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Exploring the impact of food composition and eating context on food choice, energy intake and body mass index

Exploring the impact of food composition and eating context on food choice, energy intake and body mass index

by

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Abstract

Research suggests that traditional behaviour-based weight loss approaches requiring individuals to change their dietary habits or physical activity results in only modest weight loss which often isn't maintained. Given the need to improve population health, the broader food environment (e.g., food price or composition) and the more immediate eating context have been identified as possible intervention targets.

For product reformulation to be a successful public health strategy, consumers are required to be 'insensitive' to changes to the reformulated product. This raises a more general question regarding whether humans are sensitive to food composition and whether this influences food choice and energy intake. The studies presented in Part A suggest that people are sensitive to both the energy content and macronutrient composition of food. Specifically, the results presented in chapters two to four indicate a non-linear pattern in meal caloric intake in response to meal energy density (kcal/g), and this pattern was captured in a theoretical two-component model of meal size (g, chapter five). The remaining two chapters in Part A (chapters six and seven) explore human sensitivity to food macronutrient composition. Chapter six describes the development of a new paradigm and task to assess protein discrimination by humans. Chapter seven focuses on the remaining two macronutrients, fat and carbohydrate, and demonstrates that, alongside being more liked, foods containing a combination of fat and carbohydrate are selected in larger portions than foods high in either fat or carbohydrate.

The effect of eating contexts (e.g., social or distracted eating) on acute energy intake is well-researched, but their chronic impact on energy balance is unclear. The results of chapter nine (Part B) indicated that more frequently watching TV was associated with a higher body mass index (BMI) in young adults. More generally, the work identified eating contexts as potential targets for public health messaging which could effect changes in BMI on a population level.

Together, the work presented in this thesis highlights new complexity in human dietary behaviour which presents both challenges and opportunities for successful food reformulation as a public health strategy, and it also demonstrates that the context in which we eat our meals could be leveraged to improve population-level health.

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COVID-19 Statement

This statement describes the impact of the COVID-19 pandemic and associated restrictions on the research presented in this thesis. I began my PhD in September 2019, and, in March 2020, I was running what would have been the first study of my PhD exploring the effects of macronutrient composition, specifically fat and carbohydrate, on food reward, food liking, and expected satiation. This data collection was terminated when the UK lockdown began. As it was unknown how long the COVID-related restrictions would remain in place, I decided to re-develop my PhD research agenda and utilise online studies, existing large nutritional datasets, and secondary data analyses. I had no previous experience with coding and creating online studies or with analysing large datasets (n > 1000). Therefore, I had to learn new experimental software and platforms (e.g., Gorilla, JsPsych, Prolific, and Pavlovia), and learn and develop skills in coding languages (e.g., JavaScript). Additionally, to complete the secondary data analysis and analysis of the large datasets, I had to learn R as this was the necessary data wrangling, cleaning, and analysis software package. Due to the lockdowns and restricted access to in-person resources, I did not have access to formal training in these skills and independently taught myself using various online resources.

Additionally, and related to the pause of in-person research in March 2020, I had the opportunity to lead a project which analysed existing data to explore the impact of eating contexts on body mass index (chapter nine). As it was unknown when the restrictions might lift, this was my main project immediately after the restrictions were put in place as it did not involve any in-person interactions. Despite this research project being different from the core focus of my PhD, I decided to include it in my thesis as I developed my skillset during this project, led the project through to submission for publication, and dedicated a substantial amount of my research time to it.

Author's Declaration

I declare that the work in this dissertation was carried out in accordance with the requirements of the University's Regulations and Code of Practice for Research Degree Programmes and that it has not been submitted for any other academic award. Except where indicated by specific reference in the text, the work is the candidate's own work. Work done in collaboration with, or with the assistance of, others, is indicated as such. Any views expressed in the dissertation are those of the author.

SIGNED: .... DATE: ....

Publications

Papers:

Flynn, A. N., Hamilton-Shield, J., Schneider, E., Higgs, S., Timpson, N. J., & Brunstrom, J. M. The ecology of eating: quantifying the relative impact of different eating contexts on BMI using The Avon Longitudinal Study of Parents and Children – In final preparation for submission

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Chapter 1 General introduction

1.1 A brief overview of obesity and public health strategies

It is well established that global overweight and obesity rates are increasing (GBD 2015 Obesity Collaborators et al., 2017; The GBD 2013 Obesity Collaboration et al., 2014; World Health Organization (WHO), 2023). Relatedly, the prevalence of comorbid diseases and conditions, such as high blood pressure (Mills et al., 2020; Zhou et al., 2021) or type 2 diabetes (Khan et al., 2020; Zheng et al., 2018) are also rising. In 2019, the global economic impact of overweight and obesity was estimated to be 2.19% of the global gross domestic product (GDP), and if current overweight and obesity trends continue, then, in 2060, it is estimated to increase to 3.29% (Okunogbe et al., 2022).

While the health and economic impacts of overweight and obesity are clear, the fundamental drivers and mechanisms of obesity are complex and multi-factorial, including, among others, genetic, psychological, physiological, and environmental influences (Arrone et al., 2009; Frayling, 2012; Wilding, 2012; Wright & Aronne, 2012). In addition to these factors, individual behaviours have also been considered, including frequent fast food consumption (Bowman & Vinyard, 2004; Rosenheck, 2008), eating meals outside of the home (Bes-Rastrollo et al., 2010; Gesteiro et al., 2022), snacking patterns (Gregori et al., 2011; Mattes, 2018), and physical inactivity (Menschik et al., 2008; Pietilainen et al., 2008), among others.

Given the negative health and economic costs associated with overweight and obesity, there has been a substantial effort to develop weight-loss interventions. Traditional behaviour-based weight-loss interventions largely focus on reducing an individual's energy intake and/or increasing general physical activity (Dombrowski et al., 2014). Weight loss during these interventions is often only modest (Avenell et al., 2004) and tends to not be maintained over time (Kraschnewski et al., 2010). One potential reason for the poor weight loss associated with interventions that require behaviour change (e.g., restricting food intake or increasing exercise) is that people find it difficult to both change and maintain their new dietary and exercise habits (Hammarström et al., 2014; Jakicic et al., 2008; MacLean et al., 2014; Mann et al., 2007).

Given that dietary weight-loss interventions which require individuals to change their behaviour are largely ineffective, the roles of both the broader food environment and the more immediate eating context (i.e., the setting in which individuals eat) in an individual's food choice or energy intake, and subsequently weight maintenance, have also been considered (Gressier, Swinburn, et al., 2020; Mak et al., 2012; Rauber et al., 2022). Interventions to the food environment might include changing the availability, price, or composition of foods (i.e., food reformulation) (Gressier, Swinburn, et al., 2020). In the context of improving public health, food reformulation aims to change the composition of food products such that the dietary intakes of consumers are improved without changing consumer behaviour (Gressier, Swinburn, et al., 2020). With regards to food composition, energy content, as well as the sugar, salt, fat and fibre content, have been identified as key targets for reformulation (Federici et al., 2019; Gressier, Swinburn, et al., 2020). Reformulation targeting these food characteristics is often 'silent' whereby the reformulated product maintains the sensory properties of the original product and gradually replaces the original product while maintaining consumer acceptance and liking of the reformulated product (Gressier, Swinburn, et al., 2020; Hashem et al., 2019; van Raaij et al., 2009). There have been several studies quantifying the effect of different food reformulations on population-level dietary intake, and the findings of a systematic review and meta-analysis conducted by Gressier and colleagues estimates that reformulation interventions reduced salt intake by 0.57 g/day and trans-fatty acid intake by 1.2 g/day (Gressier, Swinburn, et al., 2020). Based on the findings from their review, the authors suggest that food reformulation might be a feasible strategy to improve population health, noting that improvements in population-level monitoring of nutrient intakes and health outcomes are needed to further strengthen the evidence base on food reformulation (Gressier, Swinburn, et al., 2020).

