

1 SUPPLEMENTARY MATERIAL

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3 **Supplementary Table 1.** Energy densities and nutritional composition of experimental diets [%
4 wt:wt and % Kcal] according to analytical measures and the manufacturer's data sheet of AIN-
5 93G purified standard. SD: standard diet, RD: rice diet, WGB: whole-grain barley, PG: pearled
6 grain, BB: barley bran.

	SD	RD	WGB	BB	PG	
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8	Protein (g/100g)	17.7	15.4	16.7	17.8	16.7
9	Total carbohydrate (g/100g)	60.1	64.6	66.3*	60.1*	63.2*
10	Total fat (g/100g)	7.2	7.3	5.8	7.2	6.1
11	Kcal/g	3.8	3.8	3.8	3.6	3.0
12	Ash (%)	2.7	3.9	2.4	2.8	2.4
13	Beta-glucans (g/100g)	0	0.1	1.3	0.1	1.2
13	Arabinoxylans (g/100g)	0	0	1.6	1.1	0.8

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15 The moisture content was evaluated using the Moisture Tester MT-CA (Brabender GmbH&Co KG, Duisburg,
16 Germany). The ash content was determined by incineration (ISO 2171:2023). The Dumas combustion method
17 was used to quantify the nitrogen contents using a Nitrogen/Protein Analyzer (CN628, LECO Corporation, St.
18 Joseph, MI, U.S.A.) following AACC Method 46–30.01. The nitrogen contents were converted to crude protein
19 by multiplying with a conversion factor of 5.88 (ISO 16634-1: 2008). Soxhlet method was applied for the
20 determination of total fat, adapted by the Randall extraction method (ISO 11085:2015). Briefly, 3 g sample
21 were extracted with hexane in the Det-Gras device (JP SELECTA, Spain), the solvent was evaporated, the
22 extracted fat was dried, and the % fat was obtained by gravimetry.

23 The β -glucan content was determined using mixed-linkage β -glucan assay (K-BGLU) kit from Megazyme
24 (Wicklow, Ireland). Samples (80–120 mg) were treated with ethanol (50% v/v) and sodium phosphate buffer
25 (20 mmol/L, pH 6.5). After heating, enzymatic hydrolysis was carried out sequentially with lichenase and β -
26 glucosidase, followed by incubation with sodium acetate buffer (200 mM, pH 4.0). The samples were
27 centrifuged, and aliquots were incubated with GOPOD reagent. Absorbance was measured at 510 nm with a
28 reagent blank serving as the reference. Arabinoxylan content was quantified using the D-xylose assay kit (K-
29 XYLOSE) from Megazyme (Wicklow, Ireland). Briefly, samples (~100 mg) were hydrolyzed with 1.3 M HCl
30 at 100 °C for 1 hour, with intermittent stirring. After cooling, samples were neutralized with 1.3 M NaOH,
31 diluted to 100 mL with distilled water, thoroughly mixed, and centrifuged at 1,500 g for 10 minutes. For the
32 microplate assay, aliquots were combined with buffer, NAD⁺/ATP, hexokinase, and XDH/XMR enzymes in a
33 96-well plate. Absorbance was measured at 340 nm using a Multiscan GO spectrophotometer before and after
34 the reaction. D-xylose content was quantified using a standard and arabinoxylan content was calculated by
35 applying the following formula: Arabinoxylan content (g/100 g) = (D-xylose content (g/100 g) × 100) / (D-
36 xylose content in the polymer (g/100 g)).*Total Carbohydrate by calculation = (100-Ash%-total fat%-
37 protein%-moisture%).

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40 **Supplementary Table 2.** Daily dosage of (poly)phenols ($\mu\text{g}/\text{day}$) through the experimental
 41 diets. Standard diet (SD), rice diet (RD), whole-grain barley (WGB), barley bran diet (BB),
 42 pearled barley diet (PG) analyzed by UPLC-MS/MS.

