

## The physical state of a meal affects hormone release and postprandial thermogenesis

Maddalena Peracchi<sup>1</sup>\*, Alessandra Santangelo<sup>2</sup>, Dario Conte<sup>1</sup>, Mirella Fraquelli<sup>1</sup>, Rosalia Tagliabue<sup>1</sup>, Carlotta Gebbia<sup>1</sup> and Marisa Porrini<sup>2</sup>

<sup>1</sup>Department of Gastroenterology, Institute of Medical Science, University of Milan, Ospedale Maggiore-IRCCS, via F. Sforza 35, 20122 Milan, Italy

<sup>2</sup>Department of Food Science and Technology, Nutrition Section, University of Milan, via Celoria 2, 20133 Milan, Italy

(Received 25 May 1999 – Revised 11 October 1999 – Accepted 8 November 1999)

There is evidence that food consistency may influence postprandial physiological responses. Recently we found that homogenization of a vegetable-rich meal significantly delayed the gastric emptying rate and was more satiating than the same meal in solid–liquid form. In this present study we investigated whether homogenization also influences endocrine and metabolic responses to the meal. Eight healthy men, aged 21–28 (mean 24.5) years, were given the meal (cooked vegetables 250 g, cheese 35 g, croutons 50 g and olive oil 25 g, with water 300 ml; total energy 2.6 MJ) in both solid–liquid (SM) and homogenized (HM) form, in random order, at 1-week intervals. Variables assayed were plasma glucose, insulin and glucose-dependent insulinotropic peptide (GIP) levels for 2 h and diet-induced thermogenesis (DIT) for 5 h. Plasma glucose pattern was similar after both meals. However, HM induced significantly greater insulin, GIP and DIT responses than SM. Mean integrated areas under the curves (AUC) were 1.7 (SEM 0.38) v. 1.2 (SEM 0.33) U/l per 120 min ( $P=0.005$ ) for insulin, 19.9 (SEM 2.44) v. 16 (SEM 1.92) nmol/l per 120 min ( $P=0.042$ ) for GIP, and 237.7 (SEM 16.32) v. 126.4 (SEM 23.48) kJ/300 min ( $P=0.0029$ ) for DIT respectively. Differences between GIP-AUC after HM and SM correlated significantly with differences between insulin-AUC after HM and SM ( $r^2$  0.62,  $P=0.021$ ). These findings demonstrate that homogenization of a meal results in a coordinated series of changes of physiological gastroentero–pancreatic functions and confirm that the physical state of the meal plays an important role in modulating endocrine and metabolic responses to food.

### Food consistency: Glucose-dependent insulinotropic peptide: Insulin: Diet-induced thermogenesis

Physical and chemical properties of foods modulate the absorption and metabolism of nutrients by influencing both the gastric emptying rate and the release of gut hormones and neurotransmitters (Schirra *et al.* 1996). Thus, in non-diabetic subjects, glycaemic and insulinaemic responses to meals depend not only on the quantity of carbohydrate consumed but also on its source (Wolever & Bolognesi, 1996), and are modified by the concomitant presence of other macronutrients (Joannic *et al.* 1997). Indeed, several studies have shown that the association of protein, fat or fibre with carbohydrate can reduce the postprandial glucose response (Estrich *et al.* 1967; Collier *et al.* 1984; Gulliford *et al.* 1989; Torsdottir *et al.* 1989). Moreover, manipulations of starch, including cooking, cooling, changes in its viscosity and osmolality, and chemical modifications, can influence its glycaemic and/or insulinaemic index, and starch usually

contributes over 50 % of all carbohydrate in a mixed meal (Würsch, 1989; Björck, 1996; Raben *et al.* 1997).

Insulin plays a key role in the control of energy balance by promoting storage of ingested nutrients. However, there is some evidence that insulin may act at the level of the central nervous system, inhibiting food intake and stimulating thermogenesis (Rothwell, 1992). In addition, Landsberg & Young (1983) postulated that a meal resulting in relatively high postprandial insulin response may increase the facultative part of diet-induced thermogenesis (DIT) via stimulation of the sympathetic nervous system. Holt *et al.* (1996) reported that foods producing a higher postprandial insulin response were associated with less subsequent food intake and thus indirectly with greater satiety.