Indeed, despite the potential evidence gaps, governments and health organisations have increasingly recognised the role of food reformulation in public health policy. In 2004, the World Health Assembly approved the global strategy on diet, physical activity, and health proposed by the World Health Organization (WHO). The WHO's strategy included encouraging the food industry to "reduce the fat, sugar and salt content of processed foods and portion sizes, to increase introduction of innovative, healthy, and nutritious choices" (World Health Organization (WHO), 2004, p. 13). Government-led food reformulation strategies have mostly focussed on reducing salt and trans-fatty acids (Federici et al., 2019; Gressier, Swinburn, et al., 2020), although more recently sugar and fibre reformulation strategies have been developed (Gressier, Swinburn, et al., 2020). For example, as part of its obesity strategy, the government of the United Kingdom (UK) has set targets[1] for the food industry to reduce the salt, sugar, and calorie content of their products through food reformulation, and this set of targets forms part of the reduction and reformulation programme led by the Office for Health Improvement and Disparities (Coyle et al., 2020; Niblett et al., 2020; Office for Health Improvement and Disparities, 2017; Pyne et al., 2020). Since implementing the intake reduction targets mentioned above, average salt and sugar intakes in the UK appear to be decreasing, but the absolute intakes of both are still above their recommended values (Coyle et

[1]It should be noted that these reduction targets are not mandated by the government and the extent to which the food industry upholds these targets is debated[a,b].

[a]Action on Sugar, & Action on Salt. An evidence-based plan to prevent obesity, type 2 diabetes, tooth decay, raised blood pressure, cardiovascular disease and cancer in the UK -A benchmark for Theresa May's updated plan for action. https://www.actiononsugar.org/reformulation-/position-statements/seven-point-prevention-plan/
[b]Action on Salt. *Policy Position: UK Salt Reduction Strategy*. https://www.actiononsalt.org.uk/reformulation/position-statements/uk-salt-reduction-strategy/

al., 2020; Niblett et al., 2020). The impact of the calorie reduction programme is still unknown as, to date, the progress report is yet to be published.

Importantly, as noted previously, the success of food reformulation in improving an individual's dietary intake depends on consumers not detecting the change in the reformulated product and maintaining their acceptance of the product. Referring again to the systematic review and meta-analysis conducted by Gressier and colleagues, the findings suggest that, on the whole, consumers accepted reformulated products, although acceptance was more likely with salt-reduced products than with sugar-reduced or fibre-increased products (Gressier, Swinburn, et al., 2020). Additionally, the authors also note that compensation, did occur; however, it did not fully mitigate the effects of the reformulation (Gressier, Swinburn, et al., 2020). In the case of product reformulation, compensation (i.e., changes in consumer behaviour to offset the reformulation) is defined as either overconsumption or a change to a non-reformulated product (Gressier, Swinburn, et al., 2020)[2]. With regards to overconsumption, this could be considered a more intake-specific outcome as individuals increase their intake of the reformulated product to compensate for the reduced amount of a nutrient. Separately, consumers could also compensate for the reformulation by choosing a different, non-reformulated product. In other words, participants perceived the reformulated product to be unacceptable, and this form of compensation could be considered more of a choice-specific outcome. Critically, however, both forms of compensation (i.e., change in intake versus change in choice) result in a reduction in the efficacy of product reformulation as a strategy to improve dietary intake and population health.

[2]In experimental studies, compensation was more likely to occur when the reformulation was abrupt rather than silent[a].

[a]Gressier, M., Swinburn, B., Frost, G., Segal, A. B., & Sassi, F. (2020). What is the impact of food reformulation on individuals' behaviour, nutrient intakes and health status? A systematic review of empirical evidence. *Obesity Reviews*, *22*(2), 1-23, Article e13139. https://doi.org/10.1111/obr.13139

Given the importance of consumers being 'insensitive' to food reformulation for it to improve their dietary intake, a more general question arises regarding the extent to which humans are sensitive to the composition of food and whether this influences food choice and energy intake[3]. Exploring whether humans are sensitive to food composition has important implications for the success of food reformulation as a public health strategy and could, for example, identify types of foods which might be more amenable to reformulation or methods to promote the acceptance of reformulated products. The studies presented in the first part of this thesis (Part A) revisit questions pertaining to whether humans are sensitive to food energy content (chapters two, three, four and five) and macronutrient composition (chapters six and seven).

As mentioned previously, eating contexts, or the immediate settings in which people eat, provide another opportunity for potential public health interventions related to food choice and intake (Elliston et al., 2017; Mak et al., 2012; Rauber et al., 2022; Shams-White et al., 2021). Two eating contexts, specifically, social (Ruddock et al., 2019) and distracted eating (Robinson et al., 2013) are known to promote short-term increases in energy intake. Importantly, these eating contexts appear to occur relatively frequently at a population-level. For example, a survey of 10,287 adults residing in the United Kingdom indicated that 48% of respondents ate at least one meal per day in a social setting (i.e., with family or individuals they live with) (YouGov, 2014, as cited by Ruddock et al., 2019). The findings from this same survey suggest that 68% of adults are exposed to at least one screen (e.g., smartphone or TV screen) during the average evening meal (used either by them or someone they are eating with)

[3]A similar question was recently captured in a concept coined 'nutritional intelligence'[a] and will be discussed in further detail in chapter eight.

[a]Brunstrom, J. M., Flynn, A. N., Rogers, P. J., Zhai, Y., & Schatzker, M. (2023). Human nutritional intelligence underestimated? Exposing sensitivities to food composition in everyday dietary decisions. *Physiology & Behavior*. https://doi.org/10.1016/j.physbeh.2023.114127

(YouGov, 2014, as cited by Ruddock et al., 2019). Whilst these eating contexts appear to occur relatively frequently, the potential for social and distracted eating, as with other eating contexts (Shams-White et al., 2021), to have a chronic impact, in this case on energy balance, is unclear. The study presented in the second part of this thesis (Part B) addresses this gap and explores whether these two eating contexts associate with body mass index in a young adult cohort.

1.2 Thesis overview

As noted above, this thesis comprises two parts and explores the impact of food composition, specifically energy content and macronutrient composition (Part A), and eating contexts (Part B) on food choice energy intake and body weight. The research presented in the following chapter (chapter two) as well as chapters three and four explores whether humans are sensitive to the energy density of non-manipulated, everyday meals, and chapter five provides a discussion of the findings presented in the abovementioned three chapters. Recognising that meals and foods are comprised of more than just calories, the studies in chapter six investigate whether food protein content is detected by humans and describes the development of a new paradigm and task to assess protein discrimination. Chapter seven presents studies pertaining to the remaining two macronutrients, fat and carbohydrate, and explores whether foods comprising a more equal amount of fat and carbohydrate are less satiating, and, importantly, whether these foods are selected in larger portions (kcal). Chapter eight provides an interim summary of the results presented in Part A, discusses whether the findings provide further evidence for human ‘nutritional intelligence’ (Brunstrom et al., 2023), and provides a short introduction to Part B (chapter nine). Chapter nine investigates whether eating contexts, specifically social eating and distracted eating, might influence body mass index or weight status in young adults. Finally, in the general discussion (chapter ten), the extent to which the thesis findings can inform public health strategies pertaining to food reformulation and eating contexts is discussed. Additionally, the broader strengths and limitations of the thesis and the overlap of the findings with existing public health policy are reviewed.

Chapter 2 Energy Density: Exploring the association between meal energy density and meal calorie intake in real-world meals consumed in a controlled environment

2.1 Acknowledgements and overview

The majority of this chapter has previously been published as an original manuscript in The American Journal of Clinical Nutrition (AJCN) (Flynn, Hall, et al., 2022) and as a reply to the Letter to the Editor in AJCN (Flynn, Rogers, Hall, et al., 2023). This chapter is largely presented as portions of the published articles; however, some minor edits have been made to relate the current chapter to chapters three, four, and five and to improve its readability. I was responsible for leading research design, data cleaning and analysis, writing the first drafts of the manuscripts, editing and preparing the manuscripts for publication, and revising the manuscripts during the peer-review process. The co-authors included Dr Kevin Hall (National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, USA), Dr Amber Courville (National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, USA), Emeritus Professor Peter Rogers (University of Bristol), and Professor Jeff Brunstrom (University of Bristol). Emeritus Professor Peter Rogers and Professor Jeff Brunstrom (supervisors) provided feedback on the research design and analysis strategy, and Dr Kevin Hall and Dr Amber Courville shared the essential data and provided feedback on the analysis strategy. All co-authors provided minor edits and feedback on the manuscript text. The next paragraph will provide a brief introduction to the scientific aim of the current and following three chapters (chapters three to five).

