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HARDEN, Charlotte, RICHARDSON, J Craig, DETTMAR, Peter W, CORFE, Bernard M and PAXMAN, Jenny <http://orcid.org/0000-0003-3596-489X>

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AN IONIC-GELLING ALGINATE DRINK ATTENUATES POSTPRANDIAL GLYCAEMIA IN MALES

Charlotte J. Harden\textsuperscript{a}, J. Craig Richardson\textsuperscript{b}, Peter W. Dettmar\textsuperscript{b}, Bernard M. Corfe\textsuperscript{c*}, Jenny R Paxman\textsuperscript{a}

\textsuperscript{a} Centre for Food Innovation, Sheffield Hallam University, Howard St, Sheffield, S1 1WB, UK.
\textsuperscript{b} Technostics Limited, The Deep Business Centre, Tower Street, Kingston Upon Hull, HU1 4BG, UK.
\textsuperscript{c} Department of Oncology, The University of Sheffield, The Medical School, Beech Hill Road, Sheffield, S10 2RX, UK.

* Corresponding author: Dr Bernard Corfe, Department of Oncology, The University of Sheffield, The Medical School, Beech Hill Road, Sheffield, S10 2RX, UK,

\textit{E-mail address:} b.m.corfe@sheffield.ac.uk

\textit{Telephone:} +44 (0)114 271 3004

\textit{Fax:} +44 (0)114 271 3314
ABSTRACT

Obese individuals are at increased risk of type 2 diabetes compared to their healthy weight counterparts. Dietary fibre, such as alginate, could attenuate glycaemic disturbances associated with obesity when included in the diet.

Forty self-reported, healthy males completed this randomised, single-blinded, controlled, parallel trial to determine the glycaemic response to a controlled test-lunch of mixed composition following an ionic-gelling alginate preload drink compared to an acidic-gelling control. Individual baseline area under the curve was 52% lower (P=0.010) and peak glycaemia was 14% lower (P<0.0005) after the ionic-gelling alginate drink compared with the control. Body fatness was a predictor of postprandial glycaemia however there was no interaction effect between body fat % and treatment type.

We have shown ionic-gelling alginate can attenuate glycaemic response to set lunch of mixed composition. Functional foods that include ionic-gelling alginates may benefit those with elevated postprandial blood glucose.

KEY WORDS
Alginate; glucose; glycemia; gel; body fat
1.0 INTRODUCTION

As obesity increases, the incidence of associated co-morbidities rises concomitantly, most dramatically in relation to body mass index-related diabetes (McPherson et al., 2007). Abdominal fatness has been linked with elevated fasting blood glucose (Rezende et al., 2006). Pascot et al. (1999) showed visceral adipose tissue accumulation was accompanied by increased plasma glucose in the fasted state and after a 75g oral glucose load in young and middle aged women. In a six year prospective study Kriketos et al. (2003) showed baseline body fatness and increasing fatness over time to be strong predictors of elevated fasting plasma glucose in individuals ‘at-risk’ of type 2 diabetes.

Epidemiological evidence suggests dietary fibres may have a preventive role in the development of type 2 diabetes (Meyer et al., 2000). Several mechanisms by which soluble fibres may modulate glycaemic response have been proposed (Augustin et al., 2000). Soluble fibre ingestion reduces carbohydrate digestion rates, therefore aiding regulation of postprandial glycaemia (Augustin et al., 2000; Kimura et al., 1996; Welch, 1994).
Soluble fibres have been shown to have beneficial effects in controlling glycaemia following carbohydrate ingestion in healthy volunteers (Goñi et al., 2000; Rigaud et al., 1998; Lavin and Read, 1995). Similarly, fibre-rich foods (Flammang et al., 2006) and fibre supplementation (Sierra et al., 2002) have been shown to help attenuate postprandial glycaemic responses in type 2 diabetic adults. Kaline et al. (2007) reviewed the potential mechanisms by which diets rich in dietary fibre can be useful in diabetes prevention.

