Defining the functional properties of dietary protein and protein-rich foods in human energy expenditure

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Abstract

Food has a number of functional properties that can support the balance between energy intake and energy expenditure and, theoretically, one of these is the thermic effect of food. Including high-protein foods in meals may be advantageous in contributing to energy expenditure but, in practice, the evidence needs to relate to specific foods and normal dietary conditions. Using the human whole room calorimeter facility, we conducted three studies to examine the effects of: (i) higher and lower levels of protein on energy expenditure; (ii) high-protein meals using different foods to deliver the protein; and (iii) omnivorous and vegetarian cuisines in meal tests before and after a period of dietary intervention for weight loss. The meal effect of protein does not appear to differ between foods, and while high-protein diets might support weight loss, it may be difficult to prove a metabolic cause in the free-living environment.

Key words: energy expenditure, functional food, protein, weight management.

INTRODUCTION

Weight management is one of the key trends in the functional food market,1 aligned to the major public health problem of obesity.2 Food has a number of functional properties that can support a desirable balance between energy intake and energy expenditure,3 and one of these properties is known as the thermic effect of food (TEF). TEF is the energy cost associated with the assimilation of food in the body,4 and is largely associated with macronutrients. Compared with that of dietary carbohydrate and fat, human oxidation of dietary protein requires more energy as it involves metabolically costly physiological processes such as deamination, gluconeogenesis and urea formation.5 Foods that deliver a high proportion of protein in a diet may be associated with higher energy expenditure than alternative foods that deliver lower protein levels.5–8 High-protein diets have been shown to be efficacious in weight management,9–13 and one of the functional aspects may be greater energy expenditure for the same energy intake.

In practice, however, a high-protein diet can be loosely defined in terms of the amount of protein in a diet, and it would be helpful to know the amount and relative proportion of protein in the diet that is likely to significantly increase human energy expenditure. Further, we are beginning to appreciate that nutrition–health effects may not be due simply to single nutrients, but rather to the food and/or diet matrix.14 For example, some animal foods have been implicated in increased health risks,15 and plant-based foods are now recognised for delivering protective components in the diet.16 In addition, because protein reflects the genetic base of the organism, the amino acid composition of foods will be different and this may have implications at the metabolic level. Thus, there may be differences between foods that deliver high levels of protein, and between whole diets, or cuisines with high proportions of protein. Finally, recognising that people consume food rather than single nutrients, diet design concepts need to be translated into meals and advice which participants are able to follow. These observations formed the basis of our studies on the functional properties of protein-rich foods in relation to energy expenditure. This review summarises the key findings of three studies that assessed energy expenditure associated with: (i) higher and lower levels of protein within a meal; (ii) high-protein meals using different foods to deliver the protein; and (iii) omnivorous compared with vegetarian cuisines in meal tests before and after a period of dietary intervention for weight loss.
METHODS

Calorimetry

Each of the three studies measured energy expenditure and substrate oxidation via indirect calorimetry. This involved measurements of oxygen consumption and carbon dioxide production, corrected to standard temperature, pressure and dry and using paramagnetic O₂ and infrared CO₂ analysers (Sable System, Las Vegas, NV), from the human calorimeter facility at the University of Wollongong. Energy expenditure on the gas exchange rates. Urinary samples are collected during the stay for assessment of nitrogen excretion (estimated from urea) which is then used in assessing protein utilisation. Rates of energy expenditure and fuel utilisation are calculated from measures of in- and outflow as described in the literature. Urinary samples are collected during the stay for assessment of nitrogen excretion (estimated from urea). Energy expenditure and fuel utilisation are calculated from measures of in- and outflow as described in the literature.

The facility consists of two separate air-tight rooms (3 x 2.1 m) that are ventilated and air-conditioned, each of which contains a bed, desk, chair, hand basin, television and video/DVD player, computer, telephone and toilet. The facility is ventilated with fresh air which is measured by a solid-state gas sample drying system (Peltier dryer, Maastricht Instrument, Netherlands). The analysers are manually calibrated against a span gas and nitrogen each study day by comparing readings of O₂ and CO₂ with known values. Chamber air is sampled every two minutes. Rates of oxygen consumption and carbon dioxide production are calculated from measures of in- and outflow as described in the literature.

In the research presented here, participants stayed in the chamber for eight hours in the acute studies and 24 hours in the intervention trial. Meals were provided according to the designated time in the study protocol. Foods contained in each meal were specifically defined in the protocol with designated time in the study protocol. Foods contained in each meal were specifically defined in the protocol with dietary modelling using the FoodWorks software system (Xyris Software, Brisbane, Australia, Version 3, 2002).