The extent to which humans are sensitive to the energy content of the foods they consume is a well-researched question. However, depending on the approach taken (i.e., ad libitum meals or preload test-meal paradigm), the results can suggest both sensitivity and insensitivity to food energy content. The aim of this chapter, and the following three chapters (chapters three, four, and five), was to re-evaluate the association between energy density and energy intake within a single meal (i.e., acute effect). Importantly, the association between meal energy density and meal energy intake was assessed across a broad and continuous range of energy densities and using ‘real-world’ foods, rather than test meals where the energy density was covertly manipulated. Using data from a trial conducted by Hall et al. (2019), the current chapter presents the results of the first analysis exploring a potential association between energy density and energy intake in meals consumed in a controlled environment.

With the above-outlined scientific aim in mind, chapters two, three, and four present studies assessing evidence for sensitivity to energy density within a meal in four different datasets. It is important to note that the structure of the current chapter and the following two chapters (three and four) is very similar and will be outlined in brief. Each chapter begins with an introduction followed by the methods, results, and a short discussion. To avoid substantial repetition in the introductions of the three chapters, the current chapter presents a more detailed review of relevant literature (as published in Flynn, Hall, et al. (2022)), and the introductions of chapters three and four are brief to prevent repeating information unnecessarily. Similarly, rather than duplicate information in the discussion of the three chapters, the decision was made to include one chapter (chapter 5) which summarises and interprets the results from chapters two, three, and four.

2.2 Introduction

Food energy density (ED, kcal/g) refers to the energy content (kcal) of a specified weight of food (g) and can differ considerably between foods. For example, cucumber and pecan nuts have an ED of 0.15 kcal/g and 7.26 kcal/g, respectively. Literature over decades links excess energy intake (kcal) to an inability to ‘compensate’ for differences in meal ED by selecting smaller meals with increasing ED (Bell et al., 1998; Duncan et al., 1983; Rolls et al., 1999). Two main methodologies, (i) energy intake during ad libitum meals (Rolls, 2009) and (ii) test meals following a food or beverage preload (preload test-meal paradigm) (Rolls, 2009), assess the effects of ED on energy intake via compensatory changes in meal size. However, these two methods produce different findings regarding sensitivity to food energy content.

With respect to satiation, many well-designed ad libitum meal studies find little or no sensitivity to ED within a meal (i.e., the same weight of food is consumed irrespective of ED). In most of these studies, ED was covertly manipulated over a short period of time (e.g., < 10 total exposure days) (Bell et al., 1998; Bell & Rolls, 2001; Rolls et al., 1999). In other words, differences in meal caloric content have little impact on the amount of food ingested, and this insensitivity can persist over several days (Bell et al., 1998; Rolls et al., 1999). This effect of ED on energy intake, particularly in the case of high-fat food consumption, is sometimes referred to as 'passive overconsumption' (Blundell & MacDiarmid, 1997).

By contrast, studies of satiety (preload test-meal paradigm) provide strong evidence that calories in a preload can influence subsequent energy intake. These demonstrate a variable degree of short-term compensation in response to covertly manipulating the preload ED, even including sugar in a beverage (Almiron-Roig et al., 2013; Birch & Deysher, 1985; Hulshof et al., 1993; Louis-Sylvestre et al., 1989; Mazlan et al., 2006; Pliner, 1973; Rogers et al., 2016). These studies demonstrate that calories can influence behaviour and subsequent food intake after a meal has ended. Furthermore, partial compensation to the manipulation of ED is found in ad libitum studies that expose participants to covertly manipulated diets over long periods of time (e.g., > 10 total exposure days) (Lissner et al., 1987; Porikos et al., 1982; Stubbs et al., 1998). Compensation in these various studies represents an 'unlearned' response to the satiating effect of calories (single or first exposure) (Birch & Deysher, 1985; Booth et al., 1976) as well as a potentially learned response (repeated exposure) (Birch & Deysher, 1985; Booth, 1985; Booth et al., 1976; Yeomans et al., 2009; Yeomans et al., 2005).

In relation to these apparently contradictory findings and a separate finding of a non-linear relationship between absolute ED and its effect on behaviour, specifically portion size selection

(Brunstrom et al., 2018), the decision was made to re-evaluate the association between meal ED and meal energy intake. Importantly, this was done using a broad and continuous range of energy densities, ‘real world’ foods, rather than covertly manipulated test meals, and allowing for the possibility that the association between energy intake and ED is non-linear. To achieve this aim, data from a recent study investigating the effects of ultra-processing on energy intake over time and under controlled conditions (Hall et al., 2019) were analysed and this dataset is outlined in further detail below.

2.3 Methods

2.3.1 Overview of the Hall et al. study on food ultra-processing and energy intake

Hall et al. (2019) assessed a potential causal association between the consumption of ultra-processed foods, ad libitum energy intake, and subsequent changes in body weight. Twenty (10 male and 10 female) weight-stable adults (*M* ± *SE,* age = 31.2 ± 1.6 years, body mass index (BMI) = 27 ± 1.5 kg/m^2) resided in a metabolic ward in the National Institutes of Health Clinical Center for 28 days (ethical approval provided by the Institutional Review Board of the National Institute of Diabetes & Digestive & Kidney Diseases (ClinicalTrials.gov Identifier NCT03407053)). Participants were randomly assigned to receive either an ultra-processed or an unprocessed diet for two weeks, followed immediately by the alternate diet for another two weeks. Specific details regarding this study’s methodology, including the diet composition of the two seven-day rotating menus can be found in the Hall et al. (2019) paper.

In summary, participants were provided with three daily meals (breakfast, lunch, and dinner) plus snacks; however, this secondary analysis focuses only on the data from the meals. The decision to only analyse the data from meals was made for two reasons. Firstly, snacks tend to have a higher ED and be consumed in smaller portions (i.e., fewer calories) as compared to main meals (Chan et al., 2022; Murakami & Livingstone, 2016; Olea López & Johnson, 2016; Ovaskainen et al., 2006). Therefore, by excluding snacks, one potentially controls for a pattern which would be consistent with sensitivity to calories (i.e., smaller portions of energy-rich foods), thereby making it potentially more difficult to show non-linearity in caloric intake in response to energy density (i.e., more difficult to reject the null hypothesis). Secondly, and this is a greater concern for the analyses presented in chapters three and four, there is no single accepted definition of a snack which makes it difficult to accurately identify these eating events when analysing large nutritional datasets (Hess et al., 2016).

Returning to the Hall et al., study, the two diets presented to participants were matched for a variety of characteristics: total calories, energy density (including beverages), macronutrients, fibre, sugars, and sodium. However, the meals differed in their level of processing based on the NOVA classification scheme (Monteiro et al., 2018). Additionally, the participants rated the diets as equally pleasant and familiar, and the three daily meals plus snacks were provided in large portions (twice the individual's estimated energy requirements for weight maintenance). Importantly, on average, the 'presented meals' (i.e., the meals served to the participant) differed in their non-beverage energy density based on diet type (ultra-processed meals: 1.96 kcal/g, unprocessed meals: 1.06 kcal/g). This provided the rare opportunity to assess ad libitum energy intake across a broad and continuous range of energy densities using familiar foods in a highly controlled environment.

2.3.2 Secondary analysis of the Hall et al. dataset on food ultra-processing and energy intake (kcal)

To determine whether the relationship between meal ED and meal energy intake is linear (as would be predicted if people did not compensate for energy content by changing meal size (g)), the consumed meal caloric intake (kcal), meal size (g) and ED of each meal was calculated. Meals were collapsed across diet types (i.e., unprocessed or ultra-processed), and meals that were 'plate cleaned' (i.e., > 95% of the served portion was consumed) were excluded (*n*= 159 meals) alongside all calorie and non-calorie containing beverages. In a few meals (5%

of meals served to each participant), cereal or oatmeal were presented alongside milk. In these cases, milk was not excluded because neither cereal nor oatmeal were consumed without milk. However, the possibility that a proportion of the milk was consumed separately, as a beverage, cannot be ruled out.

To control for both individual (participant level) and 'meal type' (breakfast, lunch, and dinner) differences in energy intake, meal caloric intakes were mean centred for each participant and for each meal type across the 28 days (20 participants x 3 meal types x 28 days = 1,680 total centred meals). So, for example, for participant one, there were 28 centred meal caloric intakes for 'breakfast', 28 centred meal caloric intakes for 'lunch' and 28 centred meal caloric intakes for 'dinner'. Centred meals with Z-scores less than or greater than ± 3.29 were treated as outliers and removed (Field, 2013)[4], resulting in a final dataset with 1,519 meals (see Appendix 1 Figure 11.1 for a visualisation of the meal exclusion stages).

