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# New high-specificity fibers with strong and consistent responses across individuals

Mirian Aparecida de Campos Costa,[a,b] Xinruo Zhao,[a] Dirce Komura,[c] Elaine Rosechrer Carbonero,[d] Grethel Teresa Choque-Delgado,[e] Yunus E. Tunçil,[a,f] Thales Cipriani,[g] Yony Román-Ochoa,[g,h] Bruce R. Hamaker[a] and Thaisa M. Cantu-Jungles *[a]

Individual variability in gut microbiota responses limits the consistency of health benefits from prebiotic fiber interventions. Building on our concept of fiber hierarchical specificity, defined as the selective alignment and use of fibers by a narrow subset of gut microbes, we evaluated new putative high-specificity fibers for their ability to promote predictable and intense microbial shifts across individuals. Here, six candidate fibers (Acacia gum, Fucogalactan, Gellan gum, Guar gum, Locust bean gum, and Xylooligosaccharides) were tested *in vitro* using fecal microbiota from ten donors and compared to low-specificity (Fructooligosaccharides) and high-specificity (an insoluble glucan) reference fibers. SCFA analysis showed that Fucogalactan and Guar were strongly propiogenic, while Acacia and Locust promoted balanced SCFA production. Gellan exhibited minimal fermentability. Acacia, Fucogalactan, Guar, and Locust consistently enriched putative beneficial genera (*Eisenbergiella*, *Hungatella*, *Anaerotruncus*, and *Parabacteroides*, respectively), with strong and consistent responses across individuals, features characteristic of high-specificity fibers. In contrast, Fructooligosaccharides and Xylooligosaccharides produced more variable, and less intense responses. Our findings support Acacia, Fucogalactan, Guar, and Locust as high-specificity fibers that induce consistent, taxon-targeted shifts in the gut microbiome. These expand the repertoire of high-specificity fibers—a promising prebiotic approach for predictable microbiota modulation and related health outcomes.

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## 1. Introduction

Individual variability in gut microbiota composition and function has posed a major challenge for the consistent modulation of host health through dietary fiber interventions.[1–7] While some individuals exhibit robust microbial responses to fiber intake, others show minimal or no changes, often characterized as "non-responders".[8–10] This inter-individual variation highlights the need for more targeted strategies capable of eliciting predictable microbiota shifts. We previously introduced the concept of fiber hierarchical specificity, which refers to the degree to which a dietary fiber is selectively utilized by gut microbes.[11] In this framework, low-specificity fibers are broadly fermentable by many taxa, while high-specificity fibers are accessible to only a narrow group of microbes with specialized metabolic capabilities. We showed experimentally that specificity has important implications for both the consistency and magnitude of microbial responses.[12] Low-specificity fibers yield variable outcomes across individuals, given that many microbes can access the substrate and that the composition of these competing microbes differs between people. As a result, the outcome depends heavily on the competitive dynamics within each individual's gut microbiome.[11,12] In contrast, high-specificity fibers reduce microbial competition by limiting access to a few specialized taxa. This reduction in competitive pressure enables more consistent enrichment of target microbes across individuals, provided those taxa are present.

[a]*Whistler Center for Carbohydrate Research and Department of Food Science, Purdue University, 745 Agriculture Mall Drive, West Lafayette, IN 47907, USA. E-mail: tcantuju@purdue.edu*
[b]*Grupo de Pesquisa em Compostos Bioativos e Carboidratos (BIOCARB), Departamento de Ciência e Tecnologia de Alimentos, Avenida Peter Henry Rolfs, s/n, Viçosa, MG, 36570-900, Brazil*
[c]*Instituto Nacional de Pesquisas da Amazonia (INPA), Manaus, MA, Brazil*
[d]*Instituto de Química, Universidade Federal de Catalão, Catalão, GO, Brazil*
[e]*Departamento Académico de Ingeniería de Industrias Alimentarias, Universidad Nacional de San Agustín de Arequipa, Arequipa, Peru*
[f]*Food Engineering Department, Necmettin Erbakan University, Konya 42090, Turkey*
[g]*Departmento de Bioquimica e Biologia Molecular, Universidade Federal do Paraná, Curitiba, Paraná, Brazil*
[h]*Carbohydrate Biochemistry Research Group (BIOCAB), Department of Chemistry, Universidad de Los Andes, Bogotá, Colombia*

Moreover, the intensity of the response is often greater for high-specificity fibers, as fewer organisms compete for such fibers, funneling energy generated from fermentation toward the growth and metabolism of the target taxa.[12] Conversely, low-specificity fibers typically produce lower intensity and less targeted shifts due to resource sharing among a larger group of microbes.[12] This concept has since been referenced by others as a useful framework for understanding inter-individual variation in fiber responses.[13–25]

The specificity of a dietary fiber is primarily determined by its physicochemical structure, complexity, and the bacterial genetic requirements for its degradation. High-specificity fibers typically possess rare glycosidic linkages, uncommon monosaccharide compositions, branching patterns, or degrees of polymerization that are not widely accessible to most gut microbes.[11] Moreover, they can be present as matrices or possess other specific physical arrangements and properties (*e.g.*, insolubility, high-viscosity, *etc.*) which may restrict bacterial utilization. Only microbes that harbor specialized carbohydrate-active enzymes (CAZymes) and additional necessary machinery for accessing those carbohydrates can metabolize these complex substrates,[26,27] restricting fermentation to a narrow subset of taxa.[11,12] While a gut bacterium may exhibit alignment to a high-specificity fiber, it typically harbors a broader repertoire of CAZymes that enables it to metabolize additional, structurally distinct carbohydrates.[27] Low-specificity fibers on the other hand, have more common structural features—such as those found in fructooligosaccharides—to which many bacteria in the gut harbor the necessary degradation machinery. As a result, a diverse set of microbes can compete for and utilize these substrates. Beyond physicochemical features, commonality in the diet (*i.e.*, more bacteria have evolved to degrade them) and cross-feeding mechanisms must also be considered, as they can interfere with or reduce specificity levels.[11,12] Based on these concepts, most currently available prebiotics fall into the low-specificity category, being low-complexity polymers and oligomers (*e.g.*, inulin, FOS, GOS) and/or are common in the diet (*e.g.*, resistant starches). We have previously identified a unique 1,3-β-glucan derived from an Amazonian mushroom, which consistently enriched *Anaerostipes* spp. and *Bacteroides uniformis*[12,28] and classified it as a high-specificity fiber. However, to date, there remains a lack of data on other structurally distinct fibers that may exert similarly targeted effects on different beneficial bacterial taxa.

In this study, we evaluated a set of structurally complex candidate fibers or with diet-unusual sugar compositions and linkage patterns, for their potential classification as high-specificity fibers. We assessed whether they could induce consistent and intense modulation of gut microbiota composition across multiple individuals, which are criteria in our definition. Using *in vitro* fecal fermentation models with microbiota from ten different donors, we aimed to expand the repertoire of high-specificity fibers and deepen our understanding of how fiber structure determines community shifts across individuals.

# 2. Materials and methods

## 2.1. Donors and fecal sample collection

Fecal samples were obtained from 10 reportedly healthy donors (two males and eight females) recruited through advertisements fixed at the Purdue University campus. Eligible donors were male and female between 18 and 40 years old, with participants in the study ranging from 23 to 37 years of age and a mean age of 28 years. All donors had a normal body mass index (BMI) range (18.5–24.9 kg $m^{-2}$), with an average BMI of 22.87 kg $m^{-2}$ (range: 19.53–24.84 kg $m^{-2}$). In terms of self-reported ethnicity, 70% identified as Latino or Hispanic, while 10% identified as Asian, 10% as African American, and 10% as White. We did not include individuals who had taken antibiotics in the 3 months before the study or probiotics in the last 2 weeks; those with any gastrointestinal disorders or previous surgery in the gastrointestinal tract in the last 5 years; heavy drinkers (>4 drinks per day or >14 drinks per week for men and >3 drinks per day or >7 drinks per week for women); smokers; or pregnant and breastfeeding women. Donors were required to fast overnight the day before fecal collection (no food intake after 10 PM) and refrain from heavy physical activity outside their routine.

Fecal samples were collected in sterile plastic tubes, which were immediately sealed, stored in a refrigerator, and then transferred into an anaerobic chamber (BactronEZ Anaerobic Chamber; Shel Lab, Cornelius, OR) where *in vitro* fecal fermentation was performed. All samples were utilized for fermentation within 2 hours of collection. Human stool collection and use were performed in accordance with U.S. DHHS 45 CFR 46 regulations and institutional guidelines, and were approved by the Purdue University IRB (protocol #1510016635). Informed consent was obtained from all human participants.

## 2.2. Dietary fibers

Fibers were selected based on parameters previously described to increase fiber specificity, including complex physicochemical structure and low abundance in most diets, as we previously proposed.[11] Acacia gum [ACACIA] (TIC Pretested® Gum Arabic FT Powder), Gellan gum [GELLAN] (TICorganic® Caragum 200), Guar gum [GUAR] (TICorganic® Guar Gum 3500 F Powder), and Locust bean gum [LOCUST] (TIC Pretested® Locust Bean Gum POR A2 Powder) were sourced from TIC Gums (an Ingredion company, Belcamp, MD, USA). Fucogalactan [FUCG] was extracted and purified following the method described by Román *et al.*[29] Xylooligosaccharides [XOS] were obtained from Nutrasumma (Phoenix, AZ, USA; product code 00150). Fructooligosaccharides [FOS] from chicory root (Sigma-Aldrich, St Louis, MO, USA; CAS number F8052) were used as the low-specificity negative control.[12] The insoluble 1,3-β-glucan [GLUCAN] extracted from *Cookeina speciosa*,[28] was utilized as the high-specificity positive control as previously demonstrated.[12]

## 2.3. *In vitro* fecal fermentation

*In vitro*, fecal fermentation was performed according to the methodology described by Lebet *et al.*[30] with modifications.