We recently found that homogenization of a fat-rich and vegetable-rich meal significantly delayed the gastric

**Abbreviations:** AUC, area under the curve; DIT, diet-induced thermogenesis; GIP, glucose-dependent insulinotropic peptide; HM, meal in homogenized form; SM, meal in solid form.

\* **Corresponding author:** Dr Maddalena Peracchi, fax +39 02 55012111, email gastrbia@imiucca.csi.unimi.it

emptying rate in healthy subjects and that such a meal was significantly more satiating than the same meal given in a conventional solid–liquid form (Santangelo *et al.* 1998). The reasons for these differences are still unclear. Therefore, in this present paper we investigated whether plasma glucose, insulin and glucose-dependent insulinotropic peptide (GIP) responses to the vegetable-rich meal and DIT are influenced by homogenization of the meal.

## Materials and methods

### Subjects

Eight healthy men, aged 21–28 (mean 24.5) years, were recruited for this study which was approved by the local ethics committee. All subjects were within 10% of ideal body weight, with a BMI of 21–25 (mean 22.8) kg/m<sup>2</sup>. None of them was a smoker, was taking medication, had a family history of diabetes mellitus, suffered from gastrointestinal symptoms or any systemic disease including diabetes mellitus.

All the subjects underwent a complete endocrine evaluation, with a concomitant energy expenditure assessment in six cases.

### Procedures

The test meal consisted of (g): cooked vegetables 250, cheese 35, croutons 50 and olive oil 25 (2.6 MJ (60% energy as fat, 27% energy as carbohydrate, 13% energy as protein), 8 g fibre (16% soluble, 84% insoluble)) (Santangelo *et al.* 1998), given either in solid form (SM), with 300 ml of water to drink during the consumption, or homogenized with the addition of the same amount of water (HM).

Subjects were studied twice, at an interval of at least 1 week, with SM and HM given in randomized order. They were instructed to abstain from physical exercise the day before each test and to fast from 22.00 hours the evening before. On the study day the subjects came to the laboratory between 08.30 and 09.30 hours. During the first 15 min after arrival they rested quietly in the supine position, and a retrograde venous cannula was inserted into the left hand for blood sampling and kept patent by slow saline (9 g NaCl/l) infusion. The BMR was recorded for 30 min. The test meal was then served (time 0) and eaten in 15 min. Postprandial energy expenditure was measured continually for 5 h with three 20 min breaks. Subjects were instructed to remain awake and immobile during the measurement and were allowed to listen to music. During breaks, they could stand up and move in the laboratory or, if necessary, go to the bathroom. Blood samples were collected in ice-chilled polypropylene tubes containing EDTA (1 mg/ml) and aprotinin (500 kIU/ml) at the following times: –30, 0, 15, 30, 45, 60, 90 and 120 min. Plasma was separated immediately by centrifugation at 3000 g at 4° and stored in portions at –80° until assayed.

### Assays

Plasma insulin and GIP levels were measured by radio-immunoassay using commercially available kits (Insulin, Biodata SpA, Guidonia Montecelio, Rome, Italy; Gastric inhibitory peptide, Peninsula Laboratories Inc, Belmont,

CA, USA). Before being assayed for GIP, plasma was extracted on Sep Pak C<sub>18</sub> cartridges (Waters Corporation, Milford, MA, USA) (Peracchi *et al.* 1999). The 95% confidence detection limits were 0.2 µU/tube for insulin and 3 pg/tube for GIP, and the intra- and interassay CV were 4.5% and 5.7%, and 6.8% and 9.9% respectively. Plasma glucose levels were determined by means of a glucose autoanalyser with a hexokinase method (Beckman, Milan, Italy).

Energy expenditure was measured by indirect calorimetry using an open-circuit ventilated-hood system (Deltatrac II, Datex Instrumentarium Corp., Helsinki, Finland). O<sub>2</sub> consumption (V<sub>O<sub>2</sub></sub> (l/min)) and CO<sub>2</sub> production (V<sub>CO<sub>2</sub></sub> (l/min)) were printed out every min, and the mean values for the 30 min pre-meal measurement period and every 30 min period after the meal were calculated automatically. At each time point energy expenditure was obtained according to the standard abbreviated Weir (1949) equation. DIT was calculated as the postprandial increase in energy expenditure above the pre-meal values. The within-person day-to-day CV of pre-meal energy expenditure was 6.1%.

