The effects of pork on satiety

Report prepared for the Co-operative Research Centre for an Internationally Competitive Pork Industry

By

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Executive Summary

Given that more than half of the Australian adult population is either overweight or obese, an evidence base is required to identify foods and meal patterns that contribute to an overall reduction in energy intake. There is increasing commercial interest in the emerging scientific evidence that some foods and/or food components may support weight loss through increased satiety.

Limited evidence from one animal and one human study suggest that pork may have an important role in obesity prevention and weight loss through satiating effects, when replacing other protein foods in a mixed diet. Mechanistically, the short-term regulation of food intake is mediated via neural and humoral signals from the gastrointestinal tract to different regions in the brain. Some studies have demonstrated that consumption of a high-protein diet increases satiety through a decrease in circulating ghrelin (associated with feelings of hunger), and increased concentrations of cholecystokinin (CCK) and Peptide Y-Y (PYY) (both associated with satiety). In the longer term, regulation of food intake by hormones such as leptin from adipose tissue and insulin from the pancreas may be significant.

The aim of the present study was to demonstrate that the consumption of a pork meal has a greater effect on acute satiety than comparative meals using animal protein sources, namely beef and chicken.

Thirty non-smoking pre-menopausal women aged 19 - 45 years, with a Body Mass Index range of 19.2 – 38.3 kg/m^2 were recruited for the study. On three test days, fasting participants attended a research centre and consumed, in random order, one of three meat-containing meals (pork, chicken, beef) that were matched in energy (kJ), total protein content, palatability, and appearance. A within-subjects design was employed whereby each participant served as her own control. The primary outcome measures were: (1) amount of food consumed at a subsequent *ad libitum* buffet lunch meal; (2) amount of food consumed and macronutrient selection for the rest of the day; (3) Visual Analogue Scale (VAS) ratings for hunger and satiety; and (4) hormonal appetite and satiety signals.

No difference was found between meat groups for either energy intake, or for macronutrient profile (% energy from protein, fat or carbohydrate) of food consumed at the buffet lunch, following test meal breakfast consumption. Participants also consumed comparable quantities of food over the rest of the day on each of the three test meal days, indicating no test meal effect on later food choice. Subjective VAS scores did not differ between test meals. With the exception of a difference in PYY between chicken and pork meals ($P = 0.027$), no significant differences were found for any of the appetite hormone levels investigated (CCK, ghrelin, insulin) after consumption of pork, beef or chicken.

The study findings add a new marketing opportunity for the pork industry in the context of consumer demands for foods to improve health and wellbeing. In an obesogenic environment where high protein diets are seen to provide opportunity for better satiety and weight loss, this study positions pork in a healthy diet as being equal to lean beef or chicken in terms of its effect on satiety and release of appetite-related intestinal hormones and insulin. Where previously consumers may have only thought of red meat and chicken in terms of these benefits, pork is seen as equally effective.

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1. Introduction

Overweight and obesity are major predisposing factors for diseases such as diabetes, hypertension and coronary heart disease. Management of overweight, especially before the development of these complications, is particularly relevant for Australia where more than half of the adult population is either overweight or obese. Energy intake is an important factor in weight management. An evidence base is required to identify foods that contribute to reduction of energy intake, including those which increase satiety. Such information is important to allow the translation of messages about various foods within the context of weight management and the prevention of obesity.

There is presently considerable interest in the use of higher protein diets for weight loss when the primary source of protein is from meat-based products. In Australia, the highly successful, best-selling CSIRO "Total Wellbeing Diet" advocates a higher meat and fish intake (approximately 300g/day) than is promoted by the Australian dietary guidelines. The diet has been tested for efficacy in 100 overweight women who were randomly assigned to follow either the CSIRO Total Wellbeing Diet or a high-carbohydrate hypocaloric diet. A similar weight loss was found in both groups, but the high-protein diet provided nutritional and metabolic benefits that were either equal to or greater than those observed with a high-carbohydrate diet. (Noakes et al., 2005). It is unclear whether high-protein diets reduce body weight by reducing energy intake through satiety signals or by increasing energy expenditure or both.

Evidence supporting the health benefits of higher protein diets is increasingly being recognized by Australian agencies involved in the development of dietary guidelines and food guides for healthy eating. The National Health and Medical Research Council (NHMRC, 2005) recently published Acceptable Macronutrient Distribution Ranges (AMDR, expressed as % energy (E)) for lowering chronic disease risk generally. The NHMRC advocates that 15-25 %E be provided from protein, 45-65 %E from CHO, and 20-35 %E from fat. To specifically support weight loss, Schoeller and Buchholz (2005) recommend a higher protein intake of between 25 and 30% E, accompanied by a moderate carbohydrate intake (35-50% E) and a moderate fat intake (25-35% E) for management of the metabolic syndrome.

Compared to isocaloric meals of either fat or carbohydrate, protein has been found to be the most satiating macronutrient in both human and animal studies (Eisenstein *et al.*, 2002; Westerterp-Plantenga, 2003, (Barkeling et al., 1990; Booth et al., 1970; Geliebter, 1979; Jen et al., 1985). However, evidence regarding the effect of different types of protein on satiety is limited to only a few studies. These suggest that fish may be more satiating than red meat and eggs (Uhe et al , 1992; Holt et al 1995) but the results are not entirely consistent (eg see Borzoei et al., 2006).

There are also some studies identifying differences in satiety in relation to pork. One rat model study found that pork protein isolate increased satiety (Sufian et al., 2006). In this study protein fractions from beef, pork, chicken and eggs were tested and pork was shown to have the greatest ability to suppress appetite in rats, through direct interaction with the small intestinal cholecystokinin (CCK) cells to release greater amounts of CCK hormone. CCK is a gut-brain satiety hormone that is secreted by the enteroendocrine cells in response to the entry of nutrients in the small intestine. The role of CCK as a major determinant in

appetite control is well documented (Reidelberger 1994). One human study has reported a greater satiating effect of a high pork diet compared to either high soy or high carbohydrate diets (Mikkelsen et al., 2000), with the pork diet having the highest potential to induce negative energy balance.

These two studies provide encouraging, albeit limited, evidence that pork may have an important role in obesity prevention and weight loss through satiating effects, when replacing other protein foods in a mixed diet. Further research in this area is warranted to also address anecdotal evidence that consumers may perceive "white" meat (poultry, pork and fish) to be less satiating than red meat.

This may be reflected in the current consumption patterns of Australian consumers. Australia has a predominantly meat-centered cuisine, with muscle meat being favoured (McLennan & Podger, 1998). Despite a gradual increase in pork meat consumption since 1996 (data available from ABS reports 7215.0; 7113.0; 7121.0), per capita consumption still falls far below that of beef. The representative industry body for pork in Australia, Australian Pork Limited, has reported a general increase in pork consumption over recent years to an estimated 8.1kg/capita by July 2009 (APL, 2009). For a competitive edge, the pork meat industry will need to promote the health properties of lean fresh pork, particularly in the context of a dramatic rise in chicken consumption in the country (ABARE, 2007, cited in Charlton *et al*., 2008).

The aim of the study reported here was to demonstrate that the consumption of a pork meal is has a greater effect on acute satiety than comparative meals using animal protein sources, namely beef and chicken.

Significance

The study findings will provide evidence regarding the positioning of pork and pork products in food based dietary advice to support weight loss. Little is known about how pork meat can regulate appetite when consumed as part of a composite meal and which mechanisms may be important. Elucidation of these mechanisms would add a new marketing opportunity for the pork industry in the context of consumer demands for foods to improve health and wellbeing. In addition, regulatory agencies require scientific evidence of the health benefits of foods to underpin commercial marketing activity, such as would be determined within this study.

The primary targeted outcome of this research was to provide evidence of any relative benefits of pork consumption on appetite and satiety. The research strategy is grounded in a mechanistic understanding of the role of protein in the hormonal regulation of appetite. The benefit of pork protein over that of other animal sources has not been adequately investigated to date, however the positive findings of the previous animal study (Sufian et al., 2006) provided the basis for the study hypothesis. The study sought to address unanswered scientific questions that would allow practical application of the results to supporting healthy consumer food-related behaviour.

The significance of this project is that pork may be positioned as an optimal protein source to include in products and in diet plans aimed at weight loss and maintenance. The innovation was in determining if the protein-induced changes in appetite and satiety, and subsequent effect on food intake were dependent on the

protein source (pork). The comparison of a pork based meal with meals containing other types of animal protein would provide valuable scientific and commercial information on the role of pork as a meat protein source within the context of a weight loss diet.

2. Methodology

Study design

A within-subjects design was employed whereby each participant serves as her own control. On three test days, fasting participants attended a research centre and consumed one of three meat-containing meals (pork, chicken, beef) that were matched in energy (kJ), total protein content, palatability, and appearance. The sequence of the three meals was randomly generated to yield six possible orders of presentation of the meals, which was balanced across participants. The primary outcome measures were: (1) amount of food consumed at a subsequent *ad libitum* buffet lunch meal; (2) amount of food consumed and macronutrient selection for the rest of the day; (3) Visual Analogue Scale (VAS) ratings for hunger and satiety; and (4) hormonal appetite and satiety signals.

Participants

Sample size was calculated based on a previous study comparing the effects of two protein and two carbohydrate preloads on subsequent energy intake at a buffet meal consumed 4 hours later (Bowen et al, 2006a). For a 10% change in energy intake (which is considered to be a clinically relevant outcome) the SE is 3% for 19 subjects giving a SD of 15.1%. Based on paired t tests with a Bonferroni adjustment (for 3 comparisons taking the P value to 0.02) it was estimated that 26 participants were required for 80% power. Total sample size was increased to 30 people, to account for potential drop-outs.

Inclusion criteria: aged 18-50 years, BMI >18.5 and < 35 kg/m^2, generally well, pre-menopausal.

Exclusion criteria: major illnesses, Type 1 and Type 2 diabetes, cigarette smokers, pregnancy/lactation, abnormal scores on the eating inventory scale (dietary restraint), food allergies or habits inhibiting compliance with the study design, illiteracy and inadequate conversational English.

Because hormonal mechanisms of appetite control are stronger in women only women were recruited to the study. Limiting the study to female participants also created a homogenous sample that minimized a potentially high degree of variance associated with food intake by inclusion of both men and women. Food intake varies by the phase of the menstrual cycle (Johnson et al., 1994); therefore, all test lunches were conducted during the same phase of the menstrual cycle. Menstrual cycle phase was assessed with a semi-structured interview. Participants with very irregular menstrual cycles were excluded due to scheduling difficulty. Females taking monophasic oral contraceptives were eligible to participate in the study, but those taking other oral contraceptive regimens and prescription medications were excluded.

