

Glycaemic, gastrointestinal and appetite responses to breakfast porridges from ancient cereal grains: a MRI pilot study in healthy humans

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SHORTENED VERSION OF THE TITLE: Millet, oat, rye porridge breakfast responses (max 45 characters)

ABBREVIATIONS: FMP, finger millet porridge; PMP, pearl millet porridge; RP, rye porridge; SOP, Scottish oats porridge

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1 **Abstract**

2 Cereal grain based porridges are commonly consumed throughout the world. Whilst some data are
3 available for varieties that are popular in the Western world such as oats and rye, other ‘ancient’
4 grains used in the East and in Africa such as millets are thought to have beneficial health effects, such
5 as a suppression of post prandial hunger and circulating glucose levels. These grains, a sustainable
6 food source due to their tolerance of extreme weather and growing conditions, are commonly found
7 throughout Asia and Africa. However, knowledge of the physiological responses to these grain
8 varieties is very limited. This study aimed to collect initial pilot data on the physiological and
9 gastrointestinal responses to breakfast porridges made with two millet varieties and oats and rye
10 grains. A total of $n = 15$ completed the oats and rye, $n = 9$ the finger millet $n = 12$ the pearl millet
11 meals. MRI scans were undertaken at baseline, immediately after consumption and then hourly
12 postprandially. Blood glucose was measured at baseline, immediately after consumption and then
13 every 15 min until $t = 80$ min, then every 20 min until $t = 120$ min, followed on each occasion by
14 completion of VAS. Seven participants completed the entire protocol and were included in the final
15 analysis. A subgroup analysis with the $n = 10$ paired comparison between the same individuals that
16 completed the oats, rye and pearl millet was also considered. The gastric volume AUC was higher
17 for pearl millet than oats and rye ($n = 10$, $p < 0.001$). The incremental area under the curve (iAUC) for
18 blood glucose was not significantly different between the meals although this showed a trend to be
19 lower for pearl millet. Hunger was lower for pearl millet compared to oats and rye ($n = 10$, $p = 0.01$).
20 There was a significant correlation between total gastric volume AUC and average appetite AUC $r =$
21 -0.47 , $p < 0.010$. Isoenergetic breakfast porridges from ‘ancient’ varieties of millet grains showed
22 physiological responses that were comparable with those from common Western varieties known to
23 have beneficial health effects. Pearl millet appeared to induce lower postprandial blood glucose
24 response and appetite scores though the differences were not conclusive compared with the other

25 porridges and further work is needed. Improved knowledge of the effects of different cereal grains
26 could help direct dietary advice and ultimately improve health outcomes in the general population
27 worldwide.

28

29 **KEYWORDS:** Magnetic resonance imaging, Blood glucose, Appetite, Gastric emptying, Breakfast
30 porridges, Cereal grains .

31

32 **1 Introduction**

33 The increasing global prevalence of obesity is a growing public health problem. Obesity is associated
34 with insulin resistance, a major risk factor for chronic non-communicable diseases such as type 2
35 diabetes and cardiovascular disease. (Kim et al., 2009; Kopelman, 2007; Meigs et al., 2007). Eating
36 breakfast porridges based on whole cereal grains has been associated with a variety of health benefits
37 including lower postprandial blood glucose levels, improved insulin responses, increased satiety and
38 reduced long- term weight gain. (D. Jenkins et al., 1988; D. J. Jenkins et al., 2009; J. Slavin, 2004; J.
39 L. Slavin, Martini, Jacobs, & Marquart, 1999; Who & Consultation, 2003) Cereal grains used at
40 breakfast are a staple source of energy for many populations worldwide, with regional difference in
41 consumption tending to reflect historical patterns of crop cultivation. Oats, for example, are more
42 commonly consumed in the English-speaking countries; rye is favoured in the Scandinavian countries
43 whilst millet is very common in Asia and Africa. (In, 1995; Kyro et al., 2012; Shobana et al., 2013).
44 However nutritional value, potentially beneficially metabolic effects and other health effects may
45 vary between different cereal grains, including insulinaemic responses and serum LDL cholesterol
46 concentration. (Magnusdottir et al., 2014; Meynier, Goux, Atkinson, Brack, & Vinoy, 2015; Nilsson,
47 Östman, Granfeldt, & Björck, 2008) This may be due to differences in the inter- relationships between
48 digestion, gastric emptying and absorption. Subsequent differences may be seen in post prandial
49 glycaemia and appetite (Koh-Banerjee et al., 2004; Liu, 2003; Liu et al., 2000; Liu et al., 1999; Meyer
50 et al., 2000; Schlundt, Hill, Sbrocco, Pope-Cordle, & Sharp, 1992; J. Slavin, 2004); low glycaemic
51 index (GI) diets produce a more gradual rise in blood sugar and insulin levels. Eating food with low
52 GI may confer health advantages such as improving glycaemic control and insulin sensitivity in
53 people with diabetes and reduced risk of chronic disorders. The rise in chronic non-communicable
54 diseases in low and middle income countries such as India and China has been linked to a large shift

55 from consumption of coarse grains such as millets to consumption of rice and wheat among the
56 population (Popkin, Horton, Kim, Mahal, & Shuigao, 2001).

57 It has recently been suggested that different cereal grains, particularly millet grains, may have
58 enhanced health benefits in terms of glucose and insulin metabolism (Helnaes et al., 2016; Nambiar,
59 Dhaduk, Sareen, Shahu, & Desai, 2011; Shobana et al., 2013).

60 However, limited studies have been conducted to investigate the physiological and gastrointestinal
61 responses to these grains, particularly the blood glucose and appetite responses to the millets.

62 Magnetic resonance imaging (MRI) provides a unique tool to investigate gastrointestinal handling of
63 food (Alyami, Spiller, & Marciani, 2015). Furthermore, the small intestinal secretory and fluid
64 response to breakfast porridges is unknown. After milling, intact particles can exert effects through
65 mechanical stimulation of the small intestine, as we showed using bran and plastic particles
66 (McIntyre, Vincent, Perkins, & Spiller, 1997), and MRI provides a non-invasive means to assess
67 gastrointestinal fluid responses (Marciani et al., 2010).

68 This pilot study was therefore designed to collect initial data on postprandial glucose levels following
69 consumption of isoenergetic breakfast porridges made from finger millet, pearl millet oats and rye.

70 Secondly, the study aimed to compare postprandial gastric volumes, small bowel water content and
71 subjective appetite for these meals. It also aimed to explore possible correlations between blood
72 glucose levels, gastric volumes, and subjective appetite outcomes. We hypothesised that breakfast
73 porridges made from different varieties of cereal grains would produce different postprandial glucose
74 responses, gastric volumes and subjective appetite scores in healthy participants.

75

76 **2 Material and methods**

77 *2.1 Study participants*

78 Within the study period a total of 17 healthy participants were screened out of the planned 18. One
79 did not attend screening so 16 healthy subjects (ten female and six male, 20.9 (SD 0.9) years old,
80 BMI 22.1 (SD 2.9) kg/m² participated. A full dataset for all four meals was obtained for seven
81 participants who were then included in subsequent analysis. This included four females and three
82 males, aged 21 (SD 1.0) years old, and with a BMI of 21.8 (SD 2.1) kg/m². A subgroup analysis was
83 also considered for ten participants who consumed all of the SOP, RP and PMP meals. The remaining
84 nine participants were excluded from the analysis either because they did not attend visits or because
85 they were unable to consume all of the test meal on one or more visits (Fig. 1).

86 Participants were recruited from the student and staff population of the University of Nottingham via
87 a poster advertisement. Those who expressed interest were invited to a screening session to establish
88 whether they met the study inclusion criteria, namely: age 18 - 65 years, being healthy, BMI ≥ 18 and
89 ≤ 30 kg/m² and able to give informed written consent. Exclusion criteria included: using medication
90 which interferes with study measurements, participating in another nutritional or biomedical trial
91 three months before the pre-study examination or during the study, not being a habitual breakfast
92 consumer, not usually eating at least three meals a day, reporting participation in night shift (between
93 midnight and 6.00 am), doing strenuous exercise for >10 h/week, consuming of ≥ 21 alcoholic drinks
94 in a typical week, following a medically or self-prescribed diet during the two weeks prior to the pre-
95 study examination and until the end of the study, contraindications for MRI scanning (e.g. presence
96 of metal implants, infusion pumps and pacemakers) as assessed by standard MRI safety questionnaire,
97 pregnancy, inability to lie flat and exceeding the scanner bed weight limit of 120kg.

98 The study was conducted at the Sir Peter Mansfield Imaging Centre at the University of Nottingham.
99 Informed written consent was obtained from each participant before the trial. A site master file and
100 case report forms were kept according to good clinical practice.

101 All procedures in this study involving human participants were approved by the University of
102 Nottingham, Medical School Research Ethics Committee (F14072015). The study was registered
103 within Clinical Trials.gov (NCT02653274). The trial registration name was ‘Assessment of Millet,
104 Oat and Rye Porridge Breakfasts Glucose and Gastric Emptying (AMORE)’.

105

106 2.2 *Study design*

107 This study was randomized, four way crossover design. Participants attended the laboratory on four
108 separate days, approximately 1 week apart, in order to consume four different porridges in a
109 randomized order. Participants consumed their habitual diet between each visit. Each visit lasted from
110 8:00 am until approximately 1:00 pm. The porridge meals differed in appearance and taste hence
111 participants could not be blind although they were not informed which porridge they were consuming
112 on each visit.

113

114 2.3 *Screening*

115 All potential participants attended a screening visit to establish that they met the study inclusion
116 criteria for the study. Height and weight were measured and the Body Mass Index (BMI) was
117 calculated as weight divided by the square of height.

118

119 2.4 *Laboratory visit protocol and procedures*

120 Fig. 2 shows the study day protocol. The participants were asked to fast overnight (for at least ten
121 hours). A glass of water was permitted on waking. On arrival they completed the study eligibility
122 check questionnaire to ensure adherence to the study day restrictions such as the overnight fasting.
123 Baseline measurements (defined as $t = 0$ min) then were made which included fasting blood glucose,
124 participant completion of paper based subjective visual analog appetite score (VAS, described below)

125 and a MRI scan. The participants were then requested to eat the given porridge within a maximum
126 time of 15 min. This was followed by an immediate postprandial (defined as $t = 20$ min) measurement
127 of blood glucose, followed by VAS completion and a MRI scan. Blood glucose was subsequently
128 measured every 15 min until $t = 60$ min, then every 20 min until $t = 120$ min, followed on each
129 occasion by completion of VAS. MRI scans were undertaken hourly from $t = 15$ min up to $t = 140$
130 min. Participants were given a blank food diary and instructed to complete it over the remainder of
131 the day.

132

133 2.5 *Breakfast porridge intervention*

134 Four breakfast porridges were made from Scottish oats (Asda, United Kingdom), rye
135 (buywholefoodsonline.co.uk, Canterbury, United Kingdom), finger millet (Top-Op Foods Ltd.,
136 Stanmore, United Kingdom) and pearl millet (Herbs n Spice it, India). The oats and rye were steam
137 rolled flakes. Rye flakes were larger than the oat flakes. The millets were plain dehulled grains ground
138 to a flour using a spice grinder in our lab. The cooked products were analysed for macronutrient
139 composition and total energy by Campden BRI, Chipping Campden, Gloucestershire, UK (Table 1)
140 so that the four breakfast meals given to the participant could be made isoenergetic (220 kcal each).
141 The grains were cooked in water in two separate aliquots using two microwaves (900 watts). The aim
142 was to achieve an acceptable final product hence the grains were subjected to different cooking
143 protocols. The oats were simply heated with water, the rye was soaked for half an hour in boiled water
144 then heated; the millets were ground prior to cooking using a spice grinder for 30 s. The study meals
145 were consumed with 240 ml of water in a glass and on each of the four occasions, the participants
146 were asked to consume the entire portion with the water drink within 15 min. Other meal
147 characteristics such as appearance, volume and weight necessarily differed between meals (SOP 400
148 g; RP 297 g; FMP 432 g; PMP 310 g).

149

150 2.6 *Gastrointestinal response measured by MRI*

151 Magnetic resonance imaging (MRI) was carried out on a research-dedicated 1.5 T Philips Achieva
152 MRI scanner (Philips Healthcare, Best, The Netherlands). Participants were in the supine position
153 with a 16 element receiver coil wrapped around their abdomen. Gastric volume was measured using
154 a balanced turbo field echo (bTFE) sequence. A total of 25 axial images were acquired with the
155 following sequence parameters: the field of view (FOV) 400 mm × 320 mm × 250 mm, acquired
156 resolution $2.01 \times 1.76 \text{ mm}^2$, slice thickness 10 mm, repetition time (TR) 2.8 ms, echo time (TE) 1.4
157 ms, no slice gap, flip angle (FA) 80° and one breath hold for 10 s. Gastric volume was measured
158 manually by one operator using Analyze9 software (Mayo Foundation, Rochester, MN, USA).

159 The water content of the entire small bowel (SBWC) was measured using a single-shot, fast spin
160 echo sequence (rapid acquisition with relaxation enhancement), which shows high intensity signals
161 from areas with free mobile fluid and dark signals from other body tissues. A total of 24 coronal
162 images were obtained using the following sequence parameters: FOV 400 mm × 400 mm, acquired
163 resolution $0.78 \times 0.78 \text{ mm}^2$, slice thickness 7 mm, TR 8000 ms, TE 320 ms, no slice gap, and one
164 breath hold for 24 s.

165 The SBWC was assessed using in-house software which was previously validated (Hoad et al., 2007).
166 Briefly, bright signals from organs other than small bowel water (e.g. stomach, gall bladder) were
167 segmented out manually, and then integrating total volume over pixels with intensity values above
168 the calculated threshold. The total AUC for gastric volume and for small bowel water were calculated.

169

170 2.7 *Glycaemic response*

171 The glycaemic response was measured in capillary blood samples (Freckmann et al., 2012) using the
172 protocol described by Brouns et al. (2005) which is in line with techniques recommended by the
173 World Health Organization (WHO) / Food and Agricultural Organization (FAO 1998). The capillary
174 blood samples were collected by finger prick using single-use lancets (Unistix Owen Mumford,
175 Oxfordshire, United Kingdom). The capillary blood glucose was measured using Accu-check (Roche
176 Diagnostics, USA).

177 Participants were requested to warm their hands before the finger prick in order to increase the blood
178 flow. To extract the blood, the fingertips were gently massaged from the base of the hand, moving
179 towards the tips in order to minimise the plasma dilution. Incremental area under the glucose curve
180 (iAUC) and peak blood glucose response to the test products were calculated according to Brouns et
181 al. (2005); Wolever and Jenkins (1986). iAUC was obtained using the trapezoid rule and ignoring the
182 area beneath the baseline.