### Statistical analysis

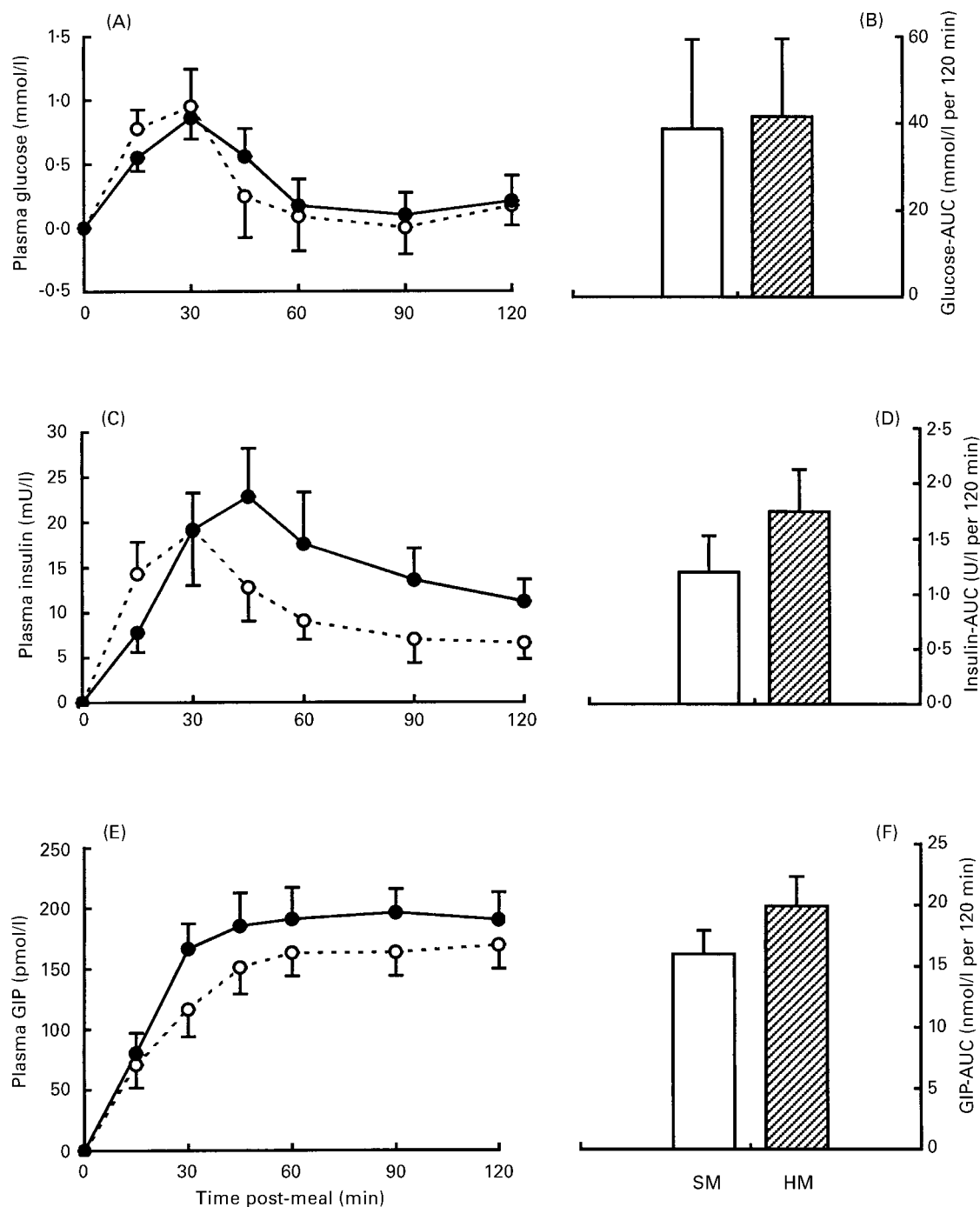
Results were given as means with their standard errors. Postprandial data were computed at each time point by subtracting basal values, and the integrated areas under these curves (AUC) were calculated using the trapezoidal rule. Data were evaluated by a repeated-measures analysis of covariance with time as covariate and type of food (SM or HM) as dependent variable; means were compared using the least significant difference test. Student's paired *t* test was employed to evaluate the difference in AUC values, basal values and peak times. Relationships between variables were assessed by linear regression analysis. *P* < 0.05 was considered statistically significant. The computer program STATISTICA for Windows (StatSoft Inc., Tulsa, OK, USA) was used for the analysis.