Alginate is an algal polysaccharide found in the cell walls of certain brown seaweed species. This fibre has been used in several relevant human intervention studies. 5.0g of sodium alginate added to a meal significantly attenuated postprandial glycaemic response in type 2 diabetics by 31% compared to the control meal (Torsdottir et al., 1991). Wolf and colleagues (2002) demonstrated that 1.5g of sodium alginate, incorporated into a 100g glucose-based preload drink with an acid-soluble calcium source (to produce an acid-induced viscosity complex), elicited a non-significant drop in peak glycaemia and a significant attenuation of incremental change from baseline area under the curve (AUC) in healthy, non-diabetic adults compared to a soluble fibre-based control. Williams et al. (2004) fed a "crispy bar"
containing 5.5g guar gum and 1.6g sodium alginate to healthy adults and measured the resultant glycaemic response compared to an alginate-free bar. Postprandial blood glucose excursions were significantly lower at 15, 30, 45, and 120 minutes and the positive incremental AUC was significantly reduced (by 33%) after consumption of the enriched “crispy bar” compared to the alginate-free bar. Paxman et al. (2008a) reported a strong positive correlation between change from individual baseline AUC glycaemia and body fat % when a hypromellose control preload was ingested prior to a test lunch. This positive correlation was not apparent following an ionic gelling sodium alginate preload, providing preliminary evidence to suggest that the enhanced glycaemic response to a meal at higher body fat could be normalised following ingestion of an alginate preload identical to the one used in the present study.

Hoad et al. (2004) fed volunteers a strong gelling (high-G) and a weaker gelling (low-G) alginate meal, a guar-based meal or a control (without added fibre) and examined the resultant gastric emptying rates. In vitro, both alginate meals formed intragastric gel 'lumps', and in the case of the strong-gelling alginate, this was reportedly associated with a feeling of fullness and a reduction in hunger. Hoad and colleagues (2004) purport that acid-gelling agents
such as alginate may be usefully incorporated into weight-reducing diets/foods in order to enhance antrum distension and/or manipulate nutrient uptake from the ileum.

Alginate is widely used in the food industry as a thickener, stabiliser and gelling agent (Brownlee et al., 2005). Its constituent sugar residues are D-mannuronic (M) and L-guluronic acid (G). Homopolymeric G blocks (comprising diaxial linkages in the $^{1}C_4$ conformation) can react with $\text{Ca}^{2+}$ and $\text{H}^{+}$ ions to yield a strong, cross-linked gel (Brownlee et al., 2005; Seal and Mathers, 2001; Kimura et al, 1996). Consequently, the gel strength of alginate and its consequent biochemical and biophysical properties are determined by its chemical structure. Specific alginates and specific alginate formulations are therefore likely to react differently within the gastrointestinal milieu.

The primary objective of the present study was to examine the effect of alginate gelled ionically compared to acidically (control) on glycaemic response to a standard meal of mixed composition. Secondary to this, we investigated how body fatness affects the postprandial glycaemic response when subjects ingest the ionic-gelling formulation compared to the acid-gelling control.
2.0 MATERIALS AND METHODS

2.1 Subjects

41 male subjects participated in the study. Only one subject was excluded, due to unusually low fasting glucose levels, leaving complete datasets for 40 participants. Subjects aged 18 to 65 years were eligible to take part providing they did not meet any of the criteria for exclusion which were; type 1 or 2 diabetes, history of, or current cardiovascular complaints (or if they had been fitted with a pacemaker or other implantable electronic device) or gastrointestinal complaints (such as irritable bowel syndrome or inflammatory bowel disorder, dumping syndrome or Cushing’s syndrome), current fibre supplement use, use of constipation-causing drugs such as codeine or morphine, bowel blockage, bowel muscle weakness or recent food poisoning. In addition, anyone with a known allergy to, or intolerance of, the foods or ingredients used in the experiment was excluded from taking part, as were vegans (due to the nature of the foods used).
Baseline pre-screening took place less than one week prior to the experimental phase, in which subjects completed a general health questionnaire and various anthropometric measures were made. Height and weight were recorded (SECA 709 mechanical column scales with SECA 220 telescopic measuring rod; SECA United Kingdom, Birmingham) and body mass index (BMI) was calculated. Bioelectrical impedance analysis was undertaken following 5 minutes of supine rest on non-conducting foam matting using a BodyStat 1500 (BodyStat Ltd., Isle of Man, British Isles). Body fat % was recorded. Subjects completed a 51-item Three Factor Eating Questionnaire (TFEQ; Stunkard and Messick, 1985) to determine eating behaviour across three pre-defined factors. Mean values for all three factors; restraint, disinhibition and hunger, were low for the group as a whole (Stunkard and Messick, 1985). Subject characteristics are reported in Table 1. This study was approved by the relevant University Ethics Committee (Ref: FIRC/2006/RE21). All subjects gave informed consent to participate.