183

184 2.8 *Subjective appetite ratings*

185 100 mm VAS were used to measure the subjective feelings of hunger, satisfaction, fullness, desire to
186 eat and prospective food consumption.(Flint, Raben, Blundell, & Astrup, 2000) When outside the
187 MRI scanner,the participants were requested to make a vertical mark on each scale at the point that
188 best matched how they felt at that time. Each end of the line was anchored by statements expressing
189 the extreme for the sensation. For example, ‘not hungry at all’ and ‘more hungry than have ever been’.
190 To avoid bias from previous answers the participants were presented only with a new VAS sheet at
191 each time point and this was removed immediately after completion. The VAS appetite ratings were
192 determined by measuring (in millimetres) the distance from the left side of the line to the vertical
193 mark.

194 The average Appetite score was calculated for each individual at each time of measurement, for each
195 test meal, using the formula: Average appetite score = [hunger + (100 – satisfaction) + (100 –
196 fullness) + desire to eat + prospective consumption]/5 (Anderson, Catherine, Woodend, & Wolever,
197 2002; Stubbs et al., 2007). The Average Appetite scores at each time point were used for the statistical
198 analysis. The range for the appetite score was between 0 and 100; 0 representing the minimum
199 appetite sensations and 100 representing the maximum appetite sensations. Total AUC for average
200 appetite score were calculated (Blundell et al., 2010).

201

202 2.9 Food diaries

203 Food diaries were given to the participants before discharge. They were requested to provide a
204 detailed record of food and beverages consumed over the remainder of the day, once they had left the
205 unit. They were required to include information such as portion sizes, product brand names, and
206 cooking and preparation methods. Furthermore, if the participants prepared composite dishes at home,
207 then they were requested to provide the recipe and portion size.

208 Nutritics software (Nutritics Ltd , Dublin, Ireland) was used to analyse the food intake from the food
209 diaries. Some food items were added manually to the database using the information on nutrition
210 labels.

211

212 2.10 Sample size and statistical analysis

213 Descriptive and statistical analyses were undertaken using Prism version 6.07 (Graph Pad Software
214 Inc., La Jolla, CA). All data are presented as mean±SE unless otherwise indicated. Data were assessed
215 for normality using the Shapiro-Wilk 's test. Normally distributed data were analysed using
216 parametric methods; non-normally distributed data were analysed using non-parametric methods.

217 This was a pilot study and we did not have own data to speculate on the sample size required. For an
218 overall estimate, using data from Nilsson et al. (2008), a 33% change in blood glucose should be
219 detectable with $\alpha=0.05$ and a power of 80% using $n=18$.

220 Differences in glycaemic response, gastric volume, SBWC and appetite score were assessed using
221 one-way repeated-measures analysis of variance followed by Tukey 's post hoc test.

222 Correlations between blood glucose, gastric volume and appetite scores were assessed using
223 Pearson's correlation. Differences were considered significantly different at $p<0.05$.

224

225 **3 Results**

226 In this pilot study, the effects of different porridges on postprandial glycaemic response,
227 gastrointestinal response (gastric volume and SBWC) and subjective appetite were measured.

228 Seven participants failed to consume all of the finger millet test meal hence they were excluded from
229 the per protocol analysis. When asked, palatability was reported as the main problem. Two more
230 subjects did not attend one of the study session hence were also excluded. The results presented are
231 thus shown as per protocol analysis with $n = 7$ (four females and three males of normal weight) who
232 consumed all of the four study meals (Table 2). Additionally, a subgroup analysis ($n = 10$ for SOP,
233 RP and PMP) was considered in this pilot study (Table 4)

234

235 *3.1 Appearance of the gastric content and total gastric volumes*

236 Fig. 3 shows the appearance of the gastric content for each of the porridges immediately after
237 consumption ($t = 20$ min). With SOP, two layers can be seen, a bottom layer providing a lower signal
238 (appearing darker in the figure) and a top layer providing higher signal (appearing brighter in the
239 figure), whilst RP remains in a distributed form in the stomach. FMP and PMP produced multiple
240 layers in the stomach.

241 There was no significant difference in fasting baseline gastric volumes between the test days as
242 expected. Gastric volumes rose on consumption of the porridges and declined with time as shown in
243 Fig. 4.

244 The immediate postprandial gastric volumes ($t = 20$ min) were significantly different between the
245 four breakfast meals ($p = 0.007$) in keeping with the initial meal volume differences. Overall AUC 2
246 h gastric volumes showed a significant difference between the study meals ($p = 0.003$). There was a
247 significant difference in gastric volume AUC between the RP and FMP ($p = 0.04$) and a difference in
248 gastric volume AUC between the RP and the PMP ($p = 0.002$). The subgroup analysis ($n = 10$) also
249 showed significant difference immediate postprandial gastric volumes ($T = 20$) and AUC gastric
250 volum between the test meals as shown in Table 4.

251

252 3.2 *Small bowel water content*

253 SBWC data are shown in Fig. 5; the mean fasted SBWC was 23.1 ml (SD 6.4) for the four study
254 porridges. All the meals induced an initial drop in SWBC after feeding followed by a rise at $t = 80$
255 min, but the differences were not statistically different.

256

257 3.3 *Glycaemic response*

258 The glycaemic responses to all the porridges were in the normal range for non-diabetic subjects. There
259 were no differences in fasting glucose values, and the expected post consumption increase in blood
260 glucose was seen in all cases. Fig. 6 shows the iAUC for all participants. The highest mean peak
261 glucose was following FMP at 7.8 mmol/ml compared with following SOP, RP and PMP at 7.1
262 mmol/ml, 6.8 mmol/ml and 6.9 mmol/ml. For the $n = 7$ analysis, glucose iAUC 0-2 h was also the
263 lowest after PMP (109.6 mmol/l 120 min) compared with following SOP, RP and FMP (131.1 mmol/l

264 120 min, 119.5 mmol/l 120 min and 145.4 mmol/l 120 min respectively. These differences were not
265 statistically significant.

266 The n = 10 subgroup analysis showed that PMP and RP had similar peak blood glucose level at 7.0
267 mmol/ml, whereas peak blood glucose of SOP was 7.1 mmol/ml. There was no significant difference
268 in IAUC glucose between the test meals (Table 4).

269

270 3.4 *Subjective appetite ratings*

271 The area under curves of the subjective appetite ratings are summarized in Table 3 and the impact of
272 the four breakfast porridges on the scores for hunger, satisfaction and average appetite are shown in
273 Fig. 7. As expected, for all interventions, the scores for hunger, desire to eat and prospective food
274 consumption decreased after consuming the breakfast before returning to baseline, whilst fullness and
275 satisfaction initially decreased and then increased again in all cases. AUC for the sense of hunger of
276 the subgroup analysis showed a significant difference between the test meals ($p = 0.017$). The average
277 appetite score was the lowest after consuming the millet, but the AUC for this score was not
278 significantly different between the three porridges (Table 5).

279

280 3.5 *Food intake record*

281 Four food diaries were not returned (1 for SOP, 2 for RP and 1 for FMP) so these data cannot be
282 presented as per protocol. Data are presented as mean and standard errors of mean. The self-reported
283 daily energy intake records following consumption of the SOP, RP, FMP and PMP were 1747 ± 158
284 kcal/d, 2332 ± 369 kcal/d, 1694 ± 100 kcal/d and 1754 ± 322 kcal/d respectively, the differences
285 being not significant.

286

287 3.6 *Correlations*

288 There was a significant correlation between total gastric volume AUC and average appetite AUC $r =$
289 -0.47 $p < 0.010$, but not between gastric volumes and iAUC glucose ($P < 0.3$). The subgroup analysis
290 with $n = 10$ showed also a similar significant negative correlation between total gastric volume AUC
291 and average appetite AUC $r = -0.465$ $p < 0.01$.

292

293 **4 Discussion**

294 This is the first pilot, *in vivo* imaging study assessing the glycaemic, gastrointestinal and appetite
295 responses to porridges made from different ‘modern’ and ‘ancient’ cereal grains in healthy, normal-
296 weight participants. The pilot project is a small-scale study conducted to gain experience and allow
297 appropriate sample size calculations for future studies (Thabane et al., 2010).

298 In this current study, some participants were unable to consume the meals in full particularly the
299 FMP. Future studies need to review the way in which the product is prepared and consumed in order
300 to ensure participants can adhere to the protocol. Exclusion of those participants who had not
301 consumed all porridges reduced the sample size to $n = 7$. Ten subject completed oats, rye and pearl
302 millet providing a second post-hoc analysis.

303 Among those who were able to consume all four porridges, isoenergetic breakfast porridges made
304 from different grains induced different gastrointestinal and physiological responses, although for the
305 sample size used in this pilot study only gastric volume were significantly different. After
306 consumption of the pearl millet porridge, there was a trend for the glucose response to decrease,
307 gastric volume to increase and appetite to increase compared with the other porridges some of which
308 failed to reach significance potentially because of small numbers. The secondary analysis with $n =$
309 10 showed similar trends with some outcome differences reaching significance such as Hunger.

310 The immediate post prandial gastric volumes were significantly different as would be expected given
311 that the isoenergetic portions had different volumes. Although the total energy of a meal has an effect

312 on the gastric emptying, in this study the four breakfast meals were isoenergetic, suggesting that the
313 gastric volume was affected by other factors such as particle size, viscosity and the meal volume,
314 which is a key regulator of gastric emptying (Calbet & MacLean, 1997). Furthermore, separation of
315 liquid and solid parts (known as sieving) could affect the gastric emptying rate as the liquid part
316 would be absorbed quickly in the early phase (Marciani et al., 2012). In this study, SOP emptied
317 faster compared with the other porridges, which may be due to the separation between liquid part and
318 oat flakes as shown in Fig. 4. On the other hand, the liquid and solid phase were combined in the
319 PMP and this may limit the sieving and delay emptying of the meal (M. Clegg, Ranawana, Shafat, &
320 Henry, 2013). Although the volume of PMP porridge was lower than that of SOP and FMP, PMP
321 emptied at a slower rate, which could account for the smaller rise in blood sugar of this millet
322 (Horowitz, Edelbroek, Wishart, & Straathof, 1993).

323 The appearance of the SBWC resembles that reported previously (Hoad et al., 2007; Marciani et al.,
324 2013). The postprandial SBWC initially fell during the ‘gastric phase’ after feeding and the rose
325 during the ‘intestinal phase’. The early decrease in the SBWC is possibly related to the absorption of
326 the readily available nutrients in the liquid phase. The later rise of the SBWC is likely to be related
327 to the increased pancreatobiliary and enterocyte secretion after a mixed liquid/ solid meal and
328 possibly also the effect of particulates (Marciani et al., 2010; McIntyre et al., 1997; Murray et al.,
329 2014).

330 All the grains had a relatively low glycaemic index (Gonzalez & Stevenson, 2012; Nambiar et al.,
331 2011; Rosén, Östman, & Björck, 2011; Shobana et al., 2013). The present study is in agreement with
332 many studies that have shown that rye is known to induce a low and prolonged blood glucose response
333 (Rosén et al., 2011; Rosen et al., 2009). Although the rye grains were soaked in water before heating
334 in the microwave in this study, which could have an effect on the gelatinisation of rye starch and as
335 a result elevated the glucose response (Zhu, 2014). The glucose response after consuming the RP

336 remained lower. The glucose response after consumption of oats was also in agreement with other
337 studies. A study on rolled oats showed that after consumption a similar peak blood glucose value of
338 7 mmol/L, suggesting that our results are in line with the literature. (Gonzalez & Stevenson, 2012)
339 However, the results in relation to the finger millet were inconsistent in terms of the higher blood
340 glucose in comparison with other studies (Shobana et al., 2013; Shukla & Srivastava, 2014). However
341 there is little information available about the physiological and gastrointestinal responses to millet
342 grains, especially pearl millet, limiting our knowledge about their potential health benefits.
343 The differences in the glycaemic response seen between these grains could be due to the processing
344 of cereals which alters the digestion of the cereal grains; this is a considered a major determinant of
345 the glycaemic response (Heaton, Marcus, Emmett, & Bolton, 1988; Mackie et al., 2017) and also of
346 the impact on appetite. (Isaksson et al., 2012). Oat and rye are steamed rolled flakes which can keep
347 the endosperm intact and ultimately limit accessibility of amylase to the starch oats (Taylor,
348 Emmambux, & Kruger, 2015). Our finger and pearl millet, on the other hand, were milled to flour
349 which offers a higher surface area to digestion. This possible explanation is supported by a systematic
350 review investigating the effects of different processing methods on glycaemic responses, in which it
351 is shown that a smaller particle size caused greater gelatinisation and a heightened glycaemic response
352 (Granfeldt, Eliasson, & Björck, 2000; Tosh & Chu, 2015). In addition, another study has found that
353 different milling methods have effects on glycaemic response of foods made with finger millets flour
354 (Jayasinghe, Ekanayake, & Nugegoda, 2013). The difference in the glucose response between the
355 two millets could be due to the different amount of carbohydrate content which is potential an
356 important determinant of the glucose response (Arvidsson-Lenner et al., 2004; Kang et al., 2013). In
357 this study, FMP had the highest carbohydrate content (53.1 g) compared with PMP (45.6 g). This
358 could explain the greater rise of FMP compared to the other porridges.

359 The current study indicated that the millet porridges may have prolonged satiating properties
360 compared with the oats and the rye porridges. The increase in satiety following the millets could be
361 related to the delay in decline of the gastric volume in these grains causing prolonged distension of
362 the stomach and delayed delivery of nutrients into the small intestine (M. E. Clegg & Shafat, 2014;
363 Kissileff, Carretta, Geliebter, & Pi-Sunyer, 2003; Mackie, Rafiee, Malcolm, Salt, & van Aken, 2013;
364 Marciani et al., 2001). Furthermore, the reduced rate on gastric emptying following consumption of
365 the PMP could account for the blunted glycaemic response of the pearl millet (Bornet, Jardy-
366 Gennetier, Jacquet, & Stowell, 2007). This study did not measure duodenal motility hence it is not
367 possible to comment on possible differences in motility between meals and the impact that this may
368 have on gastric emptying. (Teramoto et al., 2012, 2014).

369 There were several limitations to the current study including the fact that the test meals were
370 physically different, two were steam rolled flakes and two plain grains ground to a flour. The test
371 meals were cooked slightly differently to obtain a more acceptable final product which may have
372 altered the bioavailability of carbohydrates. Seven participants found the palatability of finger millet
373 poor and could not finish all the test meal. The isoenergetic portions were of different volume. Use
374 of capillary blood glucose does not represent arterial blood however it is a close approximation
375 (Brouns et al., 2005). Also, future studies should measure insulin. Some food diaries were missed
376 limiting our opportunity to assess the impact of the porridge consumed as a breakfast on 24 energy
377 intake. Appetite ratings are a proxy measure for what people will actually eat. This leads us to suggest
378 that this will be better assessed in future studies by providing lunch using 'bottom less bowl' thereby
379 providing a more accurate and objective measure of actually food consumption at midday as well as
380 more closely managing the return of diaries.