Briefly, on the day before fermentation, 20 mg of each dietary fiber was added to sterile tubes, in duplicate. A carbonate–phosphate buffer containing trace element solution and resazurin (1 mg $mL^{-1}$) was also prepared and filtered by a Nalgene® vacuum filtration system (Sigma-Aldrich) for decontamination. Cysteine hydrochloride (0.25 g $L^{-1}$ of buffer) was added as a reducing agent. The prepared buffer was then placed into the anaerobic chamber to complete buffer reduction. On the day of the experiment, fresh stool samples were mixed with carbonate–phosphate buffer in a 1 : 3 (w/v) ratio and thoroughly homogenized. The resulting mixture was then filtered through four layers of gauze to remove large particles. A 0.5 mL portion of the fecal suspension was transferred into Balch tubes (Chemglass Life Sciences, Vineland, NJ) preloaded with 20 mg of dietary fiber and 2 mL of carbonate–phosphate buffer. The tubes were sealed using butyl rubber stoppers and aluminum crimp seals (both from Chemglass Life Sciences), then incubated at 37 °C with shaking at 110 rpm for 24 hours in a MaxQ 6000 shaker incubator (Thermo Fisher, Waltham, MA). Samples collected at baseline and after 24 hours of fermentation were aliquoted and stored at −80 °C for subsequent short-chain fatty acid (SCFA) analysis (0.5 mL) and microbial DNA sequencing (1 mL). All procedures involving sample handling were carried out under anaerobic conditions and maintained with a gas mixture composed of 85% nitrogen, 5% carbon dioxide, and 10% hydrogen.

### 2.4. Short-chain fatty acids analysis

Samples for SCFA analyses were prepared as previously described[28] and analyzed using a gas chromatograph (GC-FID 7890 A; Agilent Technologies Inc.) on a fused silica capillary column (Nukon Supelco no. 40369-03A; Bellefonte, PA) under the following conditions: injector temperature at 230 °C, initial oven temperature at 100 °C, and temperature increase of 8 °C $min^{-1}$ to 200 °C with a hold for 3 min at final temperature. Helium was used as a carrier gas at 0.75 ml $min^{-1}$. Quantification was performed based on relative peak area using external standards of acetate (0.6 M), propionate (0.15 M), butyrate (0.15 M), isobutyrate (0.15 M), and isovalerate (0.15 M) (Sigma-Aldrich). An internal standard was prepared by mixing 157.5 μL of 4-methyl valeric acid ($C_6H_{12}O_2$), 1.47 mL of phosphoric acid ($H_3PO_4$) (85% v/v), and 39 mg of copper sulfate pentahydrate ($CuSO_4{\cdot}5H_2O$) diluted in 25 μL of distilled water.

### 2.5. DNA extraction and 16S rRNA gene amplicon sequencing

DNA was extracted from stored fecal samples using the QIAamp® PowerFecal® Pro DNA Kit (Qiagen, USA) following the manufacturer's instructions. For each extraction, 1 mL of fecal material was first defrosted in an icebox and then centrifuged at 15 000*g* at room temperature for 10 minutes (Microfuge® 20R, Beckman Coulter, Germany). The supernatant was discarded, and the remaining material was suspended in 700 μL of the CD1 buffer provided in the kit. This mixture was then transferred into PowerBead Pro Tubes® (Qiagen, USA), which contain microspheres, and subjected to two centrifugation cycles at room temperature using the FastPrep-24™ system (MP Biomedicals, USA) to promote cell lysis. Each cycle lasted 40 seconds, with a 5-minute interval between them.

The tubes were then centrifuged for 1 minute at 15 000*g* at room temperature (Microfuge® 20R, Beckman Coulter, Germany). 600 μL of the resulting supernatant was transferred into a new tube, and DNA extraction was completed using the QIAcube Connect automated system (Qiagen, USA), following the manufacturer's protocol.

The concentration of extracted DNA was assessed using a NanoDrop 2000c spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA). The purity of the DNA was evaluated by measuring absorbance ratios at 260/280 nm and 260/230 nm, with optimal values defined as 1.8–2.0 and 2.0–2.2, respectively. Extracted genetic material was stored at −80 °C until it was submitted for sequencing. Genomic DNA was used as a template to amplify the V4 region of the microbial 16S rRNA gene using primers 515F (GTGCCAGCMGCCGCGGTAA) and 806R (GGACTACHVGGGTWTCTAAT). Amplicons were generated using a two-stage PCR protocol, with barcoded primers introduced in the second stage. Libraries were pooled, purified using an AMPure XP cleanup protocol, and sequenced on an Illumina MiniSeq platform (2 × 153 bp paired-end reads) with a 20% phiX spike-in at the University of Illinois at Chicago Genome Research Core.

### 2.6. Statistics and bioinformatics

Short-chain fatty acid (SCFA) concentrations were analyzed using GraphPad Prism (version 10). Differences among fiber treatments were assessed using ordinary one-way analysis of variance (ANOVA) followed by Tukey's *post-hoc* test for multiple comparisons. Statistical significance was defined as $p < 0.05$.

Sequencing data were processed using the QIIME2 platform (v. 2020.2).[31] Raw reads were demultiplexed, quality filtered, denoised, and merged using the DADA2[32] plugin to generate amplicon sequence variants (ASVs). Chimeric sequences were identified and removed during denoising. Taxonomic classification was performed using a Naïve Bayes classifier trained on the SILVA 138 database (99% sequence identity threshold). Sequences were rarefied to a depth of 10 400 reads per sample prior to diversity analyses to standardize sampling effort across samples. Alpha diversity metrics, including observed ASVs (richness) and Pielou's evenness (evenness), were calculated using the diversity plugin in QIIME2. Group comparisons for alpha diversity were assessed using Kruskal–Wallis tests with Benjamini–Hochberg false discovery rate (FDR) correction. Beta diversity was evaluated using Weighted UniFrac distance matrices. Principal Coordinate Analysis (PCoA) plots were generated to visualize differences in community structure. Statistical significance of group separations was tested by pairwise PERMANOVA (999 permutations) with FDR correction to compare microbial community shifts induced by each tested fiber to those observed with the low-specificity fiber (FOS) and the high-specificity fiber (GLUCAN) using pairwise PERMANOVA tests. To assess inter-individual variability, permutational analysis of multivariate dispersions (PERMDISP)

 

was performed using Weighted UniFrac distances. Differential abundance analysis at the genus level was performed using ANCOM-II (Analysis of Composition of Microbiomes) implemented in QIIME2. For graphical representations, we generated relative abundance bar plots, Weighted UniFrac PCoA plots with trajectory arrows (illustrating compositional shifts pre- and post-fiber fermentation), and paired before-and-after $\log_{10}$-transformed abundance plots for taxa identified by ANCOM-II in R (v. 4.3.2) using the ggplot2 and phyloseq packages. Relative abundances were expressed as percentages and $\log_{10}$-transformed to facilitate visualization across several orders of magnitude. A pseudocount of 0.01% was added prior to transformation to account for zeros, corresponding to the lower detection limit shown on the axis.

## 3. Results and discussion

We performed *in vitro* fecal fermentations using gut microbiota communities from 10 different donors, comparing six new putative high-specificity fibers (Acacia gum [ACACIA], Fucogalactan [FUCG], Gellan gum [GELLAN], Guar gum [GUAR], Locust bean gum [LOCUST], and Xylooligosaccharides [XOS]) to previously identified low- and high-specificity fibers (fructooligosaccharides [FOS] and insoluble glucan [GLUCAN], respectively).

### 3.1. SCFA production profiles differ among fibers

SCFA production varied substantially among the different fibers tested. FOS and XOS led to the highest acetate concentrations, reaching 78.1 and 85.3 mM, respectively (Fig. 1A). ACACIA, FUCG, GUAR, and LOCUST also generated notable levels of acetate (65.9, 53.4, 57.0, and 5.8 mM, respectively), though generally lower than FOS and XOS (Fig. 1A). Propionate production was highest for FUCG and GUAR, which yielded 29.4 and 27.6 mM, respectively (Fig. 1B). These were followed by intermediate levels observed for FOS, LOCUST, and XOS (16.6, 20.3, and 17.6 mM, respectively). GLUCAN and ACACIA also produced measurable propionate, though to a lesser extent (10.1 and 13.9 mM, respectively) (Fig. 1B). Butyrate levels were greatest for FOS, LOCUST, and XOS (15.8, 13.3, and 14.5 mM, respectively), with GLUCAN, ACACIA, FUCG, and GUAR showing moderate production (6.9, 8.5, 7.5, and 9.4 mM, respectively) (Fig. 1C).

Notably, GELLAN consistently resulted in the lowest concentrations of all three SCFAs, with values not significantly different from the blank control, suggesting this fiber had

