

Samples were prepared for gas chromatography as described by Pylkas *et al.* [14]. Lactate, acetate, propionate, butyrate, isobutyrate, 2-methylbutyrate, isovalerate, and valerate were determined by gas chromatography using a Hewlett Packard model 6890 gas chromatograph (Hewlett Packard, Palo Alto, CA, USA) with a 4% carbowax 20M/80/120 carbopack B-DA column (Supleco, Bellefonte, PA, USA) at a temperature of 175°C. Flow rates for nitrogen, hydrogen, and air were 24, 40, and 450 mL/min, respectively.

All SCFA concentrations were corrected for the negative control concentration of SCFA at each time point. Molar ratios of SCFA were determined by dividing the number of moles of each SCFA (acetate, propionate, and butyrate) by the total moles SCFA. SCFA production rates were calculated by dividing the change in SCFA concentrations between time points by the number of hours between time points. Statistical analyses were completed with SAS statistical software package, version 8.0 (SAS Institute, Cary, NC, USA). Analysis of variance with Tukey pair-wise comparison was conducted to compare the mean SCFA concentrations and SCFA production rates between guar gum samples and between time points. Statistical significance was achieved at p values less than 0.5. Linear regression and analysis of variance was conducted to test the relationship between guar gum molecular weight and molar ratios of SCFA.

3 Results

3.1 Total SCFA Production

3.1.1 Time comparison

The batch fermentation maintained anaerobic conditions for the entire incubation, as indicated by the colorless appearance of resazurin. The bacteria were metabolically

Table 1. Total SCFA^{a)}, acetate, propionate, and butyrate production^{b)} (μ mol/mL) of four molecular-weight forms of guar gum after 0, 4, 8, 12, and 24 hours *in vitro* batch fermentation, analyzed by gas chromatography

	15 kDa	20 kDa	400 kDa	1100 kDa				
Total SCFA								
0 h	-1.1 ± 0.9	0.6 ± 0.1	-0.7 ± 0.5	0.2 ± 0.2				
4 h	7.6 ± 0.1	9.3 ± 1.9	7.7 ± 0.02	9.2 ± 1.0				
8 h	$29.2 \pm 0.3^{\circ}$	$33.5 \pm 1.3^{\circ}$	$45.7 \pm 3.2^{\text{d}}$	$31.4 \pm 0.6^{\circ}$				
12 h	51.8 ± 1.0	63.5 ± 4.0	62.0 ± 0.6	54.7 ± 11.6				
24 h	$47.9 \pm 0.6^{c)}$	$60.9 \pm 3.1^{c, d}$	$68.6 \pm 0.8^{\text{d})}$	$62.8 \pm 4.3^{c, d)}$				
Acetate								
0 h	-0.2 ± 0.1	-0.17 ± 0.04	-0.150 ± 0.1	-0.3 ± 0.1				
4 h	5.9 ± 0.4	6.2 ± 1.2	6.6 ± 0.5	7.5 ± 0.2				
8 h	$25.1 \pm 0.5^{\circ}$	$27.8 \pm 1.3^{c, d)}$	$34.2 \pm 2.2^{d)}$	$26.1 \pm 0.8^{\circ}$				
12 h	40.3 ± 0.4	46.0 ± 2.3	45.5 ± 0.1	36.1 ± 7.3				
24 h	22.6 ± 1.6	31.3 ± 2.4	34.6 ± 0.2	34.5 ± 3.3				
Propio	Propionate							
0 h		-0.1 ± 0.01	-0.05 ± 0.04	-0.09 ± 0.006				
4 h	0.5 ± 0.1	0.6 ± 0.3	0.5 ± 0.1	0.9 ± 0.04				
8 h	1.8 ± 0.3	2.6 ± 0.5	4.5 ± 0.8	3.9 ± 0.3				
12 h	11.5 ± 0.5	16.9 ± 0.9	16.8 ± 0.3	19.8 ± 4.2				
24 h	$21.7 \pm 0.6^{c)}$	$26.0\pm0.7^{\rm d)}$	$28.5 \pm 0.7^{\text{d})}$	$25.5 \pm 0.9^{c, d)}$				
Butyra	ite							
0 h	-0.08 ± 0.03	-0.07 ± 0.003	-0.03 ± 0.04	-0.06 ± 0.006				
4 h	0.2 ± 0.04	0.2 ± 0.1	0.2 ± 0.05	0.3 ± 0.004				
8 h	0.4 ± 0.3	0.5 ± 0.1	0.9 ± 0.2	0.8 ± 0.04				
12 h	2.3 ± 0.1	2.7 ± 0.1	3.3 ± 0.1	2.4 ± 0.6				
24 h	$5.0\pm0.4^{c)}$	$4.9 \pm 0.01^{c)}$	$6.8 \pm 0.1^{d)}$	$3.8 \pm 0.1^{c)}$				

a) Total SCFA: acetate, propionate, butyrate, isobutyrate, 2-methylbutyrate, isovalerate, lactate, and valerate (if detectable).

active during the entire incubation as indicated by the SCFA concentrations from the positive control, glucose (data not shown).