2.3.3 Statistical analysis

Initially, centred meal caloric intakes were plotted by consumed meal ED for visual inspection of any evidence for non-linearity and the remaining analyses were conducted in the R statistical environment (R Core Team, 2022) with several helper packages (Kassambara, 2020; Wickham et al., 2019). To quantify whether a non-linear fit may better explain the data, a Ramsey Regression Equation Specification Error Test (RESET) was conducted following the procedure outlined by Ramsey (1969) and using the R package 'lmtest' (Zeileis & Hothorn, 2002). If the Ramsey RESET returned a significant result, then a segmented regression was run on the centred meal caloric intake data following the procedure described by Muggeo (2003) and using the R package 'segmented' (Muggeo, 2008). A segmented regression or 'broken

[4]When normally distributed, 99.9% of Z-scores should occur between -3.29 and 3.29. Therefore, any Z-score occurring outside of this range would be in the extreme 0.1% and is treated as an outlier[a].

[a]Field, A. (2013). *Discovering statistics using IBM SPSS statistics*. SAGE Publications Inc.

stick' regression is an iterative approach which establishes the existence of one or multiple breakpoints. First, a simple linear model (without a breakpoint) is computed and evidence for a breakpoint is assessed. If a breakpoint is identified ($p < 0.05$), then a segmented regression is used to establish the location. The process then repeats until no further breakpoints are identified. This approach also constrains the segments to be 'continuous' (adjacent regression lines begin and end at the same location) (Muggeo, 2003). To confirm that a segmented fit is superior to a linear fit, the Akaike's and the Bayesian information criterion were used ('stats' package, (R Core Team, 2022)).

2.4 Results

Visual inspection of the plot containing centred meal caloric intake by consumed meal ED (see Figure 2.1) indicated a potential non-linear pattern in the data. Centred meal caloric intake appeared to increase with increasing ED until ~1.5 kcal/g and then decreased slightly.

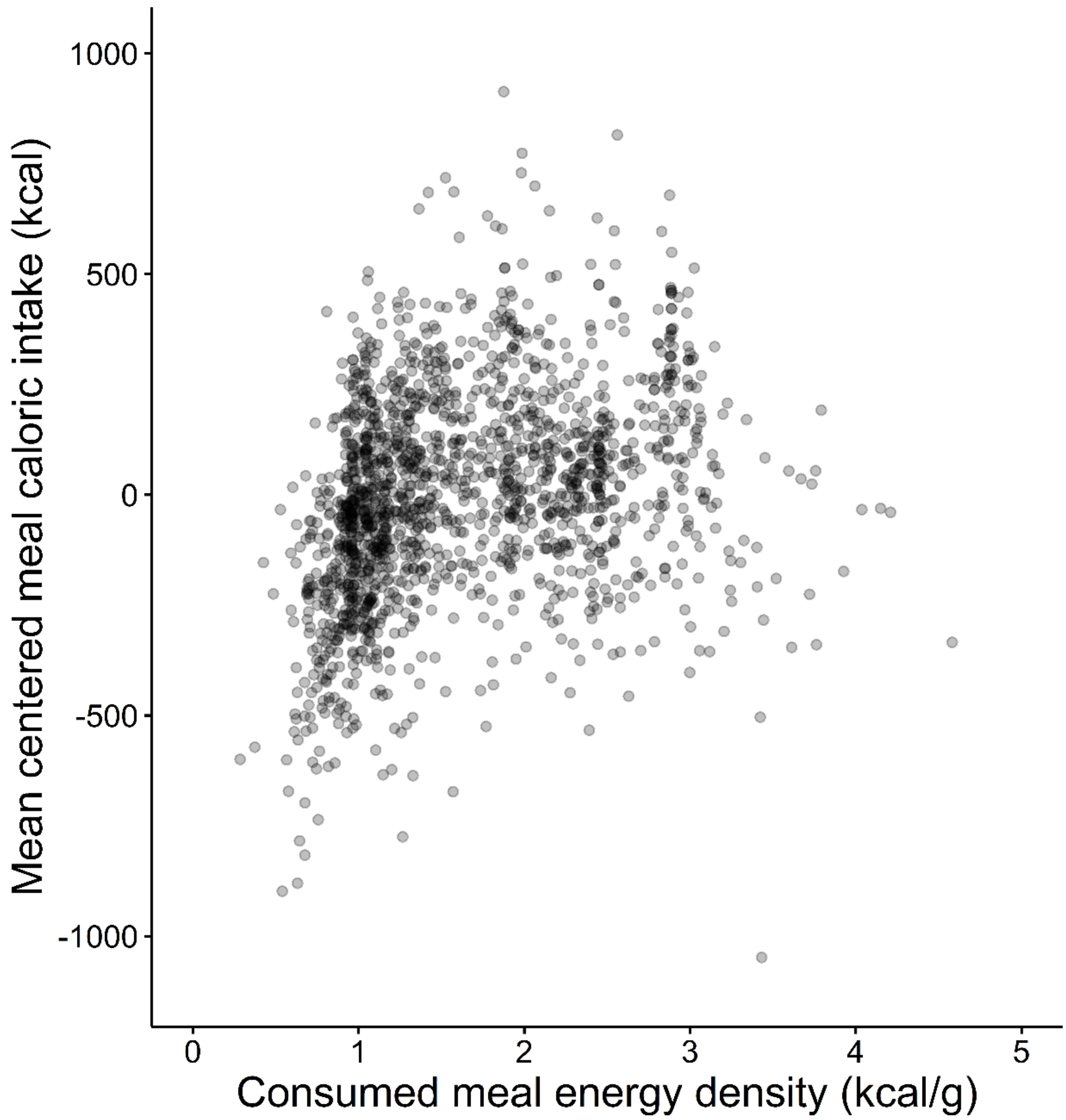


Figure 2.1 Centred meal caloric intake (kcal) by consumed meal energy density (kcal/g) in the Hall et al. dataset (*n*= 1,519).

Meals were centred within each participant and meal type. Meals which were plate cleaned (i.e. more than 95% of the served portion consumed) and meals with Z-scores < or > ± 3.29 were removed. In this scatterplot, each point represents 1 meal.

The Ramsey RESET ($F(2, 1515)= 99.32$, $p< 0.001$) indicated that a non-linear fit would better explain the data, and the segmented regression returned a one-breakpoint solution ('0 vs 2', $p< 0.001$; '1 vs 2', $p= 0.08$) at 1.41 kcal/g ($SE= 0.04$, Adjusted $R^2= 0.18$), demonstrating a significant change in the relationship between consumed meal ED and centred meal caloric intake at this point (see Table 2.1 and Figure 2.2). Respectively, a significant positive and negative association below and above 1.41 kcal/g (Table 2.1) was observed. Tests of the Akaike's and Bayesian information criteria supported a segmented fit (Appendix 1 Table 11.1).

Table 2.1 *Slope Parameter Estimates, 95% Confidence Intervals (CI), T-Values, and P-Values from a Segmented Regression Model Predicting Centred Meal Caloric Intake (kcal) from Consumed Meal Energy Density (kcal/g) in the Hall et al. Dataset (n= 1,519)*

	Slope parameter	95% CI	*t*-value	*p*-value
Slope 1 (< 1.41kcal/g)	469.13	396.58, 541.67	13.19	< 0.001
Slope 2 (> 1.41 kcal/g)	-44.42	-71.94, -16.90	-3.02	0.003