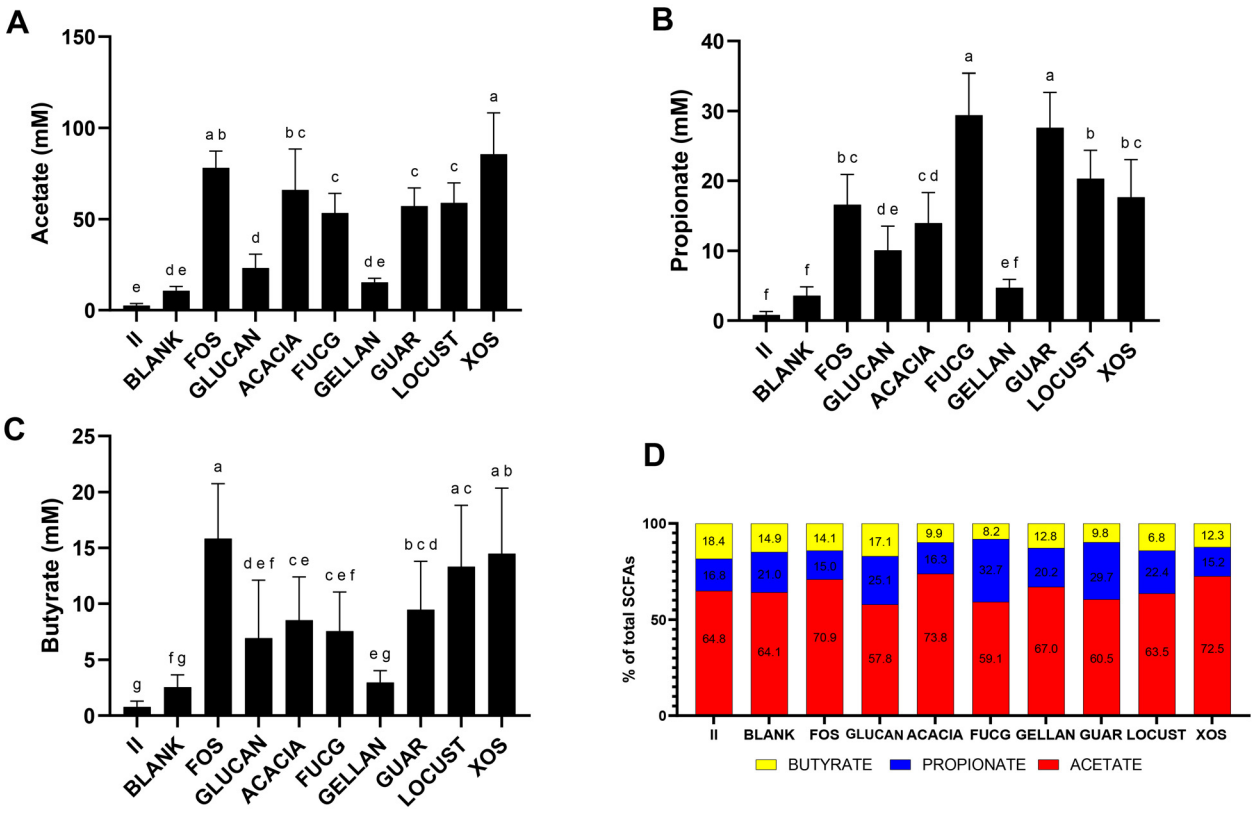


**Fig. 1** Short-chain fatty acid (SCFA) production across fibers. SCFA concentrations (mM) were measured after 24-hours *in vitro* fecal fermentation using microbiota from 10 donors, for each tested fiber. Panels show (A) acetate, (B) propionateand (C) butyrate concentrations. (D) Relative proportions (%) of each SCFA per fiber. Tested fibers included: ACACIA (acacia gum), FUCG (fucogalactan), GELLAN (gellan gum), GUAR (guar gum), LOCUST (locust bean gum), XOS (xylooligosaccharides), and the controls FOS (fructooligosaccharides), and GLUCAN (insoluble β-1,3-glucan from *Cookeina speciosa*). Bars represent mean ± SD ($n$ = 10). Statistically significant differences among groups are shown in Table S1.

poor fermentability under the tested conditions (Fig. 1A–C). Ratios among the three SCFAs helped to reveal whether fibers promoted microbial groups favoring the production of specific SCFAs over others. ACACIA, XOS, and FOS showed the highest acetate proportions (16.3%, 15.2%, and 15.0%, respectively) (Fig. 1D). FUCG, GUAR, GLUCAN, and LOCUST were highly propiogenic, with propionate comprising 32.7%, 29.7%, 25.1%, and 22.4% of total SCFAs, respectively (Fig. 1D). GLUCAN had the highest proportion of butyrate, representing 17.1% of total SCFAs across all substrates tested. This finding aligns with our previous data demonstrating the specific stimulation of a butyrogenic taxon by this fiber,[28] which was subsequently confirmed to be a high-specificity fiber,[12] reducing competitive pressure for fiber utilization.

### 3.2. Effects on microbial diversity

Changes in the gut microbial community structure in response to the tested fibers were assessed through 16S rRNA gene sequencing. Baseline diversity and composition varied substantially between donors, as illustrated in Fig. S1 and supported by the donor-level alpha-diversity metrics in Fig. 2. Alpha diversity was evaluated by the number of observed ASVs (for richness) and Pielou's evenness (for evenness) (Fig. 2). As expected, there were no substantial differences in richness among treatments (pairwise Kruskal–Wallis $q > 0.05$ for all comparisons; Table S1). In a closed system, the microbial community can utilize not only the added fiber but also residual fibers present in small amounts within the fecal inoculum, and no new taxa can enter the system, thus, changes in richness are unlikely.[33] However, differences were more apparent in community evenness as expected *in vitro* fermentations[33] (Fig. 2). GELLAN, which was poorly fermented, mirrored the diversity profile observed in the blank control, where no fiber was added. Both FUCG and GELLAN, as well as LOCUST, led to significant reductions in evenness ($q = 0.001$ for FUCG and GELLAN; $q = 0.039$ for LOCUST in pairwise Kruskal–Wallis *vs.* blank; Fig. 2), which could indicate a narrow fermentation profile that counteracts community evenness. In contrast, other fibers maintained higher levels of evenness, which could be interpreted to support a broader number of taxa or, in the case of narrow fermentation profiles, to target bacteria, which may favor the growth of less abundant taxa.[33]

Weighted UniFrac beta diversity results are visualized in comparison to the low-specificity fiber, FOS (Fig. 3, Fig. S3), and the high-specificity fiber GLUCAN (Fig. 4, Fig. S4). Compared to the low-specificity fiber FOS (in each plot), FUCG, GUAR, and LOCUST induced more consistent shifts in microbial composition across individual donors (Fig. 3, Fig. S3). This was illustrated by the directional trajectories in the ordination plots, where fermentation with these fibers led to more uniform changes in gut microbiota composition across different donors (Fig. 3). Pairwise PERMANOVA analysis further confirmed that all fibers, except XOS, led to statistically significant differences in community structure compared to FOS ($q$-values: ACACIA = 0.007, FUCG = 0.002, GELLAN = 0.026, GUAR = 0.002, LOCUST = 0.002, XOS = 0.573; Table S2), suggesting distinct microbial responses. XOS produced a shift in microbial structure that largely mirrored that of FOS, con-

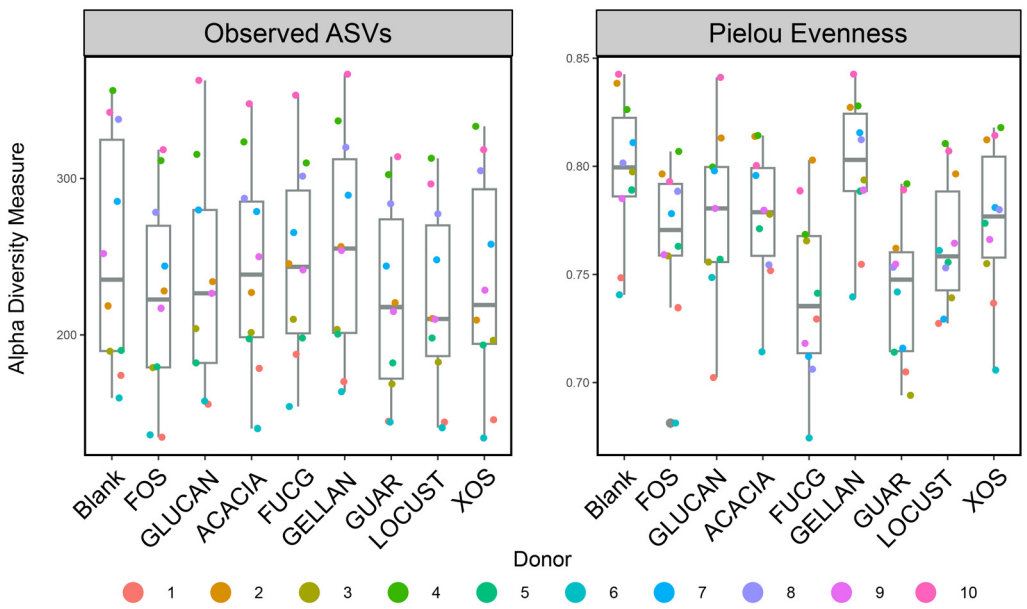


**Fig. 2** Alpha diversity of microbial communities after fiber fermentation. Number of observed amplicon sequence variants (ASVs, left) and Pielou's evenness index (right) after 24-hour fermentation of each fiber using microbiota from 10 donors. Tested fibers included: ACACIA (acacia gum), FUCG (fucogalactan), GELLAN (gellan gum), GUAR (guar gum), LOCUST (locust bean gum), XOS (xylooligosaccharides), and the controls FOS (fructooligosaccharides), and GLUCAN (insoluble β-1,3-glucan from *Cookeina speciosa*). Statistical comparisons were made using pairwise Kruskal–Wallis tests with FDR correction.