381 Although this pilot study did not demonstrate many significant differences in the physiological and
382 gastrointestinal responses after consumption of the four breakfast meals, valuable experience has been
383 gained in the implementation of the protocols and provided useful directions for further studies.
384 Finally, the use of in vivo imaging can increase our knowledge of the behaviour of these meals in the
385 gastrointestinal (GI) tract. This will facilitate an understanding of the interface between the input of
386 a given feeding stimulus and various physiological and behavioural consequences. This will help us
387 to improve our understanding of the effect of physical properties of food on digestion and appetite,
388 engineer foods with the desired in vivo behaviour and develop more relevant in vitro / in vivo food
389 digestion models.

390

391 **5 Conclusion**

392 Isoenergetic breakfast porridges from ‘ancient’ varieties of millet grains showed physiological
393 responses that were comparable with those from common Western varieties known to have beneficial
394 health effects. Pearl millet appeared to induce lower postprandial blood glucose response and appetite
395 scores though small numbers did not allow conclusive inferences against other grains and further
396 work is needed. Pearl millet is a popular ‘ancient’ and sustainable grain, and may represent a valid
397 alternative to other cereal breakfasts. Improved knowledge of the effects of different cereal grains
398 could help direct dietary advice. The breakfast porridge intervention is relatively cheap compared to
399 other interventions and could help reduce the burden of obesity and related metabolic disorders
400 worldwide.

401

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406

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410

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412

413 **Authorship:**

414 The authors' responsibilities were as follows: MAT, LM and JA designed the study with contribution
415 from: RCS on gastroenterology, AAS on statistics, PAG on imaging, IAM on metabolic physiology
416 and GPA on liver metabolism. CLH set up the MRI sequences and analysis. All authors read and
417 approved the final manuscript.

418

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420

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- 586
- 587

588 **Table1**

589 Macronutrient composition of the breakfast meals. The values are shown total for each cooked
 590 product as served

	SOP	RP	FMP	PMP
Weight (g)	400	297	432	311
Energy (kJ)	920.0	920.0	920.0	920.0
Energy (kcal)	220.0	220.0	220.0	220.0
Protein (kjeldahl, g)	7.2	4.2	3.5	5.3
Total carbohydrate (by difference, g)	42.0	49.3	53.1	45.6
Carbohydrate(Avail, g)	34.0	39.8	43.6	38.8
Total sugars (enzymic, g)	1.6	5.9	1.7	0.3
Fructose (enzymic, g)	0.4	0.3	0.4	0.3
Glucose (enzymic, g)	0.8	0.3	0.4	0.3
Maltose (enzymic, g)	0.4	0.3	0.4	0.3
Sucrose (enzymic, g)	0.8	5.9	1.7	0.3
Fat (Weibull-Stoldt, g)	4.4	2.7	1.3	3.1
Saturates (g)	0.8	0.6	0.4	0.6
MUFA (cis, g)	2.0	0.6	0.4	0.9
PUFA (cis)	1.2	1.2	0.4	1.2
Trans fatty acids (g)	0.4	0.3	0.4	0.3
Total fiber (AOAC, g)	8.0	6.5	13.8	6.8
Sodium(ICP-MS)	24.4	17.8	28.5	19.5
Moisture (Oven102°C)	345.2	240.0	372.4	255.6
Ash(@525C)	1.2	1.1	1.4	1.0
Protein N Factor	6.25	6.25	6.25	6.25
Equivalent Salt	0.4	0.3	0.4	0.3

591

592 SOP, Scottish Oats porridge; RP, Rye porridge; FMP, Finger millet porridge; PMP, Pearl millet
 593 porridge; AOAC, Association of Analytical Communities.

594

595

596 **Table 2**

597 Blood glucose, time to peak, gastric volumes, small bowel water content and average appetite sensations measured from n = 7 healthy participants who
598 were fed four different breakfast porridges.

	SOP		RP		FMP		PMP		1- ANOVA
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	
IAUC glycaemic response mmol/l (over 120 min)	131	28	119	27	145	23	110	29	0.5
Glucose peak, mmol/l	7.2	0.3	7.2	0.5	7.7	0.4	6.7	0.3	0.2
Gastric volume at T = 20	505	26	384	22	548	48	532	23	0.007
AUC Gastric volume ml/min	50324	2696	41644	2892	56606	3832	58684	3339	0.003
AUC small bowel water content ml/min	1611	429	1303	360	735	259	2157	499	0.06

599

600 (Mean values with their standards errors) n = 7

601 **Table 3**

602 Participants' (n = 7) area under the satiety curve from the visual analog scales for hunger, satisfaction ,fullness, desire to eat, prospective food
 603 consumption and average appetite score.

604

Appetite sensations variables	SOP		RP		FMP		PMP		1- ANOVA
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	
Hunger (mm/min)	6325	461.9	7717	636.5	5378	1196	5465	1030	0.08
Satisfaction (mm/min)	6877	537	5920	716	8636	1164	7849	917	0.08
Fullness (mm/min)	6881	580	6149	754	8385	1117	8238	1102	0.3
Desire to eat (mm/min)	6776	526	7643	832	5549	1349	5992	1003	0.5
Prospective food consumption (mm/min)	7092	502	7984	788	5618	1359	5938	1067	0.3
Average appetite sensations	6887	463	7855	710	5505	1220	5862	991	0.5

605 (Mean values with their standards errors) n = 7

606

607

608

609 **Table 4**

610 Blood glucose, time to peak, gastric volumes, small bowel water content and average appetite sensations measured from n = 10 healthy participants who
 611 were fed four different breakfast porridges.

612

	SOP		RP		PMP		1-
	Mean	SE	Mean	SE	Mean	SE	ANOVA
IAUC glycemic response mmol/l (over 120 min)	134	27	102	21	107	21	0.6
Glucose peak, mmol/l	7.1	0.2	7.0	0.4	7.0	0.2	0.7
Gastric volume at T = 20	535	23	407	29	544	17	0.0008
AUC Gastric volume ml/min	41519	1978	34751	2249	59454	2499	0.0001
AUC small bowel water content ml/min	1302	255	1123	332	1713	275	0.18

613

614

615 (Mean values with their standards errors) n = 10

616

617

618

619

620 **Table 5**

621 Participants' (n = 10) area under the satiety curve from the visual analog scales for hunger, satisfaction ,fullness, desire to eat, prospective food
 622 consumption and average appetite score.

623

	SOP		RP		PMP		1-
Appetite sensations variables	Mean	SE	Mean	SE	Mean	SE	ANOVA
Hunger (mm/min)	5274	673.8	6606	770.5	4996	842.7	0.01
Satisfaction (mm/min)	7989	695	7062	785.9	8121	672.4	0.22
Fullness (mm/min)	7976	733.9	7235	799.6	8502	811.9	0.43
Desire to eat (mm/min)	5737	688.3	6535	829.5	5538	835	0.18
Prospective food consumption (mm/min)	6131	690.2	6836	833	5498	887.6	0.36
Average appetite sensations	5835	672.7	6736	786.6	5482	782.1	0.22

624

625

626 (Mean values with their standards errors) n = 10

627 **Figure Legends**

628

629 **Fig. 1** Study participant flow diagram

630

631 **Fig. 2** Diagram of the study day protocol

632

633 **Fig. 3** Representative example of axial MRI images of the abdomen of a healthy participant fed with
634 , Scottish oats porridge (SOP); Rye porridge (RP); Finger millet porridge (FMP); Pearl millet porridge
635 (PMP) on four different occasions. Images were taken at t=20 min.

636

637 **Fig. 4** Plot of the volume of the gastric contents for healthy participants after they consumed the four
638 different study porridges. $\text{---}\blacktriangledown\text{---}$, Scottish oats porridge (SOP); $\text{---}\bullet\text{---}$, Rye porridge (RP); $\text{---}\blacksquare\text{---}$,
639 Finger millet porridge (FMP); $\text{---}\blacktriangle\text{---}$, Pearl millet porridge (PMP). Values are mean \pm SE, n = 15 for
640 SOP and RP, n = 9 for FMP and n = 12 for PMP. The arrow on the horizontal axis indicates the meal
641 start time.

642

643 **Fig. 5** Plot of the volume of the small bowel water content for healthy participants after they
644 consumed the four different study porridges. $\text{---}\blacktriangledown\text{---}$, Scottish oats porridge (SOP); $\text{---}\bullet\text{---}$, Rye
645 porridge (RP); $\text{---}\blacksquare\text{---}$, Finger millet porridge (FMP); $\text{---}\blacktriangle\text{---}$, Pearl millet porridge (PMP). Values are
646 mean \pm SE, n = 15 for SOP and RP, n=9 for FMP and n = 12 for PMP. The arrow on the horizontal
647 axis indicates the meal start time.

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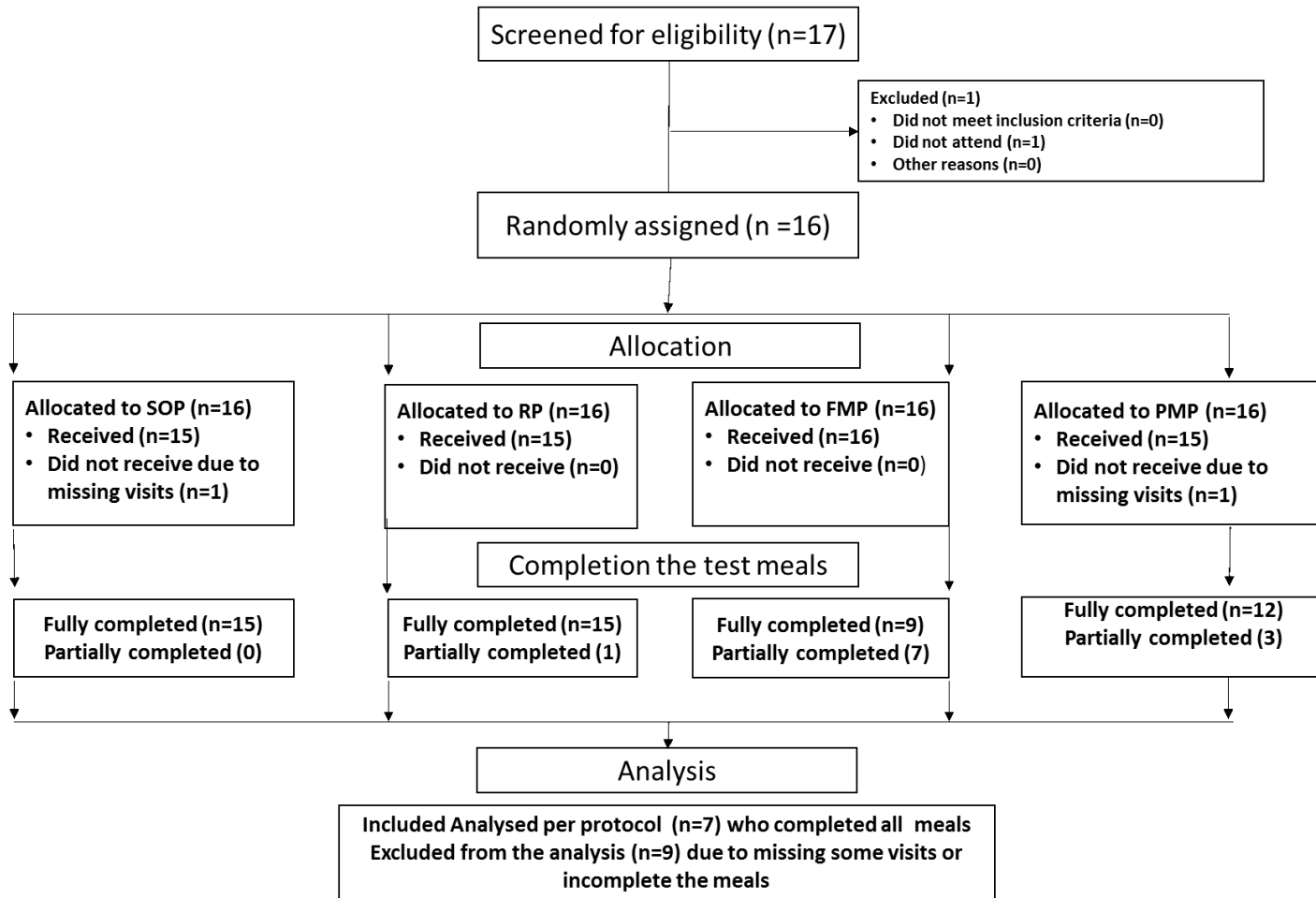
649 **Fig. 6** Incremental area under the glucose curve (iAUC) for healthy participants after they consumed
650 the four different study porridges., Scottish oats porridge (SOP); Rye porridge (RP); Finger millet

651 porridge (FMP); Pearl millet porridge (PMP). Values are mean \pm SE, n=15 for SOP and RP, n = 9
652 for FMP and n = 12 for PMP.

653

654 **Fig. 7** Plot of the average appetite sensations for healthy participants after they consumed the four
655 different study porridges. $\text{---}\blacktriangledown\text{---}$, Scottish oats porridge (SOP); $\text{---}\bullet\text{---}$, Rye porridge (RP); $\text{---}\blacksquare\text{---}$,
656 Finger millet porridge (FMP); $\text{---}\blacktriangle\text{---}$, Pearl millet porridge (PMP). Values are mean \pm SE, n = 15 for
657 SOP and RP, n = 9 for FMP and n = 12 for PMP. The arrow on the horizontal axis indicates the meal
658 start time.