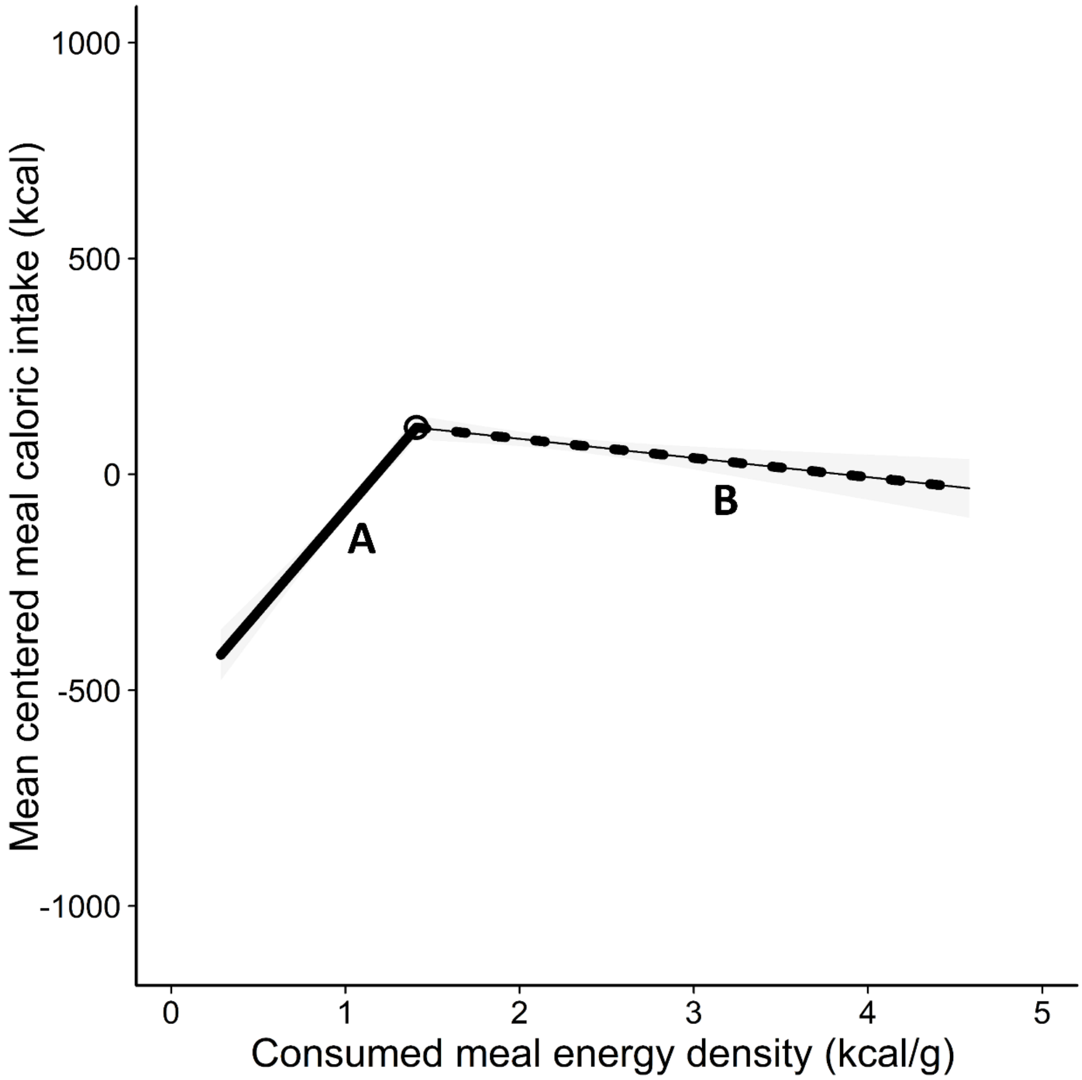


Figure 2.2 Mean centred meal caloric intakes (kcal), predicted from a segmented regression model relating consumed meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the Hall et al. dataset *(n*= 1,519).

The breakpoint located at 1.41 kcal/g (*SE*= 0.04) is represented by a circle. The dashed and solid lines represent different segments and the shading around the segments indicates 95% CIs. Segment A indicates the slope of the segment below the breakpoint (1.41 kcal/g), and segment B models the slope above the breakpoint (1.41 kcal/g).

It should also be noted that the increase in centred meal caloric intake before the 1.41 kcal/g breakpoint and subsequent decrease after the breakpoint was also observed in the raw meal caloric intake data (see Appendix 1 Figure 11.2). To assess the robustness of this evidence for non-linearity, sensitivity analyses were conducted, once including (i) plate cleaned meals, and again using (ii) presented meal ED (to account for possible spurious correlations between consumed meal ED and centred meal caloric intake). Here, for both analyses, two-breakpoint solutions were returned (i) 1.08 & 2.89 kcal/g and (ii) 1.02 & 1.84 kcal/g (see Appendix 1 Figure 11.3 (including plate cleaned meals) and Figure 11.4 (using presented ED) as well as Table 11.2 for slope parameter estimates for both sensitivity analyses). Whilst not a replication of a one-breakpoint solution as in the main analysis, evidence for non-linearity was preserved regardless.

2.5 Discussion

In data from meals consumed in a controlled setting, a non-linear association between meal caloric intake and meal ED was observed. Meal caloric intake appeared to increase with increasing ED in lower energy-dense meals (i.e., those below the breakpoint), and in higher energy-dense meals (i.e., those above the breakpoint) meal caloric intake decreased slightly as ED increased. Additionally, non-linear patterns were also observed in the raw data, when including plate cleaned meals, and when using presented ED rather than consumed ED, indicating that the non-linear pattern is a relatively robust finding. This non-linear pattern in meal caloric intake suggests a degree of sensitivity to calories. Had participants been insensitive to meal ED, then meal caloric intake should have increased linearly with energy density, and this point is further discussed in chapter five.

A methodological strength of this study is that the data were collected by trained research staff under controlled settings, eliminating the likelihood that the pattern is the result of under-reporting or misreporting by participants. However, it remained important to establish if this pattern also occurs in meals consumed under free-living conditions and in a different country. Therefore, the research presented in the next chapter explores whether the non-linear pattern replicates in meals selected and consumed by free-living participants in the United Kingdom (UK).

Chapter 3 Energy Density: Evidence of a non-linear pattern in meal caloric intake in response to meal energy density in meals consumed by free-living participants in the United Kingdom (UK) who completed the UK National and Diet Nutrition Survey (NDNS)

3.1 Acknowledgements and overview

The majority of this chapter has previously been published as an original manuscript in The American Journal of Clinical Nutrition (AJCN) (Flynn, Hall, et al., 2022) and as a reply to a Letter to the Editor in AJCN (Flynn, Rogers, Hall, et al., 2023). This chapter is largely presented as portions of the published articles; however, minor edits have been made to relate the current chapter to chapters two, four, and five. I was responsible for leading research design, data cleaning and analysis, writing the first drafts of the manuscripts, editing and preparing the manuscripts for publication, and revising the manuscripts during the peer-review process. The co-authors included Dr Kevin Hall (National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, USA), Dr Amber Courville (National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, USA), Emeritus Professor Peter Rogers (University of Bristol), and Professor Jeff Brunstrom (University of Bristol). Emeritus Professor Peter Rogers and Professor Jeff Brunstrom (supervisors) provided feedback on the research design and analysis strategy, and Dr Kevin Hall and Dr Amber Courville shared the essential data and provided feedback on the analysis strategy. All co-authors provided minor edits and feedback on the manuscript text.

As noted in section 2.1, this chapter includes a brief introduction and discussion to avoid significant repetition across chapters two, four and five. The main introduction and discussion for the three data-presenting chapters can be found in chapter two and chapter five, respectively.

3.2 Introduction

To investigate the generalizability of the non-linear pattern in meal caloric intake in response to meal ED which was observed in meals consumed under controlled settings (chapter two), the analyses were repeated in meals selected and consumed by free-living humans in the United Kingdom (UK) who completed the National Diet and Nutrition Survey (NDNS) (Food Standards Agency & Office for National Statistics, 2005).

3.3 Methods

3.3.1 Overview of the UK National Diet and Nutrition Survey (NDNS)

The 2000-2001 UK NDNS comprises dietary data obtained between July 2000 and June 2001 (Food Standards Agency & Office for National Statistics, 2005). The aim of the survey was to provide a cross-sectional record of the eating habits and nutritional status of the UK population. A multi-stage random-probability design was used to invite participants; 152 postal sectors were selected during the first stage, and from each sector, 40 addresses were randomly chosen. Individuals who were neither pregnant nor breast feeding, and those aged between 19 and 64 were eligible for inclusion. All provided written informed consent and the NDNS received ethical approval from a Multi-center Research Ethics Committee (MREC) and National Health Service Local Research Ethics Committees (LRECs).

3.3.2 Analysis of data from the UK NDNS

Participants (n= 1,724; 958 females, 766 males; $M \pm SE$, age = 42.10 ± 0.29 years; BMI = 26.83 ± 0.13 kg/m^2) used a diet diary to record all of the food and drink that they consumed over seven days. For eating events occurring at home, each food item was individually weighed and recorded, and any uneaten food was subtracted from the initial portion. For out-of-the-home eating events, participants recorded approximate amount or quantities served, and noted any leftovers. All calorie and non-calorie containing beverages were excluded using a purpose-written script in R which excluded beverages using food codes from the User Guide provided by the UK Data Archive. Remaining beverages which were not identified in the User Guide were manually removed by the lead author in five separate instances. Milk was not excluded when it was consumed with cereal or porridge or water when it was used to prepare a powdered soup. Lastly, the consumed eating event size (g), eating event caloric intake (kcal), and eating event ED were calculated for each of the 60,777 recorded eating events.