(Poly)phenolic compound	SD	RD	WGB	BB	PG
FREE (POLY)PHENOLS					
Cyanidin-3- <i>O</i> -arabinoside	n.d.	n.d.	0.07 ± 0.01	0.10 ± 0.00	0.03 ± 0.01
Cyanidin-3- <i>O</i> -glucoside	n.d.	n.d.	48.7 ± 2.73	70.9 ± 1.71	23.0 ± 1.07
Cyanidin-3- <i>O</i> -malonylglucoside	n.d.	n.d.	221 ± 11.6	230 ± 3.06	91.0 ± 2.44
Cyanidin-3- <i>O</i> -dimalonylglucoside	n.d.	n.d.	296 ± 18.0	247 ± 4.34	106 ± 4.66
Pelargonidin-3- <i>O</i> -glucoside	n.d.	n.d.	1.52 ± 0.45	1.89 ± 0.16	0.81 ± 0.11
Pelargonidin-3- <i>O</i> -malonylglucoside	n.d.	n.d.	18.9 ± 2.59	18.4 ± 0.42	6.06 ± 0.74
Peonidin-3- <i>O</i> -glucoside	n.d.	n.d.	2.32 ± 0.04	3.05 ± 0.06	1.10 ± 0.22
Peonidin-3- <i>O</i> -malonylglucoside	n.d.	n.d.	19.0 ± 0.19	18.4 ± 0.61	5.62 ± 0.26
Peonidin-3- <i>O</i> -dimalonylglucoside	n.d.	n.d.	8.51 ± 0.05	6.70 ± 0.38	1.98 ± 0.14
Delphinidin-3- <i>O</i> -glucoside	n.d.	n.d.	0.27 ± 0.05	0.16 ± 0.01	1.19 ± 0.11
Malvidin-3- <i>O</i> -glucoside	n.d.	n.d.	n.d.	0.29 ± 0.38	n.d.
Total anthocyanins	n.d.	n.d.	616 ± 33.8	597 ± 9.05	236 ± 7.35
4-Hydroxybenzoic acid	n.d.	0.00 ± 0.00	1.86 ± 0.22	1.00 ± 0.06	1.06 ± 0.09
3, 4-Dihydroxybenzoic acid	n.d.	n.d.	1.51 ± 0.38	1.04 ± 0.11	0.55 ± 0.13
4-Hydroxy-3-methoxybenzoic acid (Vanillic acid)	n.d.	0.00 ± 0.00	1.27 ± 0.09	0.95 ± 0.07	0.96 ± 0.03
3,5-Dimethoxy-4-hydroxybenzoic acid (Syringic acid)	n.d.	n.d.	1.21 ± 0.24	0.29 ± 0.05	n.d.
4'-Hydroxycinnamic acid (<i>p</i> -Coumaric acid)	1.77 ± 0.44	n.d.	0.73 ± 0.07	0.43 ± 0.04	0.36 ± 0.05
3',4'-Dihydroxycinnamic acid (Caffeic acid)	n.d.	n.d.	0.53 ± 0.03	0.43 ± 0.10	n.d.
4'-Hydroxy-3'-methoxycinnamic acid (Ferulic acid)	n.d.	0.00 ± 0.00	4.72 ± 0.33	2.35 ± 0.18	2.19 ± 0.13
3'-Hydroxy-4'-methoxycinnamic acid (Isoferulic acid)	n.d.	0.00 ± 0.00	0.63 ± 0.06	0.29 ± 0.06	0.99 ± 0.18
Total phenolic acids	1.77 ± 0.44	0.00 ± 0.00	12.4 ± 0.02	6.77 ± 0.53	6.11 ± 0.23
Catechin	n.d.	n.d.	11.4 ± 2.99	3.38 ± 0.34	12.3 ± 1.32
Catechin glucoside	n.d.	n.d.	20.1 ± 0.88	14.3 ± 1.17	13.9 ± 2.31
Procyanidin B3	n.d.	n.d.	73.8 ± 1.23	56.2 ± 7.00	38.7 ± 5.04
GC-C/prodelphinidin B4	n.d.	n.d.	75.9 ± 11.1	44.6 ± 2.61	36.8 ± 6.37
Procyanidin diglucoside	n.d.	n.d.	3.94 ± 0.53	1.70 ± 0.25	5.18 ± 1.05
Procyanidin C2	n.d.	n.d.	n.d.	1.32 ± 0.07	n.d.
Prodelphinidin C2	n.d.	n.d.	n.d.	0.97 ± 0.16	n.d.
Total flavan-3-ols	n.d.	n.d.	185 ± 6.74	122 ± 9.63	107 ± 12.3
Apigenin- <i>O</i> -glucoside	n.d.	n.d.	1.39 ± 0.08	0.56 ± 0.07	n.d.
Apigenin-6- <i>C</i> -arabinoside-8- <i>C</i> -glucoside	n.d.	n.d.	6.35 ± 0.46	4.03 ± 0.20	2.71 ± 0.17
Isovitexin- <i>C</i> -glucoside	n.d.	n.d.	6.61 ± 0.23	4.13 ± 0.47	3.26 ± 0.38
Isovitexin- <i>C</i> -rutinoside	n.d.	n.d.	1.60 ± 0.19	2.07 ± 0.36	1.69 ± 0.29
Luteolin- <i>O</i> -glucoside	n.d.	n.d.	7.81 ± 0.86	5.65 ± 0.27	3.07 ± 0.19
Isorientin	n.d.	n.d.	2.66 ± 0.08	1.03 ± 0.04	1.18 ± 0.16
Isorientin- <i>C</i> -rutinoside	n.d.	n.d.	n.d.	0.36 ± 0.01	n.d.
Isoscoparin- <i>C</i> -glucoside	n.d.	n.d.	10.8 ± 0.88	3.05 ± 0.46	8.61 ± 0.37