2.2 Study Design

In this randomised, single-blinded, controlled parallel trial subjects (n = 40) were split equally either side of the
median into haptiles by body fatness (lower body fat group: <16.10%, upper body fat group: ≥16.10%).

Following a 12 hour overnight fast, all subjects consumed a controlled breakfast at 9am (60g Kellogg’s® Hint of Honey Corn Flakes; Kellogg’s Company GB Limited, Manchester, 125ml semi-skimmed milk and 200ml ‘Drink Fresh’ orange juice; DCB Foodservice, Herts). After breakfast subjects were asked to travel to the laboratory using motorised transport to minimise energy expenditure.

From breakfast until 11am subjects consumed only bottled spring water (Highland Spring still natural mineral water with a sports cap, 2 x 500ml; Highland Spring Ltd, Perthshire, Scotland) to a maximum volume of 1 litre.

Water consumption was ad libitum but the bottles were weighed prior to the experiment and at 11am in order to determine the exact amount consumed before the test-lunch. Upon arrival at the facility for the experimental day, subjects were randomly allocated to one of two preload treatments; an ionic-gelling sodium alginate formulation (SA) or an acid-gelling excipient free control (EF).

2.3 Preload Formulations and Glycaemia

The SA formulation contained sodium alginate, calcium carbonate (CaCO₃) and buffering agents. It was specifically formulated to undergo enhanced ionic
intragastric gelation upon ingestion. This is achieved by mixing sodium alginate with an acid soluble calcium salt. Post-ingestion solubilisation of calcium salt in acidic gastric fluid liberates free calcium ions which are then available to cross-link with the sodium alginate. The SA formulation has been described in detail by Paxman et al. (2008b). The EF control is identical in composition to SA with the omission of the CaCO₃. This formulation yields a gel via acid gelation (in the absence of calcium), resulting in weaker intra-molecular hydrogen bonded mass.

Prior to preload ingestion, baseline glycaemia (11:45am, 0 minutes) was determined using capillary blood taken from the finger. A single use Accu-check® Softclix® Pro lancing device was used to obtain a single droplet sample via OneTouch® Ultra® Test Strips with FastDrawTM design. The OneTouch® Ultra® Blood Glucose Monitoring System was used to determine glycaemia (reference range 1.1 to 33.3mmol/l; Lifescan Inc., Bucks). Each preload was served at 12:00pm (15 minutes after baseline glycaemia measurements) in an opaque non-descript plastic cup in standard feeding booths in green light. The coloured light masked a very slight colour difference between preload drinks. The drinks were flavoured with vanilla to yield an orosensory match. Subjects were instructed to drink the entire product. All
preloads were consumed within 5 minutes of their initial hydration with 100ml bottled water.

Following ingestion of the product (12:15pm, 30 minutes from baseline), glycaemia was again determined following identical protocol.

2.4 Test-lunch

Volunteers ingested a controlled test-lunch of mixed composition thirty minutes after consuming the preload drink (12:30pm, 45 minutes from baseline) in standard feeding booths in natural light. The test-lunch consisted of 300g pre-cooked then chilled penne pasta (Don Mario 100% durum wheat semolina pasta quills', manufactured by Abbey Foods Ltd, PO BOX 178, Liverpool) and 100g Sacla Italia ™ vine-ripened tomato and mascarpone stir through sauce (F.lli Sacla S.p.A. Asti Italy; Sacla UK LTD, Basil House, 21 London End, Bucks). This test-lunch was heated to a temperature of at least 72°C in a microwave and was served at a temperature of between 60-65°C. Subjects were instructed to consume the entire test-lunch and all subjects adhered to protocol.

The meal provided 57%, 13% and 30% of total energy from carbohydrate, protein and fat respectively, as
analysed by NetWISP (version 3.0 for Windows, Tinuviel
Software, Anglesey, UK). The test-lunch protocol used
here has been described previously (Paxman et al.,
2008a).