Participants who reported smoking cigarettes were excluded due to the effects of nicotine upon taste and appetite (Grunberg, 1982). Those taking herbal

supplements were asked to discontinue use during the study. Participants were also be excluded if they reported any allergies to the foods used in the study or if they disliked the test food that was used at the lunch-time meal.

Recruitment and screening

Recruitment was by way of advertising in the local area, including posters and presentations, use of community notice boards and via University Staff emails. Participants were screened using three methods to confirm eligibility into the study (Fig 1).

After successfully completing a screening questionnaire via telephone which included questions related to sociodemographic characteristics, recent weight loss and willingness to eat the three test meats, participants were asked to complete the Three Factor Eating Questionnaire (TFEQ, n=55). Originally, potential participants were excluded if they had elevated scores for any of the three eating behaviour constructs of the eating inventory (EI) (Stunkard & Messick, 1988), namely: dietary restraint (score >14); disinhibition (score >12); and perceived hunger (score >12). The three items of the eating inventory instrument referred to the following constructs related to eating behaviour: dietary restraint scale measures dieting behavior and intention to diet; the disinhibition scale measures loss of control of eating; while the perceived hunger scale provides a subjective measure of hunger state (Stunkard and Messick, 1985; 1988). Due to recruitment difficulties, we later amended this eligibility criterion to only exclude participants who scored >14 on the dietary restraint eating scale, with appropriate approvals.

Eligible participants (n = 38) were then asked to complete a final face-to-face screening. During this appointment anthropometric measurements and blood pressure readings were taken. Body weight and percentage body fat was measured whilst standing using bioelectrical impedance scales (Tanita TBF-622). These scales have been validated and are thought to be a reasonable comparison with dual X-ray absorptiometry as a reference method. (Batterham *et al.,* 2002). Waist and hip circumference measurements were taken with a flexible tape measure and fasting glucose levels were measured using an Accutrend GC Meter.

Participants with very irregular menstrual cycles were excluded due to scheduling difficulty. Two participants completed three sessions during the luteal phase, while the remaining participants completed their test visits during the follicular phase (i.e. between end of menses and before ovulation).

Randomisation and Blinding

A researcher independent of the subject interface undertook the randomisation of subjects into order of test meal allocation.

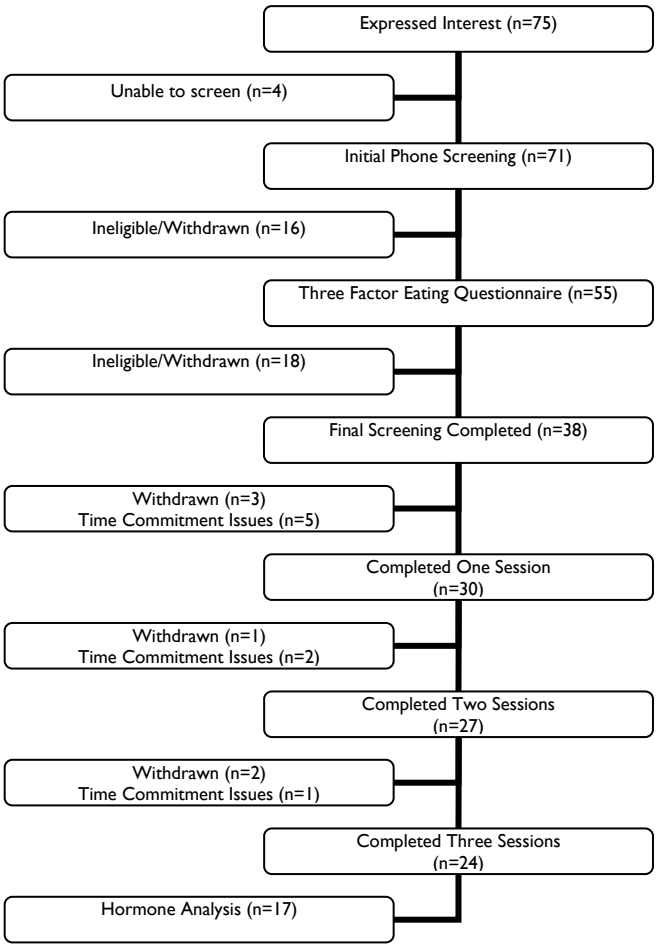


Figure 1: Flowchart of recruitment, screening and enrolment

Testing procedures

Participants were instructed to refrain from eating for 12 h (overnight), and not to consume alcohol or perform exercise for 24 h, prior to their breakfast test meals. Participants were provided with a commercially prepared frozen meal to consume for the evening meal the night before the test day. Upon arrival at the research center for the breakfast test meal, participants were questioned about adherence to these instructions. Depending on appointment scheduling, participants were required to consume the test meal between the hours of 7:00 and 9:00 am, within a 20-minute period. After consumption of the test meal, participants were provided with a 600 ml bottle of water which they could freely consume between breakfast and lunch.

Breakfast test meals

The three test meals comprised a toasted sandwich made with beef, chicken or pork. The three test breakfast meals were matched as closely as possible for taste and appearance (Table 1) as well as for nutrient content (Table 2). The meals were consumer-tested for acceptability and palatability as a breakfast meal with staff at the University of Wollongong prior to the study commencement. All meats used in the study were purchased from one supplier on the same day and frozen in vacuum-packed 140g individual portions. In order to standardize the pork meal, all portions were obtained from a single pig carcass. Similarly, all beef and chicken

meat was obtained from the same breed of cattle and poultry, and provided from a single farm, respectively.

Table 1: Food and Ingredients of test meals

Test breakfast: Meat sandwich ingredients	Buffet lunch ingredients
140g Meat (Chicken, Beef, Pork)	150g of pasta (dry weight)
3 slices of Wholegrain Bread	250g Tomato-based pasta sauce
3 tsp Polyunsaturated Margarine	100g Grated Cheese
1 tsp Soy Sauce	10 Black Olives
1 tsp Olive Oil	Salad containing: 50g lettuce, 100g tomato, 60g cucumber, 50g red capsicum.
0.5 tsp Garlic	60g salad dressing
	2 small white bread rolls (~40g each)
	20g polyunsaturated margarine
	200g fruit yoghurt
	100g grapes

Table 2: Macronutrient profiles of breakfast test meals

	Energy (kJ)	Protein (%E)	Fat (%E)	Carbohydrate (%E)	Fibre (g)
Breakfast					
Sandwich - Pork	2493.29	27.12	31.08	38.09	6.45
Sandwich – Beef	2467.92	28.61	31.13	37.70	6.45
Sandwich – Chicken	2647.29	29.27	35.53	35.51	6.45

Measurements

Subjective assessment of satiety – Visual Analogue Scales

Food intake motivation was assessed with 100 mm line visual analogue scales (VAS). Participants completed Visual Analogue Scales (VAS) to measure hunger/fullness at the same time points that blood samples were taken throughout the study protocol (ie. 0, 15, 30, 60, 90, 120, 180 minutes). The VAS questionnaire provides a subjective assessment of appetite-related sensations over time and allows the relationship between biochemical and dietary intake data to be explored. Visual analogue scales have been found to be a reliable and valid method to assess subjective states related to food intake behaviour (Flint et al., 2000; Geiselman et al., 1998).

On each of the test days, participants were asked to rate the following components relating to hunger and satiety using various questions, scored using the VAS instrument:
(1) Hunger: how hungry do you feel at this moment? (VAS1)
(2) Desire to eat: how strong is your desire to eat at this moment? (VAS5- VAS8);
(3) Fullness: how full does your stomach feel at this moment? (VAS2, VAS3);
(4) Motivation to eat: how much food do you think you could eat at this moment? (VAS4).

Table 3: shows the actual questions asked and the range of responses on the VAS scale (ranked from 1 – 100)

Item	Low (score = 1)	High (score = 100)
How hungry do you feel? VAS question 1	I am not hungry at all	I have never been more hungry
How satisfied do you feel? VAS question 2	I am completely empty	I cannot eat another bite
How full do you feel? VAS question 3	Not at all full	Totally full
How much do you think you can eat? VAS question 4	Nothing at all	A lot
Would you like to eat something sweet? VAS question 5	Yes, very much	No, not at all
Would you like to eat something salty? VAS question 6	Yes, very much	No, not at all
Would you like to eat something savoury? VAS question 7	Yes, very much	No, not at all
Would you like to eat something fatty? VAS question 8	Yes, very much	No, not at all

Blood samples

Blood samples were collected at 0 (fasting), 15, 30, 60, 90, 120 and 180mins. One tube containing EDTA was collected for gut hormone analyses and one SST tube collected for glucose and insulin analyses. Blood samples were collected by qualified phlebotomists and trained cannulation specialists. Glucose and insulin levels were analysed by the local branch of Southern IML Pathology. Gut hormone analyses were performed by Dr John Cardinal (Cardinal Bio-Research Pty Ltd) in Queensland. The EDTA tube was treated with 90µl 100mg/ml Roche Pefabloc (Cat. # 11 429 868 001). Roche Pefabloc is a protease inhibitor that prevents the natural degradation of peptides once they have been removed from in vivo.

Subsequent food intake

- *Buffet lunch*

Three hours after the breakfast test meal, participants were provided with an *ad libitum* buffet style lunch meal comprising a tomato-based pasta meal, together with a selection of other food items including fruit, yoghurt and salad (Table 2). Time allocated for the buffet lunch meal was a maximum of 30 minutes. The same meal was served at each of the lunch sessions on the three testing days. After lunch, dietitians weighed the remaining food items and calculated actual food intake.