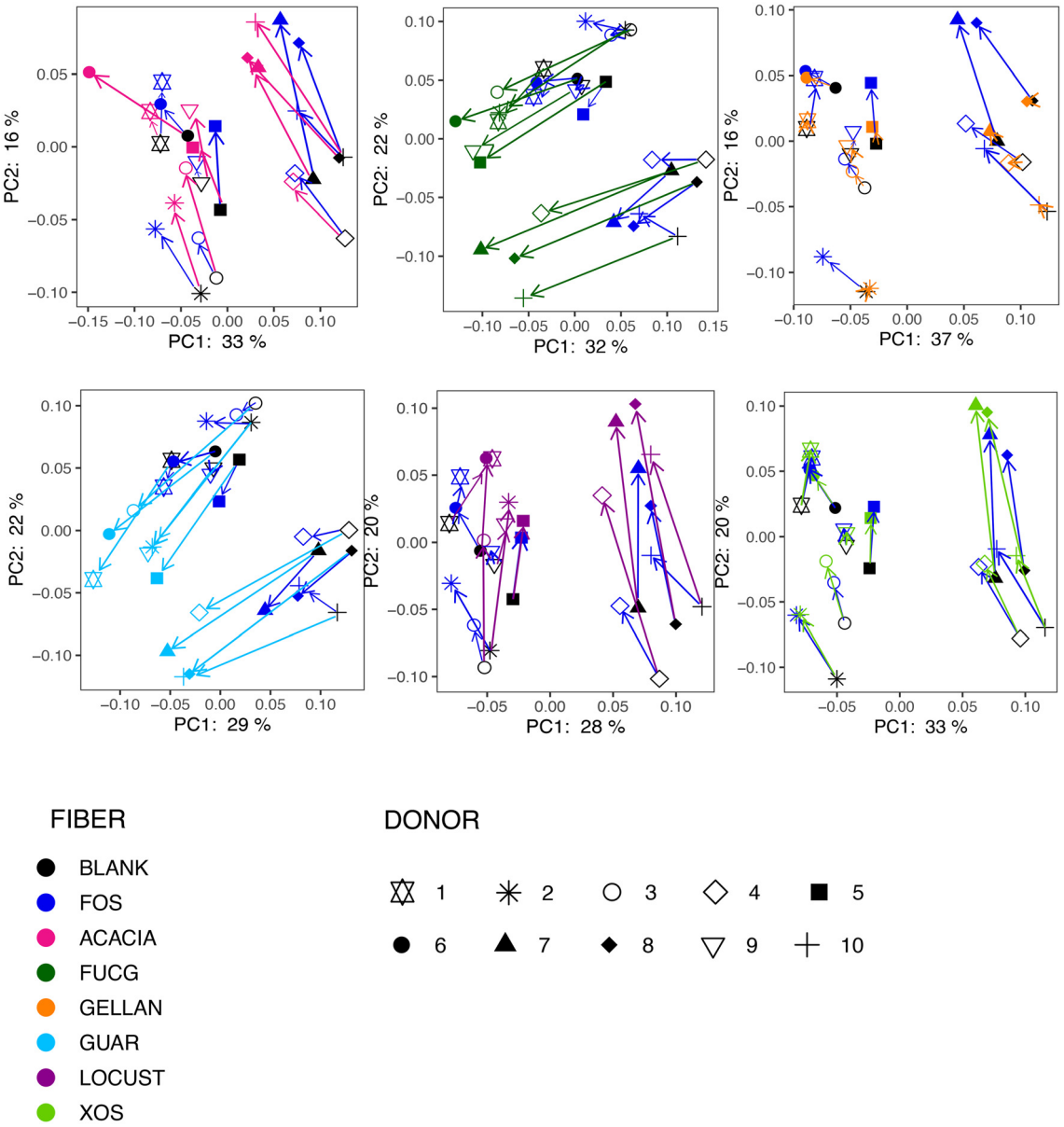


**Fig. 3** Beta diversity analysis comparing fiber treatments to the low-specificity fiber FOS. Weighted UniFrac principal coordinate analysis (PCoA) plots showing microbial community trajectories after 24-hour fermentation with each tested fiber, plotted in reference to the FOS-treated community. Samples are colored according to treatment and shapes correspond to individual donors. The connecting lines indicate the trajectory of each donor's microbiota from baseline to treatment ($n$ = 10). Tested fibers included: ACACIA (acacia gum), FUCG (fucogalactan), GELLAN (gellan gum), GUAR (guar gum), LOCUST (locust bean gum), XOS (xylooligosaccharides), and the control FOS (fructooligosaccharides).

sistent with their classification as low-specificity fibers. As anticipated from its SCFA profile indicating poor fermentability, GELLAN elicited minimal changes in community composition and closely resembled the blank control (pairwise PERMANOVA $q$ = 0.973 *vs.* blank; Table S2).

Compared to the high-specificity fiber GLUCAN, all tested fibers induced distinct shifts in community structure, as supported by pairwise PERMANOVA results (Table S2) and evidenced by differences in trajectory direction in some of the ordination plots (Fig. 4). These findings indicate that the specific directional shift in microbial composition promoted by GLUCAN was not replicated by any other tested fiber, suggesting that each fiber supports distinct bacteria or bacterial groups, as expected given their differing physicochemical structures.[34] However, when examining the magnitude and consistency of responses (reflected by the length and alignment of sample trajectories across donors [Fig. 4, Fig. S3]), ACACIA, FUCG, GUAR, and LOCUST, but not XOS, displayed a response pattern resembling that of GLUCAN, *i.e.*, with long, similarly oriented trajectories. This response profile is consistent with our previous observations of high-specificity fibers, which elicit strong and uniform shifts in microbial community

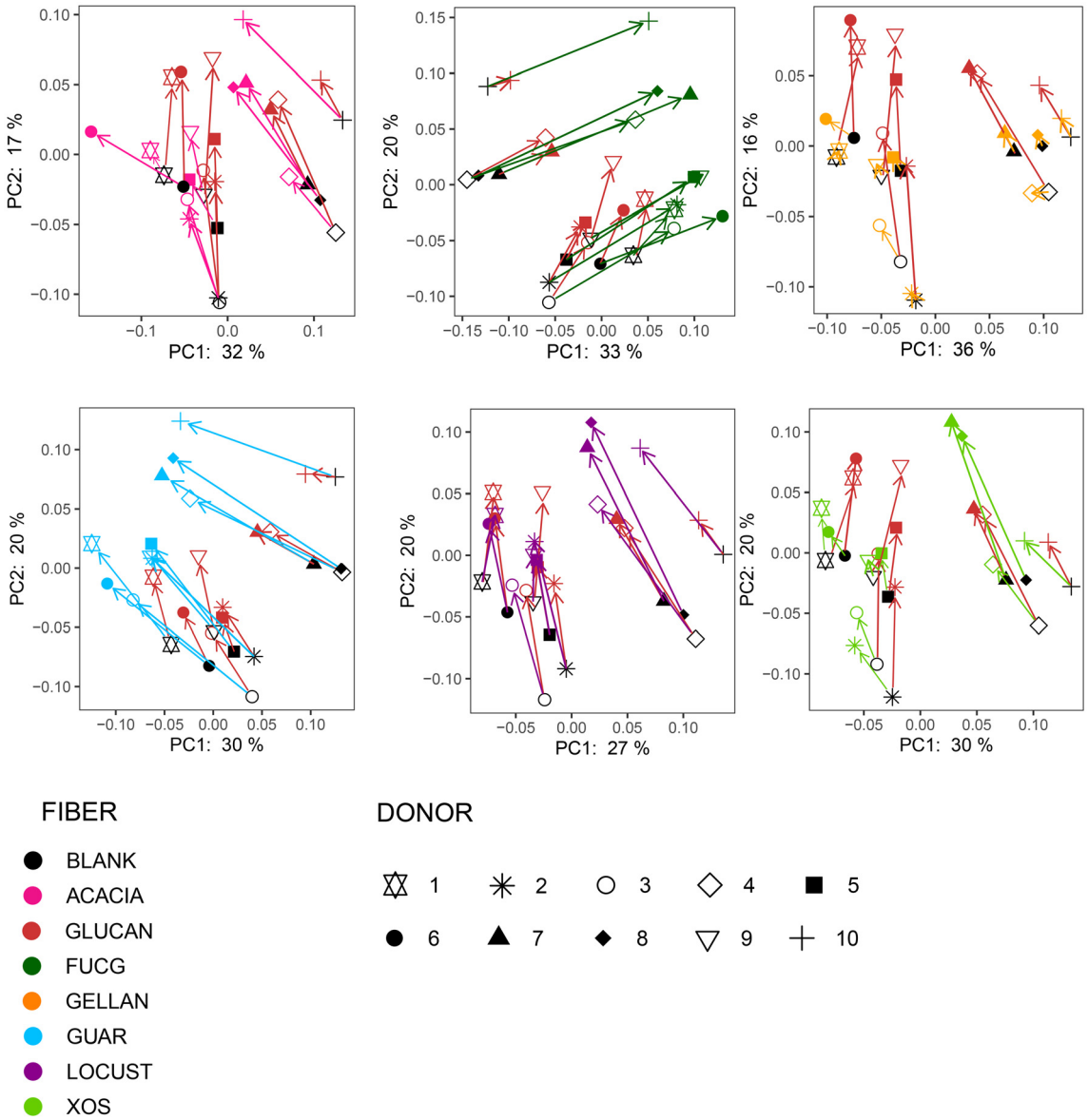


**Fig. 4** Beta diversity analysis comparing fiber treatments to the high-specificity fiber GLUCAN. Weighted UniFrac principal coordinate analysis (PCoA) plots showing microbial community trajectories after 24-hour fermentation with each tested fiber, plotted in reference to the GLUCAN-treated community. Samples are colored according to treatment and shapes correspond to individual donors. The connecting lines indicate the trajectory of each donor's microbiota from baseline to treatment ($n$ = 10). Tested fibers included: ACACIA (acacia gum), FUCG (fucogalactan), GELLAN (gellan gum), GUAR (guar gum), LOCUST (locust bean gum), XOS (xylooligosaccharides), and the control GLUCAN (insoluble β-1,3-glucan from *Cookeina speciosa*).

structure across individuals.[12] Notably, GUAR and FUCG reduced inter-individual variability in the final community: GUAR compared to Blank and FOS (pairwise PERMDISP, $q$ = 0.041 and $q$ = 0.003, respectively), and FUCG compared to Blank (pairwise PERMDISP, $q$ = 0.045) (Table S2). These corroborate the observed more uniform donor responses, reflective of their high-specificity pattern. The observed intensity and similar fiber responses on microbial structure across donors highlight the capacity of ACACIA, FUCG, GUAR, and LOCUST to elicit consistent, directional shifts in the gut microbiota, a defining feature of high-specificity fiber responses.