Isoscoparin- <i>C</i> -rutinoside	n.d.	n.d.	2.68 ± 0.07	3.22 ± 0.26	1.93 ± 0.20
Total flavones	n.d.	n.d.	39.9 ± 0.32	24.1 ± 1.80	22.4 ± 0.93
TOTAL FREE (POLY)PHENOLS	1.77 ± 0.44	0.00 ± 0.00	853 ± 36.0	750 ± 9.61	372 ± 17.4

BOUND (POLY)PHENOLS

Cyanidin-3- <i>O</i> -glucoside	n.d.	n.d.	0.56 ± 0.15	0.43 ± 0.07	n.d.
Pelargonidin-3- <i>O</i> -glucoside	n.d.	n.d.	0.18 ± 0.04	0.25 ± 0.02	n.d.
Peonidin-3- <i>O</i> -glucoside	n.d.	n.d.	0.15 ± 0.03	0.23 ± 0.01	n.d.
Delphinidin-3- <i>O</i> -glucoside	n.d.	n.d.	0.01 ± 0.00	0.03 ± 0.00	n.d.
Malvidin-3- <i>O</i> -glucoside	n.d.	n.d.	0.01 ± 0.00	0.01 ± 0.00	n.d.
Total anthocyanins	n.d.	n.d.	0.90 ± 0.22	0.95 ± 0.08	n.d.
4-Hydroxybenzoic acid	0.86 ± 0.07	0.00 ± 0.00	7.51 ± 0.84	4.38 ± 0.22	4.30 ± 0.26
Hydroxybenzoic acid	1.54 ± 0.34	1.22 ± 0.00	1.44 ± 0.14	0.29 ± 0.01	0.93 ± 0.07
3,4-Dihydroxybenzoic acid	n.d.	n.d.	0.53 ± 0.03	2.21 ± 0.38	0.32 ± 0.02
4-Hydroxy-3-methoxybenzoic acid (Vanillic acid)	0.21 ± 0.05	0.00 ± 0.00	4.05 ± 0.42	2.66 ± 0.02	2.41 ± 0.07
3,5-Dimethoxy-4-hydroxybenzoic acid (Syringic acid)	n.d.	0.19 ± 0.00	1.63 ± 0.13	0.74 ± 0.06	0.90 ± 0.14
Cinnamic acid	n.d.	n.d.	0.99 ± 0.11	0.68 ± 0.02	0.50 ± 0.10
4'-Hydroxycinnamic acid (<i>p</i> -Coumaric acid)	0.56 ± 0.05	0.01 ± 0.00	11.0 ± 0.85	10.8 ± 0.73	6.70 ± 0.22
3',4'-Dihydroxycinnamic acid (Caffeic acid)	n.d.	n.d.	0.23 ± 0.02	0.44 ± 0.13	0.11 ± 0.01
3',4'-Dihydroxycinnamic acid- <i>O</i> -glucoside (Caffeic acid glucoside)	n.d.	n.d.	0.14 ± 0.05	0.03 ± 0.00	0.07 ± 0.01
4'-Hydroxy-3'-methoxycinnamic acid (Ferulic acid)	2.82 ± 0.19	0.08 ± 0.00	340 ± 23.5	158 ± 0.89	243 ± 8.89
3'-Hydroxy-4'-methoxycinnamic acid (Isoferulic acid)	1.16 ± 0.38	56.9 ± 0.00	56.9 ± 4.48	32.9 ± 1.00	71.4 ± 1.34
3,5-Dimethoxy-4-hydroxycinnamic acid (Sinapic acid)	n.d.	0.01 ± 0.00	26.1 ± 3.96	8.42 ± 0.14	17.4 ± 2.63
3,5-Dimethoxy-4-hydroxycinnamic acid- <i>O</i> -glucoside (Sinapic acid- <i>O</i> -glucoside)	n.d.	0.00 ± 0.00	9.01 ± 2.16	5.53 ± 0.29	4.14 ± 0.59
8/5-5'-Diferulic acid	n.d.	0.01 ± 0.00	113 ± 4.31	91.0 ± 5.01	71.4 ± 2.96
Diferulic acid decarboxylated	n.d.	0.00 ± 0.00	23.1 ± 1.72	15.2 ± 1.10	14.5 ± 1.07
Triferulic acid	n.d.	0.00 ± 0.00	15.1 ± 2.51	13.0 ± 2.36	7.00 ± 0.81
Total phenolic acids	7.14 ± 0.52	58.4 ± 0.00	611 ± 39.0	346 ± 8.75	445 ± 16.2
Apigenin- <i>O</i> -glucoside	n.d.	n.d.	0.40 ± 0.16	0.05 ± 0.02	0.13 ± 0.03
Apigenin-6- <i>C</i> -arabinoside-8- <i>C</i> -glucoside	n.d.	n.d.	0.56 ± 0.06	0.15 ± 0.03	0.12 ± 0.02
Isovitexin- <i>C</i> -glucoside	n.d.	n.d.	0.45 ± 0.06	0.30 ± 0.03	0.20 ± 0.02
Isovitexin- <i>C</i> -rutinoside	n.d.	n.d.	0.39 ± 0.07	0.23 ± 0.04	0.29 ± 0.03
Luteolin- <i>O</i> -glucoside	n.d.	n.d.	0.25 ± 0.05	0.34 ± 0.04	n.d.
Isoorientin	n.d.	n.d.	n.d.	0.06 ± 0.01	n.d.
Isoorientin- <i>C</i> -rutinoside	n.d.	n.d.	n.d.	0.04 ± 0.01	n.d.
Isoscoparin- <i>C</i> -glucoside	n.d.	n.d.	0.36 ± 0.16	0.36 ± 0.07	0.21 ± 0.03
Isoscoparin- <i>C</i> -rutinoside	n.d.	n.d.	n.d.	0.09 ± 0.03	n.d.
Total flavones	n.d.	n.d.	2.41 ± 0.19	1.61 ± 0.13	0.95 ± 0.06
TOTAL BOUND (POLY)PHENOLS	7.14 ± 0.52	58.4 ± 0.00	614 ± 38.9	348 ± 8.59	446 ± 16.2
TOTAL (POLY)PHENOLS (Free + Bound)	8.91 ± 0.63	58.4 ± 0.00	1467 ± 15.8	1098 ± 5.53	818 ± 32.9

