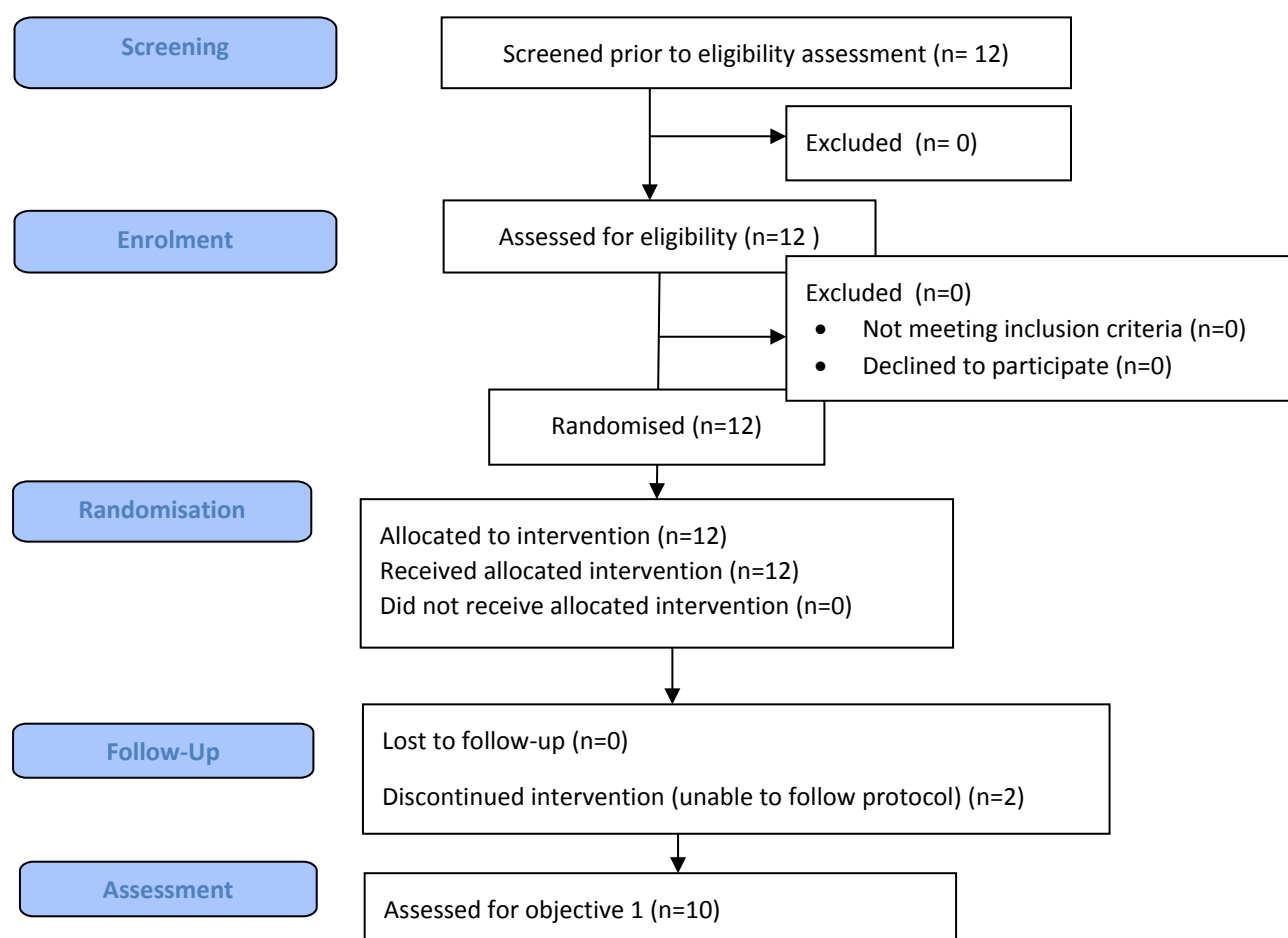


Supplementary Information

Effect of semolina pudding prepared from *starch branching enzyme IIa* and *b* mutant wheat on glycaemic response *in vitro* and *in vivo*: a randomised controlled pilot study

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Figure S1. CONSORT 2010 Pilot trial flow diagram



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Figure S2. Particle size distribution of raw semolina

