**Supplementary material:**

**Devin J. Rose**

**Impact of whole grains on the gut microbiota: the next frontier for oats?**

**Web Table 1:** Carbohydrate composition of selected whole grains (% dry basis)

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Component | Barley | Oats | Rice | Rye | Sorghum | Wheat |
| Sugars\* | 1-3 | 1-3 | 0.7-1.3 | 1.7-3.0 | 1-4 | 1-2 |
| α-Galactosyl derivatives of sucrose† | 0.16-0.56 | 0.34 | 0.75-1.0 |  | 0.14-0.60 | 0.19-0.68 |
| β-Fructosyl derivatives of sucrose | 0.3-0.8 | 0.6-1 | 0.09 | 2.4-3.1 |  | 0.5-2.5 |
| Starch | 63-65 | 46-60 | 66-77 | 55-61 | 55.6-75.2 | 71-75 |
| Arabinoxylan‡ | 4-7 (14-20) | 2.0-4.5 (10-20) | 1.2-2.4 (2-6) | 6.5-12 (30-41) | 2.5-5.6 (16-25) | 5.8-6.5 (5-12) |
| β-Glucan‡ | 2.0-11 (65-69) | 3-8 (82-88) | 0.11-0.13 (0-20) | 1.5-2.0 (20-40) | 0.6-1.4 | 0.55-1.0 (7-40) |
| Cellulose | 1-5 | 0.6-8 | 0.8 | 1.3-2.6 | 2.7 | 1.7-2.8 |
| References | (1-6) | (1,2,4,7) | (1,2,8) | (1,2,9,10) | (1,11,12) | (1,2,13,14) |

\* Glucose + fructose + sucrose.

† Raffinose + stachyose + verbascose.

‡ Parenthetic numbers refer to the percentage of the corresponding polymer that is soluble in hot water.

**Web Table 2:** *In vivo* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Test group | Treatments | Microbial analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| *Human trials - Oat bran* | |  |  |  |  |  |
| Oat bran | 23 patients with quiescent UC | 60 g oat bran products (bread, drink) per d for 12 weeks | None | Not reported | Increased fecal butyrate; decreased GI symptoms | (15) |
| Oat milk (fermented) | Healthy humans (n=18 or 20 per group) | 200 ml, 3x daily of yogurt or "yogurt" made with oat milk fermented with Lactobacillus delbrueckii subsp bulgaricus and Streptococcus salivarius subsp thermophilus or Pediococcus damnosus 2.6 (produces a b-glucan-like exopolysaccharide) | FISH: Bifidobacteria (Bif164) and total bacteria (Bact338) | 0.4 log increase in Bifidobacteria on oat-based yogurt fermented with P. damnosus only | Not reported | (16) |
| *Human trials - β-Glucan* | |  |  |  |  |  |
| Barley β-glucan | 52 Healthy humans ages 39-70 y | Randomly assigned to receive cake containing 0.75 g β-glucan or no β-glucan for 30 d | Culture: Aerobes, Coliforms, E. coli, Enterococcus, Anaerobes, Bacteroides, Lactobacillus, Clostridium perfringens, Bifidobacterium | In subjects >50 y only (who also had reduced Bifidobacteria at baseline) significant increase in Bifidobacteria (to normal levels) and Bacteroides | Not reported; no gastrointestinal side effects in subjects >50 y | (17) |
| Barley β-glucan | 20 humans that had undergone a polypectomy | Bread containing 3 g β-glucan/d for 90 d | Culture: Aerobes, Coliforms, E. coli, Enterococcus, Anaerobes, Bacteroides, Lactobacillus, Clostridium perfringens, Bifidobacterium | No effects on bacterial counts between control and treatment groups; some significant changes within group, but no trends over time | β-Glucan group showed reduced butyrate production at baseline and throughout the experiment | (18) |