- *Energy intake for the rest of the day*

Participants left the research facility after lunch and were instructed to carefully record all food and beverages consumed for the remainder of the day, using a

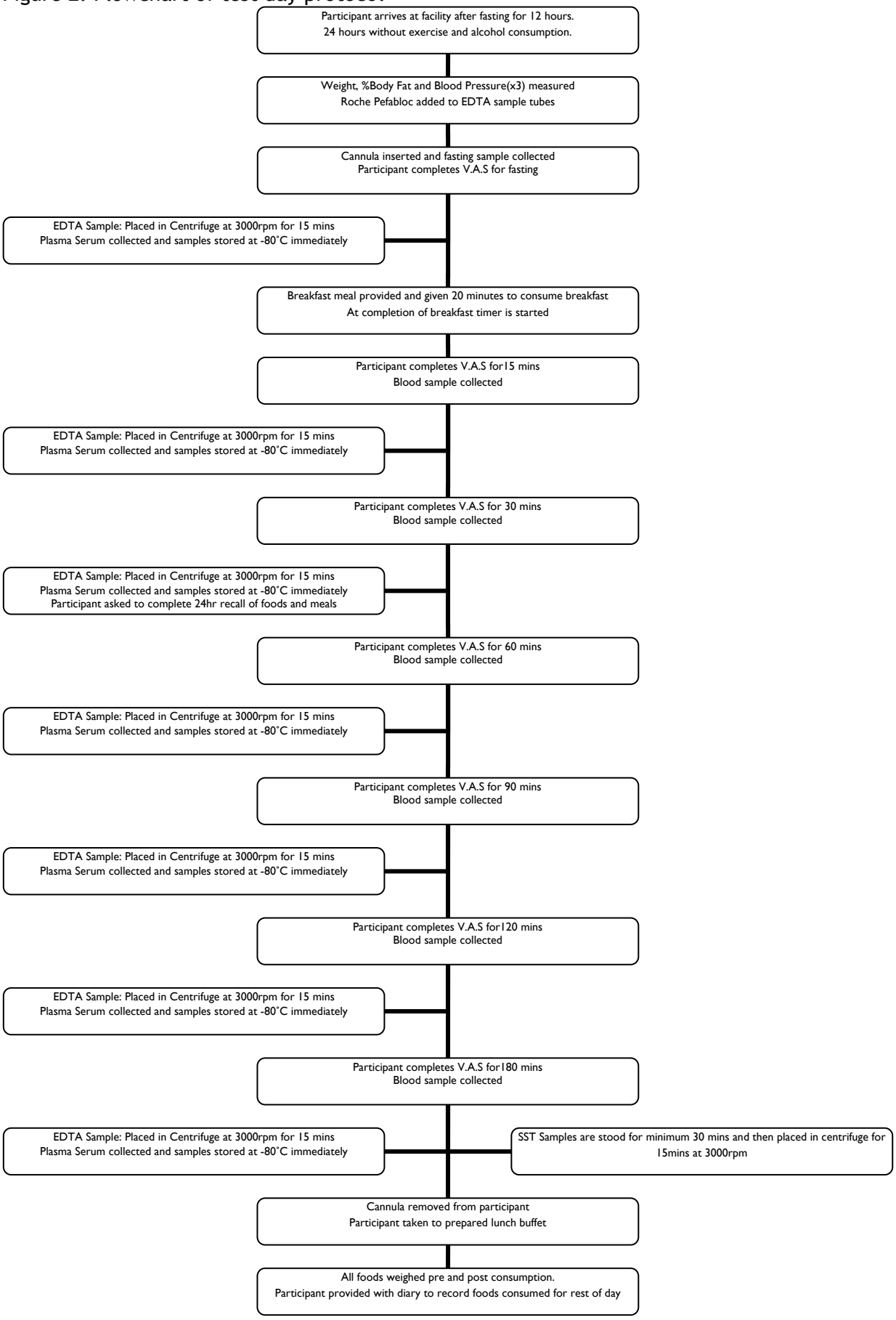
quantified food diary. The completed food diaries were returned to the research facility, either in person or by post.

All dietary data was analysed for nutrient content using FoodWorks software system (Xyris Software, Highgate Hill, GLD, Australia, Version 5, 2007). Nutrient intake data was analysed in terms of energy and macronutrients using the AusNut (Allfoods) Revision 18 database.

Statistical analyses

Statistical analysis was completed using SPSS Software (version 15, 2002: SPSS Inc, Chicago, IL, USA). The primary outcome measures were analysed using a linear mixed model. The advantage of this model is that it incorporates all available data hence partial datasets from those who do not complete the study are still included in the analysis. The Area Under the Curve was assessed using trapezoidal methods for all VAS and biochemical data and analysed using repeated measures ANOVA.

Figure 2: Flowchart of test day protocol



3. Outcomes

Participant characteristics

Of the 71 volunteers who were screened, thirty participants completed at least one of the test sessions (Table 4).

Table 4: No. of participants at each stage of study

Recruitment, Screening and Enrolment	
No. of volunteers screened	71
No. of volunteers completed Three Factor Eating Questionnaire	55
Participants allocated randomization code	39
No. of volunteers who completed final screening	38
No. of participants who withdrew post-enrolment (no sessions completed)	6
No. of participants who withdrew post-enrolment (completed 1 or 2 sessions)	3
No. of participants completed 0 sessions	2
No. of participants completed 1 sessions	30
No. of participants completed 2 sessions	27
No. of participants completed 3 sessions	24
No. of participants enrolled at study completion	29

The characteristics of the participants are shown in Table 5. The age range of the women was between 19 and 45 years, while BMI ranged from 19.2 – 38.3 and percentage body fat from 16.5 – 52.5 %. Eight participants were obese (BMI ≥30), three overweight (BMI = 25 – 29.9) and the remainder (n = 19) normal weight (BMI = 18.5 – 24.9).

Table 5: Characteristics of subjects at baseline (mean (SD)

	Women (n=30)
Age (years)	27.37 (±8.22)
Weight (kg)	72.32 (±18.85)
Height (m)	1.67 (±0.07)
BMI (kg/m2)	25.87 (±5.99)
Waist circumference (cm)	84.47 (±15.11)
Hip circumference (cm)	99.71 (±16.91)
% Body Fat (%)	32.86 (±9.94)
Blood Glucose (mmol/L)	3.13 (±1.03)
Systolic Blood Pressure (mmHg)	116.05 (±10.99)
Diastolic Blood Pressure (mmHg)	71.08 (±8.57)

The primary outcomes are reported according to differences between responses to test meals with respect to:

- Visual Analogue Scale scores (subjective hunger and satiety)
- Energy intake at subsequent meals
- Biochemical markers of appetite

Visual Analogue Scale (VAS) ratings for hunger and satiety

There was no difference in VAS ratings over 180 minutes (calculated as Area Under the Curve) for either (i) hunger (VAS1), (ii) desire to eat (VAS5 - VAS8), (iii) satiety/fullness (VAS2, VAS3) or (iv) motivation to eat (VAS4) across the three test days, as shown in Table 6.

Table 6: Visual analogue scale (VAS) measurements of subjective hunger and satiety following consumption of three meat-containing breakfast meals (AUC)*

Variable	Beef (n=26)	Chicken (n-26)	Pork (n=29)	P-Value
How hungry do you feel? VAS 1	558.37±312.41	596.58±234.15	594.59±303.10	0.730
How satisfied do you feel? VAS 2	1163.01±319.31	1094.00±274.66	1138.50±291.89	0.407
How full do you feel? VAS 3	1139.82±345.41	1088.91±249.06	1127.04±295.02	0.663
How much do you think you can eat? VAS 4	592.92±284.16	661.73±252.36	720.36±285.29	0.085
Would you like to eat something sweet? VAS 5	1141.08±506.91	1081.88±538.68	1009.69±497.00	0.404
Would you like to eat something salty? VAS 6	1485.85±250.86	1400.73±309.27	1430.07±263.93	0.179
Would you like to eat something savoury? VAS 7	1342.97±320.51	1270.37±306.83	1318.05±304.04	0.294
Would you like to eat something fatty? VAS 8	1545.95±342.13	1527.14±275.98	1540.19±338.53	0.973

*AUC – area under curve

VAS ratings over the seven time points between breakfast and buffet lunch (0 (fasting), 15, 30, 60, 90, 120 and 180mins) are shown graphically in Figures 3 – 9 [it should be noted that intervals between the plotted time points are not equal].

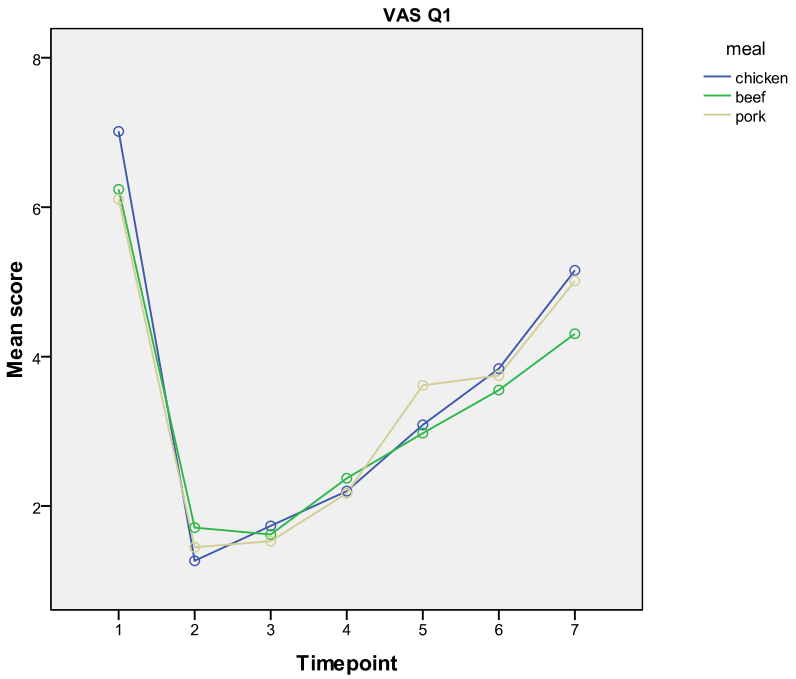


Figure 3: Visual analogue scale (VAS) measurements of subjective hunger (VAS1: **"How hungry do you feel?")**

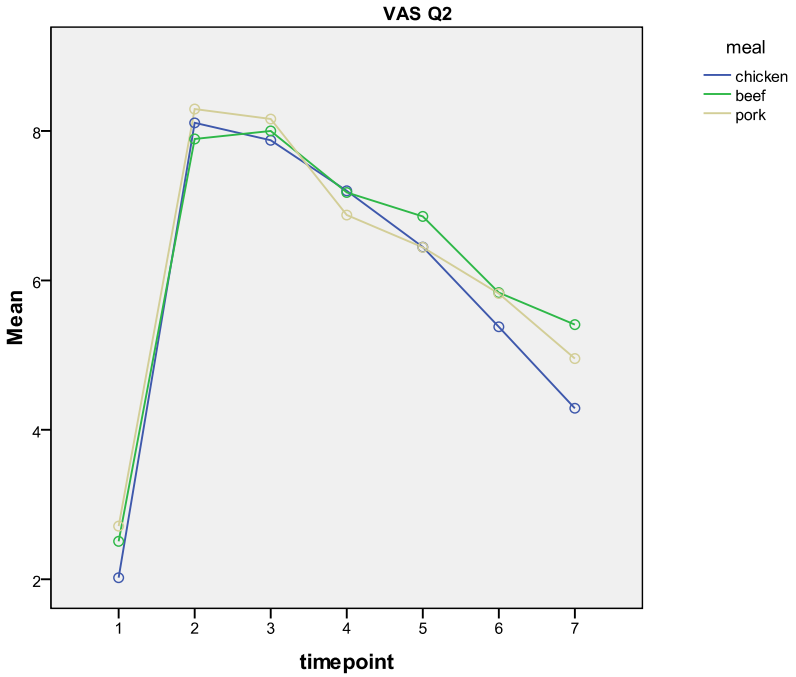


Figure 4: Visual analogue scale (VAS) measurements of satiety/fullness (VAS2: **"How satisfied do you feel?"**