## Results

Fasting plasma levels of glucose (4.6 (SEM 0.10) *v.* 4.7 (SEM 0.12) nmol/l), insulin (9.3 (SEM 1.07) *v.* 10.6 (SEM 0.91) mU/l), GIP (22 (SEM 5.4) *v.* 29 (SEM 4.1) pmol/l) and basal energy expenditure (5.7 (SEM 0.19) *v.* 5.5 (SEM 0.16) kJ/min) were not significantly different on the two test days.

Postprandial time courses of plasma glucose, insulin and GIP concentrations and their integrated AUC are shown in Fig. 1. There were no significant differences in plasma glucose increments after the SM and HM, although peak times were slightly delayed after the HM (28 (SEM 3.4) *v.* 24 (SEM 3.9) min, NS). Plasma insulin and GIP responses were significantly higher after the HM compared with the SM, both when the data were expressed as variation over time ( $F_{(1,54)} 7.183$ , *P* = 0.0097 and  $F_{(1,54)} 13.466$ , *P* = 0.0006 respectively) and as AUC (*t* 4.053, *P* = 0.005 and *t* 2.479, *P* = 0.042 respectively).

After the HM, insulin peak levels occurred significantly later than after the SM (41 (SEM 4.7) *v.* 28 (SEM 3.4) min, *t* 2.497, *P* = 0.041) whereas with both meals GIP levels peaked at 30–60 min and thereafter remained elevated

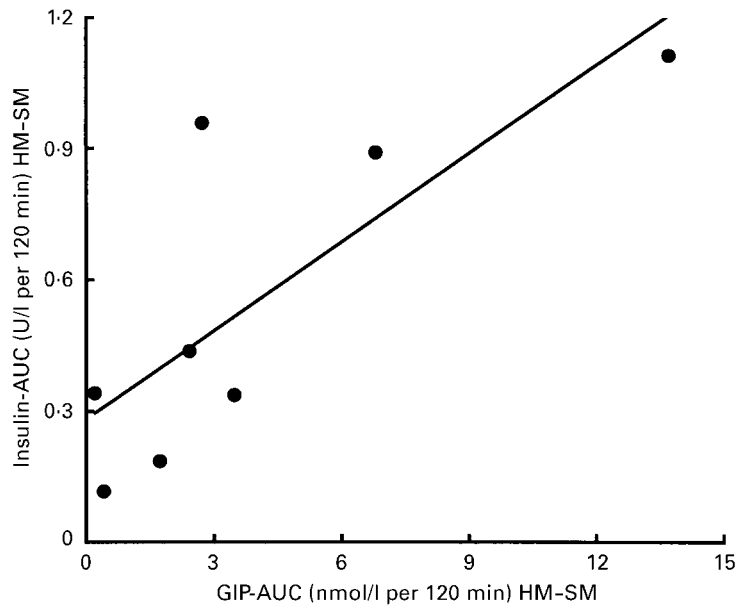


**Fig. 1.** Plasma glucose ((A), (B)), insulin ((C), (D)), and glucose-dependent insulintropic peptide (GIP) ((E), (F)) after the solid-liquid (SM; ○) and the homogenized meal (HM; ●) in eight healthy men. Results are given as variation from baseline at different time points ((A), (C), (E)) and as integrated areas under the curves (AUC) ((B), (D), (F)). Values are means with their standard errors represented by vertical bars. For (C) and (E), mean values after SM were significantly different from those after HM:  $P=0.0097$ ,  $P=0.0006$  respectively (analysis of covariance); for (D) and (F) mean values for SM were significantly different from those after HM:  $P=0.005$ ,  $P=0.042$  respectively (Student's paired  $t$  test). For details of meals and procedures see p. 624.