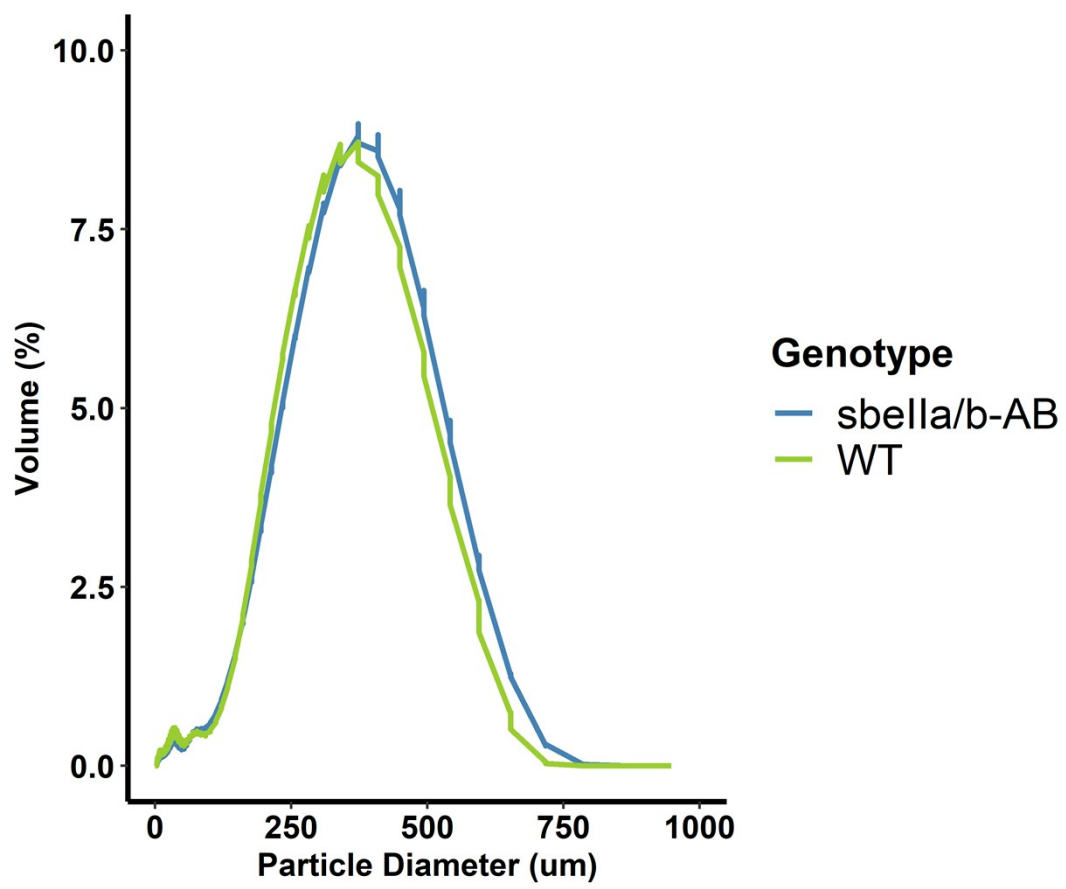


Table S1. Pudding nutritional composition on an 'as is' basis

Pudding	Protein	Total Fat	Saturated Fat	Monounsaturated Fat	Polyunsaturated Fat	Trans Fats	Total Carbohydrate	Starch	Resistant Starch	Total Sugars	Glucose	Fructose	Sucrose	Lactose	Maltose	Galactose	Ash	AOAC Fibre	Energy (kcal)	Energy (kJ)
Per serving	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)	(g)		
WT	10.296	1.08	0.28152	0.13176	0.6336	0	51.768	47.59	2.75	4.176	0.144	0	0.288	0	3.744	0.144	0.6	2.304	262.58	1113.5
<i>sbella/b</i> -AB	13.68	1.52	0.3584	0.1824	0.904	0	51.76	48.64	4.89	3.12	0.16	0.08	0.32	0	2.4	0.08	0.6	3.52	282.48	1196.9

Pudding	Sodium	Potassium	Calcium	Magnesium	Phosphorus	Iron	Zinc	Manganese
Per serving	(mg)	(mg)	(mg)	(mg)	(mg)	(mg)	(mg)	(mg)
WT	393	175.85	15.628	33.4	100.09	0.8	0.9	0.453
<i>sbella/b</i> -AB	0	192	16.853	46.613	146.4	1.2	1.4	0.858

Table S2. Capillary blood glucose concentration 0-120 minutes

<i>sbella/b</i> -AB pudding								
Participant ID	Time to consume the meal (min)	Time-point 0 (min)	Time-point 15 (min)	Time-point 30 (min)	Time-point 45 (min)	Time-point 60 (min)	Time-point 90 (min)	Time-point 120 (min)
1	15.00	4.65	4.60	6.00	6.00	5.20	4.60	4.40
2	15.00	5.60	6.70	7.90	7.00	6.30	5.30	5.20
3	8.00	4.50	5.20	6.55	6.60	5.90	5.00	4.60
4	12.40	4.60	4.20	5.10	5.40	6.10	6.10	5.70
6	7.00	4.15	3.70	6.20	6.30	6.50	6.10	5.60
7	18.00	4.45	5.10	6.75	6.90	5.70	4.30	5.30
9	20.00	5.25	4.70	7.80	5.70	5.90	5.30	5.10
10	6.30	5.05	5.40	6.45	6.70	6.90	5.70	5.50
11	9.00	4.70	5.80	8.45	8.00	6.40	4.80	4.80
12	11.30	3.85	3.70	4.65	5.10	4.50	5.00	4.40
WT pudding								
Participant ID	Time to consume the meal (min)	Time-point 0 (min)	Time-point 15 (min)	Time-point 30 (min)	Time-point 45 (min)	Time-point 60 (min)	Time-point 90 (min)	Time-point 120 (min)
1	15.00	4.35	4.40	5.25	7.70	7.30	5.80	4.40
2	14.40	4.95	5.00	7.80	7.60	6.60	5.60	4.80
3	10.00	4.50	5.20	6.70	6.20	5.60	4.20	4.00
4	6.00	5.20	6.20	7.35	8.20	7.90	7.20	6.00
6	7.00	4.20	4.30	5.90	6.50	5.80	5.60	4.80
8	18.00	5.10	3.90	6.80	6.20	6.00	5.00	4.70
9	18.00	4.75	4.60	6.45	5.50	6.10	5.40	4.20
10	6.00	5.20	5.70	6.75	6.60	7.10	4.70	4.70
11	9.00	4.75	4.50	7.35	7.70	7.70	5.60	5.30
12	11.30	4.65	5.70	6.05	6.30	6.60	6.20	5.60