Total SCFA concentrations increased during the interval 0–12 h, for all fibers (Table 1). The 15 kDa fraction resulted in significantly lower total SCFA concentrations at 24 h compared to 12 h. Total SCFA concentrations at 12 and 24 h were not significantly different for 20-, 400-, and 1100-kDa samples.

3.1.2 Fiber comparison

Total SCFA concentrations produced by the four forms of guar gum (15, 20, 400, and 1100 kDa) varied significantly at 8 and 24 h. At 8 h, the 400-kDa form produced significantly more SCFA than the forms of 15, 20, and 1100 kDa. Total SCFA production by 400-kDa form remained signifi-

Table 2. Rate^{a)} of total SCFA^{b)}, acetate, propionate, and buty-rate production from 0 to 24 h (μ mol/mL/h)

	15 kDa	20 kDa	400 kDa	1100 kDa
Total SCE	A			
0-4 h	2.2 ± 0.2	2.2 ± 0.5	2.1 ± 0.1	2.2 ± 0.2
4-8 h	$5.4 \pm 0.1^{c)}$	$6.0 \pm 0.1^{c)}$	$9.5 \pm 0.8^{d)}$	$5.6 \pm 0.4^{c)}$
8 - 12 h	5.7 ± 0.3	7.5 ± 1.3	4.1 ± 0.7	5.8 ± 2.8
12-24 h	-0.3 ± 0.0	-0.2 ± 0.1	0.6 ± 0.1	0.7 ± 1.3
Acetate				
0-4 h	1.5 ± 0.1	1.6 ± 0.3	1.7 ± 0.1	1.9 ± 0.0
4-8 h	$4.8 \pm 0.0^{c)}$	$5.4 \pm 0.0^{c, d}$	$6.9 \pm 0.7^{\text{d}}$	$4.7 \pm 0.3^{\circ}$
8 - 12 h	3.8 ± 0.2	4.5 ± 0.9	2.8 ± 0.6	2.5 ± 1.6
12-24 h	-1.5 ± 0.1	-1.2 ± 0.0	-0.9 ± 0.0	-0.1 ± 0.9
Propionate	e			
0 - 4 h	0.1 ± 0.0	0.2 ± 0.1	0.1 ± 0.0	0.2 ± 0.0
4 - 8 h	$0.3 \pm 0.0^{c)}$	$0.5 \pm 0.0^{c)}$	$1.0 \pm 0.2^{d)}$	$0.8 \pm 0.1^{c)}$
8 - 12 h	2.4 ± 0.2	3.6 ± 0.3	3.1 ± 0.1	4.0 ± 1.0
12 - 24 h	0.8 ± 0.1	0.8 ± 0.0	1.0 ± 0.1	0.5 ± 0.4
Butyrate				
0 - 4 h	0.1 ± 0.0	0.1 ± 0.0	0.0 ± 0.0	0.1 ± 0.0
4 - 8 h	0.1 ± 0.0	0.1 ± 0.0	0.2 ± 0.1	0.1 ± 0.0
8 - 12 h	0.5 ± 0.0	0.6 ± 0.1	0.6 ± 0.0	0.4 ± 0.1
12-24 h	0.2 ± 0.0	0.2 ± 0.0	0.3 ± 0.0	0.1 ± 0.1

a) Values shown are mean \pm standard error (n=2).

cantly higher than that of the 15-kDa form at 24 h, but was not different from that of the 1100- or 20-kDa forms.

3.1.3 Rate comparison

Peak total SCFA production rates (Table 2) were reached during the interval from 8 to 12 h for 15- and 20-kDa guar gum; however, these rates were not significantly different from the rates at 4–8 h. The 1100-kDa form also had peak total SCFA production at 8–12 h, but none of the rates was significantly different. The 400-kDa form reached peak total SCFA production rate at 4–8 h. Total SCFA production rate did not differ between the fibers except during the interval from 4 to 8 h. The 400-kDa gum produced total SCFA at a significantly higher rate than the 15-, 20- and 1100-kDa forms.

3.2 Acetate production

3.2.1 Time comparison

All fibers produced peak acetate concentrations at 12 h (Table 1). Acetate concentrations produced by 1100-kDa

b) Values shown are mean \pm standard error (n=2).

c,d) Values with different letters are statistically different from each other within the same row (ANOVA Tukey pairwise analysis, p < 0.05).

Total SCFA: acetate, propionate, butyrate, isobutyrate, 2-methylbutyrate, isovalerate, lactate, and valerate (if detectable).

c,d) Values with different letters are statistically different from each other within the same row (ANOVA Tukey pairwise analysis, p < 0.05).