2.5 Protocol Postprandially

Further measures of capillary glucose were obtained at
90, 120, 150, 180, 210, 240, 270 and 330 minutes from
baseline. In total, ten capillary blood samples were taken
to determine glycaemia up to 330 minutes from baseline
(270 minutes postprandially).

2.6 Statistical analysis

Blood glucose measures were converted to delta area
under the curve (AUC) using the trapezoid rule with
subtraction of basal values (NCSS; Hintze, 2004, NCSS
and PASS Number Cruncher Statistical Systems,
Kaysville, Utah). Two-way between groups ANOVAs
were performed in order to identify the main effects of
treatment and body fat haptile and any interaction effects
on glycaemia at each time point, change from individual
baseline AUC glycaemia and peak postprandial
glycaemia (SPSS; version 15.0 for Windows, SPSS Inc.,
Chicago, IL, USA). Graphical presentations were
produced using SPSS (version 15.0 for Windows, SPSS
Inc., Chicago, IL, USA) and Microsoft Excel 2003
Significance was set at $p < 0.05$. Data are presented as mean ± 1 SD.

### 3.0 RESULTS

Forty self-reported healthy male subjects (equal numbers in each treatment arm) successfully completed the experiment with no deviation from protocol.

#### 3.1 Ionic gelling sodium alginate attenuates the glycaemic response to a meal

Two-way between groups ANOVAs showed a significant effect of treatment type on glycaemia at 90 ($p < .0005$), 150 ($p = .003$), 180 ($p = .021$) and 210 ($p = .013$) minutes (see Figure 1). Overall, ingestion of SA compared to EF resulted in a significant reduction in a mean change from individual baseline AUC glycaemia ($M = 148.43 ± 148.65$ vs. $M = 312.53 ± 253.60; p = .010$) of 52.5% (see Figure 1).

Irrespective of treatment type, subjects in the lower haptile for body fat % had a reduced mean change from individual baseline AUC glycaemia ($177.68 ± 255.44$) compared to those in the upper haptile for body fat % ($283.28 ± 171.66; p = .065$; data not shown) however, this was not significant and there was no interaction effect between treatment type and body fat % grouping.
3.2 Ionic gelling sodium alginate reduces peak postprandial glycaemia

Preload type failed to affect the timing of peak glycaemia as shown in Figure 1. However, Figure 2 shows the significant 14% lower mean peak postprandial glycaemia at 90 minutes following SA versus EF (M= 6.06 ± .59 mmol/L vs. M= 6.92 ± .70mmol/L; p< .0005) for the study group as a whole. Subjects in the lower body fat haptile had a lower peak postprandial glycaemia (6.39 ± .85mmol/L) than those in the upper body fat haptile, irrespective of treatment type (6.59 ± .70mmol/L; p=.170; data not shown) however this was not significant and there was no interaction effect between treatment type and body fat % grouping.

3.3 Body fat classification determines the postprandial glycaemic response to a meal but the beneficial effects of alginate remain

Irrespective of treatment type, subjects in the upper body fat haptile had non-significantly elevated peak postprandial glycaemia and non-significantly greater mean change from individual baseline AUC glycaemia compared to those in the lower body fat haptile. In addition, body fat % grouping had a significant effect on delta glycaemia at 120 (p= .005), 150 (p=.012), 180
(p = .049) and 210 minutes (p = .046) from baseline, with subjects in the upper body fat haptile having higher mean glycaemia than those in the lower body fat haptile at these time points, irrespective of preload treatment (Figure 3). For glycaemia at each time point, change from individual baseline AUC glycaemia and peak postprandial glycaemia however, the two-way between-groups ANOVA showed no interaction effect between treatment type and body fat % grouping in each case. Subjects appeared to respond to the ionic-gelling sodium alginate (SA) treatment in a similar fashion irrespective of body fat %.

Examination of the response to treatment type by body fat % grouping showed the lower body fat haptile on SA reduced their change from individual baseline AUC glycaemia by 68.3%, and their peak postprandial glycaemia by 16.2% compared to the lower body fat haptile on EF. A slightly weaker effect was apparent in the upper body fat haptile on SA who reduced their change from individual baseline AUC glycaemia by 46.6%, and their peak postprandial glycaemia by 9.7% compared to those in the upper body fat haptile on the EF treatment type (Figure 3). This finding supports our
previous suggestions relating to altered glycaemic response and body fatness (Paxman et al., 2008a).