659 Fig 1:

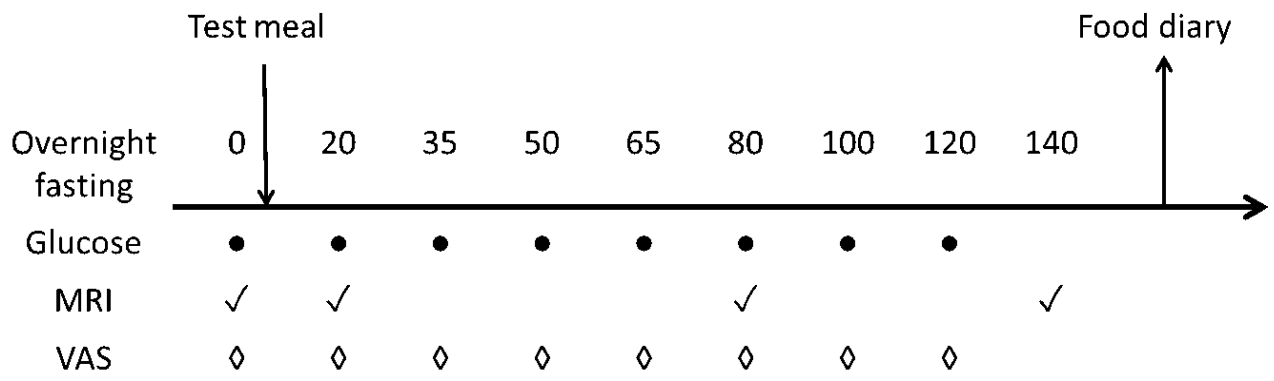


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662 Figure 2

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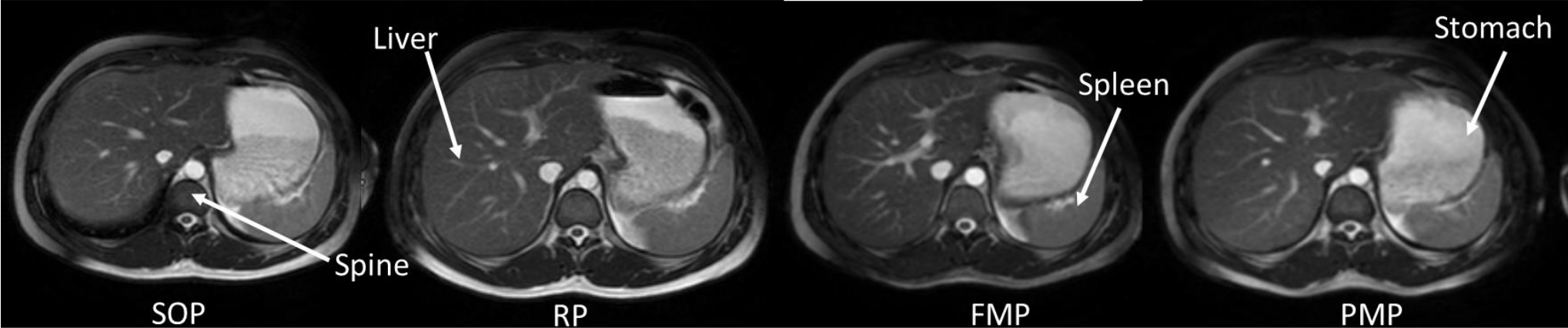
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669 Figure 3



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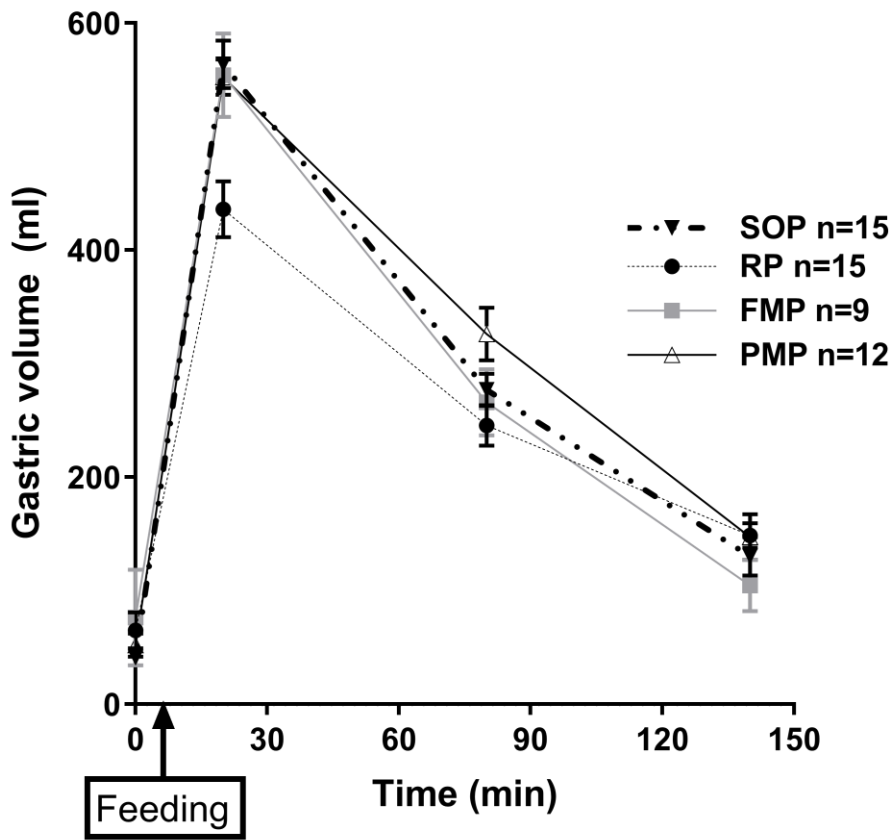
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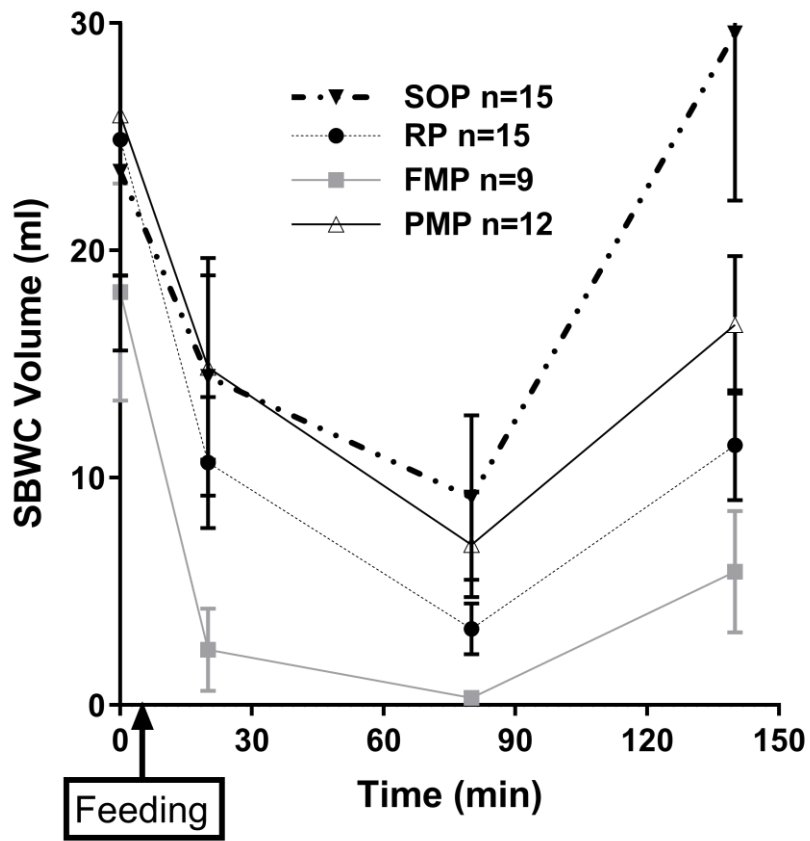
677 Figure 4

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680 Figure 5



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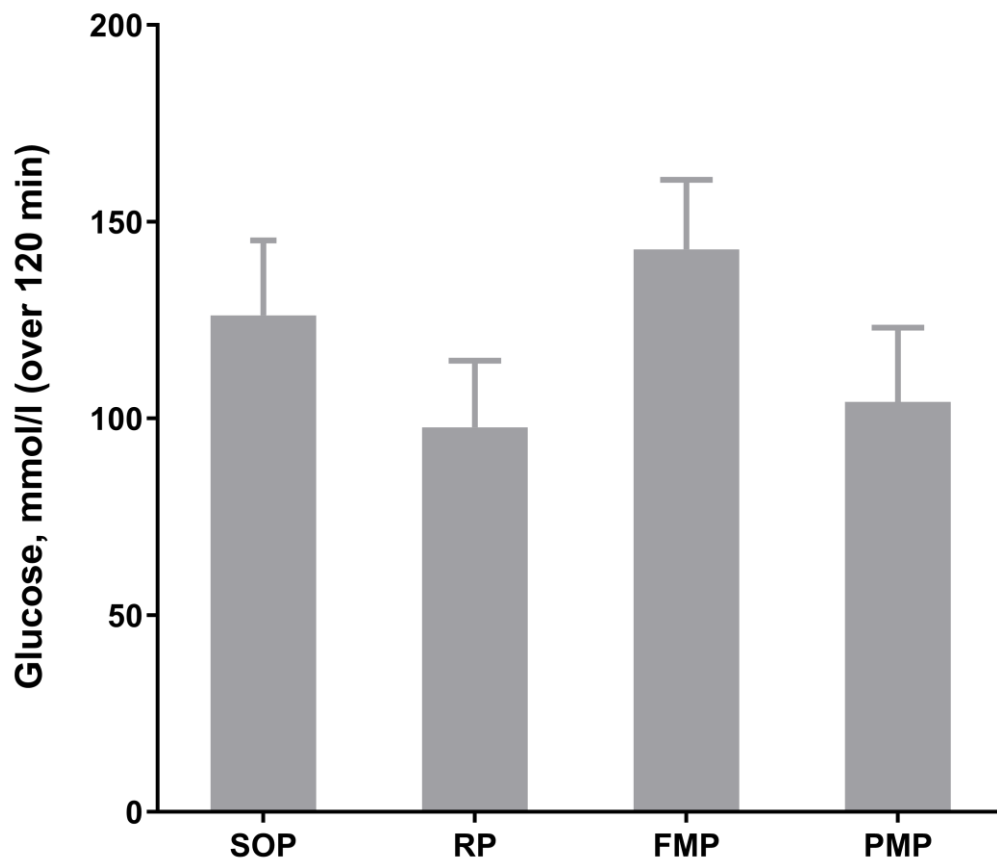
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686 Figure 6



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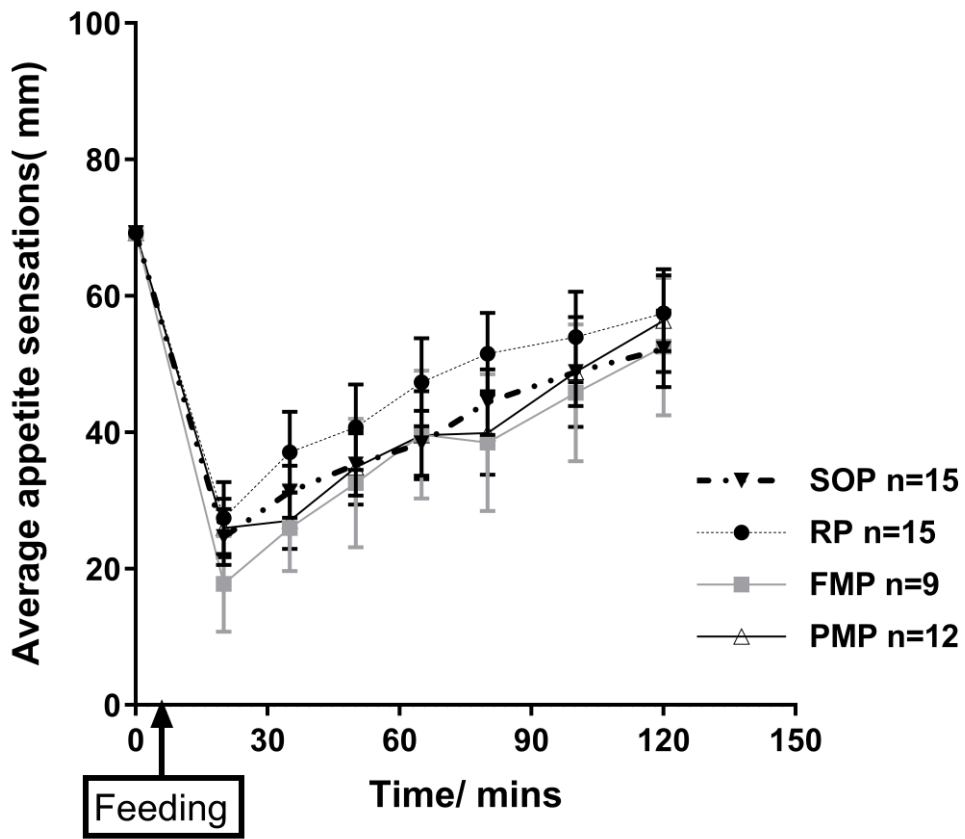
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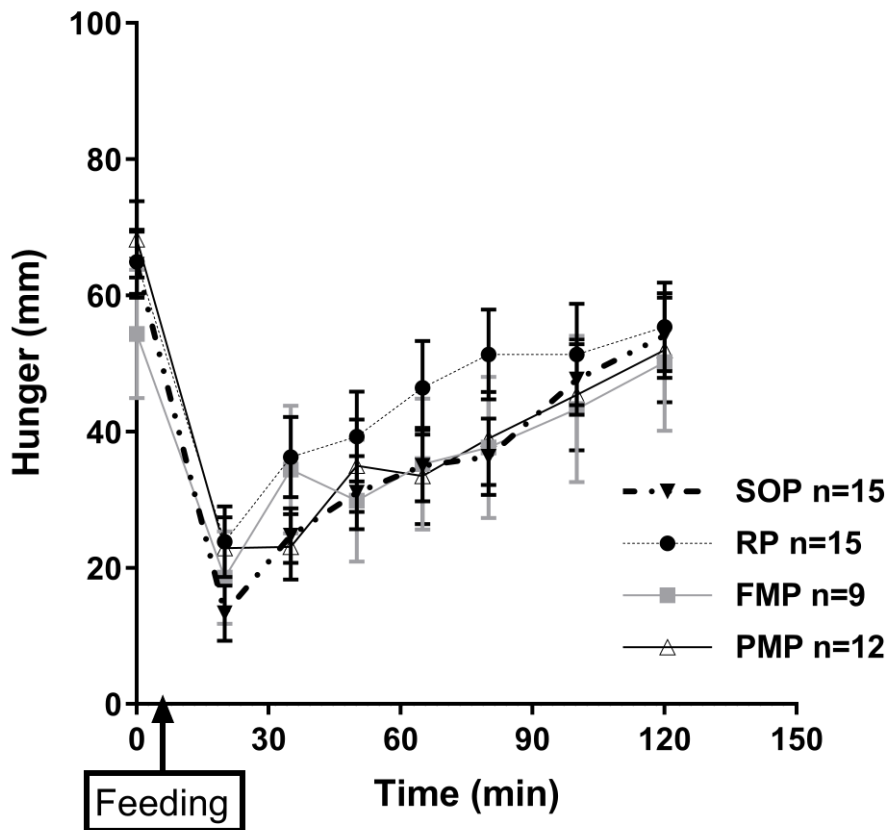
692 Figure 7



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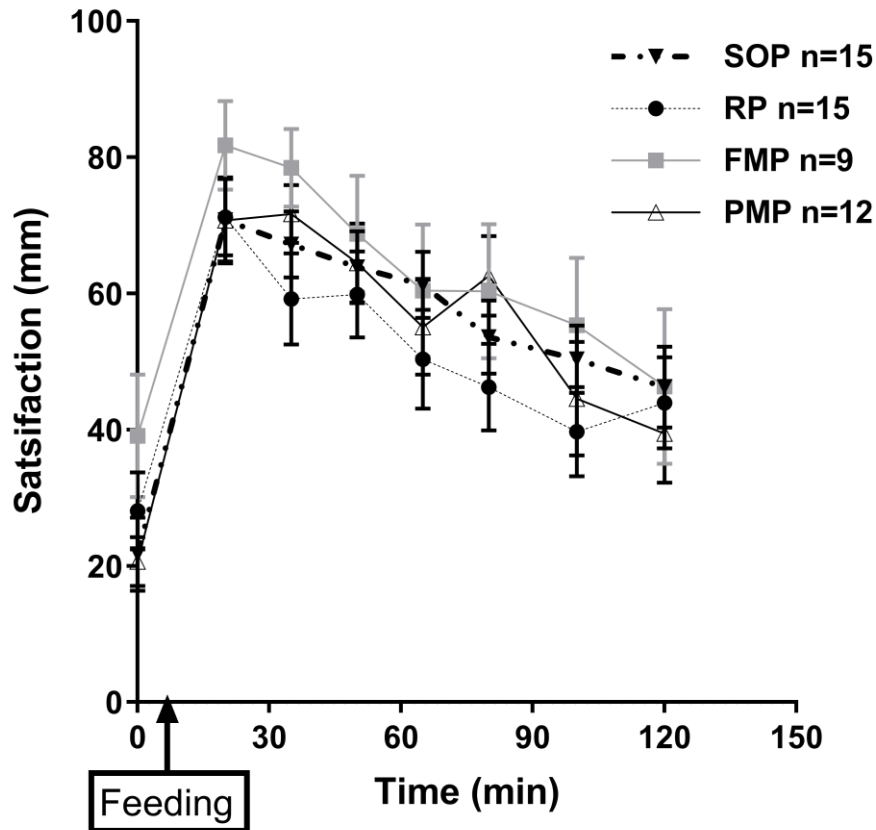
705 **Supplementary material**

706 **Supplementary Figure 1.** Plot of hunger for healthy participants after they consumed the four
707 different study porridges. -•▼-, Scottish oats porridge (SOP); -●-, Rye porridge (RP); -■-,
708 Finger millet porridge (FMP); -△-, Pearl millet porridge (PMP). Values are mean ± SE, n=15 for
709 SOP and RP, n=9 for FMP and n=12 for PMP. The arrow on the horizontal axis indicates the meal
710 start time.