### 3.4. Differential promotion of gut bacterial genera

Next, we aimed to identify which taxa were targeted by the newly identified high-specificity fibers through differential abundance analysis using ANCOM-II at the genus level. As expected, the control low-specificity fiber FOS did not differentially promote any specific taxa (Fig. 5), consistent with its previously reported low-specificity profile. In contrast, the high-specificity fiber GLUCAN significantly promoted the *Eubacterium ventriosum* group (Fig. 5). This is in agreement with our previous studies, where this taxon was also promoted

 

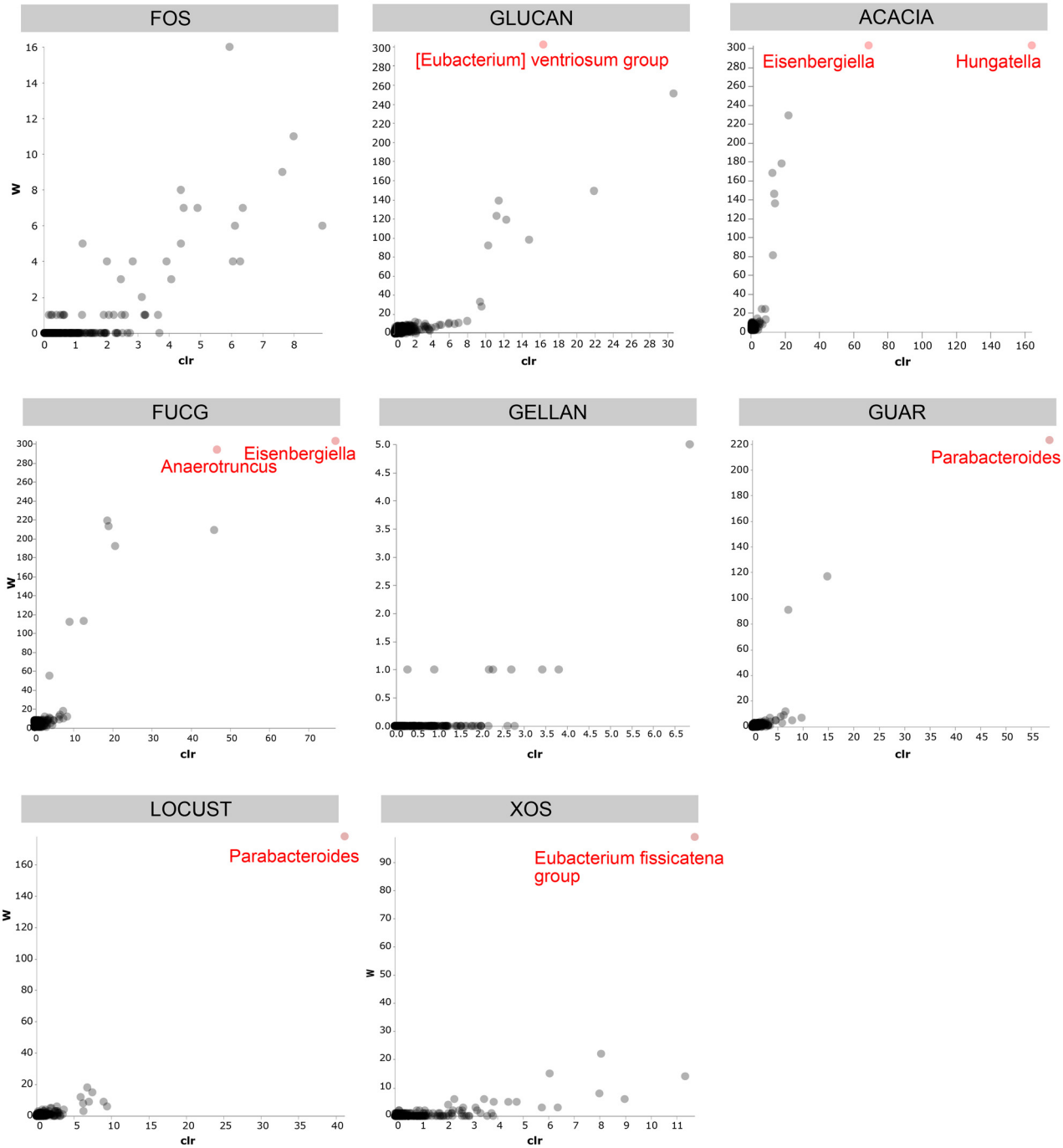


**Fig. 5** Differentially abundant genera are promoted by each fiber. Taxa identified as differentially abundant (W-statistic > threshold, identified in red) by ANCOM-II at the genus level across 10 donor fermentations. Only significant changes relative to the blank control are shown. Tested fibers included: ACACIA (acacia gum), FUCG (fucogalactan), GELLAN (gellan gum), GUAR (guar gum), LOCUST (locust bean gum), XOS (xylooligosaccharides) and the controls FOS (fructooligosaccharides), and GLUCAN (insoluble β-1,3-glucan from *Cookeina speciosa*).

(though classified as *Anaerostipes* at the time, due to differences in the classification databases used across studies). Among the newly identified high-specificity fibers, several novel targeted genera emerged. *Eisenbergiella* was promoted by both ACACIA and FUCG, while *Hungatella* was specifically targeted by ACACIA. FUCG also promoted *Anaerotruncus*, and *Parabacteroides* was supported by both GUAR and LOCUST (Fig. 5). Although XOS elicited fermentation responses resembling FOS in beta-diversity analyses (Fig. 3), with high interindividual variability and a mix of responders and non-responders, it still was detected to promote the *Eubacterium fissicatena* group. As expected, GELLAN, which showed poor

fermentability, did not result in any differentially abundant taxa.

Fig. S1 illustrates overall microbiota relative abundances across donors and treatments, confirming the expected inter-individual variation in community structure. To further evaluate whether ANCOM-II-identified taxa were promoted in a consistent and intense manner across individuals, a key feature of high-specificity fiber responses,[12] we evaluated their relative abundances ($\log_{10}$) across donors (Fig. 6). As anticipated, the high-specificity control GLUCAN led to consistent increases in *E. ventriosum* across all donors, with an average $\log_{10}$ fold change of 1.7, except in donors D7 and D8, who lacked detectable baseline levels (Fig. 6A). Similarly, ACACIA and FUCG consistently promoted *Eisenbergiella* across all donors, with average $\log_{10}$ fold changes of 2.1 and 1.9, respectively (Fig. 6B). While other fibers also induced some growth of this taxon, their effects were less consistent across individuals. ACACIA uniquely promoted *Hungatella* with a robust and consistent response across all donors (average $\log_{10}$ fold change: 3.1), an effect not observed with any other fiber (Fig. 6C). *Anaerotruncus* was promoted by multiple fibers, but only FUCG induced strong and consistent increases across all donors (average $\log_{10}$ fold change: 1.6) (Fig. 6D). *Parabacteroides* was also consistently and intensively promoted by GUAR and LOCUST, with average $\log_{10}$ fold changes of 0.7 and 0.73, respectively (Fig. 6E). While FUCG also appeared to promote *Bacteroides*, the response was not consistent—donor D6, for instance, did not respond to the fiber. This variability may reflect differences in carbohydrate utilization capabilities at the species or strain level that would need to be investigated through shotgun sequencing analysis. XOS and GLUCAN also induced some homogeneous increases in *Bacteroides*, though at much lower intensities ($\log_{10}$ fold changes of 0.3 and 0.2, respectively). Notably, ACACIA, FUCG, GUAR, and LOCUST promoted their respective bacterial targets consistently across donors (Fig. 6), despite low baseline abundances (averaging 0.3% of the community), including in donors where target taxa were initially below the sequencing detection threshold (Fig. S2). This corroborates our previous reports where a strong promotion of target taxa was achieved by the high-specificity fiber, even when making up for as low as 0.01% of the baseline community.[12] Finally, although XOS promoted *E. fissicatena*, the effect was mild ($\log_{10}$ fold change: 0.5) and inconsistent (*e.g.*, donor D2 did not respond despite being present at baseline at low levels, and even showed growth in the blank control) (Fig. 6F).

Overall, based on the consistency and intensity of responses, our findings support the classification of ACACIA as a high-specificity fiber for *Eisenbergiella* and *Hungatella*, FUCG for *Eisenbergiella* and *Anaerotruncus*, and GUAR and LOCUST for *Parabacteroides*. The mechanisms underlying this selectivity (high-specificity) are likely tied to each fiber's structural complexity and physicochemical properties, including solubility, viscosity, and glycosidic linkage patterns.[11]

ACACIA promoted consistent enrichment of *Eisenbergiella* and *Hungatella*. Structurally, acacia gum is a highly branched arabinogalactan–protein complex, consisting mainly of β-(1→3)-linked galactose backbone with side chains containing galactose, arabinose, rhamnose, and glucuronic acid.[35] This extensive branching and incorporation of uncommon sugar moieties require a specialized enzymatic repertoire, limiting microbial access to only a few taxa.[36] *Eisenbergiella*, part of the *Lachnospiraceae* family in the Clostridium Cluster XIVa, is a butyrate producer associated with gut barrier maintenance, anti-inflammatory effects, and reduced risk of irritable bowel syndrome.[37–39] *Hungatella*, also enriched by ACACIA, contains genes encoding the acetyl–CoA pathway for butyrate production[40] and has been linked to anti-inflammatory dietary patterns and improved bowel function,[41] as well as protective roles in Crohn's disease and ulcerative colitis.[42] Furthermore, *Hungatella hathewayi* was noted to be enriched in long-lived individuals and associated with neuroactive metabolites and brain connectivity, suggesting a role in cognitive health and aging *via* the gut–brain axis.[43,44]

FUCG similarly enriched *Eisenbergiella*, but also uniquely promoted *Anaerotruncus*. FUCG is composed of an α-(1→6)-linked galactopyranosyl backbone similar to ACACIA, which could be linked to the promotion of *Eisenbergiella*. Moreover, it is a fiber partially methylated at the O-3 position and substituted at O-2 with α-L-fucopyranose or α-D-galactopyranose residues.[29] The presence of rare α-linkages and fucose branches, as well as methylation, might be related to its observed high-specificity. *Anaerotruncus*, a butyrate-producing genus in Clostridium Cluster IV[45] plays an important role in intestinal barrier integrity and immune regulation.[39,46] It has been inversely associated with intestinal inflammation in murine colitis models[47–49] and was shown to decline in overweight and obese individuals,[50] suggesting a protective role.