Table 3. Molar ratios of SCFA at 24 h

Type of Guar Gum	15 kDa	20 kDa	400 kDa	1100 kDa
Acetate	46	50	49	54
Propionate	44	42	41	40
Butyrate	10	8	10	6

gum at 12 h were not statistically different from those at 8 and 24 h, but were numerically greater. The 15-, 20-, and 400-kDa forms all produced the statistically greatest acetate concentrations at 12 h.

3.2.2 Fiber comparison

Similar to total SCFA production, the 400-kDa form produced the significantly greatest concentration of acetate at 8 h compared to the other samples. Peak acetate concentrations were reached at 12 h; however, no samples were significantly different from one another. Acetate concentrations dropped at 24 h, and no differences between samples were statistically significant. Molar ratios of SCFA produced varied slightly in molecular weight (46-54) and are shown in Table 3. When analyzed with a linear regression model, the molar ratio of acetate increased as the molecular weight of the guar gum increased $(R^2 = 0.7077, p = 0.1633)$.

3.2.3 Rate comparison

Acetate production rate peaked during the interval from 4 to 8 h for all fibers. The acetate production rates of the 15-and 400-kDa forms were significantly higher from 4 to 8 h compared with all other time intervals. Acetate production rate from 4 to 8 h was numerically greater than all other values, but only significantly higher than at 0-4 h and 12-24 h. The 1100-kDa fiber form did not have statistically different production rates during the entire 24-h incubation. Acetate production rate differed between the fibers only during the interval from 4 to 8 h. The 400-kDa fiber produced acetate at the highest rate during this time interval, but was not significantly different from that of the 20-kDa form.

3.3 Propionate production

3.3.1 Time comparison

Propionate concentrations increased over the entire time period for all guar gums (Table 1). The 15-, 20- and 400-kDa fibers produced the significantly highest propionate concentrations at 24 h. Peak propionate concentration with the 1100-kDa form was reached at 24 h, but was not significantly different than the propionate concentration produced at 12 h.

3.3.2 Fiber comparison

No samples were significantly different from one another at 4, 8, or 12 h. At 24 h, the 20- and 400-kDa gum fibers produced significantly more propionate than the 15-kDa form, but were not different than 1100-kDa form or each other. Molar ratios of propionate ranged from 40 to 44 (see Table 3). The molar ratio of propionate decreased as the molecular weight of guar gum increased ($R^2 = 0.6651$, p = 0.1846).

3.3.3 Rate comparison

During the 24-h incubation, propionate production rate peaked from 8 to 12 h and was the statistically greatest rate for all fibers (Table 2). Propionate production rate differed between fibers only during the interval from 4 to 8 h. The 400-kDa form produced propionate at a significantly higher rate than the 15-, 20-, and 1100-kDa fractions.

3.4 Butyrate production

3.4.1 Time comparison

Butyrate concentrations also increased over time for all guar gums (Table 1). The statistically greatest butyrate concentrations were reached at 24 h for all fibers.

3.4.2 Fiber comparison

The 400-kDa form produced more butyrate than the 15-, 20- and 1100-kDa forms at 24 h. The 15-, 20-, and 1100-kDa fibers were not significantly different from one another at 24 h. Molar ratios of butyrate ranged from 6 to 10 (see Table 3). The molar ratio of butyrate as not correlated to the molecular weight of the guar gum ($R^2 = 0.5566$, p = 0.2539).

3.4.3 Rate comparison

Butyrate production rate peaked from 8 to 12 h for all fibers. The 15-, 20-, and 400-kDa forms all had the statistically greatest butyrate production rate at this time interval. Butyrate production rates did not differ significantly for 1100-kDa gum fibers at any of the time intervals. Between the four fibers, the butyrate production rate did not differ significantly at any of the time intervals.

4 Discussion

Dietary fiber intakes are known to be protective against chronic disease and potential mechanisms include byproducts of fiber fermentation in the gut, especially SCFAs. Fiber intakes remain low, and food manufacturers are interested in adding fiber sources into foods and beverages. The viscosity of soluble fibers, such as guar gum, limits the ability to add these fibers into foods and beverages. These soluble fibers can be hydrolyzed to improve their functional properties in foods, but it is essential that these altered fibers still provide physiological benefits.

Batch fermentation systems with human fecal inoculum were designed to mimic the human digestive tract. In general, digesta enter the large intestine about 12 h after ingestion, and total gastrointestinal transit time is between 1 and 3 days. Of course, gastrointestinal transit times are highly variable with diarrhea causing fast transit and constipation being associated with very slow transit. In the batch model system, fermentation is measured from zero to 24 h with the expectation that between 12 and 24 h would be the time points most relevant to the large intestine.