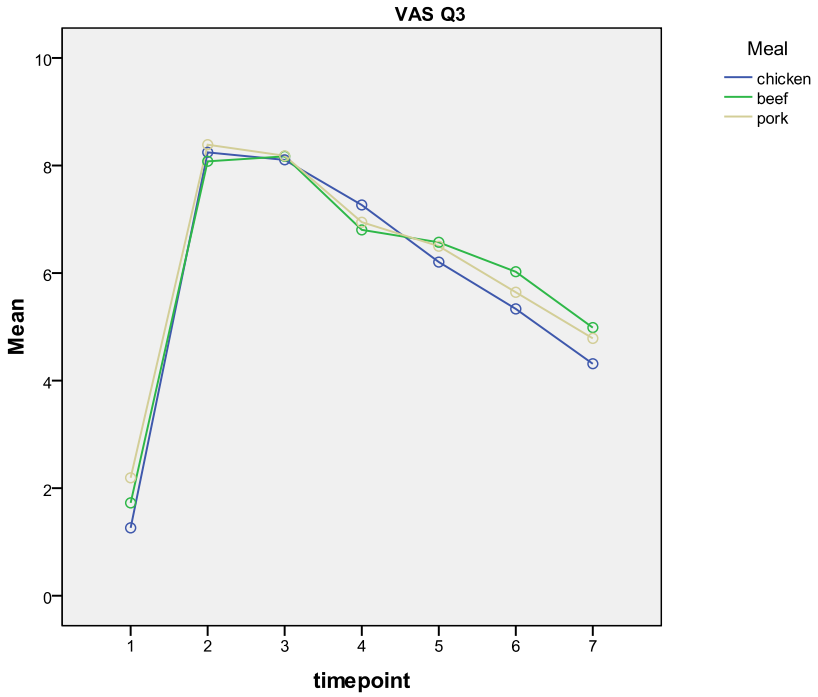


Figure 5: Visual analogue scale (VAS) measurements of satiety/fullness (VAS3: **"How full do you feel?"**

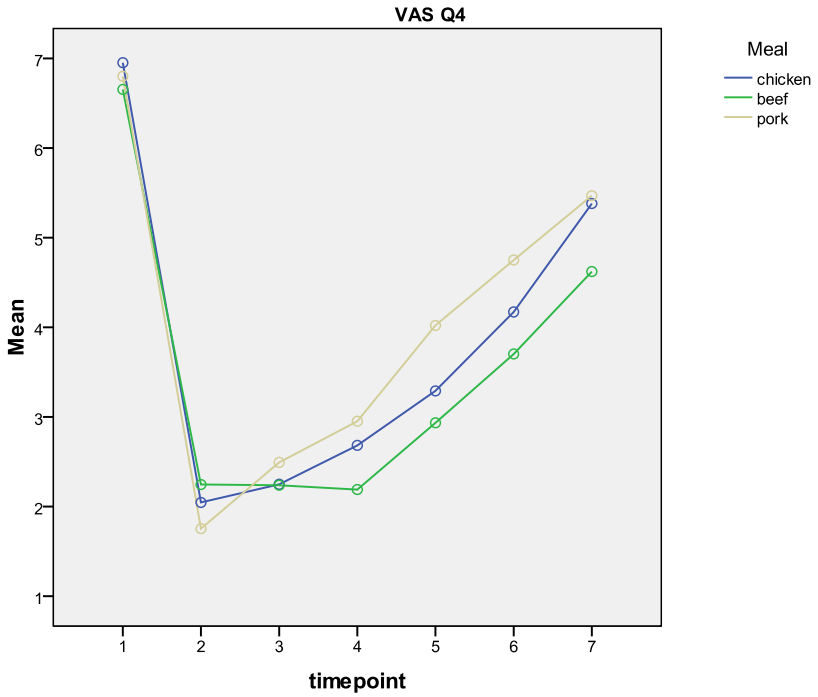


Figure 6: Visual analogue scale (VAS) measurements of motivation to eat (VAS4: **"How much do you think you can eat?"**

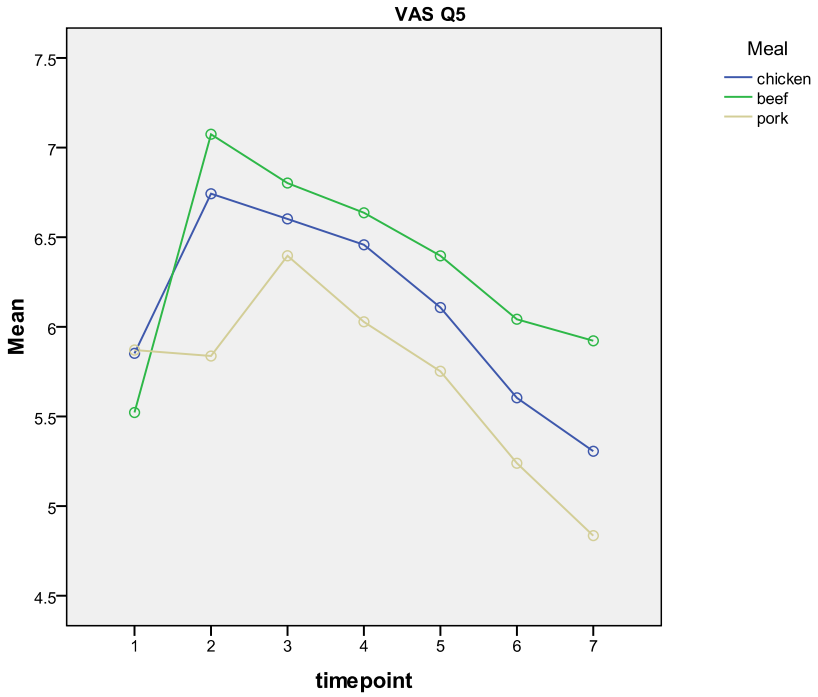


Figure 7: Visual analogue scale (VAS) measurements of desire to eat (VAS5: **"Would you like to eat something sweet?"**

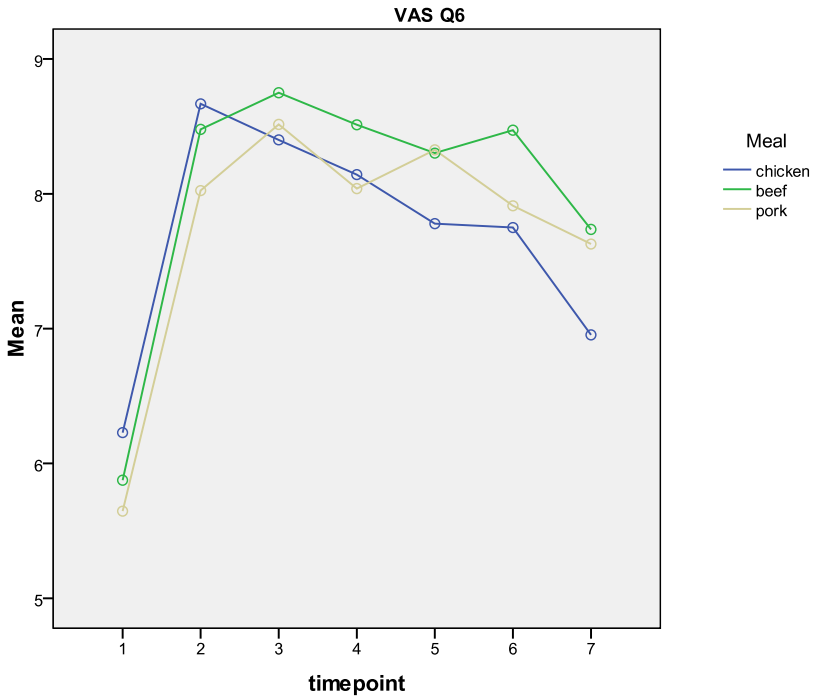


Figure 8: Visual analogue scale (VAS) measurements of desire to eat (VAS6: **"Would you like to eat something salty?**

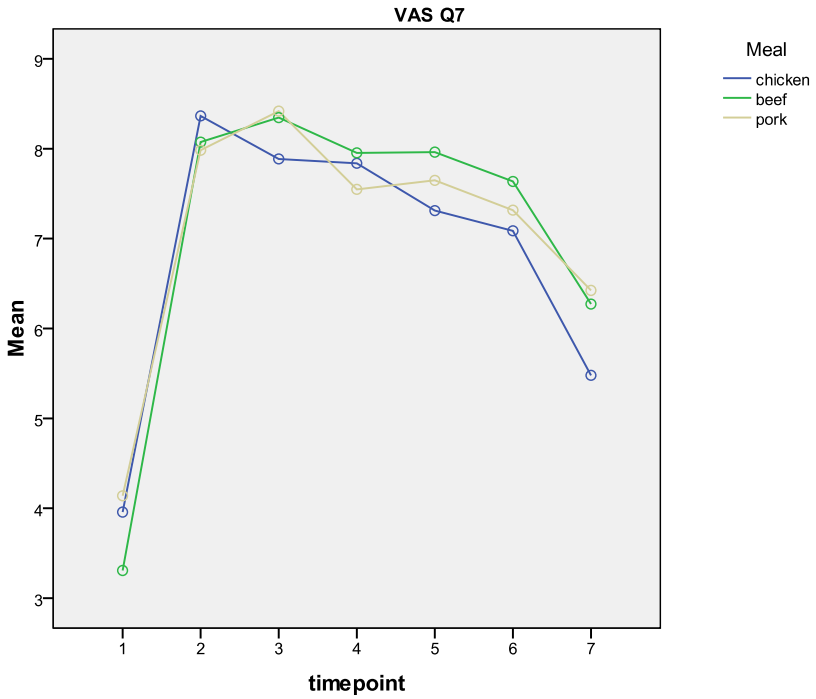


Figure 9: Visual analogue scale (VAS) measurements of desire to eat (VAS7: **"Would you like to eat something savoury?"**

