

Supplementary Materials

The bioavailability of polyphenols following acute consumption of pigmented barley and wheat



A. Purple Barley



B. Purple Wheat



C. Blue Wheat



D. Regular Wheat

Figure S1. Images of whole grain seeds used in study cut in cross sections, Pictures taken with an optical microscope (Leica DM 750 Microsystems, Wetzlar, Germany)

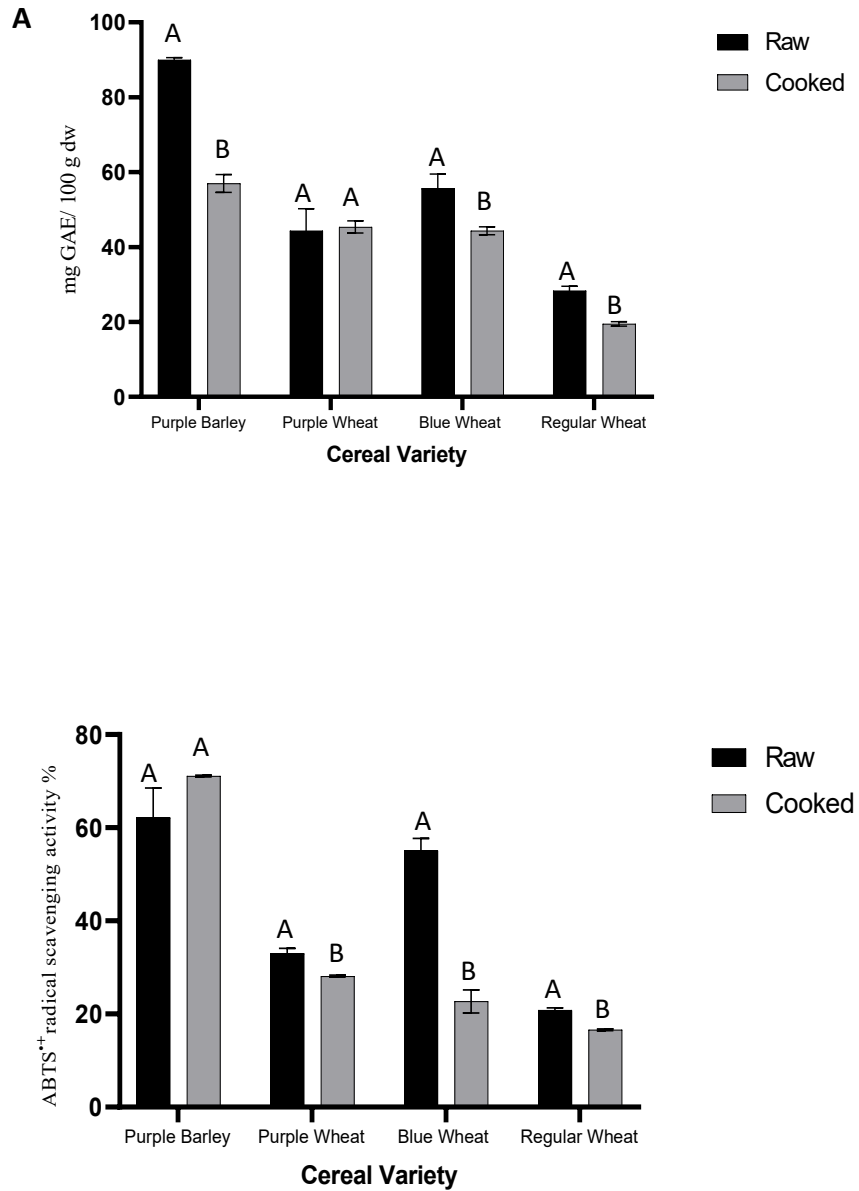
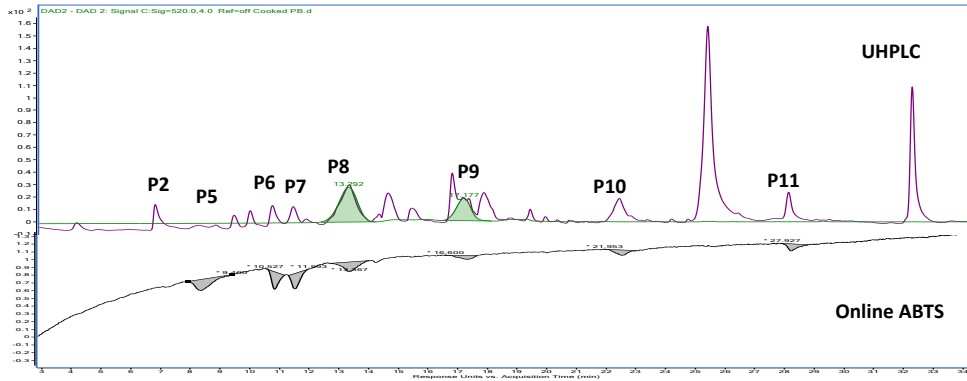
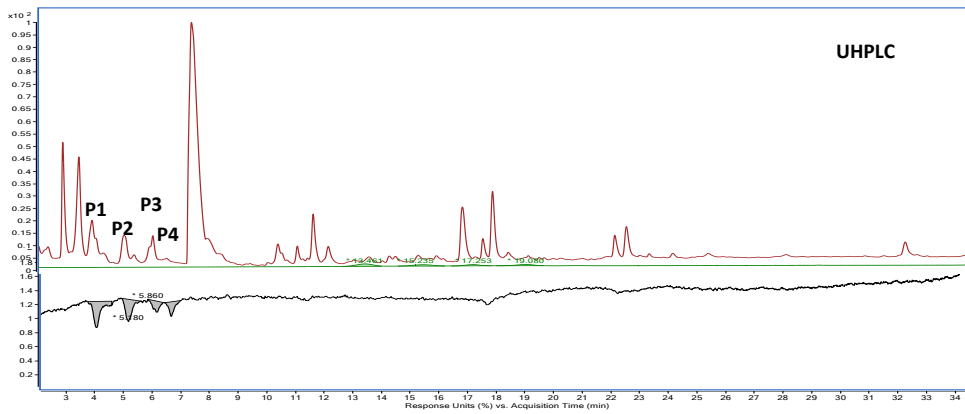