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44 Free (poly)phenolic compounds were extracted by vortexing 100 mg of the sample with 5 mL of
45 methanol/water/formic acid (79/20/1, v/v/v) for 15 min, followed by centrifugation at 8784 g for 10 min. The
46 extraction was repeated, and the supernatants were combined, filtered through a 0.22 µm PVDF filter, and
47 stored for chromatographic analysis. Bound (poly)phenolic compounds were extracted from the residue after
48 free (poly)phenol extraction by alkaline hydrolysis with 4 mL of 2 M NaOH for 24 hours at room temperature.
49 After centrifugation (8784 g for 10 min), the supernatant was acidified with HCl to pH 2 and processed using
50 micro-elution solid-phase extraction (µSPE) with OASIS HLB cartridges. The cartridges were preconditioned
51 with methanol and acidified water, and the bound (poly)phenolic compounds were eluted with methanol.

52 Chromatographic analyses were conducted on an AcQuity Ultra-Performance™ liquid chromatography system
53 coupled with a tandem mass spectrometry (UPLC-MS/MS) detector (Waters, Milford, MA, USA). Two
54 methods were used: one for analyzing anthocyanins (ACNs) and their metabolites, and another for the
55 remaining (poly)phenolic compounds. The UPLC-MS/MS conditions were the same as those used in our
56 previous study [16]. Tandem MS analysis was performed using a triple quadrupole mass spectrometer (Waters,
57 Milford, MA, USA) equipped with a Z-spray electrospray interface.

58 This paper uses the metabolite nomenclature recommended by several authors (C. D. Kay, M. N. Clifford, P.
59 Mena, G. J. McDougall, C. Andres-Lacueva, A. Cassidy, D. Del Rio, N. Kuhnert, C. Manach, G. Pereira-Caro,
60 A. Rodriguez-Mateos, A. Scalbert, F. Tomás-Barberán, G. Williamson, D. S. Wishart and A. Crozier,
61 Recommendations for standardizing nomenclature for dietary (poly)phenol catabolites, *Am. J. Clin. Nutr.*,
62 2020, 112, 1051–1068).

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82 **Supplementary Table 3.** Cumulative body weight gain at the end of the experiment after the sustained
83 supplementation of different diets, and total feed intake expressed in grams (g) after six weeks. SD:
84 standard diet, RD: rice diet, WGB: whole-grain barley, PG: pearled grain, BB: barley bran.

Group	Male body weight gain (g)	Female body weight gain (g)	Male feed intake (g)	Female feed intake (g)
SD	5.82 ± 0.66	3.75 ± 0.48	615	544
RD	5.90 ± 0.75	3.16 ± 0.42	628	508
WGB	4.62 ± 0.52	3.21 ± 0.41	589	514
BB	3.66 ± 1.23	2.87 ± 0.41	685	560
PG	4.17 ± 1.02	4.08 ± 0.44	704	568

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106 **Supplementary Table 4.** Mouse serum protein levels of inflammatory biomarkers considering both
 107 males and females together. Interferon-gamma (IFN- γ), C-reactive protein (CRP), Interleukine-4 (IL-4),
 108 Tumor necrosis factor- alfa (TNF- α), Interleukine-2 (IL-2), Lipopolysaccharide binding protein (LBP);
 109 SD: standard diet, RD: rice diet, WGB: whole-grain barley, PG: pearled grain, BB: barley bran. The
 110 results are shown as the mean \pm SEM. Significant differences (p-value<0.05) are expressed as * compared
 111 with SD.

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Diet	IFN- γ (pg/ml)	CRP (ng/ml)	IL-4 (pg/ml)	TNF- α (pg/ml)	IL-2 (pg/ml)	LBP (μ g/ml)
SD	2.02 \pm 0.33	621.9 \pm 76.64	1.05 \pm 0.40	6.99 \pm 1.12	8.94 \pm 3.19	1294.14 \pm 196.52
RD	1.99 \pm 0.57	771.33 \pm 89.88	1.75 \pm 0.78	7.52 \pm 1.94	7.22 \pm 1.48	1132.24 \pm 244.98
WGB	2.76 \pm 1.06	1992.57 \pm 747.64	0.65 \pm 0.23	7.10 \pm 1.60	3.05 \pm 1.01	1026.69 \pm 204.73
BB	0.96 \pm 0.34	773.07 \pm 212.77	0.33 \pm 0.17	2.65 \pm 0.30*	5.54 \pm 0.51	1444.35 \pm 209,14
PG	2.28 \pm 0.72	1798.71 \pm 790.93	1.59 \pm 0.67	3.49 \pm 0.57	8.44 \pm 2.53	339.80 \pm 79.64 *