The aim was to make the NDNS dataset comparable to the Hall et al. dataset which only included data from meals. Therefore, eating events where less than 200 kcal had been consumed and where the eating event ED was greater than 4 kcal/g were excluded (Murakami & Livingstone, 2016). The 200 kcal cut-off corresponds with previous research using the NDNS dataset suggesting the average caloric content of snacks to be approximately 200 kcal (Olea López & Johnson, 2016). The NDNS provides no information about meal type (i.e., breakfast, lunch, or dinner) and so the term 'meal' refers to all eating events that were not excluded. Meal caloric intakes were mean centred within each individual and centred meals with *Z*-scores less than or greater than ± 3.29 were removed from the analyses. The final dataset comprised 32,162 meals (see Appendix 1 Figure 11.5 for a visualisation of the meal exclusion stages).

3.3.3 Statistical analysis

As in the previous chapter, centred meal caloric intakes were plotted by consumed meal ED for visual inspection of any evidence for non-linearity, and the remaining analyses were conducted in the R statistical environment (R Core Team, 2022) with several helper packages (Kassambara, 2020; Wickham et al., 2019). To quantify whether a non-linear fit may better explain the data, a Ramsey Regression Equation Specification Error Test (RESET) was conducted following the procedure outlined by Ramsey (1969) and using the R package 'lmtest' (Zeileis & Hothorn, 2002). Again, if the Ramsey RESET returned a significant result, then a segmented regression was run on the centred meal caloric intake data following the procedure described by Muggeo (2003) and using the R package ‘segmented’ (Muggeo, 2008). As in chapter two, to confirm that a segmented fit is superior to a linear fit, Akaike’s and the Bayesian information criterion were used (‘stats’ package, (R Core Team, 2022)).

3.4 Results

Figure 3.1 shows centred meal caloric intake by consumed meal ED. Unlike in Figure 2.1, the large number of superimposed datapoints made it difficult to determine non-linearity and potential breakpoints from simple visual inspection. However, the Ramsey RESET test demonstrated a non-linear pattern ($F(2, 32{,}158) = 852.77$, $p < 0.001$), and two breakpoints ('0 vs 2', $p < 0.001$; '1 vs 2', $p = 0.046$) were identified at 1.75 kcal/g ($SE = 0.02$, Adjusted $R^2 = 0.06$) and 2.94 kcal/g ($SE = 0.15$), respectively (Figure 3.2, Table 3.1). Again, a significant positive association below the first breakpoint as well as a negative association between the breakpoints and above the second breakpoint were observed (see Table 3.1 below) and both the Akaike’s and the Bayesian information criterion were met (see Appendix 1 Table 11.1).

Table 3.1 *Slope Parameter Estimates, 95% Confidence Intervals (CI), T-Values, and P-Values from a Segmented Regression Model Predicting Centred Meal Caloric Intake (kcal) from Consumed Meal Energy Density (kcal/g) in the NDNS dataset (n= 32,162)*

	Slope parameter	95% CI	*t*-value	*p*-value
Slope 1 (< 1.75 kcal/g)	174.86	162.78, 186.94	31.60	< 0.001
Slope 2 (1.75 kcal/g – 2.94 kcal/g)	-107.91	-118.99, -96.84	-15.54	< 0.001
Slope 3 (> 2.94 kcal/g)	-59.19	-81.12, -37.26	-5.56	< 0.001

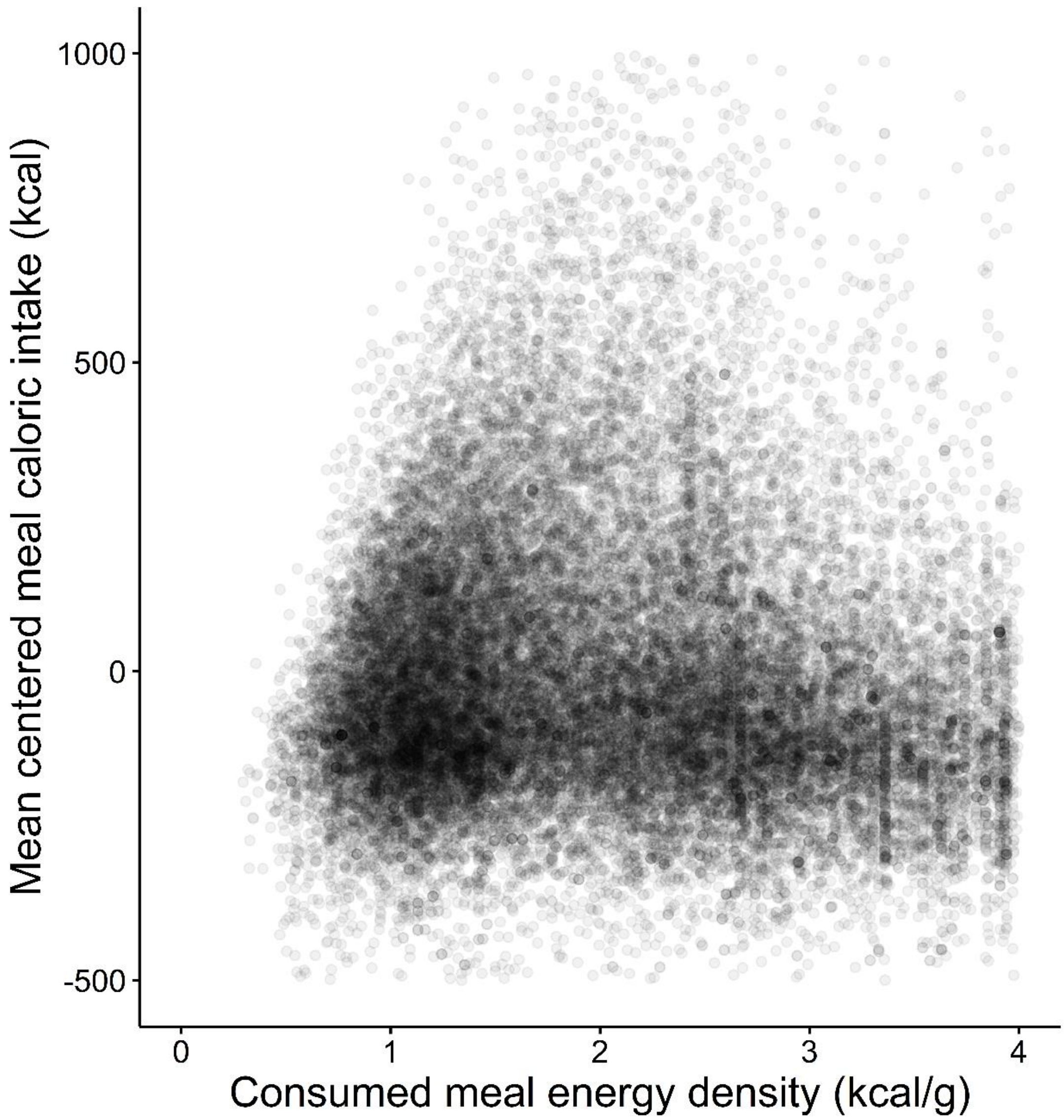


Figure 3.1 Centred meal caloric intake (kcal) by consumed meal energy density (kcal/g) in the NDNS dataset *(n*= 32,162).

Meals were centred within each participant and meals with *Z*-scores < or > ± 3.29 were removed. In this scatterplot, each point represents 1 meal. To aid graphical illustration, centred meal caloric intakes above 1,000 kcal or below –500 kcal are excluded from this figure (0.51% of total meals). They were, however, included in the reported analyses.