Figure S2. Comparison of total phenolic content (A) and total antioxidant capacity (B) between methanol extracts of raw and cooked grain measured using Folin-Ciocalteu and ABTS radical scavenging assay. Data are expressed as mg GAE/ 100g dry cereal flour and as percentages for the total phenolic content and total antioxidant activity respectively. Mean \pm SD; n=3. Different letters represent significant differences between the grains ($p < 0.05$)

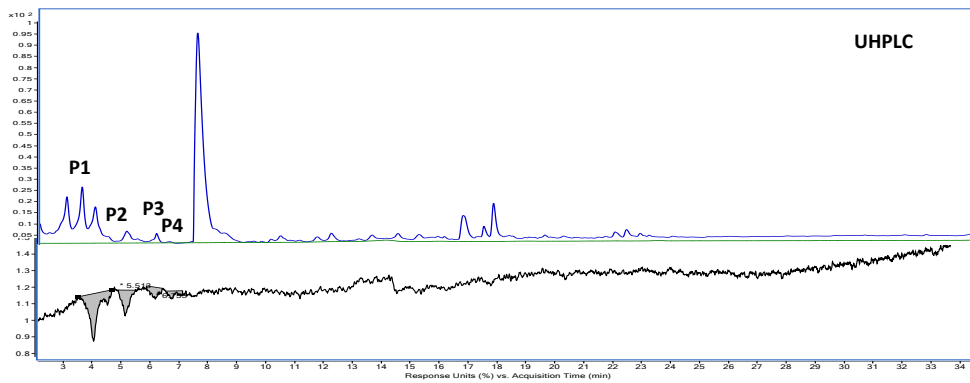
A. Cooked Purple Barley



B. Cooked Purple wheat



C. Cooked Blue wheat



D. Cooked Regular wheat

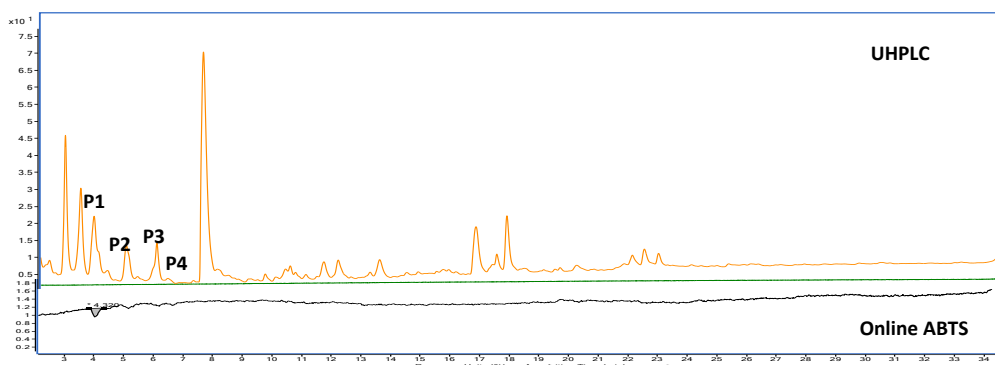


Figure S3. UHPLC – online ABTS mapping of extracts from cooked grain; Cooked purple barley (A), cooked purple wheat (A), cooked blue wheat (C) and cooked regular wheat (D). Green line: anthocyanin profile; Black line: online ABTS profile; Purple line: cooked purple barley phenolic profile; Red line: cooked purple wheat phenolic profile; Blue line: cooked blue wheat phenolic profile; Orange line: cooked regular wheat phenolic profile

Table S1. Baseline characteristics influencing anthropometric, biochemical, and haematological profiles.

Parameter	Mean	SD	Reference range
Age	28.8	5.9	
Height (m)	1.7	0.1	
Weight (Kg)	74.6	9.8	
BMI (Kg/m ²)	23.9	1.4	
Systole BP	119.5	11.3	
Diastole BP	73.7	6.9	
Pulse	12.5	12.5	
Hb (g/L)	140.4	81	115–180
RCC (10 ¹² /L)	4.67	0.7	3.9–5.7
HCT (L/L)	0.42	0.04	0.36–0.54
RDW (%)	14.6	0.6	11–15
MCV (fL)	90.4	3.4	82–98
WCC (10 ⁹ /L)	6.63	2.1	3.7–9.5
PLT (10 ⁹ /L)	243.7	81	150–400
Urea (mmol/L)	4.6	2.0	3.0–10.0
Glucose (mmol/L)	4.1	1.2	3.0–5.4
Cholesterol (mmol/L)	5.4	1.3	3.0–5.5
Calcium (mmol/L)	2.4	0.3	2.10–2.60
Magnesium (mmol/L)	0.67	0.2	0.70–1.10

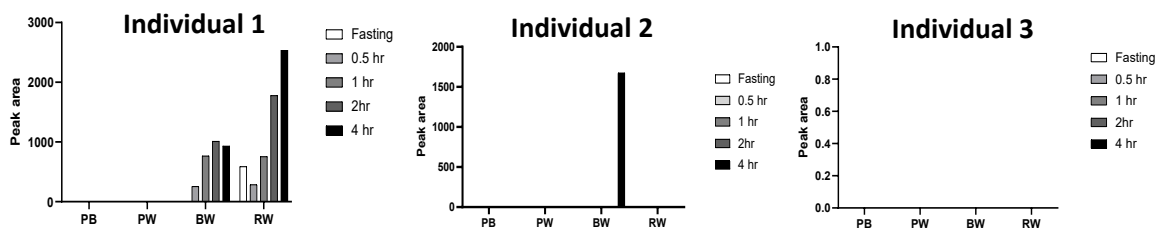
Values are presented as mean \pm SD, n = 6. Blood pressure: BP; body mass index: BMI; mean corpuscular volume: MCV; platelets, PLTS; red cell count, RCC; red cell distribution width, RDW; white cell count, WCC; N/A: Not Applicable

Baseline anthropometric, full blood count and biochemical parameters were measured and found to be within the normal reference ranges established by the Royal College of Pathologist of Australasia the baseline parameters are presented in Table 2.