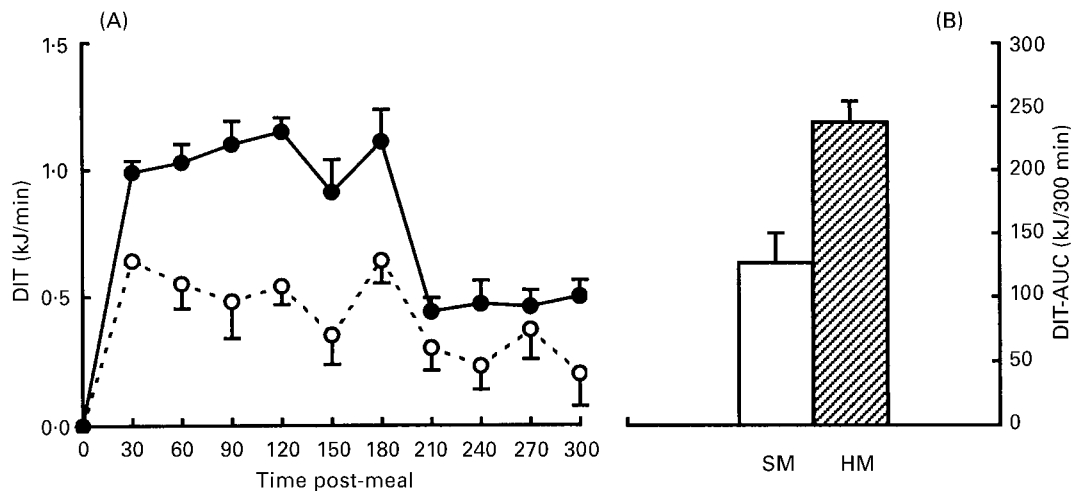
throughout the study period. With both meals, insulin and glucose peak times correlated significantly ( $y=7.295+0.546x$ ,  $r^2=0.488$ ,  $P=0.003$ ) but GIP peak times did not correlate with those of insulin or glucose. However, as shown in Fig. 2, differences between GIP-AUC after the HM and SM correlated significantly with differences

between insulin-AUC after the HM and SM ( $y=0.284+0.067x$ ,  $r^2=0.618$ ,  $P=0.021$ ).

DIT after the HM was significantly higher than after the SM (Fig. 3), both when the data were expressed as variation over time ( $F_{(1,58)} 87.351$ ,  $P<0.0001$ ) and as AUC ( $t 5.394$ ,  $P=0.0029$ ). Considering only the first 120 min of recording,



**Fig. 2.** Correlation between differences in glucose-dependent insulinotropic peptide (GIP) areas under the curve (AUC) after the homogenized (HM) and the solid-liquid meal (SM) and differences in insulin AUC after HM and SM ( $y=0.284+0.067x$ , SE of estimate 0.255;  $r^2$  0.618,  $P=0.021$ ) in eight healthy men. For details of meals and procedures see p. 624.



**Fig. 3.** Diet-induced thermogenesis (DIT) after the solid-liquid (SM;  $\circ$ ) and the homogenized meal (HM;  $\bullet$ ) in six healthy men. Results are shown as variation from baseline at different time-points (A) and as integrated areas under the curves (AUC) (B). Values are means with their standard errors represented by vertical bars. For (A) mean values after SM were significantly different from those after HM:  $P < 0.0001$  (analysis of covariance); for (B) mean values for SM were significantly different from those after HM:  $P = 0.0029$  (Student's paired  $t$  test). For details of meals and procedures see p. 624.

the difference was highly significant ( $F_{(1,22)} 76.494$ ,  $P < 0.0001$ ). Over this 2 h period mean energy expenditure increase above the basal values was 17% (0.92 (SEM 0.16) kJ/min) after the HM and 9% (0.48 (SEM 0.18) kJ/min) after the SM.