With the exception acetate production at 24 h, the 400-kDa fiber form consistently produced significantly more SCFA than the 15-kDa form at 24 h. No additives were present in any of the guar gums, so differences in fermentability suggest bacterial preference for specific molecular weights. Longer fermentation may result in greater SCFA production from the 1100-kDa form, the largest guar gum chain; however, it may not represent the actual fermentation in the human colon as well as the 24-h length.

Decreased acetate concentrations at 24 h may indicate that metabolic pathways converting acetate to butyrate were favored late in the fermentation period. The metabolic pathway utilizing butyryl CoA:acetyl CoA transferase converts acetate to butyrate. Duncan *et al.* [16] previously reported that 85% of butyrate was generated from acetate, while only 15% of butyrate was generated directly from glucose or Embden-Meyerhof-Parnas (EMP) pathway. Once monosaccharides are liberated from the guar gum chain, both galactose and mannose can be metabolized into EMP pathway intermediates for SCFA production.

The relationships between molar ratio and molecular weight indicate that molecular weight of guar gum could, perhaps, dictate metabolic pathways utilized by the bacteria. Molar ratios of acetate ranged from 46-54. These data are similar or slightly greater than molar ratios reported by other authors (range 25-62) [6-13]. Propionate molar ratios ranged from 40-44, which fell above the range of values reported from other sources (range 1-36) [6–14]. Molar ratios of butyrate reported by other groups ranged greatly (range 9-71) but included the molar ratios reported here (range 6-10) [6-14]. Differences in molar ratios between lab groups are common in these types of experiments. Similar fermentation procedures were used in all other reports; however, one factor remained variable: the microflora composition. Microbial species were not identified nor quantified in this protocol. The inoculum is estimated to be a representative microflora sample, but may

differ based on individual fecal donors. Variations in SCFA proportions between fermentation batches are a result of varying microflora composition.

The 400-kDa guar gum is the optimal molecular weight of guar gum if an increased total SCFA profile is desired. It was more fermentable than the 15-kDa form, but not statistically different from the 1100- and 20-kDa forms at 24 h. Increased SCFA concentrations have been linked to depletion of pathogenic bacteria due to decreased pH and decreased diarrhea due to increased water absorption [17, 18]. The large molecular mass of 400 kDa may make it difficult to incorporate into a western diet, due to decreased solubility and increased viscosity, compared with lower molecular weight guar gums. The lower molecular weights of guar gum were shown to be fermentable and may be more desirable dietary interventions based on palatability.

The 400-kDa guar gum produced more butyrate at 24 h than the 15-, 20-, and 1100-kDa samples. Compared with other fermentable fibers (inulin and oat beta-glucan), guar gum produced low concentrations of butyrate (Stewart and Slavin, unpublished data). Butyrate improves colonic health by serving as the main fuel for colonocytes. Additionally, butyrate has been linked to decreased carcinogenesis in the colon. Mechanisms such as influencing gene expression via non-competitive inhibition of histone deacetylation, activating NF-κB to decrease pro-inflammatory cytokines, activating glutathione S-transferase, inhibiting growth, promoting cell differentiation, or inducing apoptosis may contribute to butyrate's anti-cancer effect [19–22]. To obtain improved colon health, the 400-kDa form would be a better dietary intervention than all other samples.

Rates of SCFA production varied between time points and between fibers. The time interval from 4 to 8 h had the greatest rate of acetate production, while the interval from 8 to 12 h had the greatest rate of propionate and butyrate production. Bacterial metabolic pathways favored acetate production initially and switched to propionate and butyrate production later during fermentation. Alternatively, bacteria that produce acetate may be more active than those producing propionate or butyrate early in the fermentation. This may have implications for individuals with short colonic transit times; fiber will not ferment long enough to produce the butyrate needed for good colon health.

Consistent with SCFA concentrations, the 400 kDa had significantly higher total SCFA, acetate, and propionate rates of production. Interestingly, the 400-kDa fibers produced the significantly highest butyrate concentration at 24 h, but production rates were not significantly different from 0 to 24 h. Higher SCFA production rates may have been the result of increased enzyme activity or increased enzyme concentration. The 400-kDa form may have a bioactive capability to stimulate gene transcription or enzyme activity

that the other molecular-weight forms do not have. This hypothesis has not yet been tested but is one direction for future work.

In conclusion, molecular weight affects fermentation patterns of guar gum, with higher molecular weight guar gums producing more acetate and less propionate than lower molecular weight guar gums. Across all SCFAs measured, the 400-kDa fibers were more fermentable than the 15-kDa forms, which may suggest that an optimal molecular weight of guar gum exists. A similar study should be conducted using more molecular-weight forms of guar gum to determine if a specific molecular weight for optimal fermentability indeed exists. The next step of this study would be to identify if these differences in fermentability are great enough to exert different physiological effects in humans.

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