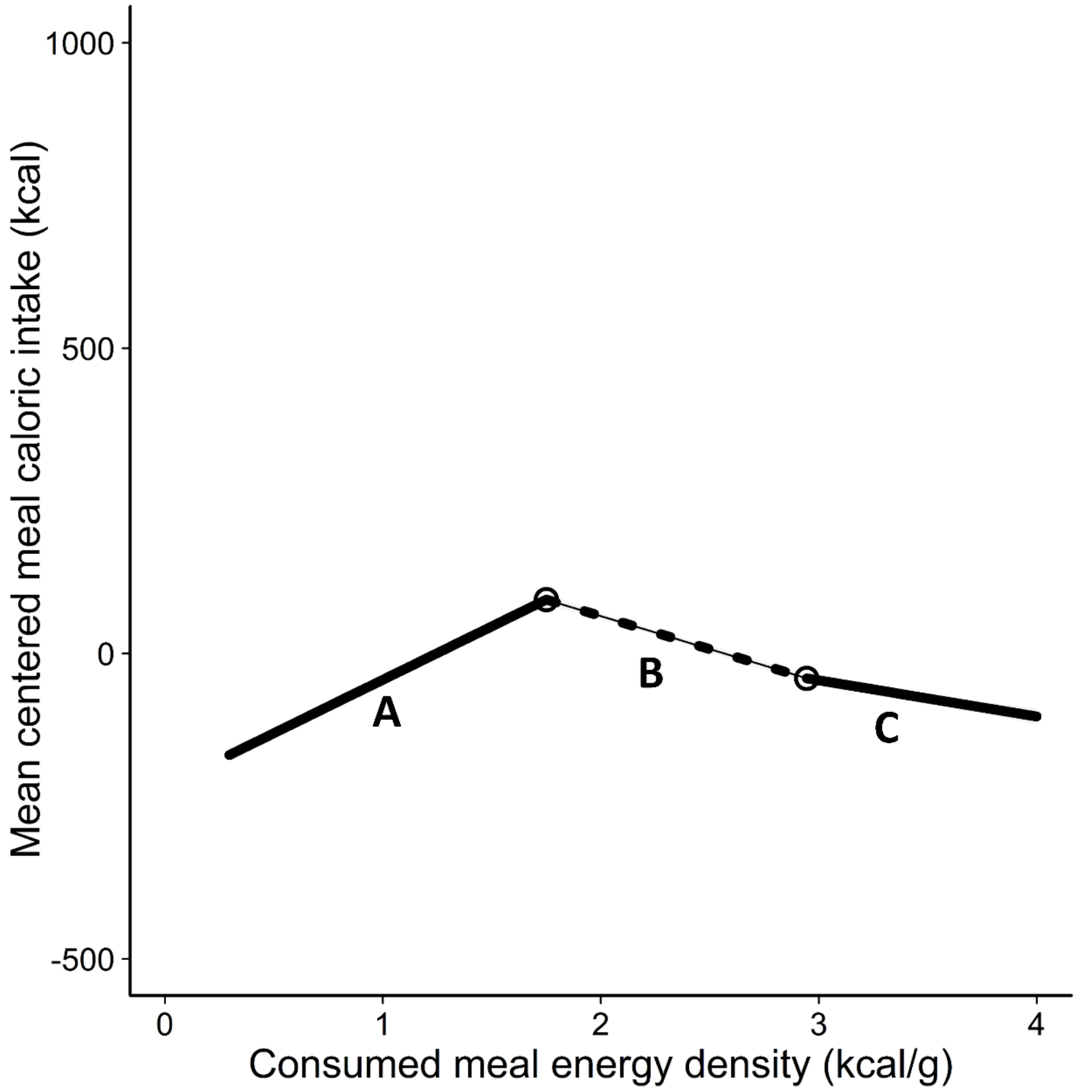


Figure 3.2 Mean centred meal caloric intakes (kcal), predicted from a segmented regression model relating consumed meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the NDNS data set *(n*= 32,162).

The breakpoints located at 1.75 kcal/g (*SE* = 0.02) and 2.94 kcal/g (*SE* = 0.15) are represented by circles. The dashed and solid lines represent different segments and the shading around the segments indicates 95% CIs. Segment A indicates the slope of the segment below the first breakpoint (1.75 kcal/g), segment B indicates the slope of the segment between the 2 breakpoints (1.75 kcal/g & 2.94 kcal/g), and segment C models the slope above the second breakpoint (2.94 kcal/g).

Regardless of the calorie cut-off used for the inclusion criteria (e.g., meal caloric intake > 600 kcal), the patterns of results were broadly similar; meal caloric intake increased until the first breakpoint and then decreased. A two-breakpoint solution was returned when the inclusion criterion for meals was set at both 400 and 600 kcal (i.e., meals < 400 or 600 kcal were excluded). However, when the criterion was set at 800, 1,000, and 1,200 kcal, a one breakpoint solution was returned. Regardless of whether a one or two breakpoint solution was selected, the first breakpoint occurred between 1.75 kcal/g and 2.30 kcal/g (see Appendix 1 Table 11.3 for these sensitivity analyses).

3.5 Discussion

A non-linear association between meal caloric intake and ED was replicated in meals selected and consumed by free-living participants in the UK. Again, meal caloric intake increased with energy density in lower energy-dense meals (~ < 1.75 kcal/g) and then decreased in higher energy-dense meals (~ >1.75 kcal/g). More generally, the non-linear pattern observed in these data is consistent with the pattern observed in data from meals consumed under controlled conditions (chapter two) and provides further evidence for human sensitivity to calories. Importantly, the non-linear pattern in meal caloric intake in response to meal ED has only been observed in data from participants living in the US or UK. It remained important to establish whether this non-linear pattern generalises to other cultures consuming different diets, and this is the aim of chapter four.

Chapter 4 Energy Density: Evidence for a non-linear pattern in meal caloric intake extends to data from free-living participants in Argentina and Malaysia

4.1 Acknowledgements and overview

The majority of this chapter has previously been published as an invited publication for the Society for the Study of Ingestive Behavior (SSIB) special issue in the journal Physiology & Behavior (Flynn, Rogers, & Brunstrom, 2023), and the Argentinean data were presented as an oral presentation (authors include Flynn, Rogers, and Brunstrom) at the 2022 annual meeting of the Society for the Study of Ingestive Behavior (Porto, Portugal). This chapter is largely presented as the published article; however, minor edits have been made to relate the current chapter to chapters two, three, and five and to improve its readability. I was responsible for leading research design, data cleaning and analysis, writing the first draft of the manuscript, editing and preparing the manuscript for publication, and revising the manuscript during the peer-review process. The co-authors included Emeritus Professor Peter Rogers (University of Bristol) and Professor Jeff Brunstrom (University of Bristol). The co-authors (supervisors) provided minor edits and feedback on the manuscript text.

As noted in section 2.1, this chapter includes a brief introduction and discussion to avoid significant repetition across chapters two, three, and five. The main introduction and discussion for the three data-presenting chapters can be found in chapter two and chapter five, respectively.

4.2 Introduction

As mentioned in the previous chapter, it remained important to establish whether the non-linear pattern in meal caloric intake in response to meal ED generalizes to other cultures and other diets. To that end, datasets from Argentina (Primer estudio sobre el estado nutricional y los hábitos alimentarios de la población adulta de Rosario) and Malaysia (Malaysia Lipid Study

2012/2013) which are part of the Food and Agriculture Organization of the United Nations/World Health Organization Global Individual Food consumption data Tool (FAO/WHO GIFT) were used and the analyses from chapters two and three were repeated.

4.3 Methods

4.3.1 Overview of the Food and Agriculture Organization of the United Nations/World Health Organization Global Individual Food consumption data Tool (FAO/WHO GIFT)

Both the Argentinean and Malaysian datasets are part of the Food and Agriculture Organization of the United Nations/World Health Organization Global Individual Food consumption data Tool (FAO/WHO GIFT). Briefly, in 2014, the FAO and WHO began a collaboration to develop an open-access platform which stores individual quantitative food consumption data from a variety of countries, including low- and middle-income nations (Leclercq et al., 2019). Food consumption data have been standardized across the various datasets using a globally adapted version of the FoodEx2 system developed by the European Food Safety Authority (European Food Safety Authority [EFSA], 2015; Leclercq et al., 2019). This allows researchers to compare datasets across countries and to match the food intake data with food composition data (European Food Safety Authority [EFSA], 2015; Leclercq et al., 2019; Roe et al., 2013). Currently, 44 datasets are available and data can be accessed via the FAO/WHO GIFT website.

The decision to analyse the Argentinean and Malaysian datasets was made for several reasons: 1) the aim was to explore whether the non-linear pattern replicated in data from different countries, 2) both datasets used a quantitative diet recall during the data collection procedure, 3) both datasets comprised only adult participants and there was a wide age range, and 4) both datasets included data from males and females.