**Web Table 2 (cont.):** *In vivo* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Test group | Treatments | Microbial analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| *Animal trials - Whole grain oats/oat bran* | | |  |  |  |  |
| Oat bran | Wistar rats (n=12-13/group) | Modified AIN-93G; contained 100g/kg rye, wheat, or oat bran, inulin, or no fiber (control) | None | Not reported | Total SCFA and butyrate were higher in the inulin and rye-bran groups compared to non-fiber and wheat-bran groups; SCFA profile in the oat-bran group did not differ significantly from SCFA profile in the other groups. | (19) |
| Two oat varieties (one normal and one high β-glucan); common hulled barley, and four hulless barley varieties | Pigs (n=8/group) | Diets containing barley and oats ranging from 41-84 g/kg bβ-glucan | DGGE, qPCR | Higher b-glucan was associated with higher numbers of butyrate-producing bacteria | Not reported | (20) |
| Oat flour & bran | Wistar rats (n=10/group) | Diets contained oat flour, oat flour/resistant starch, oat bran, oat bran/resistant starch, or autclaved oat bran (4-128 g/kg resistant starch, 30-92 g/kg b-glucan, and 122-304 g/kg dietary fiber) | Culture: Aerobes, anaerobes, coliforms, Bacteroides, Lactobacilli, Bifidobacteria | Aerobes and Bifidobacteria increased by about 0.5 log on all treatments except oat flour/resistant starch; coliforms decreased in all treatments except autoclaved flour | Treatments reduced caecal and colonic pH and increased SCFA; total SCFA highest in bran; butyrate highest in bran/resistant starch; all treatments significantly increase bile acid excretion | (21) |

**Web Table 2 (cont.):** *In vivo* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Test group | Treatments | Microbial analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| Oat bran | BioBreeding control rats | Control, 3g dietary fiber from oat bran+2 g cellulose, or 5g dietary fiber from wheat bran | DGGE, qPCR | Major change occurred within Clostridium coccoides group and C. leptum subgroup; few differences between oat and wheat | Not reported | (22) |
| Oat flour or wheat flour containing equivalent β-glucan | Pigs (n=8/group) | Oat-based diet; wheat based diet+added β-glucan (from oat) | Culture: Bifidobacteria, lactobacilli, E. coli | Higher Bifidobacteria and lacotbacilli on oat-diet compared with wheat+β-glucan diet | More total SCFA in oat-based diet; less butyrate on oat-diet compared with wheat+β-glucan diet | (23) |
| Oat flour or wheat flour containing equivalent β-glucan | Pigs (n=8/group) | Oat-based diet; wheat based diet+added β-glucan (from oat); both supplied with or without enzyme (β-glucanase and xylanase) | Culture: Bifidobacteria, lactobacilli, E. coli | Higher Bifidobacteria, lacotbacilli, and E. coli on oat-diet compared with wheat+β-glucan diet | More total SCFA in oat-based diet compared with wheat+β-glucan diet | (24) |
| Oat or barley-based diet | Pigs (n=8/group) | Oat or barley-based diet (oat diet contained less total (20 vs. 26 g/kg) and soluble (6 vs 18 g/kg) β-glucan than barley diet | Culture: Bifidobacteria, lactobacilli, E. coli | Oats showed higher Bifidobacteria, Lactobacilli, and E. coli compared to barley-based diet | Not reported | (25) |
| Barley flour, oat flour, cellulose, or barley β-glucans of high (HV) or low viscosity (LV) | BioBreeding rats (n=6/ group) | Diets contained treatment fibers at 8% of total dietary fiber (diets contained about 8% total dietary fiber) | DGGE, FISH: Lactobacilli (Lab158, Lba 23S) | HV barley β-glucan increased Lactobacilli | Not reported | (26) |

**Web Table 2 (cont.):** *In vivo* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Test group | Treatments | Microbial analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| *Animal trials - β-Glucan* | |  |  |  |  |  |
| Oat β-Glucan | C57BL/6 mice | Intraperitoneal administration of oat b-glucan | Fluorescent labeling | Oral or parenteral administration of oat β-glucan enhanced resistance to S. aureus or E. vermiformis infection | Not reported | (27) |
| Oat β-Glucan | Pigs (n=8/group) | Diets containing low or high Ca/P supplemented with 8.95% oat b-glucan | q-PCR: Lactobacillus, Enterococcus, Streptococcus, Bifidobacterium, clostridium cluster I, clostridium cluster IV, clostridium cluster XIVa, Bacteroides-Prevotella-Porphyromonas, Enterobacteriaceae | b-glucan increased Bifidobacteria and Lactobacilli and decreased clostridium cluster XIVa in colon | β-glucan increased total SCFA, propoinate, butyrate, and BCFA | (28) |
| Microcrystalline cellulose, carboxymethylcellulose or oat β-glucans of high (HV) or low viscosity (LV) | Duroc-Landrace pigs (n=8) crossover design | Cornstarch/casein-based diets supplemented with 5% of treatment dietary fiber | q-PCR: Lactobacillus, Enterococcus, Streptococcus, Bifidobacterium, clostridium cluster I, clostridium cluster IV, clostridium cluster XIVa, Bacteroides-Prevotella-Porphyromonas, Enterobacteriaceae | No differences in Bifidobacteria or Lactobacilli among cellulose and β-glucans; within β-glucans, HV resulted in higher Enterococcus and clostridial cluster I compared with LV | No differences in SCFA between β-glucans of different molecular weights; β-glucans resulted in higher BCFA production than either cellulose | (29) |