In summary, glycaemic response to the test-meal was reduced following ingestion of the ionic-gelling sodium alginate drink (SA) compared to the acid-gelling excipient-free formulation (EF) throughout the 330 minute measurement period. Body fatness influenced postprandial glycaemic response but the effect of the ionic-gelling alginate drink was maintained.

4.0 DISCUSSION

The literature suggests soluble fibre can alter subjective hunger and fullness ratings (Peters et al., 2011), gastric emptying rate and intestinal nutrient absorption, though the extent and subsequent effect on glycaemia is poorly established (Wolf et al., 2002; Delargy et al., 1997; Fairchild et al., 1996). Contradictory reports are most likely explained by the type, dose, homogeneity and physicochemical properties of fibres used, and differing participant characteristics between studies.

The physicochemical properties of alginate have particular potential in terms of attenuating postprandial glycaemic...
response and improving diabetic control (Williams et al., 2004; Wolf et al., 2002; Torsdottir et al., 1991). Highly viscous solutions are unpalatable; solutions which form solid gel particles in the gastric lumen may provide a more feasible alternative for controlling gastric emptying and nutrient uptake. In order to establish an optimum formulation for delivery of a glycaemia-modulating alginate, the physiological response to ionic- and acid-gelling alginates were compared in males of differing body fatness. Physiologic data show greater glucose intolerance among the obese and numerous prospective studies support such associations between measures of obesity and type 2 diabetes risk (Carey et al., 1997). Such differences are postulated to be connected with body fatness.

Our data show that the ionic-gelling sodium alginate drink (SA) reduced early-phase and peak postprandial glycaemia and flattened the postprandial glycaemic curve in comparison to the acid-gelling control (EF). From baseline to 30 minutes, the EF preload drink resulted in a slight elevation of blood glucose, most likely due to the 7g fructose contained within the formulation. In comparison, the SA preload treatment elicited no change in glycaemia during this period despite containing the same amount of fructose. The difference between these responses can
most probably be attributed to the addition of calcium carbonate in the SA formulation. The acid-soluble calcium salt was expected to facilitate intra-gastric ionic gelation of the drink (Kimura et al., 1996). When alginate formulations are pH dependent there is a known time lag of 25-40 minutes before gelation occurs (Mattes, 2007). The inhibition of a glycaemic response to the SA formulation could have resulted from immediate fructose entrapment, delayed gastric emptying or both. Torsdottir et al. (1991) reported delayed glucose delivery and reduced glycaemic peak in type 2 diabetics by the addition of alginate to meals. They attributed this response solely to delayed gastric emptying, measured by aspirated radioactive stomach contents. There is evidence to suggest alginate ingestion results in 'gel lump' formation, which alters nutrient transport to the small intestine (Hoad et al., 2004). In this study the glycaemic response to a test-lunch of mixed composition following the SA drink was consistently lower throughout the investigation, thus it seems likely that nutrients were captured within the gel matrix to some degree.

Hoad et al. (2004) used serial magnetic resonance imaging (MRI) to gather in vivo measurements of guar gum and weak and strong gelling alginates that had been incorporated into milk-based drinks. MRI images showed
heterogeneous distribution of alginate formulations in the stomach with the formation of 'lumps', compared to the homogenous distribution of guar gum. Initial 'gel lump' formation was observed 10 minutes postprandially, other 'lumps' developed over time, compatible with the pH decrease normally observed following ingestion of a meal. In addition, the strong gelling alginate resulted in significantly increased intragastric gel 'lump' production compared to the weak gelling. Data from 'lump' classification showed liquid filled 'lumps' were formed predominantly with the strong gelling alginate; the researchers hypothesise this gel strength is sufficient to allow layer formation which resist break forces caused by stomach motion.