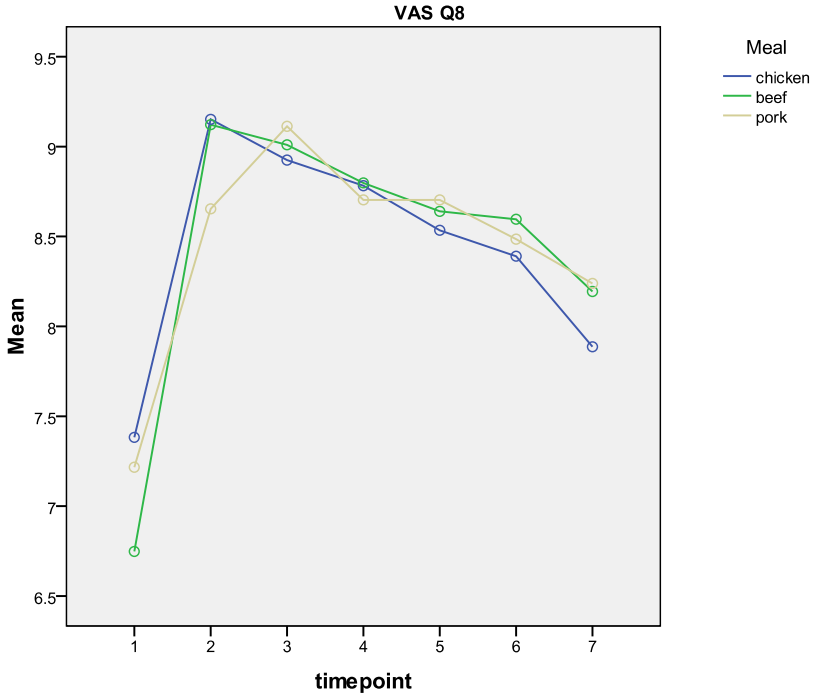


Figure 10: Visual analogue scale (VAS) measurements of desire to eat (VAS8: **"Would you like to eat something fatty?"**

Food consumed at a subsequent ad libitum buffet lunch meal and for the rest of the day

No difference was found between meat groups for either energy intake, or for macronutrient profile (% energy from protein, fat or carbohydrate) of food consumed at the buffet lunch, following test meal breakfast consumption (Table 7). Body weight remained stable over the three test days.

Similarly, participants consumed comparable quantities of food over the rest of the day on each of the three test meal days, indicating no test meal effect on later food choice (Table 7).

Table 7: Energy and macronutrient intakes at buffet lunch and for the rest of the day following consumption of three meat-containing breakfast meals

Variable	Beef (n=26)	Chicken (n-26)	Pork (n=29)	P-Value
Weight (kg)	72.57±19.11	72.67±17.40	73.16±18.17	0.568
Buffet lunch				
Buffet Lunch Energy (kJ)	2581±869	2730±881	2678±1029	0.617
Buffet Lunch Protein (%E)	14.37±1.51	14.58±1.37	14.19±2.38	0.596
Buffet Lunch Fat (%E)	21.17±6.12	21.42±5.94	21.80±7.67	0.684
Buffet Lunch CHO (%E)	62.47±6.0	62.01±6.27	62.15±9.07	0.687
Rest of the day intake				
Rest of the day Energy (kJ)	4179±1588	4039±2177	3589±1733	0.348
Rest of the day Protein (%E)	19.00±7.96	18.98±6.77	21.12±11.73	0.577
Rest of the day Fat (%E)	33.71±13.62	29.76±11.31	36.17±10.44	0.132
Rest of the day CHO (%E)	43.27±16.40	43.89±13.41	39.66±13.83	0.434

Appetite hormones

Change in blood concentrations of gut hormones associated with hunger and satiety (CCK, PYY, ghrelin), as well as for glucose and insulin, over the 180 minutes following consumption of the test meal are shown, for each of the three test meals, in Table 8 (shown as Area Under the Curve, AUC)and in Figures 11 – 15 (for time points 0 (fasting), 15, 30, 60, 90, 120 and 180mins; intervals between the plotted time points are not equal). No between-meal differences were found for calculated area under the curve (AUC) measurements for glucose, insulin, CCK and ghrelin. For PYY, a higher AUC was found for the pork compared to chicken test meal (Figure 11). Despite the pork group having a higher AUC for both CCK

and ghrelin compared to the other two groups, the differences between groups was not significant (P=0.587 and P = 0.148, respectively).

Table 8: Gut hormone measurements following consumption of three meat-containing breakfast meals

Variable (AUC)	Beef (n=26)	Chicken (n-26)	Pork (n=29)	P-Value
PYY (n=17)	46250±18268	39040±11639	46949±13269	0.027*
CCK (n=17)	1935±961	1917±1068	2110±1231	0.587
Ghrelin (n=17)	6082±4926	5173±4373	7787±8004	0.148
Glucose (n = 17)	832.68±137.84	827.48±102.24	819.47±147.28	0.495
Insulin (n = 17)	5381.83±2392.13	5510.80±1882.21	6088.38±2661.46	0.354