### Discussion

In this present study insulin and GIP release and DIT were higher after the HM compared with the SM whereas the plasma glucose pattern was similar in the two tests. The

positive correlation ( $r$  0.79) observed between differences in GIP-AUC and insulin-AUC after the two meals underlines the importance of GIP in determining postprandial insulin release. GIP's role as an incretin is well known and depends on a sufficient postprandial increase of circulating glucose levels (Andersen *et al.* 1978). The role of other incretin hormones, mainly glucagon-like peptide 1 (7–36) amide is still being investigated (Fehmann *et al.* 1995; Ørskov *et al.* 1996; Schirra *et al.* 1996) and there is evidence that the glycaemic and insulinaemic responses to meals ultimately depend on the integrated interaction of glucose delivery to

the duodenum, and GIP and glucagon-like peptide 1 (7–36) amide release (Schirra *et al.* 1996). In the presence of similar glycaemic responses the extent of GIP secretion is usually related to the total energy intake (Beck *et al.* 1984). However, in this present study we used two isoenergetic, isovolumetric meals which differed only in physical state (HM v. SM). The meal included 250 g cooked vegetables, which are a good source of dietary fibre. As evidence exists that fibre can modulate the GIP response to a standard meal (Morgan *et al.* 1990), we may suppose that homogenization of the meal influenced its physical and physiological properties by disrupting the structure of vegetables and fibre (Anderson & Chen, 1979; Holt & Miller, 1994; Gustafsson *et al.* 1995). Interestingly, in line with our previous findings indicating that homogenization delays the gastric emptying time (Santangelo *et al.* 1998), in this present study glucose and insulin levels peaked later after the HM than after the SM whereas GIP levels peaked 30–60 min after both the HM and SM. This observation is consistent with previous studies showing that GIP release is not directly related to gastric emptying but is governed by intestinal glucose and fat absorption (Fehmann *et al.* 1995; Schirra *et al.* 1996). In addition, the half-life of GIP in the circulation is remarkably long, approximately 20 min (Sarson *et al.* 1982), and this could well account for the finding that in our subjects GIP levels remained elevated throughout the study period.

On the other hand, it has already been suggested (Fried *et al.* 1991) that in healthy subjects gastric meal-emptying correlates closely with insulin release. Our results seem to confirm these findings as our HM, which was emptied more slowly than the SM, induced an initially slower postprandial secretion of insulin, which then increased over SM values. Even though we did not measure the early insulin response to the meals, the pattern of insulin release after the HM could suggest a loss of the cephalic phase of postprandial insulin response with a subsequent compensatory hyperinsulinaemia during the digestive phase (Calles-Escandon & Robbins, 1987). The cephalic phase of insulin release occurs before the nutrient absorption and the rise in arterial glucose levels, and is initiated by sensory stimulation produced by the sight, smell, taste and texture of food (LeBlanc *et al.* 1991, 1996; Teff *et al.* 1991). However, there were no differences in the palatability and acceptability scores of HM and SM (Santangelo *et al.* 1998), even though the texture of the meals was clearly different. On the other hand, the loss of cephalic phase of insulin release is usually associated with blunted DIT (Calles-Escandon & Robbins, 1987), whereas our results show that the HM caused a higher DIT than the SM. This event could be related to the different pattern of insulin release after the two meals, as insulin is known to participate in the control of facultative DIT component (Landsberg & Young, 1983). However, we cannot exclude a role for an activation of the sympathetic nervous system and catecholamine release by the different meal textures (LeBlanc *et al.* 1991).

In addition, Habas & Macdonald (1998), in their study of isoenergetic high-carbohydrate mixed-nutrient liquid and solid test meals, concluded that the physical form of a test meal affects the physiological responses to nutrient ingestion. However, they found that postprandial energy expenditure

(measured for 2 h after the meals), blood glucose and serum insulin concentrations were significantly higher after the solid meal. The discrepancy with our present results could depend on the completely different food composition of the two meals used by them (liquid meal: semi-skimmed milk, glucose polymer, a powdered protein and double cream; solid meal: cereal, semi-skimmed milk, toasted white bread, jam, butter and white sugar) and, furthermore, on the absence of food rich in fibre in the liquid meal.

Finally, as we have previously found that the vegetable-rich meal was significantly more satiating when homogenized, and homogenization increases GIP release, it is interesting that some lines of evidence suggest that GIP and other incretin hormones could modulate appetite control in animals and humans (Flatt *et al.* 1984; Raben *et al.* 1996; Lavin *et al.* 1998; Näslund *et al.* 1998).