GUAR and LOCUST, both galactomannans, selectively promoted Parabacteroides. GUAR gum features a β-(1→4)-linked mannose backbone with α-(1→6)-galactose branches at a ~2 : 1 mannose-to-galactose ratio, resulting in high solubility and extremely high viscosity.[51,52] This viscosity may act as a physical barrier, slowing substrate diffusion and favoring taxa with machineries adapted to access fibers in highly viscous solutions. LOCUST, with a lower branching frequency (mannose : galactose ~4 : 1),[53] exhibits lower viscosity, but the reduced branching is also related to a less soluble polymer, a feature that also could contribute to increasing specificity by narrowing the number of taxa with machineries to access it. Despite differences in viscosity and solubility, both fibers enriched *Parabacteroides*, a genus known for its anti-inflammatory effects and beneficial metabolic benefits.[54–58] Notably, *Parabacteroides distasonis* has been shown to improve insulin sensitivity, reduce gut inflammation, and produce beneficial metabolites like succinate and secondary bile acids, as well as modulate immune responses and detoxify xenobiotics.[54–58]

The observed specificity of fiber-driven microbiota modulation could differ at lower taxonomic levels, as carbohydrate utilization machineries have been shown, in some microbes, to vary among species and strains. These differences could not be captured in this study due to the limited taxonomic resolu-

 

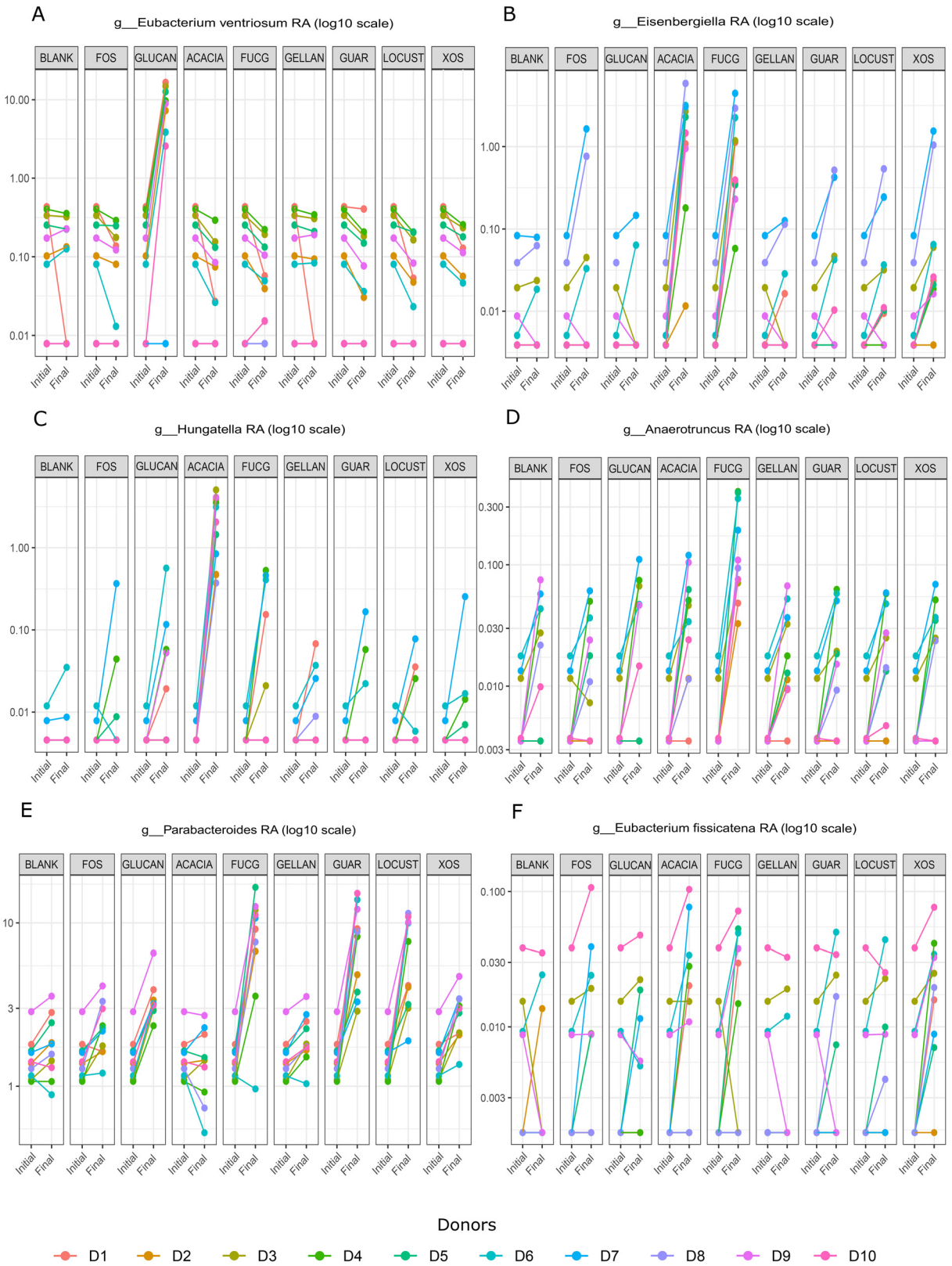


**Fig. 6** $Log_{10}$ relative abundance changes of enriched genera across donors. Individual donor-level responses ($n$ = 10) for each fiber and target taxon (A–F) identified by ANCOM-II. Values represent $log_{10}$-transformed relative abundances (%), with a pseudocount of 0.01% applied to handle zeros. Tested fibers included: ACACIA (acacia gum), FUCG (fucogalactan), GELLAN (gellan gum), GUAR (guar gum), LOCUST (locust bean gum), XOS (xylooligosaccharides), and the controls FOS (fructooligosaccharides), and GLUCAN (insoluble β-1,3-glucan from *Cookeina speciosa*).

tion of 16S rRNA sequencing. However, for many taxa, carbohydrate utilization capabilities appear to be conserved at the species level and may sometimes extend to the genus level,[59] although this might be restricted to broader structural classes of polysaccharides rather than fine physicochemical features. Targeting microbial traits shared across multiple strains and species may increase the likelihood of consistent responses across individuals, who typically harbor different strain types. However, broader targeting at higher taxonomic levels (*e.g.*, family or order) can introduce increased competitive pressures for substrate utilization, potentially diminishing fiber specificity.[11,12] While ACACIA, FUCG, GUAR, and LOCUST were placed in this study at the high end of the specificity spectrum, we recognize that fiber specificity is not a binary concept. Instead, it exists along a continuum ranging from low to intermediate to high, depending on how many taxa can access and degrade a given substrate.[11] At present, standardized or quantitative metrics to define where fibers fall within this hierarchical specificity classification remain to be developed.

With regard to dietary occurrence, several of the fibers tested here are uncommon in the human diet or consumed only at low levels. Fucogalactan, for example, is found in certain algae and mushrooms and not typically present in large amounts in Western diets, while acacia, guar, locust bean, and gellan gums occur primarily as food additives used for texturization and stabilization, leading to only minor habitual exposure. In contrast, XOS are naturally derived from xylan, a major hemicellulosic component of cereals and other plant-based foods, and thus represent structures to which the gut microbiota is frequently exposed. As we proposed previously,[11,12] fibers that are common in the diet are more likely to be utilized by a broad range of gut microbes and therefore tend to display lower specificity, whereas fibers with minimal dietary exposure are accessible to fewer microbes, eliciting more specific and consistent microbial responses. In line with this concept, it is reasonable that ACACIA, FUCG, GUAR, and LOCUST showed higher specificity than XOS in our study. Gellan, although also of low dietary exposure, did not fall into this categorization due to its very limited fermentability.

Finally, whereas here we evaluated which targets were promoted by high-specificity fibers, future studies should be conducted to evaluate how to intentionally design or select high-specificity fibers for microbes that one desires to promote. High-specificity prebiotics hold the potential to be used as precision microbiome strategies. Like oral probiotics, they can selectively augment the growth of beneficial taxa across individuals, provided that these taxa are present, even if at negligible abundance (as we showed for some targets here). For example, *Faecalibacterium prausnitzii*, a butyrate producer associated with intestinal and systemic health, is commonly found in most individuals but is often reduced in disease states.[60–62] High-specificity fibers designed to target such taxa could restore microbial balance with greater predictability than low-specificity prebiotics, offering a novel dietary strategy for microbiota-directed health interventions.

# 4. Concluding remarks

Our findings demonstrate that ACACIA, FUCG, GUAR, and LOCUST selectively enriched beneficial microbial taxa with consistency and intensity across diverse donor profiles, supporting their assignment as high-specificity fibers in our previously proposed hierarchical fiber classification scheme. That was in contrast to low-specificity fibers like FOS and XOS, which led to non-specific and variable inter-individual outcomes. Ultimately, high-specificity fibers structurally tailored to promote specific microbes present a promising strategy for targeted dietary interventions aimed at promoting beneficial microbes and improving host health through the gut microbiome.

## Author contributions

T. C.-J. conceptualized the study, supervised the experimental work, analyzed the data, and wrote the manuscript. M. A. C., X. Z., G. T. C. D., and Y. E. T. conducted the experiments. M. A. C. C. also contributed to manuscript writing. D. L. K. and E. R. C. were responsible for the collection, extraction, and purification of the Cookeina speciosa glucan (GLUCAN), and Y. R. O. and T. C. were responsible for the extraction and purification of the fucogalactan (FUCG) used in the study. B. R. H. conceptualized the study, supervised the experimental work, and edited the manuscript. All authors reviewed and approved the final version of the manuscript.

## Conflicts of interest

T. C-J. and B. H. are co-founders of the startup RiteCarbs LLC. M. A. C. C., X. Z., D. K., E. R. C., G. T. C.-D., Y. T., T. C. and Y. R. O. have no conflicts.

## Data availability

The raw sequencing data have been deposited in the NCBI Sequence Read Archive (SRA) under BioProject number PRJNA1331769.

Supplementary information is available. See DOI: https://doi.org/10.1039/d5fo02728d.

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## References

1 S. M. Gibbons, T. Gurry, J. W. Lampe, A. Chakrabarti, V. Dam, A. Everard, A. Goas, G. Gross, M. Kleerebezem, J. Lane, J. Maukonen, A. L. B. Penna, B. Pot, A. M. Valdes, G. Walton, A. Weiss, Y. C. Zanzer, N. V. Venlet and M. Miani, Perspective: Leveraging the Gut Microbiota to Predict Personalized Responses to Dietary, Prebiotic, and Probiotic Interventions, *Adv. Nutr.*, 2022, **13**, 1450–1461.