AUC – area under curve
**significant difference between chicken and pork only*

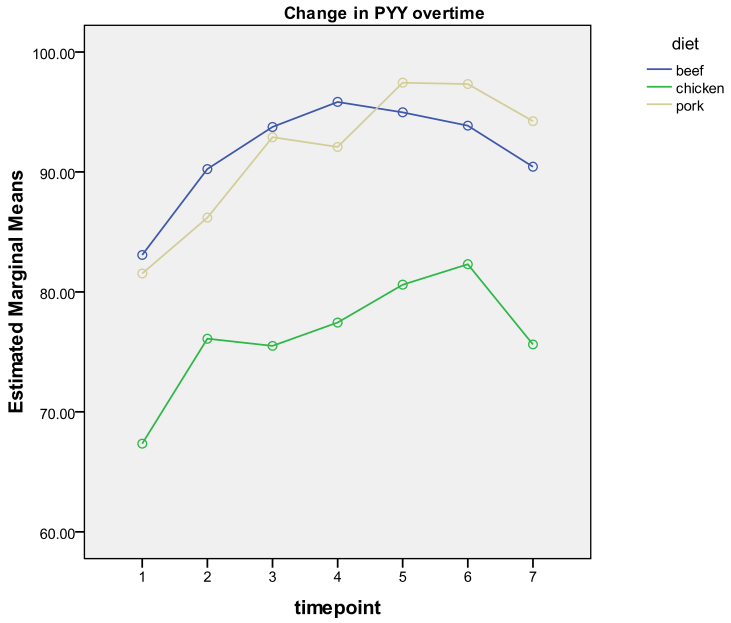


Figure 11**: Change in PYY concentrations over time for the three test meals**

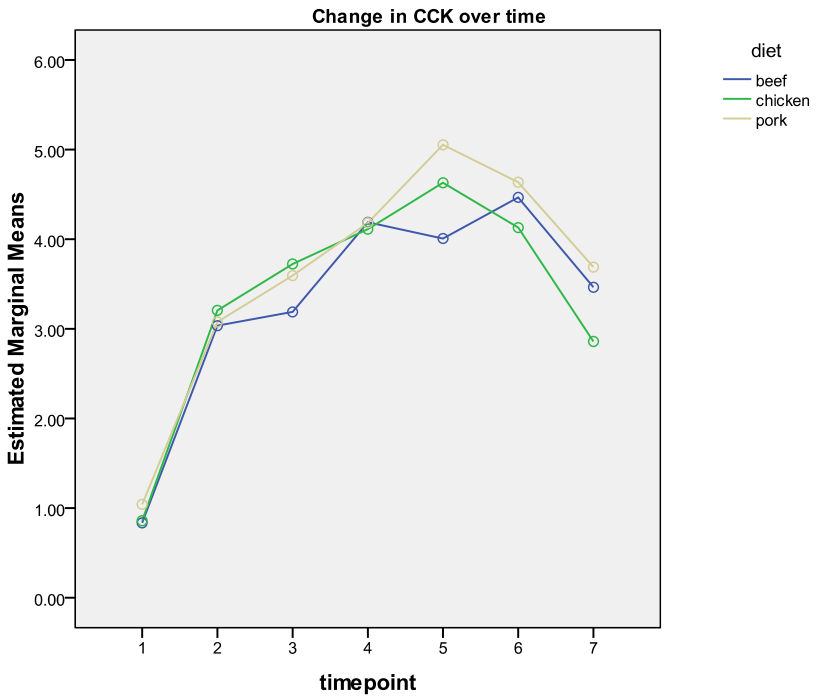


Figure 12: Change in CCK concentrations over time for the three test meals

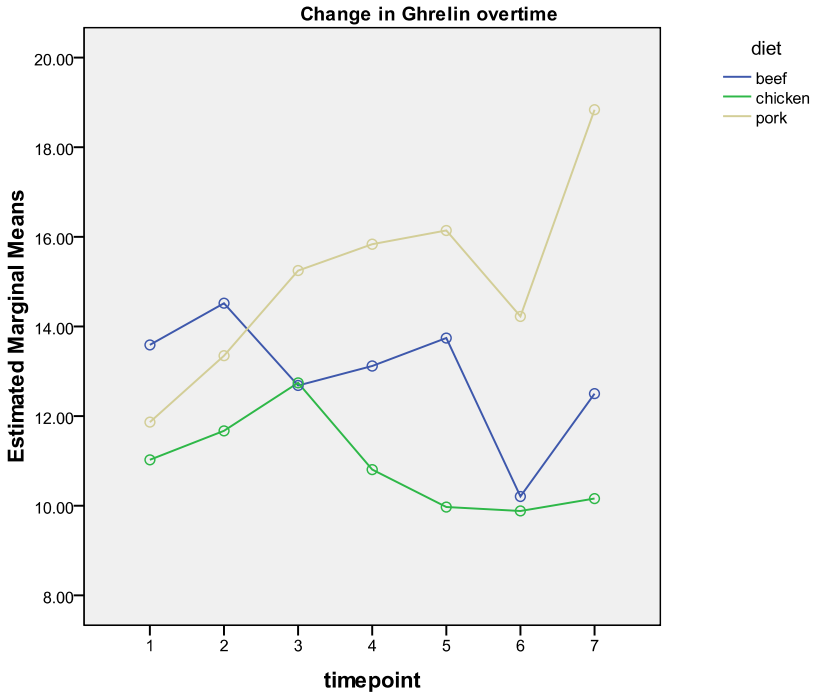


Figure 13: Change in ghrelin concentrations over time for the three test meals

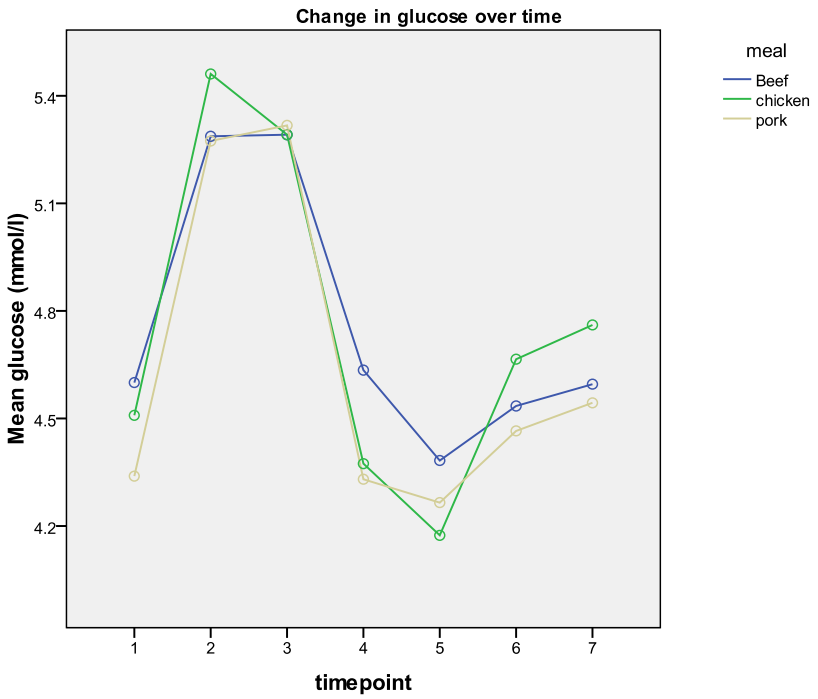


Figure 14: Change in glucose concentrations over time for the three test meals

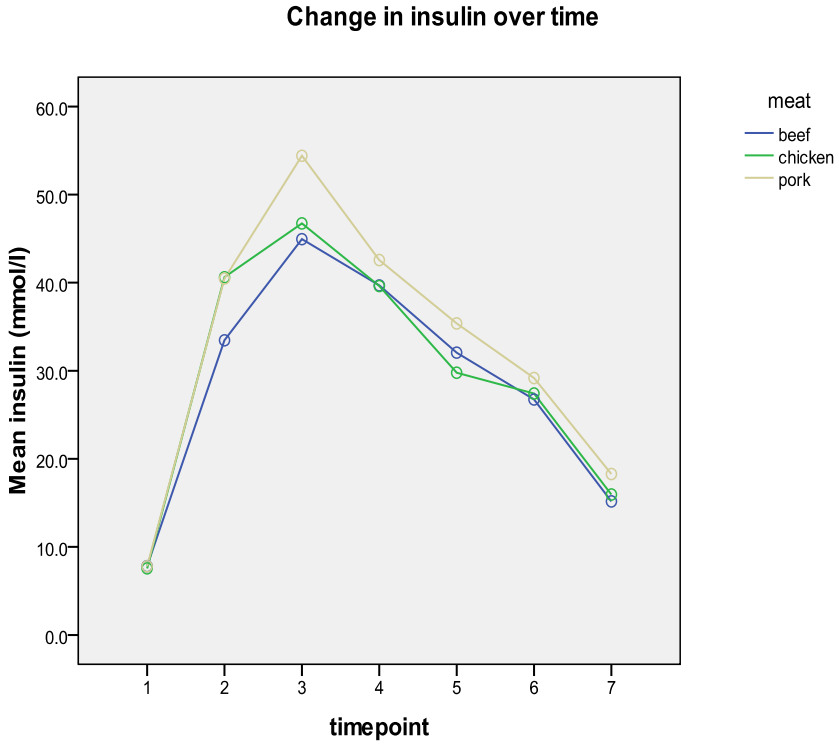


Figure 15: Change in insulin concentrations over time for the three test meals

4. Application of Research

In this study we were not able to show that pork is more satiating than beef or chicken. The question was investigated by comparing various outcome measures of satiety, namely Visual Analogue Scales (VAS) for subjective satiety, subsequent energy consumption for evidence of hunger (buffet lunch meal and rest of day) and appetite hormones (CCK and PYY to indicate satiation; and ghrelin and insulin to indicate hunger levels). This followed previous research demonstrating that a fish protein meal produced a greater short-term satiety compared to either a beef or chicken meal (Uhe *et al.* (1992). A further study found that fish protein resulted in higher subjective satiety ratings than other protein-rich food items tested, such as beef steak and eggs. In this study 38 different food items providing 1000 kJ each were consumed and rated for subjective satiety every 15 min over a 2-hour period (Holt *et al.,* 1995). Another study detected no significant differences in subjective ratings of satiety or hunger associated with fish protein compared to beef protein meals in healthy young males. However, an 11% reduction in energy intake at the subsequent evening meal was associated with the fish meal (Borzoei et al., 2006). These results suggested it might be worthwhile considering the effects of pork meals.

We measured acute satiety responses associated with eating pork as a breakfast meal. No differences were reported in subjective feelings of hunger and/or appetite over a 3-hour period after consuming pork, beef or chicken. Similarly, compared to either beef or chicken, pork consumption did not appear to result in a decreased dietary intake (energy or protein) at the subsequent meal (lunch) nor for the rest of the day. These findings related to acute (short-term) satiety and cannot be extrapolated to the longer term. Indeed longer term studies also appear to suggest a value in high protein diets but no differentiation between protein sources. For example, Mahon et al. (2007) found that both higher protein energy restricted diets (using chicken and beef) resulted in lower energy intakes and greater weight loss and fat mass loss than an isocaloric high carbohydrate diet. These findings were supported by another study (Melanson et al., 2003) in which both chicken and lean beef-containing diets achieved a similar weight loss and similar improvements in lipid profiles.

Mechanisms of action

The short-term regulation of food intake is mediated via neural and humoral signals from the gastrointestinal tract to different regions in the brain (Salmenkallio-Marttila et al., 2009). Consumption of different macronutrients (carbohydrates, protein, fibre and fat) from foods affects the release of satiety-related peptides from the stomach and different parts of intestine. For example, the release of glucacon-like peptide 1 (GLP-1) and peptide YY (PYY) from the intestine soon after eating a meal increases satiety. In the longer term, regulation of food intake by hormones such as leptin from adipose tissue and insulin from the Pancreas may play a significant role. In terms of satiety regulation, some studies support the concept that the consumption of a high-protein diet decreases circulating ghrelin, an orexigenic gut peptide associated with feelings of hunger, whereas it increases the concentrations of the anorexic gut peptides (associated with satiety) such as CCK and GLP-1 (Blom et al., 2006; Bowen, et al., 2006; Tannousdit et al., 2006). However, other studies suggest that the satiety induced

by high-protein diets is unrelated to changes in circulating ghrelin (Weigle et al., 2005; Moran et al., 2005). Genetic evidence also supports a role for another hormone, the anorexic peptide YY (Pyy) in protein-mediated reduction in food intake (Batterham et al., 2006).

In the present study, with the exception of a difference in PYY between chicken and pork meals (P = 0.027, see Table 8) no significant differences were found for any of the appetite hormone levels investigated (CCK, ghrelin, insulin) after consumption of pork, beef or chicken. The difference in PYY levels suggests that consumers choosing between white meats may have some kind of physiological advantage by choosing pork over chicken but it is not possible to translate into observable differences in overall dietary consumption, which is the level of information that is relevant to consumers.

The role of CCK as a major determinant in appetite control through increasing satiety in response to the entry of nutrients into the small intestine is well documented (Reidelberger 1994). The CCK results show expected trends from food consumption but no differentiating effects from the different foods. This may be related to the experimental food form with protein type having a lesser differentiating effect than other food components such as fat or fibre.

The ghrelin trends are interesting. As expected from the main physiological effect of ghrelin being an initiation of the next eating episode (through increased hunger sensations), we observed a substantive increase in levels of this hormone approximately 120 minutes after the breakfast meal and this was similar across all three meats tested, with no difference between groups. The trends in shifting ghrelin levels was consistent with the initiation pattern of meals/snacks in regular eaters, but our results suggest no differential effects from any meat type on changes in ghrelin levels. Other studies have found ghrelin to decrease after high protein mixed meals, which include yoghurt as the primary protein source (Al-Awar et al. 2005, Blom et al. 2006). It has been suggested that the higher satiety associated with the consumption of high protein foods may partially be mediated by prolonged ghrelin suppression (Bowen et al. 2006b) as well as the type of protein ingested (Aziz and Anderson 2007). However, there are conflicting findings that demonstrated a rise in ghrelin levels to a plateau after the ingestion of a high-protein turkey meal (Erdmann et al. 2003). These may be due to differences in the time of digestion and absorption of the test food (Bowen et al. 2006a).

It is also known that different protein sources have different effects on the release of insulin (Gannon et al., 1988; Westphal et al., 1990), and this may be important to the mechanism of action of the enhanced satiety (Latner and Schwartz 1999). Greater releases of insulin, although not always considered positive for metabolic control, are also often associated with decreased intake at subsequent meals. In our study, no difference was found between the three meats for either insulin or glucose release over time (AUC). Previous research tends to support a link between rapid changes in blood glucose with appetitive sensations (Arumugama et al., 2008). However, the greater the insulin response, the more likely it is that appetite will be suppressed (Flint et al., 2006). Fasting plasma insulin and insulin responses to a meal are both correlated with body adiposity (Havel, 1999). Thus, the inclusion of subjects of varying BMI may have resulted in greater variation in glucose and insulin responses to food intake, and thereby the potential to produce variation in measures of appetite such as visual analogue scales. Nevertheless,

severely obese subjects (BMI >35 kg/m^2) who would be at high risk of insulin resistance, were excluded from the sampling frame.