In conclusion, the present findings confirm that the physical state of the meal has an important role in metabolic responses to food.

### Acknowledgements

This study was supported in part by MURST and by Associazione Amici della Gastroenterologia del Padiglione Granelli, Milan, Italy.

### References

- Andersen DK, Elahi D, Brown JC, Tobin JD & Andres R (1978) Oral glucose augmentation of insulin secretion. Interactions of gastric inhibitory polypeptide with ambient glucose and insulin levels. *Journal of Clinical Investigation* **62**, 152–161.
- Anderson JW & Lin Chen WJ (1979) Plant fiber. Carbohydrate and lipid metabolism. *American Journal of Clinical Nutrition* **32**, 346–363.
- Beck B, Villaume C, Chayvialle JA, Gariot P, Ulmer M, Desalme A & Debry G (1984) Influence of caloric intake on gastric inhibitory polypeptide, VIP and gastrin release in man. *Peptides* **5**, 403–406.
- Björck I (1996) Starch: nutritional aspects. In *Carbohydrates in Food*, pp. 505–553 [AC Eliasson, editor]. New York, NY: Marcel Dekker Inc.
- Calles-Escandon J & Robbins DC (1987) Loss of early phase of insulin release in humans impairs glucose tolerance and blunts thermic effect of glucose. *Diabetes* **36**, 1167–1172.
- Collier G, McLean A & O’Dea K (1984) Effect of co-ingestion of fat on the metabolic responses to slowly and rapidly absorbed carbohydrates. *Diabetologia* **26**, 50–54.
- Estrich D, Ravnik A, Schlierf G, Fukayama G & Kinsell L (1967) Effects of co-ingestion of fat and protein upon carbohydrate-induced hyperglycemia. *Diabetes* **16**, 232–237.
- Fehmann HC, Göke R & Göke B (1995) Cell and molecular biology of the incretin hormones glucagon-like peptide-I and glucose-dependent insulin releasing polypeptide. *Endocrine Reviews* **16**, 390–410.
- Flatt PR, Bailey CJ, Kwasowski P, Page T & Marks V (1984) Plasma immunoreactive gastric inhibitory polypeptide in obese hyperglycemic (ob/ob) mice. *Journal of Endocrinology* **101**, 249–256.
- Fried M, Schwizer W, Beglinger C, Keller U, Jansen JB & Lamers CB (1991) Physiological role of cholecystokinin on postprandial insulin secretion and gastric meal emptying in man. Studies with the cholecystokinin receptor antagonist loxiglu-mide. *Diabetologia* **34**, 721–726.