**Web Table 3:** *In vitro* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Microbial inoculum | Treatments | Microbiota analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| *Whole grain oats/oat bran* | |  |  |  |  |  |
| Thin/thick oat flakes | Fecal microbiota from 3 healthy individuals in United Kingdom (not specified whether fecal samples were pooled or not) | In vitro batch fermentation with pH control (6.7-6.8); in vitro digestion performed prior to fermentation | FISH: Bifidobacterium genus (Bif164), Lactobacillus-Enterococcus group (Lab158), Bacteroides-Prevotella group (Bac303), Clostridium histolyticum subgroup (His 150), Ruminococcus-Eubacterium-Clostridium cluster (Erec482), Atopobium cluster including most Coriobacteriaceae species (Ato 291) | Thick oat flakes resulted in about 0.5 log increase in Bifidobacteria (same as oligofructose); no significant changes on thin oat flakes; authors suggest higher resistant starch in thick oat flakes was responsible for the bifidogenic effect | Thick oat flakes resulted in high propionate and butyrate proportion; thin oat flakes resulted in high propionate proportion; authors suggest high butyrate production in thick flakes was due to high resistant starch | (30) |
| Oat flour, bran, and fractions thereof | Fecal microbiota from 2 healthy individuals in Spain (not specified whether fecal samples were pooled or not) | In vitro batch fermentation without pH control (starting pH 7.3); no in vitro digestion prior to fermentation | Culture: lactobacilli (MRS), bifidobacteria (BIM25), clostridia (SPS), anaerobes (Wilkins Chalgren), Enterobacteria (MacConkey) | >2-fold increase in bifidobacteria; >1 log increase in lactobacilli; oat bran showed a higher "prebiotic index" than oat flour, but lower than oligofructose | Propionate and butyrate production similar to olgiofructose on both oat substrates | (31) |
| Three oat lines containing 5.2-7.7% β-glucan | Fecal microbiota from 3 healthy individuals in US (not pooled) | In vitro batch fermentation without pH control (starting pH about 6.4); in vitro digestion performed prior to fermentation | None | Not reported | Propionate higher and acetate lower than lactulose (positive control) | (32) |

**Web Table 3 (cont.):** *In vitro* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Microbial inoculum | Treatments | Microbiota analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| Two oat lines containing 4.7 and 5.3% β-glucan and a purified oat β-glucan | Fecal microbiota from 2 healthy individuals in US (pooled | In vitro batch fermentation without pH control (starting pH about 6.8); in vitro digestion performed prior to fermentation | None | Not reported | >80% of total carbohydrate was depleted in 24 h for oat lines; all of the β-glucan was depleted within 8 h; purified β-glucan resulted in higher propionate:acetate and butyrate:acetate ratios than oat lines | (33) |
| Oat bran, wheat bran, β-glucan | Fecal microbiota from 3 healthy individuals (pooled) | In vitro batch fermentation without pH control (starting pH about 6.7); in vitro digestion performed prior to fermentation for brans only | None | Not reported | All soluble carbohydrate fermented after 4 h of fermentation; purified β-glucan resulted in the highest SCFA production and higher butyrate and equivalent propionate production compared with oat bran | (34) |
| Oat bran, wheat bran, rye bran | Pooled fecal microbiota from 3 healthy individuals in Finland | In vitro batch fermentation without pH control (starting pH about 7.2); in vitro digestion performed prior to fermentation | None | Not reported | Oat bran was more completely fermented with more SCFA than other brans; all brans showed high proportion of propionate and low butyrate compared to inulin | (35) |
| Oat flour, bran, and fractions thereof | Lactobacillus plantarum, L. reuteri, L. acidophilus | In vitro batch fermentation | Optical density | β-Glucan was fermented by Lactobacilli; greatest cell growth was L. plantarum | Not reported | (36) |