Strength of study design

The strength of the study design relates to the tightly controlled and standardized background diet whereby participants arrived at the research centre for each of the testing days fasting, after having consumed the same frozen meal the evening before. Eligibility criteria and careful screening procedures ensured that only non-smoking women, who were neither underweight nor severely obese participated in the study. To further reduce potential confounding factors on the outcomes of interest, test meal days were planned to coincide with the same phase of the menstrual cycle for each participant and individuals with abnormal eating habits with regard to dietary restraint were excluded. In order to minimize variability in nutrient content, all meat used for the test meals was purchased at the same time from a single supplier and was provided from the same farm for chicken, pork and beef, respectively.

This study was adequately powered for differences in the primary outcome measures, changes between groups were not of clinical or statistical significance and the risk of type II error is excluded. Previous appetite studies that have used biochemical indices (CCK) as the primary outcome showed statistical differences between CCK release and visual analogue scale (VAS) measures of satiety with as few as fifteen subjects (Burton-Freeman et al., 2002). In that study, both men and women were included and sex-group analyses were conducted in even smaller numbers (n = 7 men; n = 8 women). Similarly, an inverse relationship between satiety and CCK release was demonstrated in a dietary fibre study that had only seven subjects (Holt, 1992).

Application of the research findings in the commercial world

This study demonstrated that fresh pork meat, consumed as part of a mixed meal, has all the appetite advantages of other high protein foods, such as red meat (lean beef) and chicken.

Commercialization/Adoption Strategies

This research refers to the general use of fresh pork rather than benefits to the cost of pork production. The commercialization benefit of the research relates to increasing consumer demand for fresh pork, which is one of the six key strategic objectives making up the Strategic Plan (2005–2010) of Australian Pork Limited (APL, 2009). One of the major proposed strategies to achieve this objective is to improve consumer attitudes and perceptions towards pork. Our data supports the marketing of pork as a nutritious, high protein, low fat meat that could replace red meat or chicken in the diet without compromising an ability to satisfy hunger.

Pork will need to compete in the market place with the much more affordable chicken meat. It has been estimated that beef cost increased by an average 39% between 1998 and 2002, compared to a 19% increase for chicken (ABS 4306, various years). Whilst the retail cost of beef, lamb and pork has steadily increased, particularly since 2000, the cost of chicken has remained remarkably stable (ABS, no. 6401.0; ABS No. 6403.0). The success of increased poultry meat

consumption appears to be determined by its health image (Verbeke and Vaine, 1999). Compared to other meat types, chicken is perceived as healthier in terms of fat content and is considered to be a lean, low-fat food, particularly in the case of chicken breast fillets (Kennedy et al., 2004).

Despite its favourable nutritional profile, pork is perceived by some consumers as having a high fat content (Australian Pork Limited, Annual report 2008-09). Over the past decade or so, the pork industry has reduced the fat content of fresh pork, as evidenced by the award of the Heart Foundation Tick of Approval for 15 pork cuts. Data available on the Australian Pork Limited (APL) website documents that trimmed lean pork is the lowest in total and saturated fat content compared to red meats such as lean beef or lamb, and similar in fat content to skinless chicken breast. Fresh pork meat has an advantage over chicken meat in terms of a higher thiamine and iron content. Despite having a lower iron content than red meats, pork has a much higher selenium content which may provide opportunities for research in cancer prevention.

Potential competitors in the appetite control for weight loss include the increasing range of vegetable protein ingredients (pulses, cereal protein, fungi, nuts, vegetables) for the manufacture of meat substitute products that are becoming available. Among the health benefits of mycoprotein and tofu that are promoted are their positive satiety effects (Rodger 2001, Sadler 2004), at least compared to chicken (Williamson et al. 2006). It may be worthwhile partnering with these foods to develop combinations of meals that demonstrate increased satiety.

Qualitative studies from European consumers have found that freshness, sensory quality and perceived healthfulness are the most important drivers of product choice with regard to meat consumption (Munoz, 1998; Verbeke and Viane, 1999). Poultry tends to be perceived more favourably than beef or pork in terms of these attributes (Verbeke and Viane, 1999). Such data is not available for Australian consumers. Consumers are not always presented with accurate dietary information with which they can make a balanced judgement of the healthiness of foodstuffs such as meat (Lea & Worsley, 2001). The results of this study suggest that pork is at least as satiating as red meat and chicken and this message may be useful to convey.

In recent years, there has been an increasing rise in vegetarianism in certain sectors of the population. Even in those who are not vegetarians, the choice of occasional meat-free meals as part of a varied diet appears to be increasing (Sadler 2004). A study of Australian vegetarians and semi-vegetarians found that the most important predictors of the belief that meat is intrinsically unhealthy were the perceived benefits of vegetarian diets. Three main themes emerged, namely: the perceived links between vegetarianism, peace and increased contentment; animal welfare and environmental benefits; and health benefits associated with vegetarianism (Lea & Worsley, 2001). Messages related to pork in this regard are on another level and relate to production practices and the environment.

Impact of the research

This study positions pork in the appetite arena as equivalent to comparable low fat meats. The study findings refute the anecdotal perception that red meat is more filling than white meat (chicken, pork and fish). Our study findings support those of Uhe *et al.* (1992) who demonstrated that the acute postprandial satiety

response to either beef or chicken did not differ. The benefits associated with fish in that study were attributed to differences in protein digestion and plasma concentrations and profiles of amino acids between fish and other animal proteins. A comparison between pork and other meats with regard to satiety has not previously been undertaken. The advantage of pork over either high soy or high carbohydrate diets to be able to induce negative energy balance (Mikkelsen et al., 2000) probably relates to the differing amino acid composition of the predominantly animal vs. plant-protein based diets.

The results on the effects of different macronutrients on satiety and weight control, particularly when manipulated through whole foods in a mixed meal (rather than as protein isolate preloads (Bowen *et al.*, 2006b)), are not necessarily conclusive. Nevertheless, our study comparing three high protein meats, used similar in macronutrient contents and this may be the reason no difference in effects were seen.

5. Conclusion

In this study we were not able to show that, in the short term, pork is more satiating than beef or chicken. Under tightly controlled conditions, no differences were demonstrated in subjective measures of appetite and hunger over a 3-hour period following consumption. Similarly, no differences in release of intestinal hormones associated with appetite and hunger signaling were observed. The lack of differences in effect between pork, lean beef and chicken consumption suggests that high protein meats that are similar in fat content exert similar effects on short-term appetite control. Further studies investigating the role of pork in place of other meats in the diet on food choice and weight control would be informative. Our findings confirm the position of pork in a healthy diet with regard to its ability to result in feelings of satiety that are similar to either red meat or chicken.

In an obesogenic environment where high protein diets are seen to provide opportunity for better satiety and weight loss, this study puts pork on the table. Where previously consumers may have only thought of red meat and chicken, pork is seen as equally effective. Combining pork with high volume satiating vegetables in recipes that lend themselves well to stir-fry cuts, pork may have a culinary advantage.

6. Limitations/Risks

Whilst there are minimal risks to the application of the research findings, there are a number of limitations to the study design that should be clarified.

The inclusion of participants with a wide range of BMI raises a question of whether a dilution of effects may be evident, as compared to results obtained from a more homogenous sample of either those of desirable weight, overweight or obese people alone. Much of the research in this area investigates obese and normal weight subjects separately. Generally, the appetite effects are altered with BMI so the closer the BMI ranges of the study sample, the less variation in responses.

However, some recent research indicates that for certain of the key outcome measures, a difference in test meal responses is more likely to be found in a normal weight population.

In relation to gender differences and differences in other hormone responses in subjects of varying BMI there are likely to be differences. Although the levels of hormones such as PYY are altered with obesity states, it seems that appetite-induced responses to these hormones still exist (Batterham et al., 2003) albeit somewhat attenuated (Le Roux, 2005). Therefore, the inclusion of normal weight participants as well as overweight individuals may actually improve meal-related differences in PYY responses (and thus show an effect where we may not necessarily have found one should the BMI be limited to overweight people only). It has been demonstrated that the greater the BMI, the higher post-prandial glucose and insulin response, and the smaller the meal-related associated ghrelin response (Carroll et al., 2007). For ghrelin, this means that if there are differences in ghrelin release between the test meals, inclusion of normal weight subjects may actually help show statistical significance in outcomes.

Importantly, use of the repeated measures study design where each participant acts as her own control, means that weight status is less likely to impact on outcome measures than if a parallel study design was used. Other variables that are probably more important influences on the outcomes of interest, and which we tightly controlled for during screening and recruitment, include: (1) smoking (only non-smokers eligible); (2) abnormal attitudes to eating (the Three-Factor Eating Questionnaire was used during screening to determine abnormal responses on the dietary restraint scale) and (3) menstrual cycle (we standardized this for each participant to ensure that all three laboratory testing days were conducted within the same phase of their menstrual cycle).

In the present study, pork was provided as a toasted sandwich meal. The synergistic role of other food components consumed at the same time as pork on appetite was not investigated. Current research on the role of fat quality and glycaemic index, as well as quantity and type of fibre (such as B-glucan), on the regulation of weight, satiety and hunger is actively ongoing and some promising results are being reported (Beck et al., 2009a; Beck et al., 2009b). The physical properties of food may also be important in terms of the satiating effect of food due to effects of the macrostructure on the rate of digestion and absorption of nutrients. Physiological evidence of this is demonstrated through observed differences in blood glucose and insulin responses, both of which may affect the feeling of satiety. It would be interesting to examine the satiating properties of pork consumed within different cuisines, such as Asian noodle-style pork meals compared to a more traditional Eurocentric pork roast with potatoes and vegetables.

Various individual amino acids, such as dietary tryptophan which acts as a precursor for the production of the neurotransmitter serotonin (5-hydroxytryptophan, 5-HTP), have been shown to be of potential importance in regulating food intake through a lowered appetite. In a placebo-controlled trial, Cangiano et al. (1992) observed significant weight loss in obese subjects who had 5-HTP before a meal, whether energy intake was restricted or not. The 5-HTP group significantly decreased their ingestion of carbohydrates and satiety was

reached early during meals. Our study compared the effects of composite meats, rather than individual amino acids within foods. There may be some commercial benefit with regard to further investigations into methods to increase the tryptophan content of pork meat.

7. Recommendations

As a result of the outcomes in this study the following recommendations have been made:
Fresh pork meat can be promoted as being equal to lean beef or chicken in terms of its effect on satiety and release of appetite-related intestinal hormones and insulin. Opportunities for using pork in more recipes and cuisines with high volume vegetables may provide a practical way of extending the satiety opportunity further.

8. References

Australian Bureau of Agricultural and Resource Economics, ABARE. (2007). *Australian Commodity* Statistics, 2007. ABS, Livestock products, Australia, cat no. 7215.0. Canberra: Australian Bureau of Statistics.