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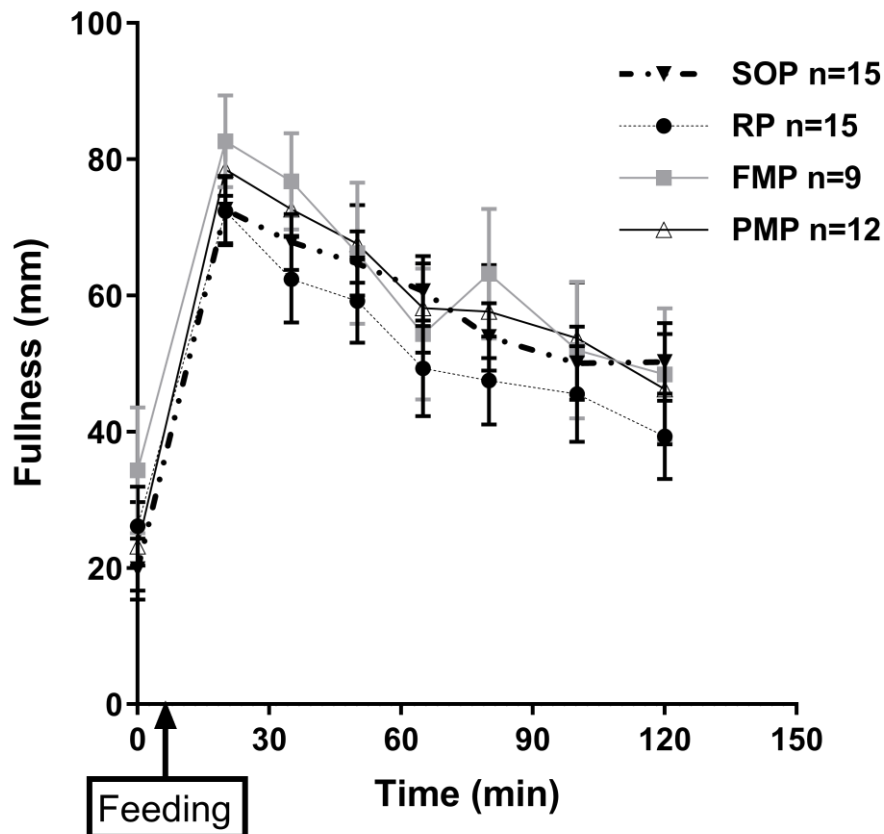
716 **Supplementary Figure 2.** Plot of satisfaction for healthy participants after they consumed the four
717 different study porridges. -·▼-, Scottish oats porridge (SOP); ···●···, Rye porridge (RP); —■—,
718 Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=15 for
719 SOP and RP, n=9 for FMP and n=12 for PMP. The arrow on the horizontal axis indicates the meal
720 start time.



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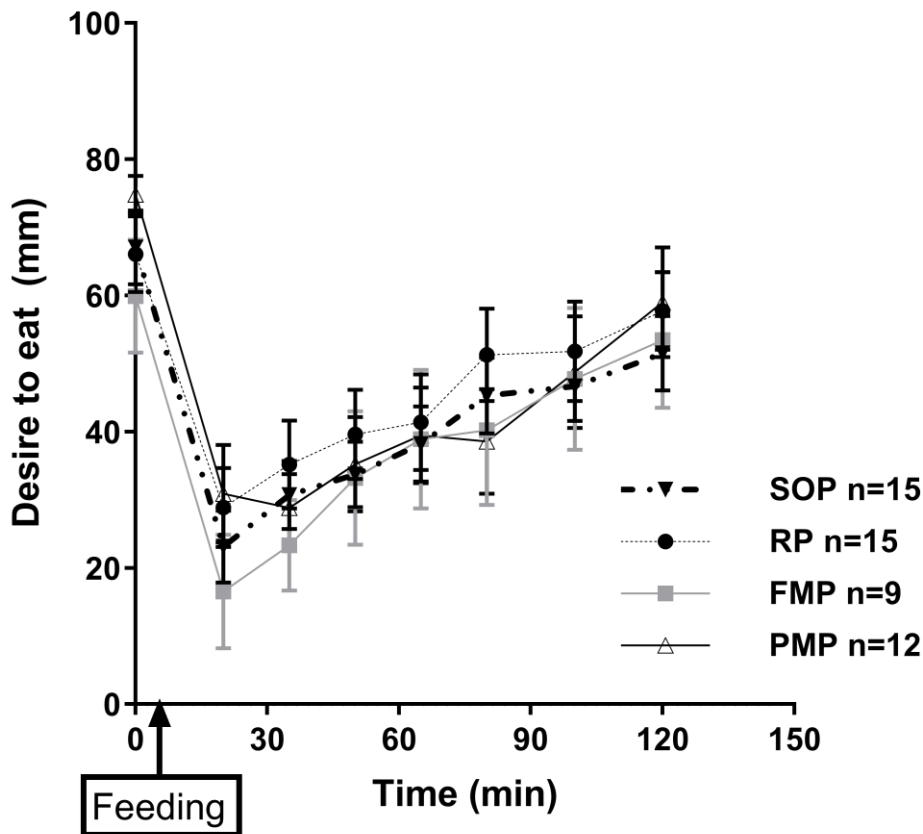
723 **Supplementary Figure 3.** Plot of fullness for healthy participants after they consumed the four
724 different study porridges. -·▼-, Scottish oats porridge (SOP); ···●···, Rye porridge (RP); —■—,
725 Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=15 for
726 SOP and RP, n=9 for FMP and n=12 for PMP. The arrow on the horizontal axis indicates the meal
727 start time.



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735 **Supplementary Figure 4.** Plot of desire to eat for healthy participants after they consumed the four
736 different study porridges. -·▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —■—,
737 Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=15 for
738 SOP and RP, n=9 for FMP and n=12 for PMP. The arrow on the horizontal axis indicates the meal
739 start time.

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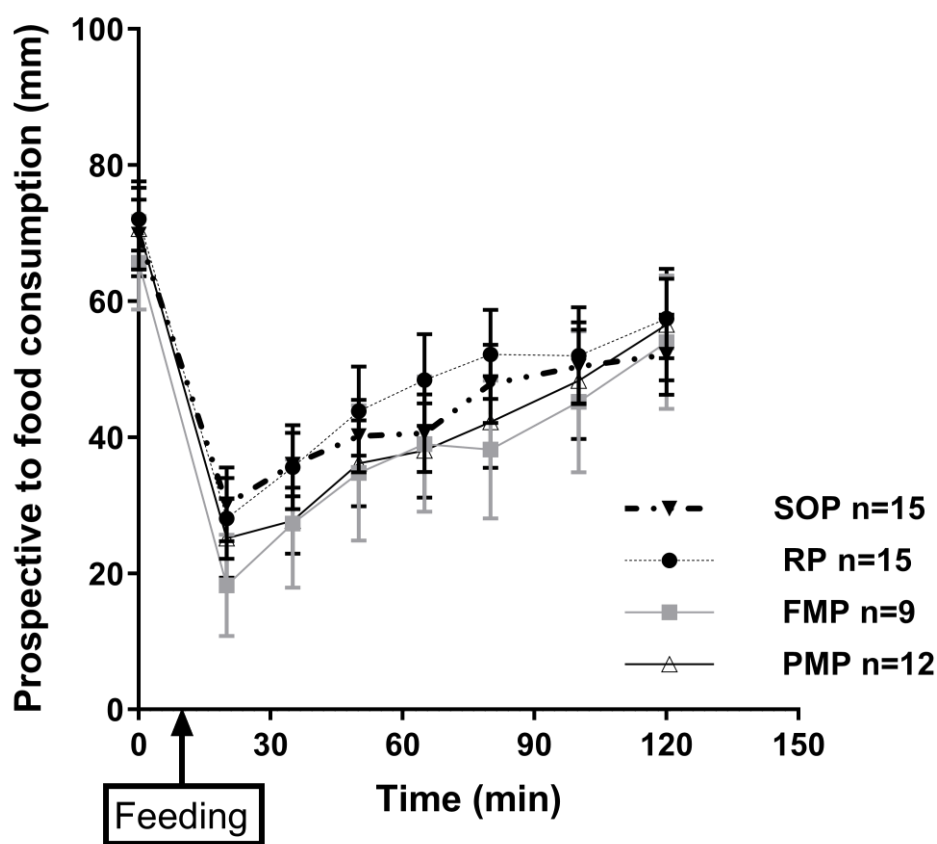
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743 **Supplementary Figure 5.** Plot of prospective food consumption for healthy participants after they
744 consumed the four different study porridges. -•-•-, Scottish oats porridge (SOP);●....., Rye
745 porridge (RP); —■—, Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are
746 mean \pm SE, n=15 for SOP and RP, n=9 for FMP and n=12 for PMP. The arrow on the horizontal axis
747 indicates the meal start time.

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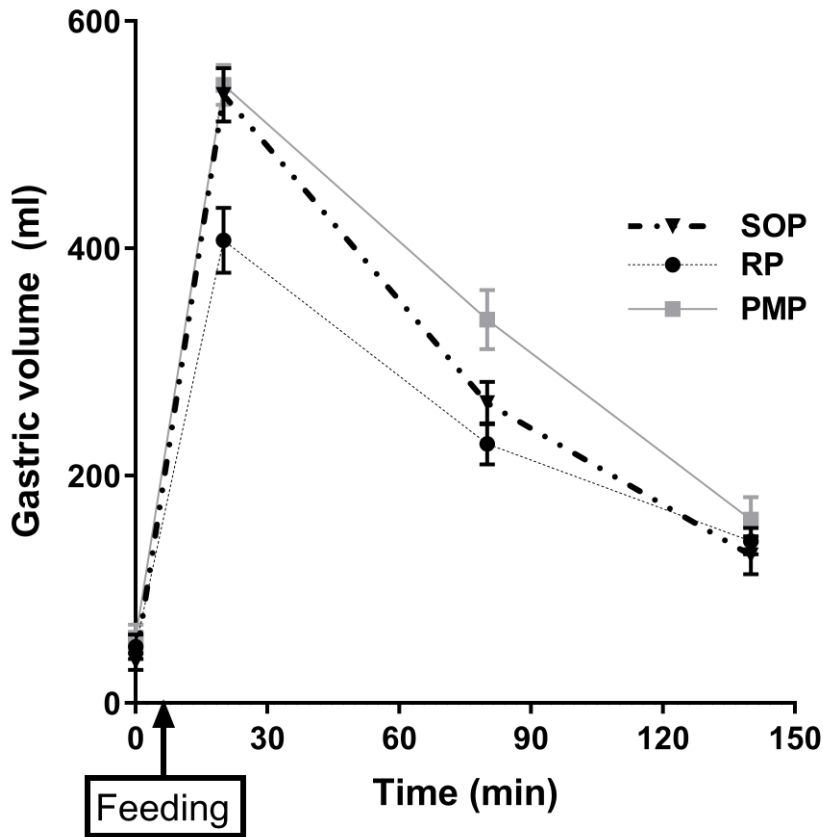
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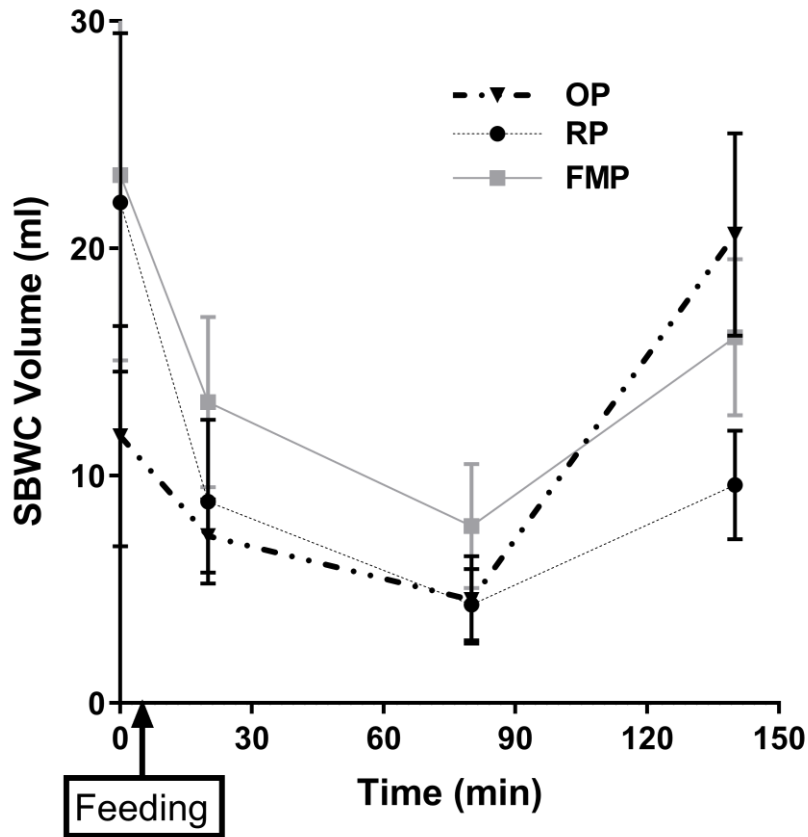
754 **Supplementary Figure 6.** Plot of the volume of the gastric contents for healthy participants after
755 they consumed the three different study porridges. -·▼-, Scottish oats porridge (SOP);●....., Rye
756 porridge (RP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the
757 horizontal axis indicates the meal start time.