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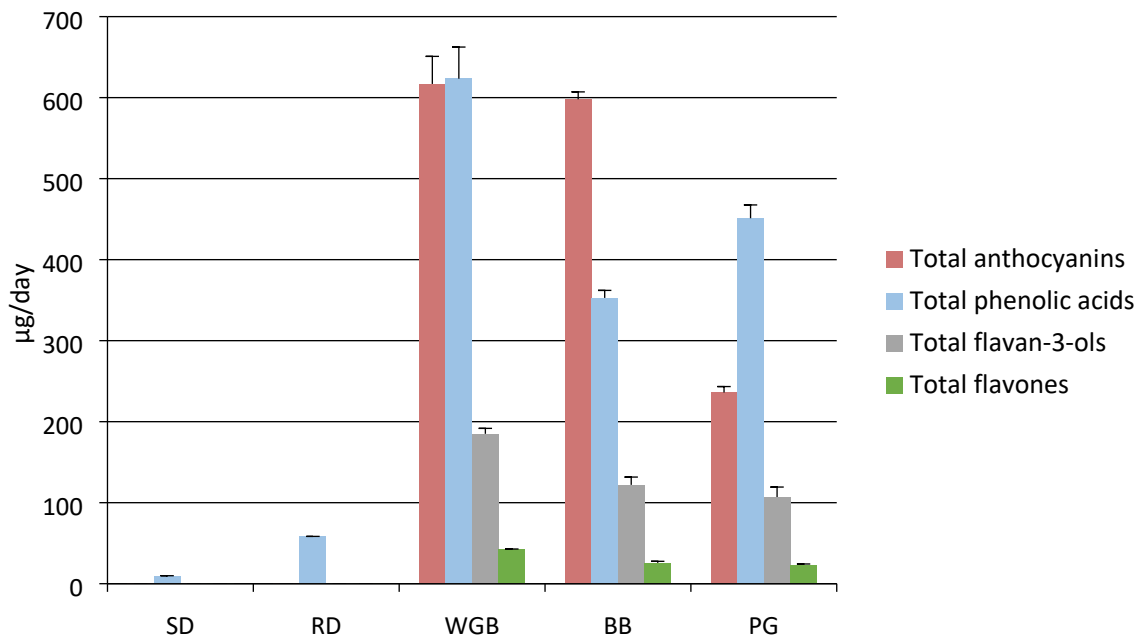
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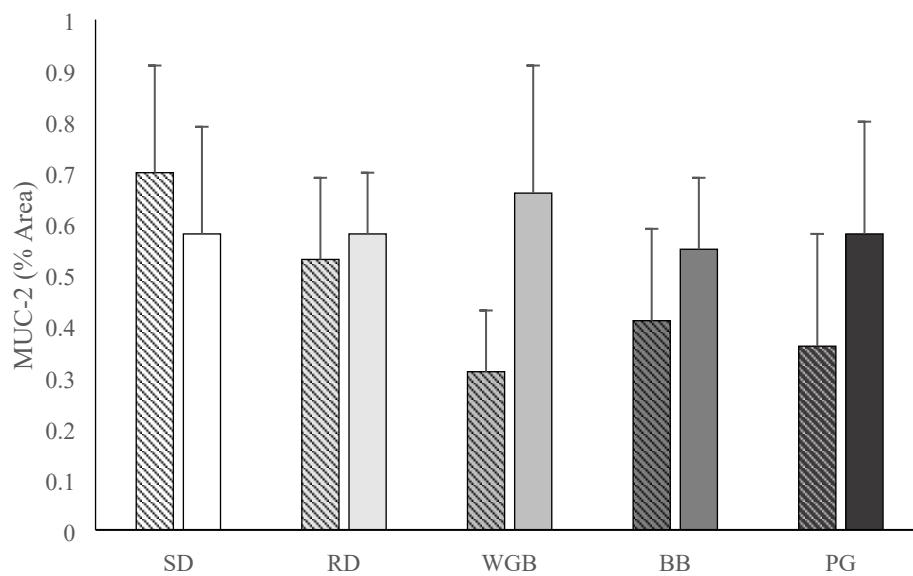
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