Subject recruitment and baseline measurements

In all cases, subject eligibility was assessed through completion of a screening questionnaire eliciting health and demographic information. Recruitment was through local media. Study criteria were aged 18 years and above, generally well, not taking any regular medications, non-smokers, not allergic to any food and indicated that they liked protein-rich foods. Participants attended the initial assessment session where baseline dietary intake, bodyweight (Tanita scale model UM019, Tanita, IL) and body composition (tetrapolar bioelectrical impedance, BodyStat 1500 or dual-energy X-ray absorptiometry (DEXA)) measurements were taken. This was followed by the eight-hour or 24-hour stay in the human calorimeter facility.

Study 1: Effects of higher dietary protein on energy expenditure

This was a randomised cross-over feeding study where healthy adults who were either of normal weight, overweight or obese were given a control meal (14% protein, Glycemic Index, GI = 65) and test meals (33% protein, GI = 74; or 35% protein, GI = 45) at least one week apart for men and one month apart for women (to control for the effects of the menstrual cycle). Results were compared using a repeated-measure ANOVA with a customised post hoc analysis in a linear model of the form; y = α + β₁ × lean body mass (kg) + β₂ × log, fat mass (kg).

Study 2: Effects of source of protein on energy expenditure

This was another randomised cross-over feeding study, this time comparing energy expenditure in high-protein meals with different food sources. During the eight-hour stay in the calorimeter, participants were provided with isoenergetic high-protein breakfast and lunch meals from three protein sources: meat (beef and pork), dairy or soy. Meals were calculated to meet 65% daily energy requirements (30% protein, 30% fat, 40% carbohydrate) using the FoodWorks software system (Xyris Software, Brisbane, Australia, Professional Edition). Energy expenditure of the three stays was compared using the repeated-measure ANOVA using the SPSS v15.0 (SPSS, Chicago, IL).

Study 3: Effects of type of cuisine on weight loss and energy expenditure

The present study was a three-month parallel randomised, single-blinded, controlled trial comparing weight loss and energy expenditure from the consumption of omnivorous and vegetarian high-protein cuisines. Modelling of the high-plant-protein diet was a challenging task as plant foods have lower-protein content compared with animal foods (Figure 1). A secondary aim was to assess cuisine implications in high-protein diet strategies. Participants were randomised to receive meals in the calorimeter and advice on a free-living energy-deficit diet relating to either a control (18% protein), or two high-protein diet models (30% or 35% protein). The latter based on animal (HAP, omnivorous), or plant (HPP, vegetarian) sources, respectively. In the present study, body composition was assessed using DEXA. Participants were seen by dietitians on a monthly basis to ensure dietary compliance. Statistical analyses were performed using the repeated-measure ANOVA using the SPSS v15.0.

RESULTS

Data analysis

Study 1: Eighteen healthy adults (39.8 ± 12.9 years) with a body mass index (BMI) range of 21.4–36.0 kg² completed...
Body composition played a key role in response to the diets. The negative relationship between body fat and fat oxidation disappeared with the high-protein meals ($\alpha = -4.7$, $\beta_2 = 2.23$, $P < 0.01$) and between body fat and energy expenditure, seen in the control meals ($\alpha = -1.5$, $\beta_2 = 0.63$, $P < 0.05$). No GI effect was observed.

Study 2: A total of 31 participants were recruited and 24 completed all three stays. Pending data cleaning for individual values, preliminary analyses for the first six participants are reported (all male, mean age of 26 years). Bodyweight and body fat were consistent through the three stays and energy expenditure was therefore not adjusted for body composition. Bearing in mind the small sample size, no significant differences were found between groups with varying dietary sources of protein (Table 1).

Study 3: A total of 61 participants were recruited and 35 completed the study. Again, pending data cleaning for individual values, preliminary analyses for the first 15 participants are reported (six male, nine female; mean age 44 years, mean BMI 32.6 kg$^{-2}$). In all three groups, mean BMI and bodyweight reduced significantly after the three-month intervention (Table 2); however, there were no between-group differences, nor was there a time or diet effect on energy expenditure (per kilogram fat-free mass). From the dietary modelling perspective, some commercial soy products were found to contain high-protein levels but, even including such products in the diet, it was difficult to reach the 30% E protein levels. To achieve the dietary target, we fortified the diet with the use of soy protein powder in the vegetable protein group.