There is a prevailing assumption that BMI measurement is strongly associated with body fatness and consequent morbidity and mortality (Gallagher et al., 2000). Increased postprandial blood glucose is independently related to the risk of cardiovascular disease and all-cause mortality in newly diagnosed type 2 diabetics. Some individuals classified overweight by BMI do not have high % body fat. Conversely, others who have normal or healthy BMIs have a relatively high body fat %. Individuals who are misclassified by BMI are reportedly uncommon relative to the UK population as a whole but
since body fatness is a stronger predictor of increased fasting glucose than BMI (Kriketos et al., 2003) it is more appropriate and meaningful to divide subjects in the present study by body fat %. In support of this, the present study clearly shows subjects in the upper body fat haptile had comparatively elevated early-phase glycaemic excursion to those in the lower body fat haptile.

5.0 CONCLUSIONS

We conclude that an ionic-gelling sodium alginate drink can significantly attenuate postprandial glycaemic response in self-reported healthy males in comparison to an acid-gelling control. This effect persisted in subjects in both the lower and upper haptiles of body fatness. The benefits of optimising glycaemic control through the use of ionic-gelling sodium alginate products in patients with morbidity related to body fatness (including type 2 diabetic and metabolic syndrome patients) warrant further investigation.

ROLE OF THE FUNDING SOURCE
This work was financially supported by Technostics Ltd., UK, who also had input into the overall design, write up and submission of this work.

REFERENCES


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### TABLES

**Table 1**

**Subject characteristics**

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| **BMI (kg/m²)**      | 40| 18.6 – 39.4 | 26.02 ± 4.41 |
| Sodium Alginate      |   |           |           |
| Lower BF%            | 9 | 21.7 – 24.7 | 23.34 ± 1.07   |
| Upper BF%            | 11| 22.7 – 35.2 | 29.07 ± 3.03   |
| **TOTAL**            | 20| 21.7 – 35.2 | 26.50 ± 3.72   |
| Excipient Free       |   |           |           |
| Lower BF%            | 11| 18.6 – 26.0 | 22.32 ± 2.33   |
| Upper BF%            | 9 | 23.0 – 35.6 | 29.47 ± 4.72   |
| **TOTAL**            | 20| 18.6 – 39.4 | 25.54 ± 5.06   |

| **Body Fat (BF) %**  | 40| 7.1 - 35.6 | 17.54 ± 7.05 |
| Sodium Alginate      |   |           |           |
| Lower BF%            | 9 | 7.1 – 11.9 | 10.31 ± 1.56   |
| Upper BF%            | 11| 16.8 – 31.7| 22.58 ± 4.94   |
| **TOTAL**            | 20| 7.1 – 31.7 | 17.06 ± 7.29   |
| Excipient Free       |   |           |           |
| Lower BF%            | 11| 9.2 – 15.4 | 12.76 ± 1.82   |
| Upper BF%            | 9 | 18.1 – 35.6| 24.47 ± 5.09   |
| **TOTAL**            | 20| 9.2 – 35.6 | 18.03 ± 6.96   |
Figure Captions

Figure 1

Mean delta AUC glycaemia (±1SD)

Following ingestion of the SA preload (solid line, filled diamonds), mean delta AUC glycaemia was reduced by 52.5% when compared to the EF preload (broken line, open squares). There was a significant effect of preload treatment type on mean delta AUC ($p = .010$). In addition, preload treatment type had a significant effect (*) on mean delta glycaemia at 90 minutes ($p < .0005$), 150 minutes ($p = .003$), 180 minutes ($p = .021$) and 210 minutes ($p = .013$).

Figure 2

Mean peak postprandial glycaemia (±1SD)

There was a significant effect of preload treatment type on mean peak postprandial glycaemia (SA solid bars; $M = 6.06 \pm .59 \text{ mmol/L}$ compared to EF shaded bars; $M = 6.92 \pm .70 \text{ mmol/L}$; *$p < .0005$).

Figure 3
Mean delta AUC glycaemia by body fat haptile

When subjects were split by haptiles of body fat % (solid line = upper body fat haptile, broken line = lower body fat haptile) there was a significant effect of body fat % classification ($^\dagger$) on mean delta glycaemia at 120 minutes ($p = .005$) 150 minutes ($p = .012$) 180 minutes ($p = .049$) and 210 minutes ($p = .046$), irrespective of treatment type (solid diamonds = sodium alginate, open squares = excipient free).
Figure 1
Figure 2

Mean Peak Postprandial Glycaemia (mmol/L)

Preload Treatment Type
Figure 3