Australian Bureau of Agricultural and Resource Economics, ABARE. (2007). *ABS, Agriculture Australia, cat. No. 7113.0. Canberra: Australian Bureau of Statistics.*

Australian Bureau of Agricultural and Resource Economics, ABARE. (2007). *ABS, Agricultural Commodities, Australia, cat. No. 7121.0. Canberra: Australian Bureau of Statistics.*

Australian Pork Limited Annual Report 2008–09. ABN 83 092 783 278: page 23. Available at: http://www.apl.au.com/pages/images/Annual%20Report%200809.pdf. [Accessed 22nd February 2010]

Arumugama V, Leea JS, Nowaka JK, Pohlea RJ, Nyropa JE, Leddyb JL and Pelkman CL. A high glycaemic meal pattern elicited increased subjective appetite sensations in overweight and obese women. *Appetite.* 2008; 50: 215-222.

Barkeling B, Rossner S, Bjorvell H, Effects of a high-protein meal (meat) and a high-carbohydrate meal (vegetarian) on satiety measured by automated computerized monitoring of subsequent food intake, motivation to eat and food preferences, International Journal of Obesity 1990; 14: 743–751.

Batterham, R. L, Cohen, M. A, Ellis, S. M, Le Roux, C. W. et al. Inhibition of food intake in obese subjects by peptide 3-36. *The New England Journal of Medicine* 2003; 349: 941-948.

Batterham RL, Heffron H, Kapoor S, Chivers JE, Chandarana K, et al. Critical role for peptide YY in protein-mediated satiation and body-weight regulation. Cell Metab. 2006;4:223–33.

Beck EJ, Tosh SM, Batterham MJ, Tapsell LC, Huang X-F. Oat *b*-glucan increases postprandial cholecystokinin levels, decreases insulin response and extends subjective satiety in overweight subjects. *Mol Nutr Food Res* 2009; 53: 1343 – 1351.

Beck EJ, Tapsell LC, Batterham MJ, Tosh SM ,Huang X-F. Increases in peptide Y-Y levels following oat β-glucan ingestion are dose-dependent in overweight adults. *Nutrition Research* 2009; 29: 705–709.

Blom WA, Lluch A, Stafleu A, Vinoy S, Holst JJ, et al. Effect of a high-protein breakfast on the postprandial ghrelin response. Am J Clin Nutr 2006; 83:211–20

Booth DA, Chase A, Campbell AT. Relative effectiveness of protein in the late stages of appetite suppression in man, Physiology and Behavior 1970; 5: 1299–1302.

Borzoei S, Neovius M, Barkeling B, Teixeira-Pinto A, Rössner S.A comparison of effects of fish and beef protein on satiety in normal weight men. Eur J Clin Nutr. 2006; 60(7): 897-902.

Bowen J, Noakes M, Trenerry C, Clifton PM. Energy intake, ghrelin, and cholecystokinin after different carbohydrate and protein preloads in overweight men. *J. Clin. Endocrinol. Metab* 2006a; 91(4):1477–1483.

Bowen J, Noakes M, Clifton PM. Appetite regulatory hormone responses to various dietary proteins differ by body mass index status despite similar reductions in ad libitum energy intake. *J. Clin. Endocrinol. Metab*. 2006b; 91:2913–19.

Burton-Freeman B, Davis PA and Schneeman B. Plasma cholecystokinin is associated with subjective measures of satiety in women. *American Journal of Clinical Nutrition* 2002: 76: 659-667

Carroll, J. F, Kaiser, K. A, Franks, S. F, Deere, C and Cafrey, J. L (2007) Influence of BMI and gender on postprandial hormone responses. *Obesity*. 15, 2974-2983.

Charlton KE, Probst YC, Tapsell LC, Blackall P. Food, Health and Nutrition: Where does chicken fit?" *Journal of the Home Economics Institute of Australia* 2008; 15 (2): 5-17.

Eisenstein J, Roberts SB, Dallal G, Saltzman E. High-protein weight-loss diets: are they safe and do they work? A review of the experimental and epidemiologic data. *Nutr Rev* 2002; 60: 189–200. |

Flint A, Moller BK, Raben A, Sloth B, Pedersen D, Tetens I, Hoist JJ and Astrup A. Glycaemic and insulinemic responses as determinants of appetite in humans. *American Journal of Clinical Nutrition* 2006; 84: 1365-73.

Flint A, Raben, A, Blundell JE, Astrup A, Reproducibility, power and validity of visual analogue scales in assessment of appetite sensations in single test meal studies, *International Journal of Obesity and Related Metabolic Disorders* 2000; 24: 38–48.

Gannon MC, Nuttall FQ, Lane JT, Burmeister LA. Metabolic response to cottage cheese or egg white protein, with or without glucose, in type II diabetic subjects. *Metabolism* 1992; 41:1137–45

Geiselman PJ, Anderson AM, Dowdy ML, West DB, Redmann SM, Smith SR, Reliability and validity of a macronutrient self-selection paradigm and a food preference questionnaire, *Physiology and Behavior* 1998; 63: 919–928

Geliebter, AA. Effects of equicaloric loads of protein, fat, and carbohydrate on food intake in the rat and man, *Physiology and Behavior* 1979; 22: 267–273.

Grunberg NE, The effects of nicotine and cigarette smoking on food consumption and taste preferences, *Addictive Behaviors* 1982; 7: 317–331

Havel PJ, Townsend R, Chaump L and Teff K. High-fat meals reduce 24-h circulating leptin concentrations in women. *Diabetes* 1999; 48: 334-341.

Holt SHA, Brand-Miller JC, Petocz P, Farmakalidis E. A satiety index of common foods, *Eur J Clin Nutr* 1995; 49: 675-690/

Holt S, Brand J, Soveny C and Hansky J. Relationship of satiety to postprandial glycaemic, insulin, and cholecystokinin responses. *Appetite* 1992; 18: 129-141

Jen KL, Bodkin NL, Metzger BL, Hansen BC. Nutrient composition: Effects on appetite in monkeys with oral factors held constant, Physiology and Behavior 1985; 34: 655–659.

Johnson WG, Corrigan SA, Lemmon, CR, Bergeron KB, Crusco AH. Energy regulation over the menstrual cycle, *Physiology and Behavior* 1994; 56: 523–527.

Latner JD, Schwartz M. The effects of a high-carbohydrate, high-protein or balanced lunch upon later food intake and hunger ratings. *Appetite* 1999; 33(1):119-28.

Le Roux, C. W, Batterham, R. L, Aylwin, S.J.B, Patterson, M, Borg, C.M, et al. Attentuated peptide YY release in obese subjects is associated with reduced satiety. *Endocrinology*, 2005; 147: 3-8.

Lea E & Worsley A. The cognitive contexts of beliefs about the healthiness of meat. *Public Health Nutrition* 2001; 5(1): 37-45.

Martin GS, Tapsell LC, Batterham, MJ, Russell KG. Relative validity of a diet history interview in an intervention trial manipulating dietary fat in the management of type 2 diabetes mellitus. Prev Med 2003; 36: 420-8.

McLennan W, Podger A. *National Nutrition Survey. Nutrient intakes and physical measurements. Australia 1995*. Canberra: Australian Bureau of Statistics, 1998.

Mikkelsen PB, Toubro S, Astrup A. Effect of fat-reduced diets on 24-h energy expenditure: comparisons between animal protein, vegetable protein, and carbohydrate. Am J Clin Nutr 2000; 72: 1135-1141.

Moran LJ, Luscombe-Marsh ND, Noakes M, Wittert GA, Keogh JB, Clifton PM. The satiating effect of dietary protein is unrelated to postprandial ghrelin secretion. J. Clin. Endocrinol. Metab. 2005; 90: 5205–11.

National Health and Medical Research Council. Nutrient Reference Values for Australia and New Zealand, including recommended dietary intakes. Canberra: NHMRC, 2006.

Reidelberger RD. Cholecystokinin and control of food intake. J Nutr. 1994; 124(8 Suppl):1327S-1333S.

Salmenkallio-Marttila M, Anette Due A, Gunnarsdottir I, Karhunen L, Saarela M, Lyly M. Satiety, weight management and foods. Literature review. VTT Technical Research Centre of Finland. Espoo, Finland, 2009.

Schoeller DA, Buchholz AC. Energetics of obesity and weight control: does diet composition matter? *J Am Diet Assoc* 2005; 105(5 Suppl 1):S24-28.

Stunkard AJ, Messick S. The three-factor eating questionnaire to measure dietary restraint, disinhibition and hunger, *Journal of Psychosomatic Research* 1985; 29: 71–83.

Stunkard AJ, Messick S. Eating inventory manual (The Psychological Corporation), Harcourt Brace & Company, San Antonio, TX , 1988.

Sufian MKNB, Hira T, Miyashita K, Nishi T, Asano K, Hara H. Pork peptone stimulates cholecystokinin secretion from enteroendocrione cells and suppresses appetite in rats, *Biosci Biotechnol Biochem* 2006; 70: 1869-74.

Tannous dit EK, Obeid O, Azar ST, Hwalla N. Variations in postprandial ghrelin status following ingestion of high-carbohydrate, high-fat, and high-protein meals in males. *Ann. Nutr. Metab*. 2006; 50:260–69

Uhe AM, Collier GR, O'Dea K. A comparison of the effects of beef, chicken and fish protein on satiety and amino acid profiles in lean male subjects. *J Nutr* 1992; 122: 467–472.

Verbeke W, Viane J. Beliefs, attitude and behaviour towards fresh meat consumption in Belgium; empirical evidence from a consumer study*. Food Quality and Preference*. 1999;Vol. 10:437-45.

Weigle DS, Breen PA, Matthys CC, Callahan HS, Meeuws KE, et al. A high-protein diet induces sustained reductions in appetite, ad libitum caloric intake, and body weight despite compensatory changes in diurnal plasma leptin and ghrelin concentrations. *Am. J. Clin. Nutr.*2005; 82:41–48

Westerterp-Plantenga MS. The significance of protein in food intake and body weight regulation. *Curr Opin Clin Nutr Metab Care* 2003; 6: 635–638.

Westphal SA, Gannon MC, Nuttall FQ. Metabolic response to glucose ingested with various amounts of protein. *Am J Clin Nutr.* 1990; 52(2):267-72.

9. Appendix 1 - Notes

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If a Final Report contains Confidential Information:

- the Researcher must indicate on the cover of the final Report that the Final Report contains Confidential Information
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