Table S3. Monosaccharides profiles.

Total monosaccharides									
Sample	Genotype	Arabinose		Galactose		Xylose		Mannose	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE
Semolina	<i>sbella/b</i> -AB	11.4535	0.728124	3.888567	0.541709	17.26183	0.909625	1.266833	0.62608
	WT	7.304467	0.325402	2.442633	0.242645	11.041	0.29733	0.81915	0.57925
Pudding	<i>sbella/b</i> -AB	9.95941	0.295793	3.048276	0.085165	14.21245	0.449171	0.979076	0.04943
	WT	7.201238	0.650758	2.154119	0.273856	10.35535	0.894504	0.937471	0.113535
Water extractable monosaccharides									
Sample	Genotype	Arabinose		Galactose		Xylose		Mannose	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE
Semolina	<i>sbella/b</i> -AB	1.8308	0.2746	1.7315	0.1463	1.2823	0.1978	0.1712	0.0707
	WT	1.3966	0.0146	1.2811	0.0333	0.9701	0.0513	0.0552	0.0078
Pudding	<i>sbella/b</i> -AB	1.844905	0.049324	1.953886	0.035205	1.601395	0.026227	0.435219	0.017454
	WT	1.27039	0.055	1.486367	0.027577	1.265081	0.051052	0.283267	0.009228

Mean of 3 independent replicates of bulked semolina. Mean of 7 independent replicates of freeze-dried pudding

Table S4. Total (TOT) and Water Extractable (WE) Arabinoxylans (AX) of *sbella/b*-AB pudding compared to the wild-type control (WT).

Parameter	Pudding WT control	Pudding <i>sbella/b</i> -AB
WE-AX (mg/g sample)	1.33 ± 0.03	1.84 ± 0.02
TOT-AX (mg/g sample)	14.28 ± 1.39	19.61 ± 0.73

Means ± SEMs, freeze-dried pudding *n* = 7 replicates prepared independently.

Arabinoxylans are calculated as the sum of Arabinose and Xylose corrected for arabinogalactan (AGP) content as in^{1-3} and multiplied by 0.89 to convert monosaccharide values, for arabinose and xylose, to that for the polysaccharide, arabinoxylan.

Table S5. AXOS of *sbella/b*-AB pudding compared to the wild-type control (WT).

Parameter	Pudding WT control	Pudding <i>sbella/b</i> -AB
TOT-AXOS [†]	11.84 ± 0.13	14.56 ± 0.09
AXOS Unsubstituted [†]	6.76 ± 0.09	7.93 ± 0.06
AXOS Substituted [†]	5.07 ± 0.05	6.63 ± 0.04
MLG (G3+G4) [†]	2.58 ± 0.04	3.11 ± 0.03

Means ± SEMs, freeze-dried pudding *n* = 7 replicates prepared independently from the same batch of bulked semolina.

[†]Sum of peaks relative to the internal standard

1. N. Anders, K. Stott, P. R. Shewry, J. C. Mortimer, P. Dupree, T. Weimar, A. Lovegrove, R. A. C. Mitchell, J. Freeman, M. D. Wilkinson, T. Tryfona, M. Defoin-Platel, J. M. Baker and T. K. Pellny, Glycosyl transferases in family 61 mediate arabinofuranosyl transfer onto xylan in grasses, *Proceedings of the National Academy of Sciences*, 2012, **109**, 989-993.
2. N. Anders, M. D. Wilkinson, A. Lovegrove, J. Freeman, T. Tryfona, T. K. Pellny, T. Weimar, J. C. Mortimer, K. Stott, J. M. Baker, M. Defoin-Platel, P. R. Shewry, P. Dupree and R. A. C. Mitchell, Glycosyl transferases in family 61 mediate arabinofuranosyl transfer onto xylan in grasses - Supporting Information, *Proceedings of the National Academy of Sciences*, 2012, DOI: 10.1021/jo101696z, 1-10.
3. H. N. Englyst, M. E. Quigley and G. J. Hudson, Determination of dietary fibre as non-starch polysaccharides with gas-liquid chromatographic, high-performance liquid chromatographic or spectrophotometric measurement of constituent sugars, *The Analyst*, 1994, **119**, 1497-1509.