2 J. S. Lichtman, J. L. Sonnenburg and J. E. Elias, Monitoring host responses to the gut microbiota, *ISME J.*, 2015, **9**, 1908–1915.

3 A. A. Kolodziejczyk, D. Zheng and E. Elinav, Diet–microbiota interactions and personalized nutrition, *Nat. Rev. Microbiol.*, 2019, **17**, 742–753.

4 G. R. Healey, R. Murphy, L. Brough, C. A. Butts and J. Coad, Interindividual variability in gut microbiota and host response to dietary interventions, *Nutr. Rev.*, 2017, **75**, 1059–1080.

5 A. Leshem, E. Segal and E. Elinav, The Gut Microbiome and Individual-Specific Responses to Diet, *mSystems*, 2020, **5**, 10–1128.

6 L. Dethlefsen and D. A. Relman, Incomplete recovery and individualized responses of the human distal gut microbiota to repeated antibiotic perturbation, *Proc. Natl. Acad. Sci. U. S. A.*, 2011, **108**, 4554–4561.

7 S. A. Smits, A. Marcobal, S. Higginbottom, J. L. Sonnenburg and P. C. Kashyap, Individualized Responses of Gut Microbiota to Dietary Intervention Modeled in Humanized Mice, *mSystems*, 2016, **1**, 10–128.

8 S. X. Mingat, T. Ehara, H. Nakamura and K. Miyaji, Comparative Study of Prebiotics for Infants Using a Fecal Culture System: Insights into Responders and Non-Responders, *Nutrients*, 2024, **16**, 3347.

9 G. Reid, E. Gaudier, F. Guarner, G. B. Huffnagle, J. M. Macklaim, A. M. Munoz, M. Martini, T. Ringel-Kulka, B. R. Sartor, R. R. Unal, K. Verbeke and J. Walter, Responders and non-responders to probiotic interventions, *Gut Microbes*, 2010, **1**, 200–204.

10 M. N. Ojima, K. Yoshida, M. Sakanaka, L. Jiang, T. Odamaki and T. Katayama, Ecological and molecular perspectives on responders and non-responders to probiotics and prebiotics, *Curr. Opin. Biotechnol*, 2022, **73**, 108–120.

11 T. Cantu-Jungles and B. Hamaker, New view on dietary fiber selection for predictable shifts in gut microbiota, *mBio*, 2020, **11**, e02179-19.

12 T. Cantu-Jungles, N. Bulut, E. Chambry, A. Ruthes, M. Iacomini, A. Keshavarzian, T. A. Johnson and B. R. Hamaker, Dietary fiber hierarchical specificity: the missing link for predictable and strong shifts in gut bacterial communities, *mBio*, 2021, **12**, e0102821.

13 M. Xiang, L. Qiao, Q. Han, Y. Zha, X. Sui and Q. Wang, Effects of Supplementation With Different Specificities of Dietary Fiber on Health-Related Indicators in Adults With Overweight or Obesity: A Systematic Review and Meta-analysis of Randomized Controlled Trials, *Nutr. Rev.*, 2025, **83**, e1544–e1563.

14 C. Li and Y. Hu, New definition of resistant starch types from the gut microbiota perspectives – a review, *Crit. Rev. Food Sci. Nutr.*, 2023, **63**, 6412–6422.

15 P. Van den Abbeele, S. Deyaert, R. Albers, A. Baudot and A. Mercenier, Carrot RG-I Reduces Interindividual Differences between 24 Adults through Consistent Effects on Gut Microbiota Composition and Function Ex Vivo, *Nutrients*, 2023, **15**, 2090.

16 C. Yu, S. Ahmadi, X. Hu, J. Wu, D. Wu, Z. Hou, H. Pan, H. Xiao, X. Ye and S. Chen, Individual Difference in the Capacity of Gut Microbiota to Ferment Four Complex Carbohydrates from Normal to Overweight People: An In Vitro Study, *J. Agric. Food Chem.*, 2024, **72**, 20513–20526.

17 V. Agamennone, T. J. van den Broek, A. de Kat Angelino-Bart, F. P. M. Hoevenaars, J. W. van der Kamp and F. H. J. Schuren, Individual and Group-Based Effects of In Vitro Fiber Interventions on the Fecal Microbiota, *Microorganisms*, 2023, **11**, 2001.

18 C. Jian, N. Sorensen, R. Lutter, R. Albers, W. de Vos, A. Salonen and A. Mercenier, The impact of daily supplementation with rhamnogalacturonan-I on the gut microbiota in healthy adults: A randomized controlled trial, *Biomed. Pharmacother.*, 2024, **174**, 116561.

19 C. Li and Y. Hu, Align resistant starch structures from plant-based foods with human gut microbiome for personalized health promotion, *Crit. Rev. Food Sci. Nutr.*, 2023, **63**, 2509–2520.

20 X. Guo, X. Zhang, X. Ying, A. Ma, Z. Li, H. Liu and Q. Guo, Fermentation properties and prebiotic potential of different pectins and their corresponding enzymatic hydrolysates, *Food Hydrocolloids*, 2023, **143**, 108878.

21 S. Fan, Z. Zhang, Y. Zhao, M. Daglia, J. Zhang, Y. Zhu, J. Bai, L. Zhu and X. Xiao, Recent advances in targeted manipulation of the gut microbiome by prebiotics: from taxonomic composition to metabolic function, *Curr. Opin. Food Sci.*, 2023, **49**, 100959.

22 A. Mercenier, L. D. Vu, J. Poppe, R. Albers, S. McKay and P. Van den Abbeele, Carrot-Derived Rhamnogalacturonan-I Consistently Increases the Microbial Production of Health-Promoting Indole-3-Propionic Acid Ex Vivo, *Metabolites*, 2024, **14**, 722.

23 S. Fan, Z. Zhang, Q. Nie, M. Ackah and S. Nie, Rethinking the classification of non-digestible carbohydrates: Perspectives from the gut microbiome, *Compr. Rev. Food Sci. Food Saf.*, 2024, **23**, e70046.

24 V. Ratanpaul, R. Stanley, C. Brennan and R. Eri, Manipulating the kinetics and site of colonic fermentation with different fibre combinations – a review, *Int. J. Food Sci. Technol.*, 2023, **58**, 2216–2227.

25 D. Bajic, F. Wiens, E. Wintergerst, S. Deyaert, A. Baudot and P. Van den Abbeele, HMOs Impact the Gut Microbiome of Children and Adults Starting from Low Predicted Daily Doses, *Metabolites*, 2024, **14**, 239.