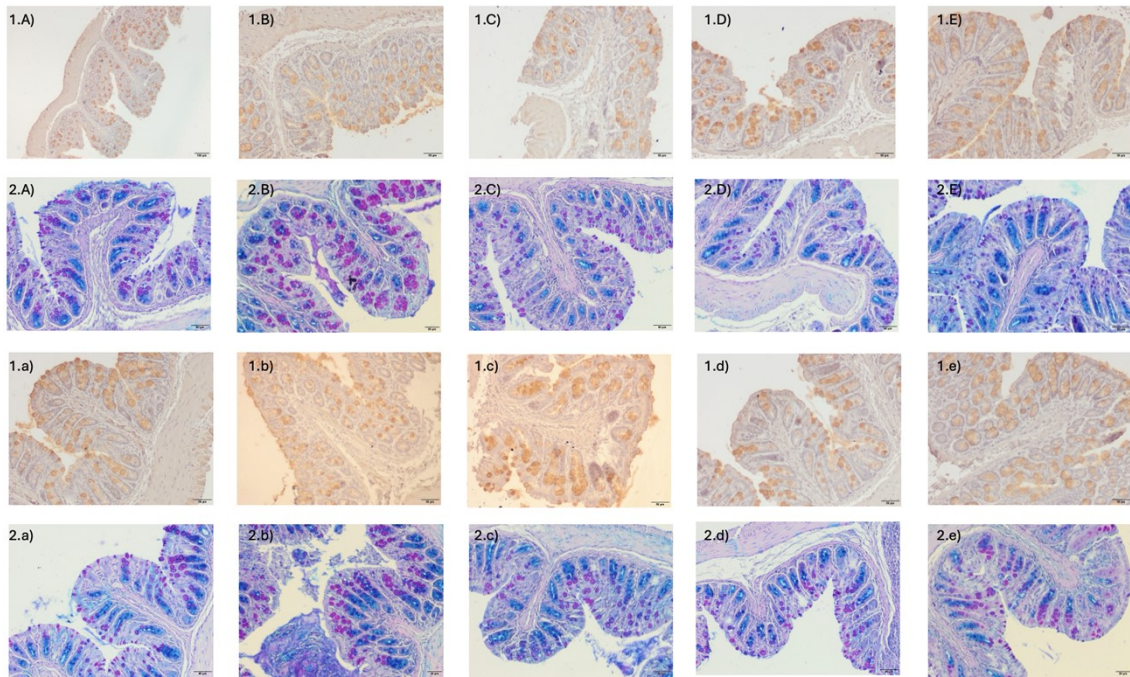
129 **Supplementary Figure 1.** Daily dosage of (poly)phenolics ($\mu\text{g}/\text{day}/\text{mouse}$) ingested through the SD,
130 RD, WGB, BB and PG diets analyzed by UPLC-MS/MS.