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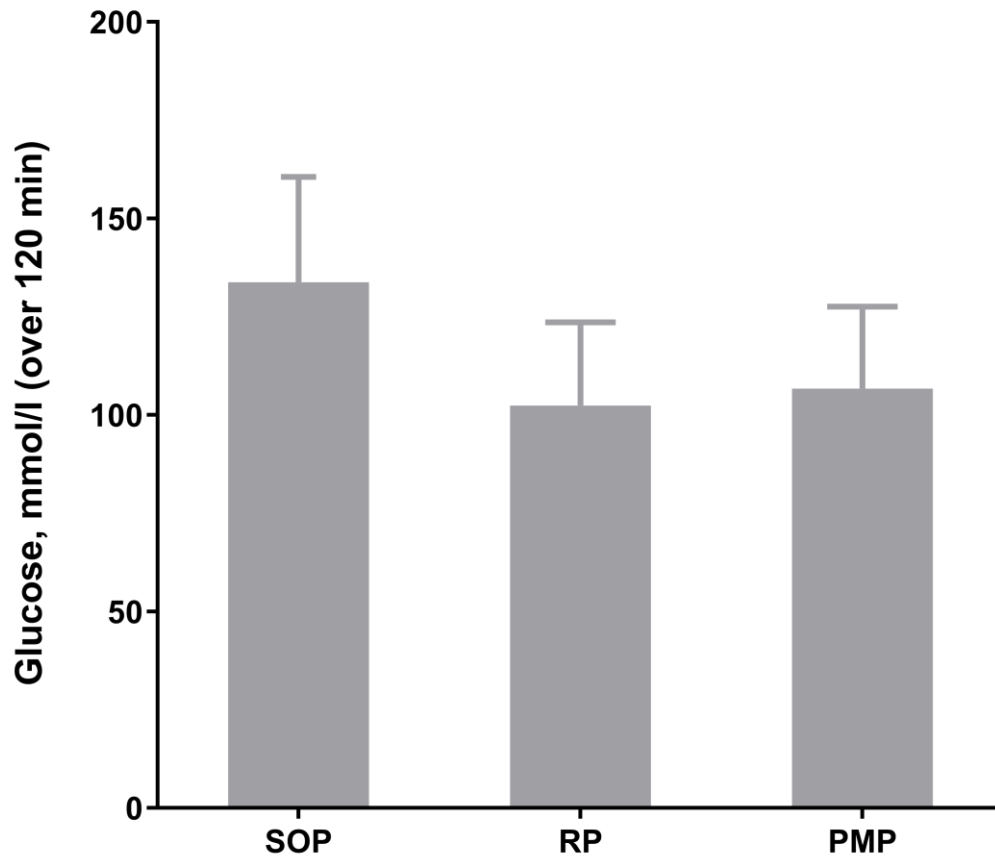
760 **Supplementary Figure 7.** Plot of the volume of the small bowel water content for healthy
761 participants after they consumed the three different study porridges. -•▼-, Scottish oats porridge
762 (SOP); -●-, Rye porridge (RP); -△-, Pearl millet porridge (PMP). Values are mean ± SE, n=10.
763 The arrow on the horizontal axis indicates the meal start time.



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766 **Supplementary Figure 8.** Incremental area under the glucose curve (iAUC) for healthy participants
767 after they consumed the three different study porridges., Scottish oats porridge (SOP); Rye porridge
768 (RP); Pearl millet porridge (PMP). Values are mean \pm SE, n=10

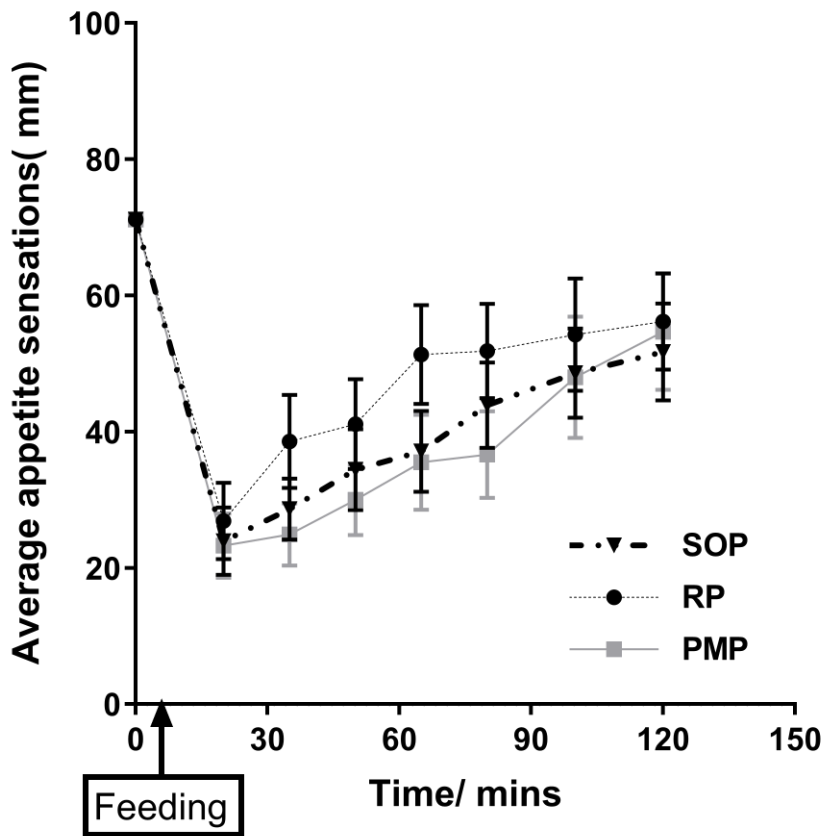


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771 **Supplementary Figure 9.** Plot of the average appetite sensations for healthy participants after they
772 consumed the three different study porridges. -•-•-, Scottish oats porridge (SOP);●....., Rye
773 porridge (RP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the
774 horizontal axis indicates the meal start time.

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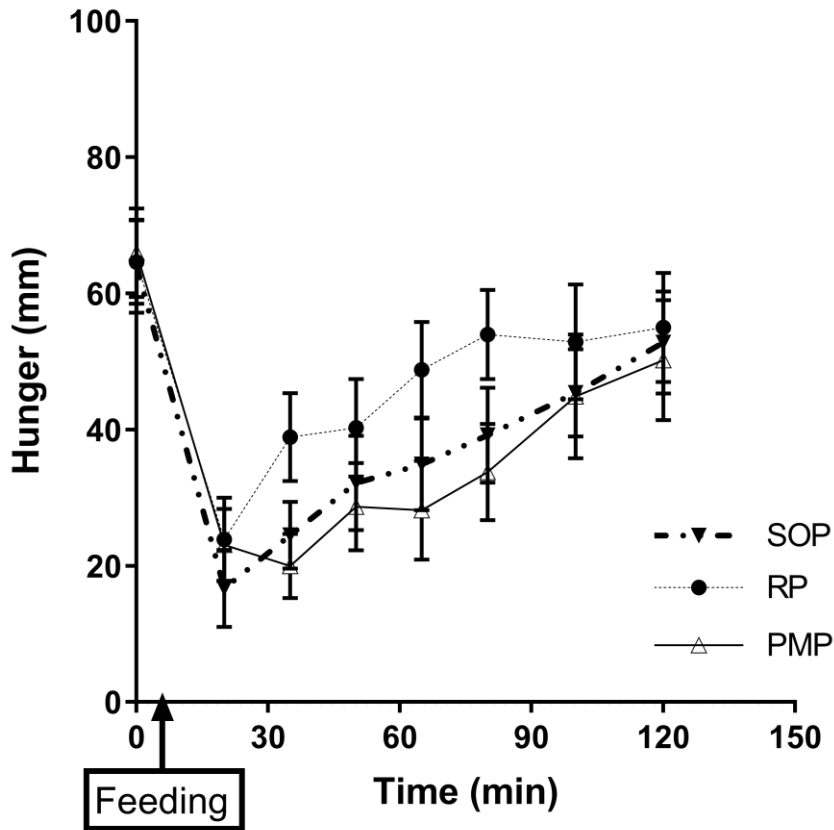


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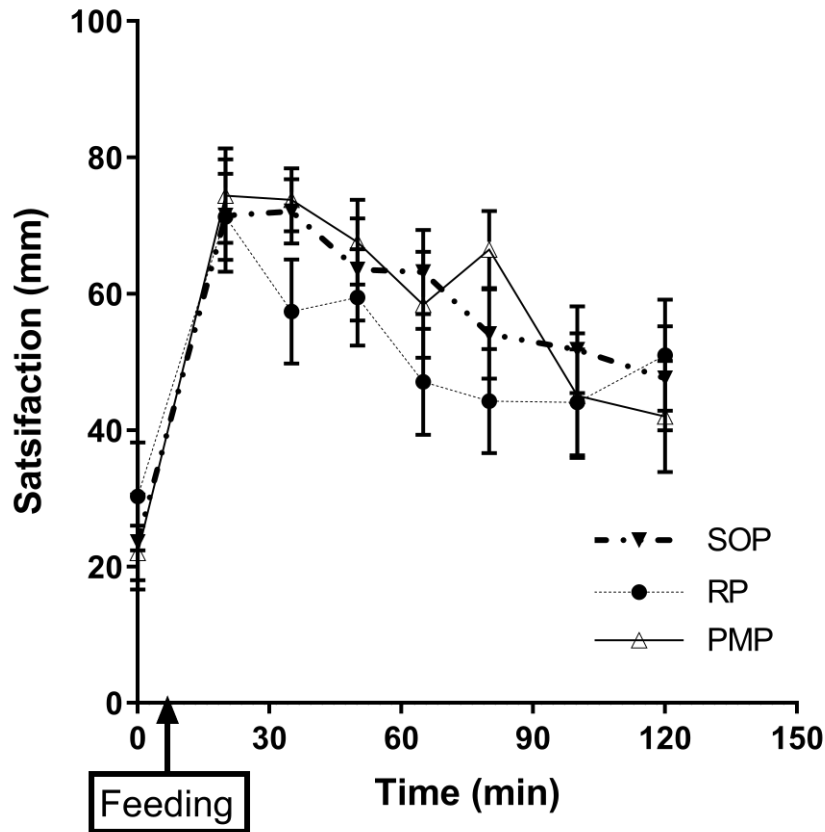
779 **Supplementary Figure 10.** Plot of hunger for healthy participants after they consumed the three
780 different study porridges. -·▼-, Scottish oats porridge (SOP); ···●···, Rye porridge (RP); —△—,
781 Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the horizontal axis indicates
782 the meal start time.



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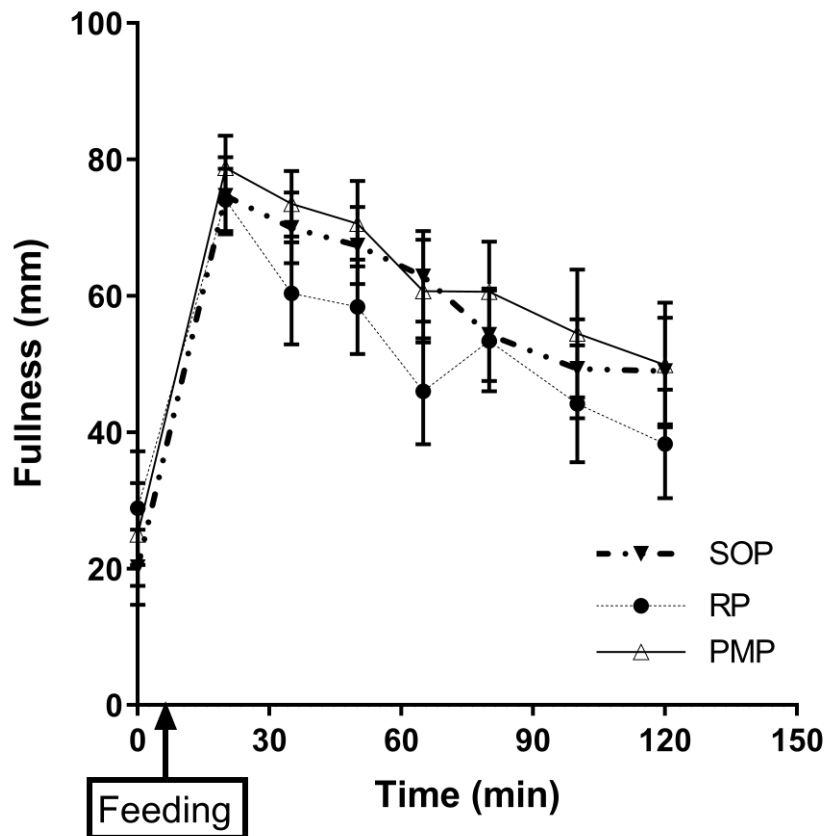
785 **Supplementary Figure 11.** Plot of satisfaction for healthy participants after they consumed the three
786 different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —△—,
787 Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the horizontal axis indicates
788 the meal start time.



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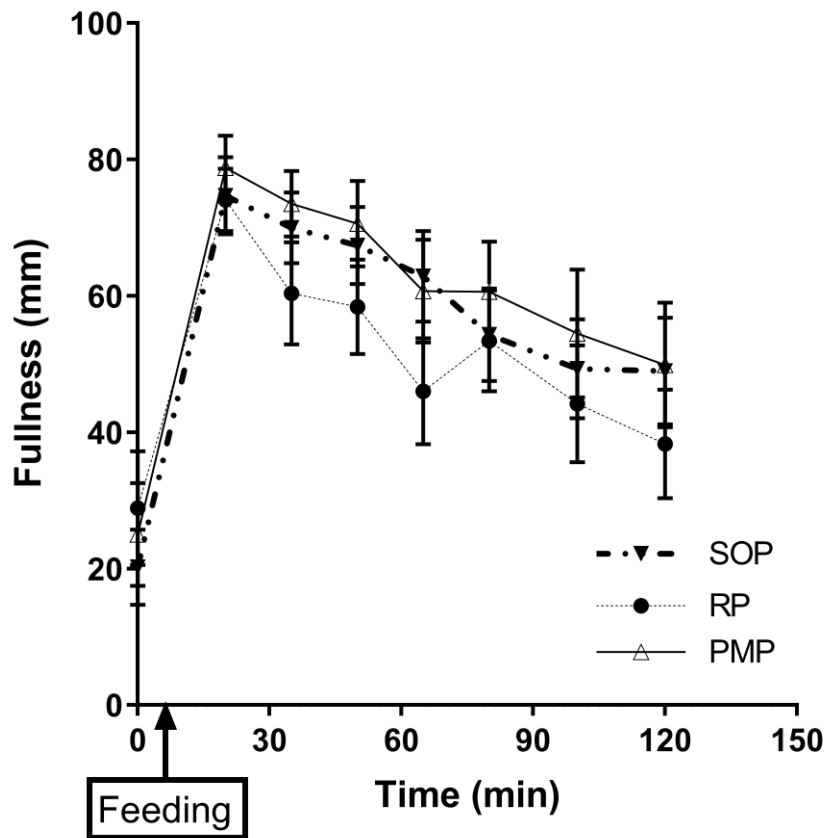
791 **Supplementary Figure 12.** Plot of fullness for healthy participants after they consumed the three
792 different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —△—,
793 Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the horizontal axis indicates
794 the meal start time.