- Gulliford MC, Bicknell EJ & Scarpello JH (1989) Differential effect of protein and fat ingestion on blood glucose responses to high- and low-glycemic-index carbohydrates in non insulin-dependent diabetic subjects. *American Journal of Clinical Nutrition* **50**, 773–777.
- Gustafsson K, Asp NG, Hagander B & Nyman M (1995) Satiety effects of spinach in mixed meals: comparison with other vegetables. *International Journal of Food Sciences and Nutrition* **46**, 327–334.
- Habas ME & Macdonald IA (1998) Metabolic and cardiovascular responses to liquid and solid test meals. *British Journal of Nutrition* **79**, 241–247.
- Holt SHA, Brand Miller JC & Petocz P (1996) Interrelationships among postprandial satiety, glucose and insulin responses and changes in subsequent food intake. *European Journal of Clinical Nutrition* **50**, 788–797.
- Holt SHA & Miller B (1994) Particle size, satiety and the glycaemic response. *European Journal of Clinical Nutrition* **48**, 496–502.
- Joannic JL, Auboiron S, Raison J, Basdevant A, Bornet F & Guy-Grand B (1997) How the degree of unsaturation of dietary fatty acids influences the glucose and insulin responses to different carbohydrates in mixed meals. *American Journal of Clinical Nutrition* **65**, 1427–1433.
- Landsberg L & Young JB (1983) The role of the sympathetic nervous system and catecholamines in the regulation of energy metabolism. *American Journal of Clinical Nutrition* **38**, 1018–1024.
- Lavin JH, Wittert GA, Andrews J, Yeap B, Wishart JM, Morris HA, Morley JE, Horowitz M & Read NW (1998) Interaction of insulin, glucagon-like peptide 1, gastric inhibitory polypeptide, and appetite in response to intraduodenal carbohydrate. *American Journal of Clinical Nutrition* **68**, 591–598.
- LeBlanc J, Diamond P & Nadeau A (1991) Thermogenic and hormonal responses to palatable protein and carbohydrate rich food. *Hormone and Metabolic Research* **23**, 336–340.
- LeBlanc J, Soucy J & Nadeau A (1996) Early insulin and glucagon responses to different food items. *Hormone and Metabolic Research* **28**, 276–279.
- Morgan LM, Tredger JA, Wright J & Marks V (1990) The effect of soluble- and insoluble-fibre supplementation on postprandial glucose tolerance, insulin and gastric inhibitory polypeptide secretion in healthy subjects. *British Journal of Nutrition* **64**, 103–110.
- Näslund E, Gutniak M, Skogar S, Rössner S & Hellström PM (1998) Glucagon-like peptide 1 increases the period of postprandial satiety and slows gastric emptying in obese men. *American Journal of Clinical Nutrition* **68**, 525–530.
- Ørskov C, Wettergren A & Holst JJ (1996) Secretion of the incretin hormones glucagon-like peptide-1 and gastric inhibitory polypeptide correlates with insulin secretion in normal man throughout the day. *Scandinavian Journal of Gastroenterology* **31**, 665–670.
- Peracchi M, Basilisco G, Tagliabue R, Terrani C, Locati A, Bianchi PA & Velio P (1999) Postprandial gut peptide plasma levels in women with idiopathic slow-transit constipation. *Scandinavian Journal of Gastroenterology* **34**, 25–28.
- Raben A, Andersen K, Karberg MA, Holst JJ & Astrup A (1997) Acetylation of or  $\beta$ -cyclodextrin addition to potato starch: beneficial effect on glucose metabolism and appetite sensations. *American Journal of Clinical Nutrition* **66**, 304–314.
- Raben A, Holst JJ, Christensen NJ & Astrup A (1996) Determinants of post-prandial appetite sensations: macronutrient intake and glucose metabolism. *International Journal of Obesity* **20**, 161–169.
- Rothwell NJ (1992) Hypothalamus and thermogenesis. In *Energy Metabolism: Tissue Determinants and Cellular Corollaries*, pp. 229–245 [JM Kinney and HN Tucker, editors]. New York, NY: Raven Press Ltd.
- Santangelo A, Peracchi M, Conte D, Fraquelli M & Porrini M (1998) Physical state of meal affects gastric emptying, cholecystokinin release and satiety. *British Journal of Nutrition* **80**, 521–527.
- Sarson DL, Hayter RC & Bloom SR (1982) The pharmacokinetics of porcine glucose-dependent insulinotropic polypeptide (GIP) in man. *European Journal of Clinical Investigation* **12**, 457–461.
- Schirra J, Katschinski M, Weidmann C, Schäfer T, Wank U, Arnold R & Göke B (1996) Gastric emptying and release of incretin hormones after glucose ingestion in humans. *Journal of Clinical Investigation* **97**, 92–103.
- Teff KL, Mattes RD & Engelman K (1991) Cephalic phase insulin release in normal weight males: verification and reliability. *American Journal of Physiology* **261**, E430–E436.
- Torsdottir I, Alpsten M, Andersson H & Einarsson S (1989) Dietary guar gum effects on postprandial blood glucose, insulin and hydroxyproline in humans. *Journal of Nutrition* **119**, 1925–1931.
- Weir JBDeV (1949) New method for calculating metabolic rate with special reference to protein metabolism. *Journal of Physiology* **109**, 1–9.
- Wolever TMS & Bolognesi C (1996) Source and amount of carbohydrate affect postprandial glucose and insulin in normal subjects. *Journal of Nutrition* **126**, 2798–2806.
- Würsch P (1989) Starch in human nutrition. *World Review of Nutrition and Dietetics* **60**, 199–256.