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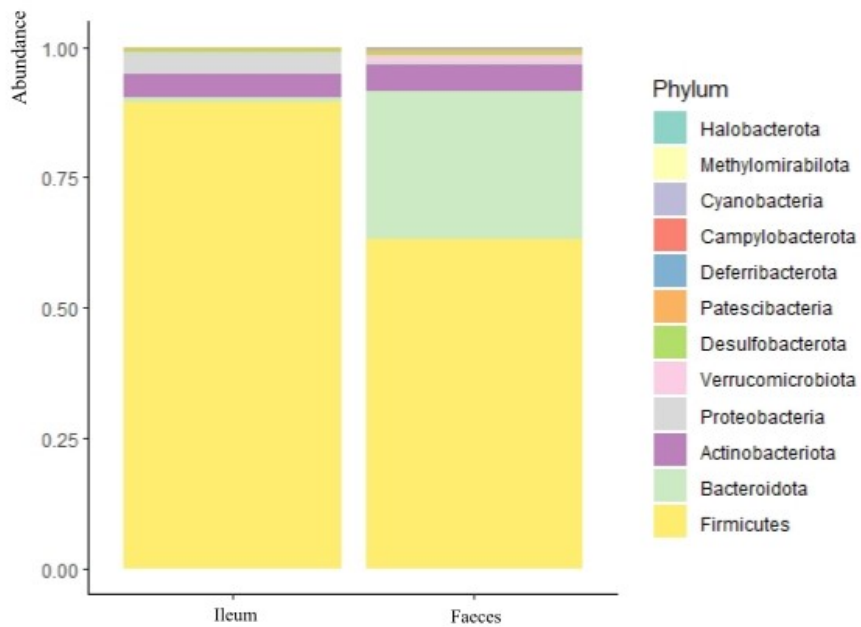
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133 **Supplementary Figure 2.** Graphical representation of the area percentage of mucin MUC2. SD: standard
134 diet, RD: rice diet, WGB: whole-grain barley, PG: pearled grain, BB: barley bran. Males with lines and
135 females without them.