**Web Table 3 (cont.):** *In vitro* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Microbial inoculum | Treatments | Microbiota analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| *β-Glucan* |  |  |  |  |  |  |
| Oat β-glucan hydrolysate | 2 *Lactobacillus* strains, 3 *Bifidobacterium* strains, *Clostridium* *diffcile*, *E. coli* | In vitro batch fermentation | Disappearance of carbohydrate | β-Glucan hydrolysates were used by most Bifidobacteria and Lactobicilli, but not nearly as well as fructans | Not reported | (37) |
| Oat β-glucan, flaxseed gum, and fenugreek gum | Pig cecal and colonic microbiota pooled from 4 pigs in Canada | In vitro batch fermentation with subculturing | PCR-DGGE with identification of 31 bands | β-Glucan exhibited the least selective pressure on the gut community | β-Glucan resulted in enhanced lactate and propionate and suppressed acetate and butyrate production comapared with other gums | (38) |
| β-Glucan from barley and oats and hydrolysates thereof | Fecal microbiota from 3 healthy individuals in United Kingdom (not pooled) | In vitro batch fermentation with pH control (6.8); β-glucan hydrolysates with 3-4 monomer units were not pH controlled | FISH: Bifidobacterium genus (Bif164), Lactobacillus-Enterococcus group (Lab158), Bacteroides-Prevotella group (Bac303), Clostridium histolyticum subgroup (His 150), Ruminococcus-Eubacterium-Clostridium cluster (Erec482), Atopobium cluster including most Coriobacteriaceae species (Ato 291), clostridial cluster IX (Prop853) | Significant increases in Bacteroides-Prevotella group and C. histolyicum subgroup on most β-glucan fractions; β-glucan hydrolysates with 3-4 monomer units significantly increase Bifidobacteria and Lactobacilli but only about half as much as inulin | All β-glucans showed greater propionate and butyrate production (with proportionally less acetate) than inulin | (39) |

**Web Table 3 (cont.):** *In vitro* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Microbial inoculum | Treatments | Microbiota analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| β-Glucan from barley | 17 *Lactobacillus* strains, 18 *Bifidobacterium* strains, 3 *Enterococcus* strains, 8 *Bacteroides* strains, 1 *Clostridium* strain, 3 *E. coli* strains (originated from Finland) | In vitro batch fermentation | Optical density | β-Glucan was fermented by Bacteroides and Clostridium beijerinckii but not by lactobacilli, bifidobacteria, enterococci, or E. coli | Not reported | (40) |
| Barley β-glucan, inulin, psyllium, alkali-soluble arabinoxylan (AS-AX), resistant potato starch (RS3), starch-entrapped microspheres | Pooled fecal microbiota from 3 healthy individuals in US | In vitro batch fermentation without pH control (starting pH about 6.8) | None | Not reported | β-Glucan resulted in among the highest butyrate (same as RS3) and total SCFA (same as RS3 and AS-AX) production | (41) |
| Barley β-glucan, wheat arabinoxylan of different molecular weights | Pooled pig feces from 5 pigs consuming a diet low in dietary fiber (starch and fish meal) in Australia | In vitro batch fermentation | None | Not reported | β-Glucan and AX resulted in similar SCFA profiles (enriched in propionate; reduced butyrate); molecular weight did not affect fermentation of soluble polysaccharides | (42) |

**Web Table 3 (cont.):** *In vitro* trials on changes in the colonic environment that may affect symptoms related to gut health with oats, oat products, and β-glucan.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Samples | Microbial inoculum | Treatments | Microbiota analysis method | Effects on bacteria | Effects on microbial metabolites | Reference |
| β-Glucan from barley, mushroom, bacteria, seaweed | *Bifidobacterium infantis, B. longum, and B. adolescentis* (originated from China) | In vitro batch fermentation (starting pH 6.5) | Optical density | Most β-glucans increased cell numbers by >1 log; increase in bifidobacteria comparable to inulun; B. infantis showed greatest increase | B. infantis showed higher proportion of propionate and butyrate on β-glucan than on inulin; other bifidobacteria showed similar SCFA production to inulin | (43) |
| β-Glucan from oats, guar gum, inulin | Fecal microbiota from 3 healthy individuals in US (pooled) | In vitro batch fermentation without pH control | None | Not reported | β-Glucan resulted in similar SCFA production to inulin, but more propionate and less butyrate | (44) |

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