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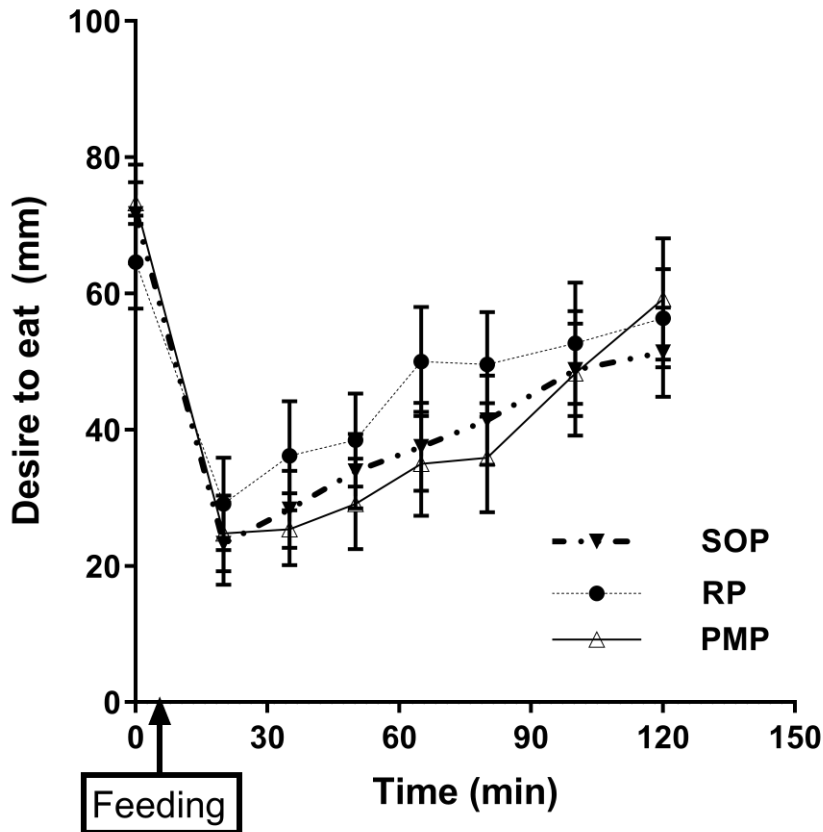
797 **Supplementary Figure 12.** Plot of fullness for healthy participants after they consumed the three
798 different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —△—,
799 Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the horizontal axis indicates
800 the meal start time.



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803 **Supplementary Figure 13.** Plot of desire to eat for healthy participants after they consumed the three
804 different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP), —△—,
805 Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the horizontal axis indicates
806 the meal start time.

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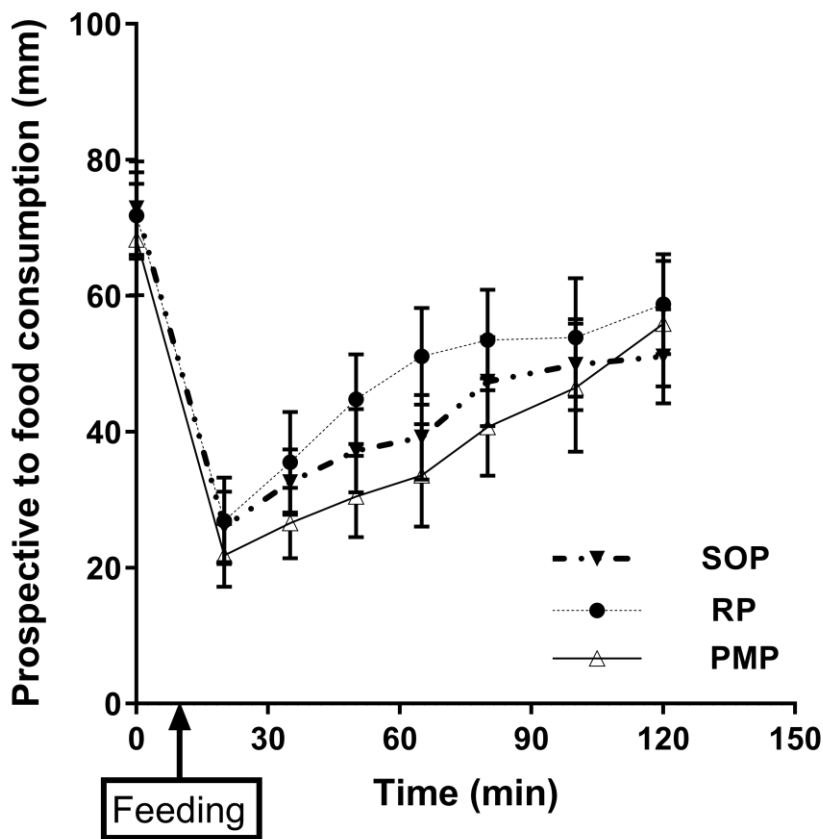
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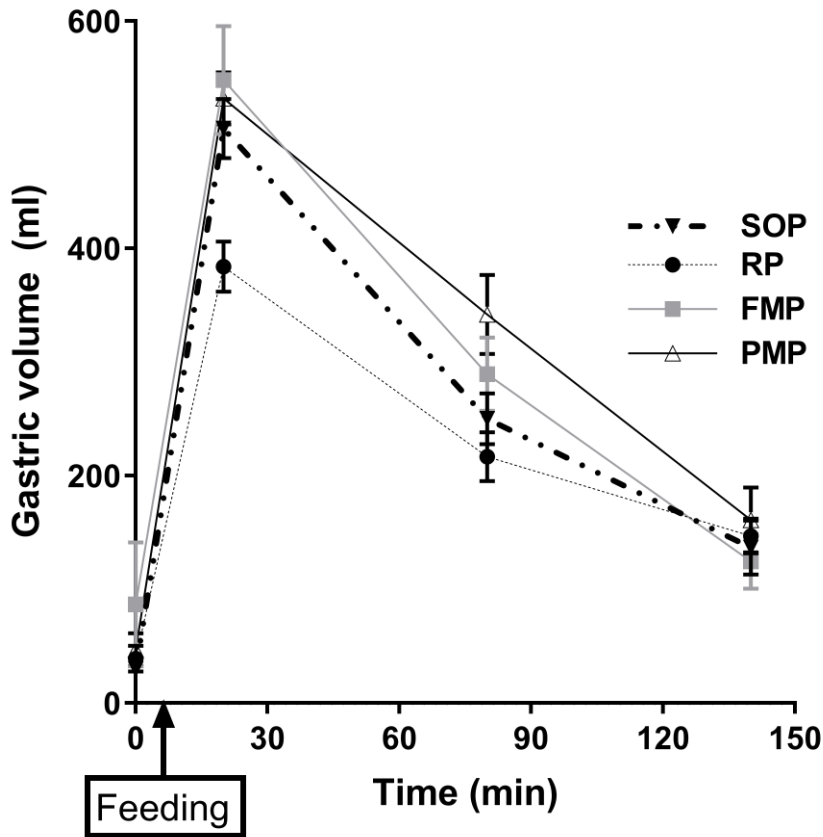
813 **Supplementary Figure 14.** Plot of prospective food consumption for healthy participants after they
814 consumed the three different study porridges. -•-•-, Scottish oats porridge (SOP);●....., Rye
815 porridge (RP), —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=10. The arrow on the
816 horizontal axis indicates the meal start time.

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824 **Supplementary Figure 15.** Plot of the volume of the gastric contents for healthy participants after
825 they consumed the four different study porridges. -·▼-, Scottish oats porridge (SOP); ····●····, Rye
826 porridge (RP); —■—, Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are
827 mean ± SE, n=7. The arrow on the horizontal axis indicates the meal start time.

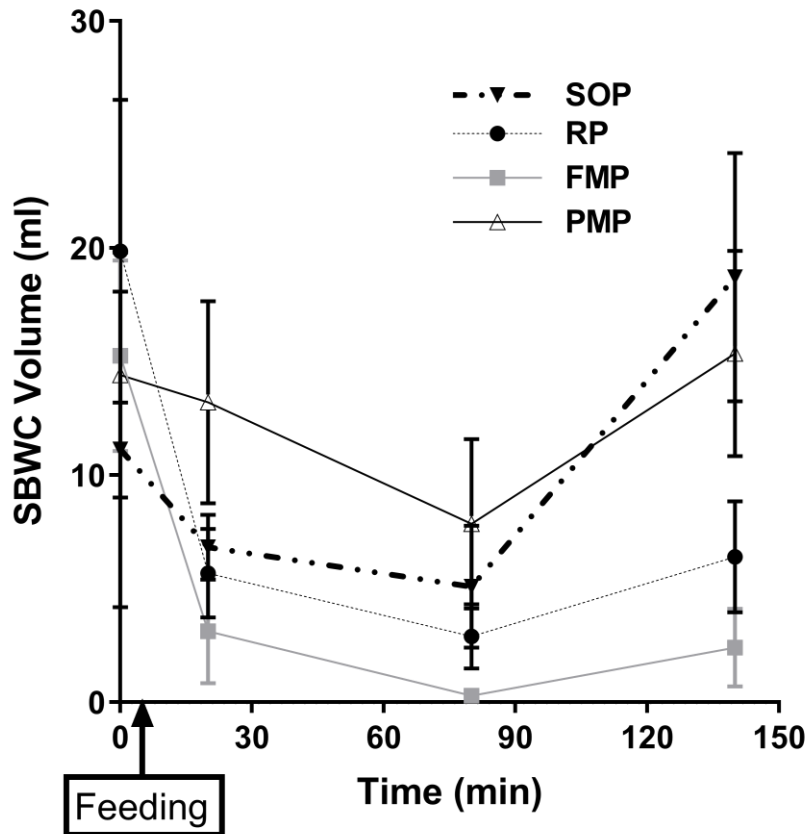


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831 **Supplementary Figure 16.** Plot of the volume of the small bowel water content for healthy
832 participants after they consumed the four different study porridges. $\text{---}\blacktriangledown\text{---}$, Scottish oats porridge
833 (SOP); $\text{---}\bullet\text{---}$, Rye porridge (RP); $\text{---}\blacksquare\text{---}$, Finger millet porridge (FMP); $\text{---}\triangle\text{---}$, Pearl millet porridge
834 (PMP). Values are mean \pm SE, n=7. The arrow on the horizontal axis indicates the meal start time.

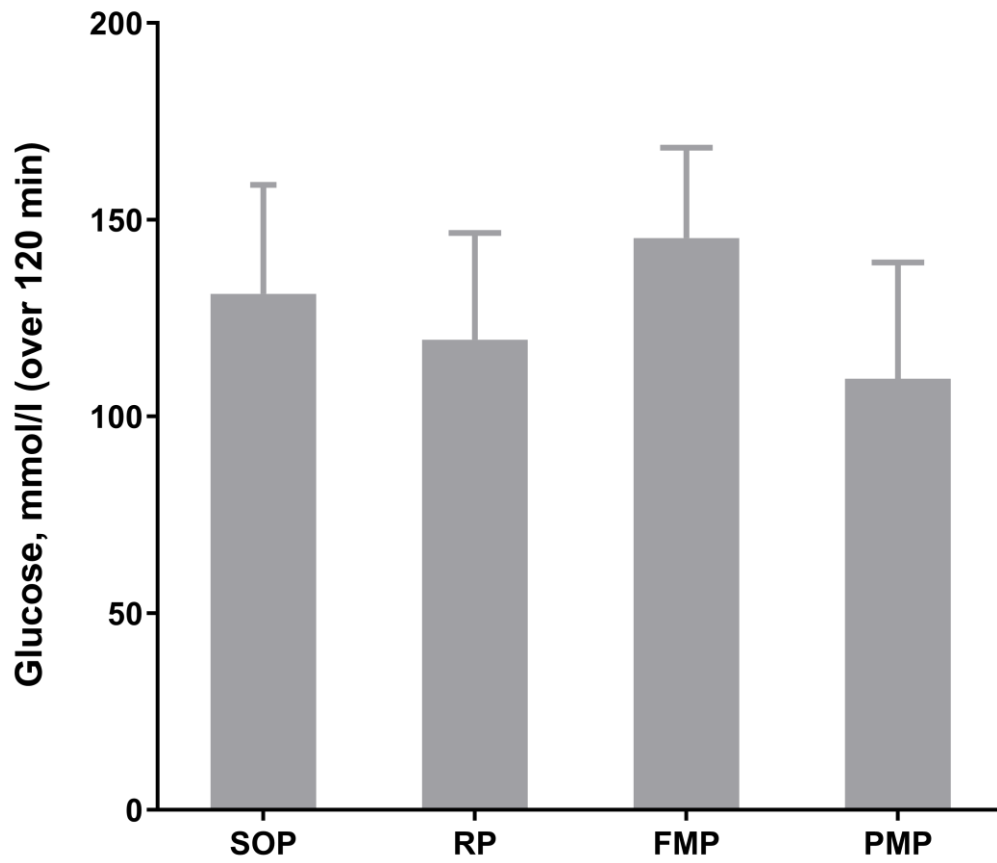


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837 **Supplementary Figure 17.** Incremental area under the glucose curve (iAUC) for healthy participants
838 after they consumed the four different study porridges., Scottish oats porridge (SOP); Rye porridge
839 (RP); Finger millet porridge (FMP); Pearl millet porridge (PMP). Values are mean \pm SE, n=7

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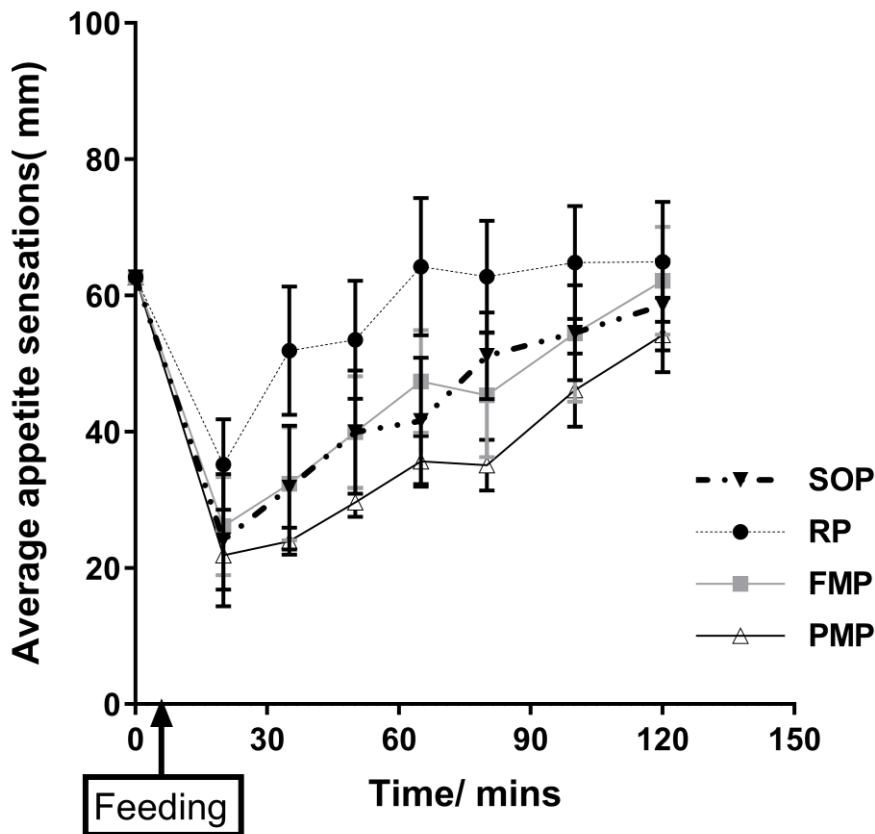
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844 **Supplementary Figure 18.** Plot of the average appetite sensations for healthy participants after they
845 consumed the four different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye
846 porridge (RP); —■—, Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are
847 mean ± SE, n=7. The arrow on the horizontal axis indicates the meal start time.