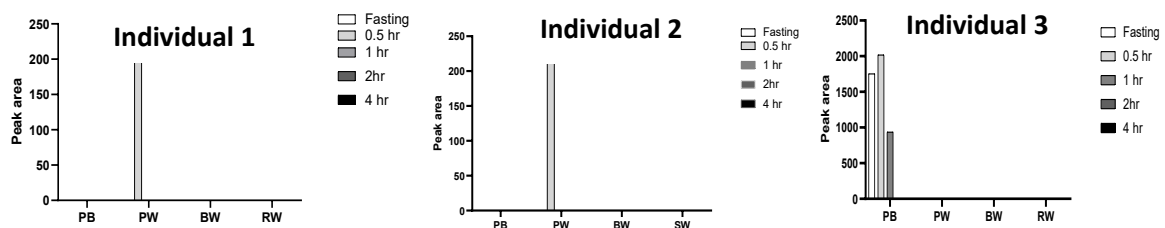
Table S2. Recovery of phenolic standards in plasma

Standard	matrix	Spiked concentration	% recovery
Ferulic acid	plasma	500	96.2
Cyanidin 3 O- glucoside	plasma	500	82.1
Hippuric acid	urine	500	100

A. Plasma protocatechuic acid levels



B. Plasma caffeic acid levels



C. Plasma hippuric acid levels

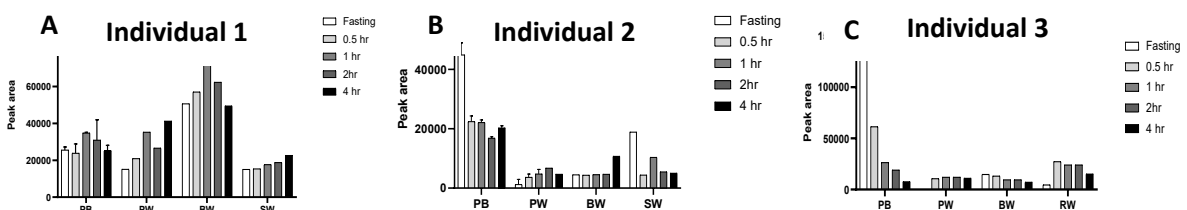


Figure S4 Plasma levels of protocatechuic acid (A), caffeic acid (B) and hippuric acid (C) compared to baseline fasting levels after consumption of the cereal grain varieties by 3 individuals.

Table S3: Selected metabolites detected from mass spectrometry analysis of urine

Preliminary identification	RT (min)	m/z
Hydroxyhippuric acid	6.7	194.0555
4-Hydroxyhippuric acid sulphate	7.0	194.0555
Ferulic acid 4- O sulphate	8.4	273.2500
Dihydrocaffeic acid 3-O sulphate	7.6	261.2370
N-feruloylglycine	7.8	250.2300

Table S4. Percentage of phenolic compound absorbed and excreted.

Phenolic acid	% Bioavailability (Plasma)				% Urinary Excretion			
	PB	PW	BW	RW	PB	PW	BW	RW
Protocatechuic acid	N/A	N/A	1.8	5.9	1.5	4.1	44.9	88.5
Caffeic acid	1.5	1.3	N/A	N/A	16.1	17.2	63.0	94.3

Values are average percentages calculated by dividing the amount of phenolic detected in biofluid by the amount found in the cooked cereal grain extract. N/A: Not Applicable