**Table 1** Anthropometric characteristics and energy expenditure values of participants in study 2 (mean ± SD)

<table>
<thead>
<tr>
<th></th>
<th>Meat ($n = 6$)</th>
<th>Soy ($n = 6$)</th>
<th>Dairy ($n = 6$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bodyweight (kg)</td>
<td>80.2 ± 16.1</td>
<td>79.5 ± 16.2</td>
<td>80.5 ± 15.3</td>
</tr>
<tr>
<td>Body fat (%)</td>
<td>17.3 ± 6.8</td>
<td>17.2 ± 5.9</td>
<td>17.4 ± 6.4</td>
</tr>
<tr>
<td>Energy expenditure (kcal/day)</td>
<td>2592.0 ± 317.0</td>
<td>2863.4 ± 640.9</td>
<td>2717.6 ± 438.9</td>
</tr>
</tbody>
</table>

**DISCUSSION**

Gaining any advantage in the energy expenditure stakes is one of the targets of contemporary bodyweight management. While physical exercise is the main source of energy expenditure, understanding the energy dynamics of food is still a useful pursuit, contributing to a better understanding of food in general and the specifics of its role in health promotion. The studies described here focused on one of the mechanisms by which food may play a role in weight management.

Weight management is an important component of diabetes control. Despite this, studies have failed to show the enhancing effect of high-protein diets on weight loss in people with hyperinsulinemia and Type II diabetes. Overweight, insulin resistance and Type II diabetes may represent a metabolic spectrum in which bodyweight itself may affect metabolism. In turn, the TEF may be related to the extent of overweight. Our finding that body composition is a significant factor in metabolic responses to high-protein meals appears to suggest that this is the case. This information may enable more targeted dietary advice for different groups in the population based on degree of overweight and insulin resistance.

The type of food that delivers the protein may also be important. In a study comparing the thermic effect of meals prepared with pork and soy, the pork meal produced a higher thermic effect than soy, but both meals produced significantly higher effects than a lower-protein meal. The increased TEF may have been influenced by the amino acid
content of the diets; the pork diet contained 27% more histidine, 14% more methionine and cysteine, and 5% more leucine than the soy diet. Our results, on the other hand, suggest that the thermic effects argued for high-protein meals may be driven by the relative amount of macronutrients rather than the type of protein in the meal. It is worth noting that the difference in effects between high-protein diets in the former study was relatively small. Indeed, when our intervention was extended to allow for a period of time in the ‘free living environment’, the effect of food differences appeared to simply ‘wash out’. While these were observations from preliminary analyses, it might also suggest that the increased energy expenditure of meals may occur at the acute level, but the differential may not be sufficiently large enough to have an impact in the long term, or at least to be measurable in the experimental sense. This has implications for both understanding the nature of dietary intervention and the position of various forms of dietary experimentation.

The most common design found in the literature for studies of thermic effects of food is an acute feeding experiment. Some of these studies used a liquid meal method, and others applied very high proportions of protein, but we aimed to reflect a feasible dietary pattern based on foods and meals usually consumed by the average Australian.

The laboratory environment provides better control than the free-living environment over dietary variables and, as such, is essential in developing theoretical positions for food-based research. The calorimeter provides excellent control over food and nutrient consumption as well as control over activity levels and ambient temperature, all of which affect the measurement of energy expenditure. In the free-living environment, there is less experimental control over food and nutrient consumption, so the shift from the calorimeter to the three-month intervention presented its own challenges. We applied a system of advice based on food groups with known macronutrient distribution ranges, so that participants could eat foods readily accessible to them, but keep within the dietary targets. Emphasis was given to the protein-rich food group to ensure compliance. While the theoretical position for studying the effects of food in energy expenditure focuses on macronutrients (and we have displayed our preliminary results in those terms), the translation to food and interpretation for practice will be critical. This will be partly addressed by separate analyses of food consumption patterns in the studies presented here, using previously described methods.

**CONCLUSION**

The three studies represented here display the way in which we can pursue questions on food from the theoretical to the clinical context. Given the strength of the theoretical position on protein and thermogenesis, it was appropriate to compare the effect of realistic meals, with higher and lower levels of protein. Moving on to compare the effects of high-protein meals using different foods to deliver the protein is a logical step supporting various forms of practice, some of which involves making health claims. The preliminary
results suggest that the translation of the effect to whole foods in normally consumed meals is valid, with the effect attributed to the protein content of the food (study 2). Progression to investigating whole diets (cuisines) is relevant, given that weight management is a ‘whole of diet’ issue, and it comes closer to the reality of consumer eating patterns, although at a cost to experimental control. This represents further challenges for research, not just in methodology, but also in risk assessment associated with taking knowledge from one context of research and translating it to another.

ACKNOWLEDGEMENTS

All studies were co-funded by the National Centre of Excellence in Functional Foods and the University of Wollongong.

CONFLICT OF INTEREST

No conflict of interest has been declared by S.Y. Tan, L. Tapsell, M. Batterham or K. Charlton.

REFERENCES

8 Raben A, Agerholm-Larsen L, Flint A, Holst JJ, Astrup A. Meals with similar energy densities but rich in protein, fat, carbohydrate, or alcohol have different effects on energy expenditure and substrate metabolism but not on appetite and energy intake. Am J Clin Nutr 2003; 77: 91–100.
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