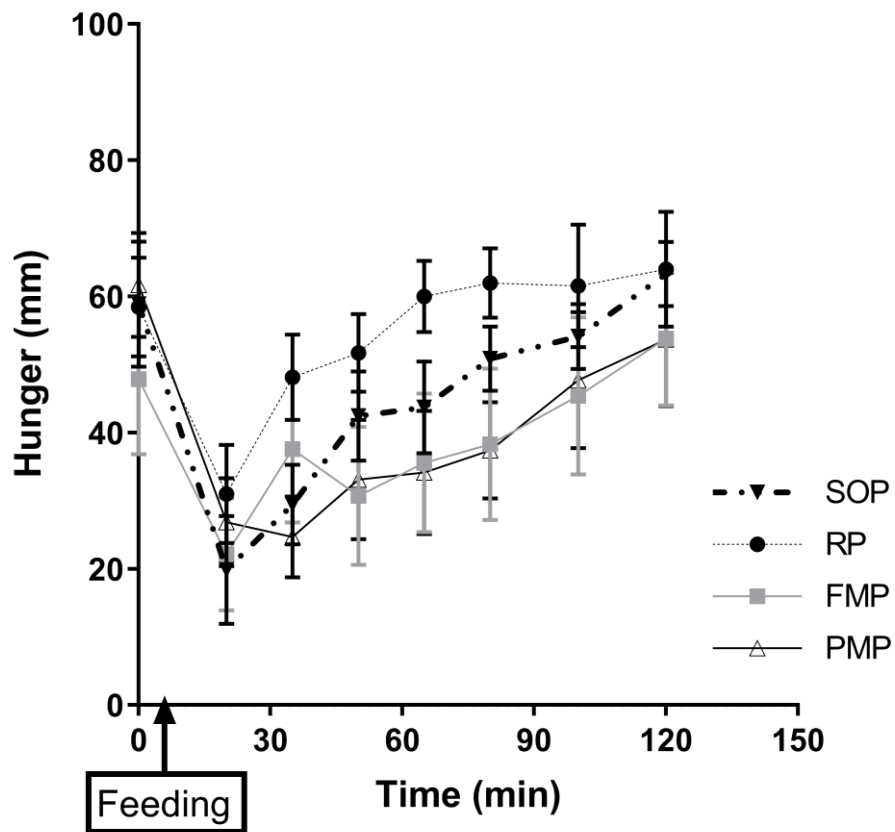
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851 **Supplementary Figure 19.** Plot of the hunger for healthy participants after they consumed the four
852 different study porridges. -•▼-, Scottish oatsporridge (SOP);●....., Rye porridge (RP); —■—,
853 Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=7. The
854 arrow on the horizontal axis indicates the meal start time.



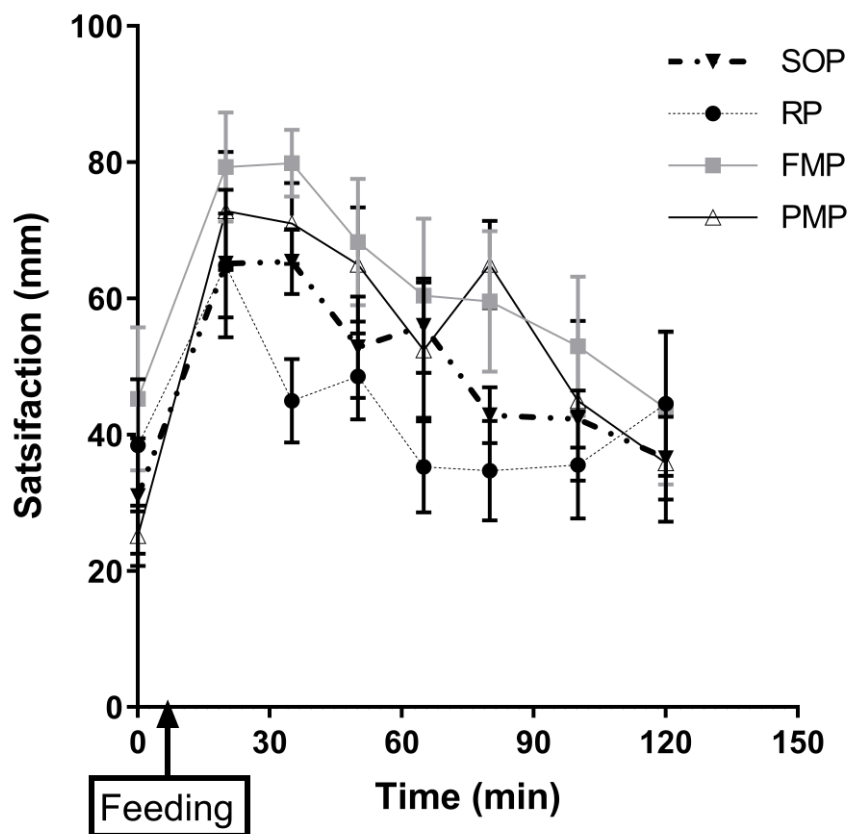
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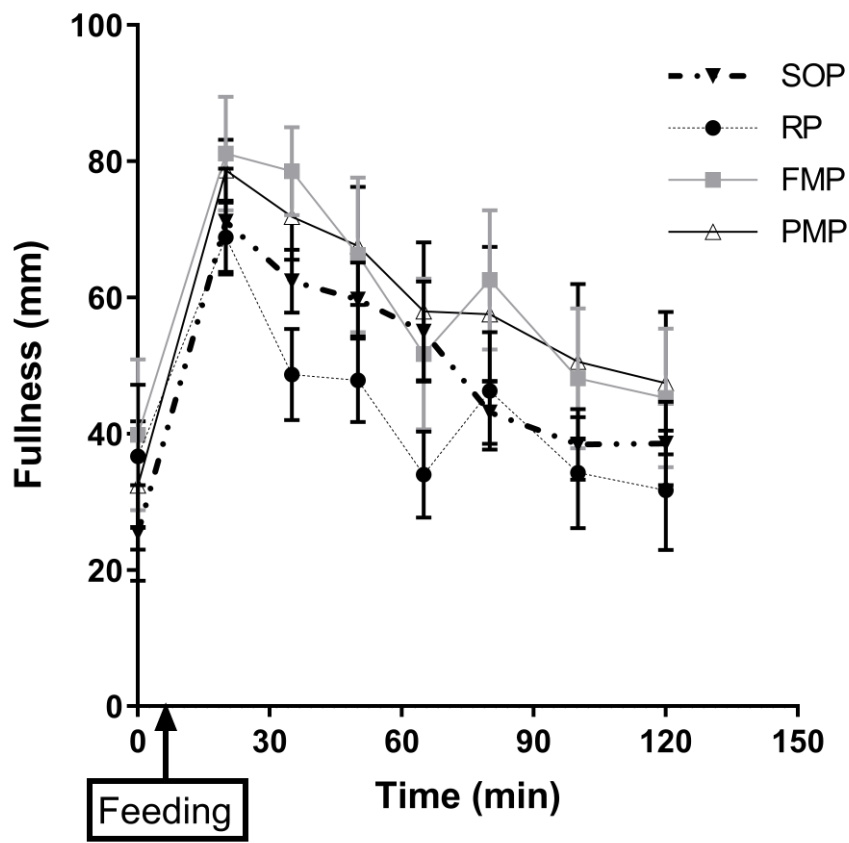
864 **Supplementary Figure 20.** Plot of the satisfaction for healthy participants after they consumed the
865 four different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —■—
866 , Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=7. The
867 arrow on the horizontal axis indicates the meal start time.



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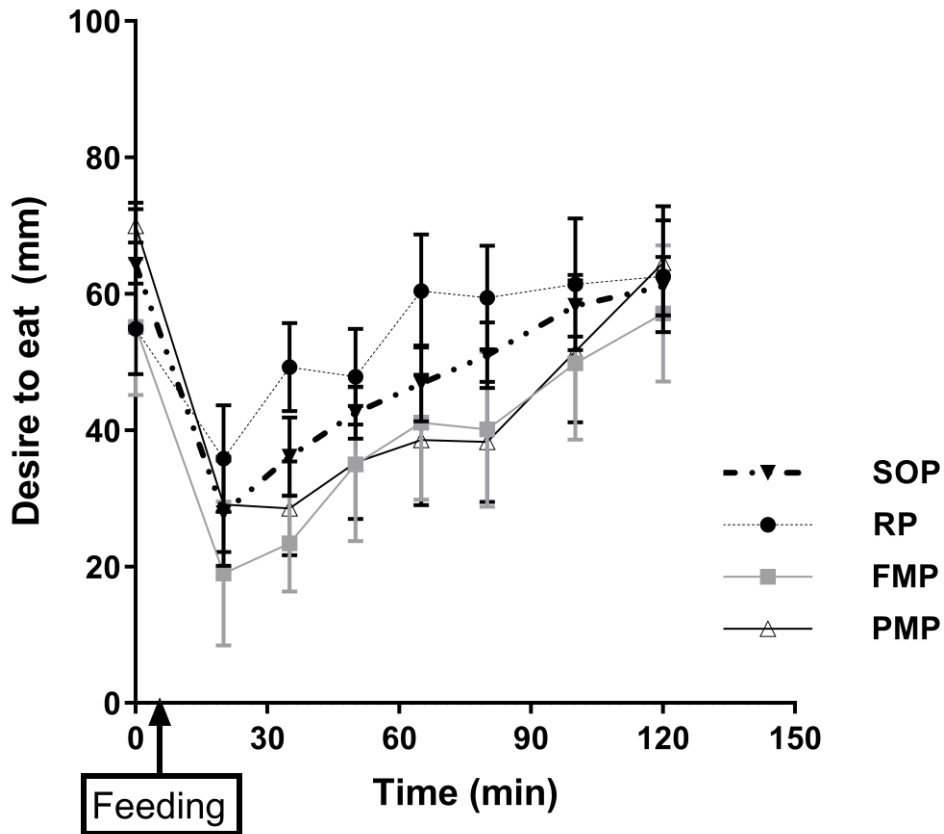
874 **Supplementary Figure 21.** Plot of fullness for healthy participants after they consumed the four
875 different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —■—,
876 Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=7. The
877 arrow on the horizontal axis indicates the meal start time.

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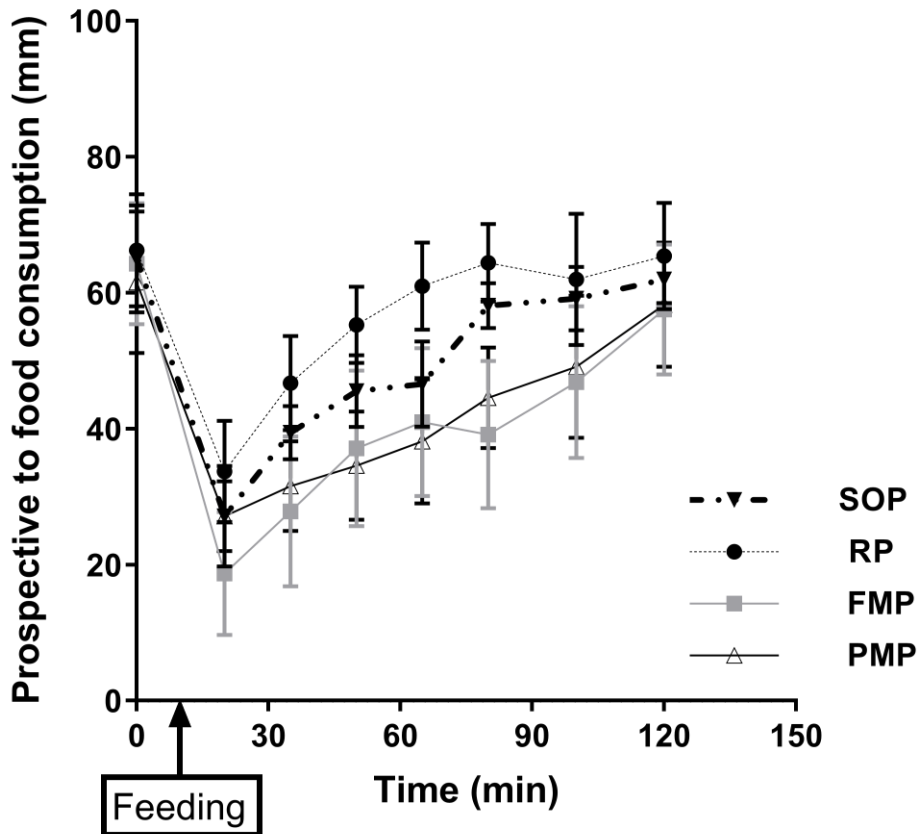
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882 **Supplementary Figure 22.** Plot of desire to eat for healthy participants after they consumed the four
883 different study porridges. -•▼-, Scottish oats porridge (SOP);●....., Rye porridge (RP); —■—,
884 Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are mean ± SE, n=7. The
885 arrow on the horizontal axis indicates the meal start time.



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892 **Supplementary Figure 23.** Plot of prospective food consumption for healthy participants after they
893 consumed the four different study porridges. -·▼-, Sc Scottish oats porridge (SOP);●....., Rye
894 porridge (RP); —■—, Finger millet porridge (FMP); —△—, Pearl millet porridge (PMP). Values are
895 mean ± SE, n=7. The arrow on the horizontal axis indicates the meal start time.
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