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137 **Supplementary Figure 3.** 1) Immunohistochemical images of mucin MUC2 and 2) Alzian blue from
 138 descending colon samples. (A) SD; (B) RD; (C) WGB; (D) BB; (E) PG; In capital letters for males and
 139 in lowercase for female. X10 microscope objective.

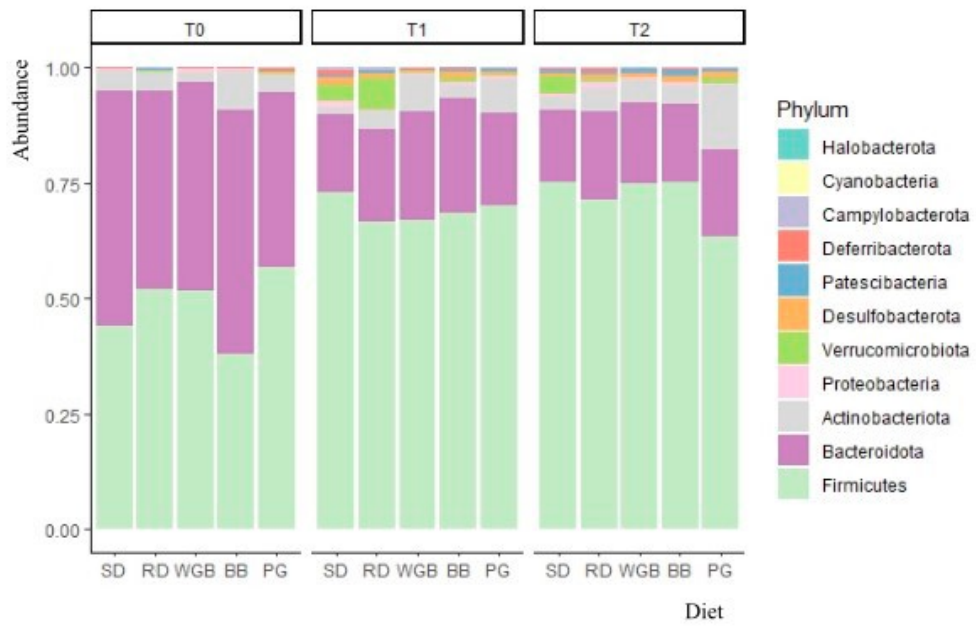


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141 **Supplementary Figure 4.** Relative abundance of identified phyla across ileum and faecal samples.

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146 **Supplementary Figure 5.** Taxonomic composition of mice faecal microbiota at phylum level

147 expressed as relative abundance. SD: standard diet, RD: rice diet, WGB: whole-grain barley, PG:

148 pearled grain, BB: barley bran.

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