26 H. J. Flint, K. P. Scott, S. H. Duncan, P. Louis and E. Forano, Microbial degradation of complex carbohydrates in the gut, *Gut Microbes*, 2012, **3**, 289–306.
27 J. F. Wardman, R. K. Bains, P. Rahfeld and S. G. Withers, Carbohydrate-active enzymes (CAZymes) in the gut microbiome, *Nat. Rev. Microbiol.*, 2022, **20**, 542–556.
28 T. M. Cantu-Jungles, A. C. Ruthes, M. El-Hindawy, R. B. Moreno, X. Zhang, L. M. C. Cordeiro, B. R. Hamaker and M. Iacomini, In vitro fermentation of Cookeina speciosa glucans stimulates the growth of the butyrogenic Clostridium cluster XIVa in a targeted way, *Carbohydr. Polym.*, 2018, **183**, 219–229.
29 Y. Román, H. P. de Oliveira Barddal, M. Iacomini, G. L. Sassaki and T. R. Cipriani, Anticoagulant and antithrombotic effects of chemically sulfated fucogalactan and citrus pectin, *Carbohydr. Polym.*, 2017, **174**, 731–739.
30 V. Lebet, E. Arrigoni and R. Amadò, Measurement of fermentation products and substrate disappearance during incubation of dietary fibre sources with human faecal flora, *LWT–Food Sci. Technol.*, 1998, **31**, 473–479.
31 E. Bolyen, J. R. Rideout, M. R. Dillon, N. A. Bokulich, C. C. Abnet, G. A. Al-Ghalith, H. Alexander, E. J. Alm, M. Arumugam, F. Asnicar, Y. Bai, J. E. Bisanz, K. Bittinger, A. Brejnrod, C. J. Brislawn, C. T. Brown, B. J. Callahan, A. M. Caraballo-Rodríguez, J. Chase, E. K. Cope, R. Da Silva, C. Diener, P. C. Dorrestein, G. M. Douglas, D. M. Durall, C. Duvallet, C. F. Edwardson, M. Ernst, M. Estaki, J. Fouquier, J. M. Gauglitz, S. M. Gibbons, D. L. Gibson, A. Gonzalez, K. Gorlick, J. Guo, B. Hillmann, S. Holmes, H. Holste, C. Huttenhower, G. A. Huttley, S. Janssen, A. K. Jarmusch, L. Jiang, B. D. Kaehler, K. Bin Kang, C. R. Keefe, P. Keim, S. T. Kelley, D. Knights, I. Koester, T. Kosciolek, J. Kreps, M. G. I. Langille, J. Lee, R. Ley, Y. X. Liu, E. Loftfield, C. Lozupone, M. Maher, C. Marotz, B. D. Martin, D. McDonald, L. J. McIver, A. V. Melnik, J. L. Metcalf, S. C. Morgan, J. T. Morton, A. T. Naimey, J. A. Navas-Molina, L. F. Nothias, S. B. Orchanian, T. Pearson, S. L. Peoples, D. Petras, M. L. Preuss, E. Pruesse, L. B. Rasmussen, A. Rivers, M. S. Robeson, P. Rosenthal, N. Segata, M. Shaffer, A. Shiffer, R. Sinha, S. J. Song, J. R. Spear, A. D. Swafford, L. R. Thompson, P. J. Torres, P. Trinh, A. Tripathi, P. J. Turnbaugh, S. Ul-Hasan, J. J. J. van der Hooft, F. Vargas, Y. Vázquez-Baeza, E. Vogtmann, M. von Hippel, W. Walters, Y. Wan, M. Wang, J. Warren, K. C. Weber, C. H. D. Williamson, A. D. Willis, Z. Z. Xu, J. R. Zaneveld, Y. Zhang, Q. Zhu, R. Knight and J. G. Caporaso, Reproducible, interactive, scalable and extensible microbiome data science using QIIME 2, *Nat. Biotechnol.*, 2019, **37**, 852–857.
32 B. J. Callahan, P. J. McMurdie, M. J. Rosen, A. W. Han, A. J. A. Johnson and S. P. Holmes, DADA2: High-resolution sample inference from Illumina amplicon data, *Nat. Methods*, 2016, **13**, 581–583.
33 T. M. Cantu-Jungles and B. R. Hamaker, Tuning Expectations to Reality: Don't Expect Increased Gut Microbiota Diversity with Dietary Fiber, *J. Nutr.*, 2023, **153**, 3156–3163.
34 B. R. Hamaker and Y. E. Tuncil, A perspective on the complexity of dietary fiber structures and their potential effect on the gut microbiota, *J. Mol. Biol.*, 2014, **426**, 3838–3850.
35 S. P. Nie, C. Wang, S. W. Cui, Q. Wang, M. Y. Xie and G. O. Phillips, The core carbohydrate structure of Acacia seyal var. seyal (Gum arabic), *Food Hydrocolloids*, 2013, **32**, 221–227.
36 M. H. Rawi, H. Y. Tan and S. R. Sarbini, Identification of acacia gum fermenting bacteria from pooled human feces using anaerobic enrichment culture, *Front. Microbiol.*, 2023, **14**, 1245042.
37 I. Amir, P. Bouvet, C. Legeay, U. Gophna and A. Weinberger, Eisenbergiella tayi gen. nov., sp. nov., isolated from human blood, *Int. J. Syst. Evol. Microbiol.*, 2014, **64**, 907–914.
38 X. Wu, N. Xu, Z. Ye, Q. Zhao, J. Liu, J. Li, M. Wu, Y. Zheng, X. Li, W. Li, T. Zhang, X. Hu and Q. Zhang, Polysaccharide from Scutellaria barbata D. Don attenuates inflammatory response and microbial dysbiosis in ulcerative colitis mice, *Int. J. Biol. Macromol.*, 2022, **206**, 1–9.
39 L. R. Lopetuso, F. Scaldaferri, V. Petito and A. Gasbarrini, Commensal Clostridia: leading players in the maintenance of gut homeostasis, *Gut Pathog.*, 2013, **5**, 23.
40 Y. C. Heng and S. Kittelmann, Proposal for reclassification of the species Hungatella xylanolytica as Lacrimispora xylanisolvens nom. nov. and transfer of the genus Hungatella to the family Lachnospiraceae, *Int. J. Syst. Evol. Microbiol.*, 2024, **74**, 006417.
41 L. M. Costa, M. M. Mendes, A. C. Oliveira, K. G. Magalhães, N. Shivappa, J. R. Hebert, T. H. M. da Costa and P. B. Botelho, Dietary inflammatory index and its relationship with gut microbiota in individuals with intestinal constipation: a cross-sectional study, *Eur. J. Nutr.*, 2022, **61**, 341–355.
42 F. Li, C. Yu, Q. Zhao, Z. Wang, Z. Wang, Y. Chang, Z. Xu, X. Han, H. Li, Y. Liu, S. Hu, S. Chang, T. Tang and Y. Li, Exploring the intestinal ecosystem: from gut microbiota to associations with subtypes of inflammatory bowel disease, *Front. Cell. Infect. Microbiol.*, 2023, **13**, 1304858.
43 S. Chen, Z. Zhang, S. Liu, T. Chen, Z. Lu, W. Zhao, X. Mou and S. Liu, Consistent signatures in the human gut microbiome of longevous populations, *Gut Microbes*, 2024, **16**(1), 2393756.
44 B. Jiao, Z. Ouyang, Q. Liu, T. Xu, M. Wan, G. Ma, L. Zhou, J. Guo, J. Wang, B. Tang, Z. Zhao and L. Shen, Integrated analysis of gut metabolome, microbiome, and brain function reveal the role of gut-brain axis in longevity, *Gut Microbes*, 2024, **16**(1), 2331434.
45 P. A. Lawson, Y. Song, C. Liu, D. R. Molitoris, M. L. Vaisanen, M. D. Collins and S. M. Finegold, Anaerotruncus colihominis gen. nov., sp. nov., from human faeces, *Int. J. Syst. Evol. Microbiol.*, 2004, **54**, 413–417.

 

46 T. Grenda, A. Grenda, P. Domaradzki, P. Krawczyk and K. Kwiatek, Probiotic Potential of Clostridium spp.—Advantages and Doubts, *Curr. Issues Mol. Biol.*, 2022, **44**, 3118.

47 Y. F. Tang, W. Y. Xie, H. Y. Wu, H. X. Guo, F. H. Wei, W. Z. Ren, W. Gao and B. Yuan, Huaier Polysaccharide Alleviates Dextran Sulphate Sodium Salt-Induced Colitis by Inhibiting Inflammation and Oxidative Stress, Maintaining the Intestinal Barrier, and Modulating Gut Microbiota, *Nutrients*, 2024, **16**, 1368.

48 N. Wang, W. Chen, C. Cui, Y. Zheng, Q. Yu, H. Ren, Z. Liu, C. Xu and G. Zhang, The Peanut Skin Procyanidins Attenuate DSS-Induced Ulcerative Colitis in C57BL/6 Mice, *Antioxidants*, 2022, **11**, 2098.

49 H. M. Xu, H. L. Huang, Y. Di Liu, J. Q. Zhu, Y. L. Zhou, H. T. Chen, J. Xu, H. L. Zhao, X. Guo, W. Shi, Y. Q. Nie and Y. J. Zhou, Selection strategy of dextran sulfate sodium-induced acute or chronic colitis mouse models based on gut microbial profile, *BMC Microbiol.*, 2021, **21**, 1–14.

50 Y. Lv, X. Qin, H. Jia, S. Chen, W. Sun and X. Wang, The association between gut microbiota composition and BMI in Chinese male college students, as analysed by next-generation sequencing, *Br. J. Nutr.*, 2019, **122**, 986–995.

51 D. Mudgil, S. Barak and B. S. Khatkar, Guar gum: processing, properties and food applications—A review, *J. Food Sci. Technol.*, 2014, **51**, 409.

52 S. Gupta and P. S. Variyar, Guar Gum: A Versatile Polymer for the Food Industry, in *Biopolymers for Food Design*, 2018, pp. 383–407.

53 M. Petitjean and J. R. Isasi, Locust Bean Gum, a Vegetable Hydrocolloid with Industrial and Biopharmaceutical Applications, *Molecules*, 2022, **27**, 8265.

54 T. R. Wu, C. S. Lin, C. J. Chang, T. L. Lin, J. Martel, Y. F. Ko, D. M. Ojcius, C. C. Lu, J. D. Young and H. C. Lai, Gut commensal Parabacteroides goldsteinii plays a predominant role in the anti-obesity effects of polysaccharides isolated from Hirsutella sinensis, *Gut*, 2019, **68**, 248–262.

55 K. Wang, M. Liao, N. Zhou, L. Bao, K. Ma, Z. Zheng, Y. Wang, C. Liu, W. Wang, J. Wang, S. J. Liu and H. Liu, Parabacteroides distasonis Alleviates Obesity and Metabolic Dysfunctions via Production of Succinate and Secondary Bile Acids, *Cell Rep.*, 2019, **26**, 222–235.

56 W. Wei, C. C. Wong, Z. Jia, W. Liu, C. Liu, F. Ji, Y. Pan, F. Wang, G. Wang, L. Zhao, E. S. H. Chu, X. Zhang, J. J. Y. Sung and J. Yu, Parabacteroides distasonis uses dietary inulin to suppress NASH via its metabolite pentadecanoic acid, *Nat. Microbiol.*, 2023, **8**, 1534–1548.

57 Q. Zhao, M. Y. Dai, R. Y. Huang, J. Y. Duan, T. Zhang, W. M. Bao, J. Y. Zhang, S. Q. Gui, S. M. Xia, C. T. Dai, Y. M. Tang, F. J. Gonzalez and F. Li, Parabacteroides distasonis ameliorates hepatic fibrosis potentially via modulating intestinal bile acid metabolism and hepatocyte pyroptosis in male mice, *Nat. Commun.*, 2023, **14**, 1–18.

58 Y. Sun, Q. Nie, S. Zhang, H. He, S. Zuo, C. Chen, J. Yang, H. Chen, J. Hu, S. Li, J. Cheng, B. Zhang, Z. Zheng, S. Pan, P. Huang, L. Lian and S. Nie, Parabacteroides distasonis ameliorates insulin resistance via activation of intestinal GPR109a, *Nat. Commun.*, 2023, **14**, 1–16.

59 R. López-Mondéjar, V. Tláskal, U. N. da Rocha and P. Baldrian, Global Distribution of Carbohydrate Utilization Potential in the Prokaryotic Tree of Life, *mSystems*, 2022, **7**(6), e00829-22.

60 S. Miquel, R. Martín, O. Rossi, L. G. Bermúdez-Humarán, J. M. Chatel, H. Sokol, M. Thomas, J. M. Wells and P. Langella, Faecalibacterium prausnitzii and human intestinal health, *Curr. Opin. Microbiol.*, 2013, **16**, 255–261.

61 H. E. Leylabadlo, R. Ghotaslou, M. M. Feizabadi, S. Farajnia, S. Y. Moaddab, K. Ganbarov, E. Khodadadi, A. Tanomand, E. Sheykhsaran, B. Yousefi and H. S. Kafil, The critical role of Faecalibacterium prausnitzii in human health: An overview, *Microb. Pathog.*, 2020, **149**, 104344.

62 X. He, S. Zhao and Y. Li, Faecalibacterium prausnitzii: A Next-Generation Probiotic in Gut Disease Improvement, *Can. J. Infect. Dis. Med. Microbiol.*, 2021, 6666114.