4.3.2 Details pertaining to original data collection for the Argentinean dataset: *Primer estudio sobre el estado nutricional y los hábitos alimentarios de la población adulta de Rosario*

Dietary intake and nutritional status data were collected from the adult population (individuals between 18 and 70 years of age) residing in the city of Rosario Argentina between October 2012 and June 2013 (Zapata, 2014). A stratified (sex, age, and district of the City of Rosario) convenience sampling strategy was used to recruit participants at Centros Municipales de Distrito (CMD). Individuals who were pregnant or lactating were excluded resulting in a total sample size of 1,200 individuals. Among other measures, and with the help of trained nutrition students, participants completed an in-person paper-based 24-hour diet-recall survey. Every food and beverage consumed the day before the survey was recorded by consumption occasion and portion size quantification was aided with a list of normative portions which were described in the Argentinean dietary guidelines ((Lema et al., 2003) as cited by Zapata (2014)). Each eating event (i.e., breakfast, lunch, dinner, or snack) was coded separately, and if composite or mixed dishes were consumed, then they were separated into ingredients using individual report or recipes. Under- or over-reporting participants were identified using dietary reference intakes for adults of > 4,013 kcal/d) or < 803 kcal/d for men and > 3,511 kcal/d or < 502 kcal/d for women ((Willet, 1998) as cited by Zapata (2014)). Ethical approval was obtained by the study from the Comité de Ética en Investigación de la Secretaría de Salud Pública de la Municipalidad de Rosario (Res Nº 1816/2010) and authorized by the Sub Secretaría General de la Municipalidad de Rosario (Zapata, 2014). Further information regarding the data collection procedure, additional survey measures, and participant details can be found on FAO/WHO GIFT website as well as in the survey's related publication by Zapata (2014).

4.3.3 Current analysis of data from Argentina

Two features of the Argentinean dataset differ slightly from the NDNS and Hall et al. datasets. Firstly, for several of the food items the reported nutritional information is for the uncooked value. Secondly, beverages, such as red wine, milk, or water, were incorporated into the meal (not coded separately), and for milk and water, it was not possible to determine whether this liquid was consumed as a beverage or used to prepare the meal (such as for making a dough or soup from scratch). Therefore, to make the dataset comparable to the NDNS and Hall et al. datasets and to remedy the above issues, the data were restructured in the following ways.

4.3.3.1 Cooked weight of pasta and rice

For some of the food items, specifically pasta, lentils, and rice, the dry (i.e., uncooked) weight of the foods was recorded. The dry weight artificially inflates the ED of the food item (for example, the ED of the uncooked pasta was ~3.71 kcal/g) as it does not consider the water which is absorbed during the cooking process. Therefore, using the water uptake ratios for pasta (1.8; 1.5 - 2.3) and rice (2.5; 2.3 - 2.8, used as a proxy value for lentils), which is the amount of water absorbed during the cooking process, the wet (cooked) weights of the foods were calculated (dry weight * water uptake ratio value) (van Dooren et al., 2019). This ‘wet’ weight replaced the original dry weight of the pasta, rice, or lentils and the calorie content remained the same.

4.3.3.2 Beverages, milk, and water

In chapters two and three, all calorie and non-calorie containing beverages were removed as the focus was on the association between food ED and energy intake. Therefore, to maintain consistency across the analyses of the different datasets, it was decided to exclude calorie and non-calorie containing beverages.

However, as previously stated, in this dataset beverages tended to be coded as part of the eating event (i.e., coffee recorded alongside croissant in the same eating event). Due to this coding structure, beverage (identified via food codes beginning with ‘B’) removal was achieved in several stages. Firstly, coffee (with/without milk and/or sugar), tea (with/without milk and/or sugar) and maté (a South American caffeine-containing drink; with or without sugar) were excluded. This was done by first identifying whether a meal contained coffee using the unique ingredient code. Next, this process was repeated for milk and sugar. The dataset was then temporarily split depending on whether the meal contained coffee. In the dataset with meals containing coffee, coffee, milk, and sugar were removed if they were present in the meal. The two datasets were then remerged. This process was completed for both tea (with/without milk and/or sugar) and maté (with or without sugar). Had milk and sugar been removed without first specifying whether coffee, tea or maté was present in the meal, then these ingredients might have unnecessarily been removed from a meal, for example milk and cereal.

This concern of arbitrarily removing milk (or water) from meals where it is a core component, such as in the preparation of a dough or milk consumed with cereal, resulted in the decision to only include water and milk when it was included in a meal with key ingredients. Specifically, milk was only included in a meal if the meal also contained flour or cereal and water was only included if the meal also contained flour, cereal or powdered soup. Importantly, water was not included if the meal contained pasta, rice or lentils as the wet weight of these ingredients (i.e., the weight of the ingredient after cooking) had previously been calculated.

Lastly, all other beverages (identified on three separate occurrences via visual examination of the data, specifically the food code), such as red wine, soft drinks, or juice, were removed. While this approach might not remove all beverages, or on occasion remove milk (or water) when it should have been kept, the aim of this process was to remove as many obvious beverages as possible whilst attempting to maintain the integrity of the dataset (i.e., what comprises a realistic meal). It also allows the analysis of this dataset to be comparable to that of the NDNS and Hall et al. datasets.

4.3.3.3 Filtering of meals in dataset

Reflecting on the aim to maintain consistency across the analyses of the different datasets, the same filtering criterion that was applied in the analysis of the NDNS data (chapter three) was used. Therefore, eating events where less than 200 kcal had been consumed or where the eating event ED was greater than 4 kcal/g were excluded. The aim of this filtering was to, as in chapter three, remove any eating events which might have been snacks, and the 200 kcal cut-off reflects the average caloric content of snacks in the NDNS (Olea López & Johnson, 2016). Additionally, as the data from Argentina were collected during a single 24-hour dietary recall, there was only one entry per meal type (i.e., breakfast, lunch or dinner). Therefore, caloric intakes were mean centred within each individual and centred meals with *Z*-scores less than or greater than ± 3.29 were removed from the analyses. The final dataset comprised 2,738 meals (see Appendix 1 Figure 11.6 for a visualisation of the meal exclusion stages).

4.3.4 Details pertaining to original data collection for the Malaysian dataset: *Malaysia Lipid Study 2012/2013*

The Malaysia Lipid Study invited free-living adults between the ages of 20 and 65 years of age to complete a quantitative 24-hour dietary recall among several other clinical measures to assess dietary practices and metabolic outcomes (Karupaiah et al., 2019; *The Malaysia Lipid Study*, 2013). The survey was completed on three separate days, two weekdays and one weekend day, and data were collected between October 30th 2012 and November 28th 2013 using a convenience sampling strategy (community health screenings) in urban and suburban areas covering the Klang Valley, which includes Kuala Lumpur and Petaling Jaya.

The inclusion criteria were participants being between the ages of 20 and 65, being free-living, and having no medical conditions. Exclusion criteria included taking cholesterol lowering medication, smoking heavily (more than 10 cigarettes per day), consuming more than two standard alcoholic drinks per day or currently attempting to lose weight or following a muscle building regime. Women who were either pregnant, breast feeding, or currently experiencing menopause, were also excluded. The study received ethical approval from the Medical Ethics Committee of the National University of Malaysia (UKM 1.5.3.5/138/NN-047-2012). In total, 598 participants completed the clinical measures, including the dietary survey.

Of these, 577 were eligible for analysis after removing participants who reported being on an extreme diet or who under-reported in their dietary recall (EI:BMR< 0.9, Goldberg cut-off method). The 24-hour diet recalls followed the methodology cited by NHANES (Center for Disease Control and Prevention [CDC]) and portion sizes were estimated with the aid of household measures as well as a food atlas containing food photographs (Karupaiah et al., 2019). Further information regarding the data collection procedure, additional survey measures, and participant details can be found on FAO/WHO GIFT website as well as in the survey's related publication by Karupaiah et al. (2019).

4.3.4.1 Current analysis of data from Malaysia

As with the Argentinean dataset, beverages, such as beer, soft drinks, water or milk, were included alongside the food in an eating event, rather than being coded as a separate event. Therefore, following the same approach to the Argentinean data, to make the analysis of this dataset comparable to the analysis of the UK NDNS and Hall et al. datasets, all calorie and non-calorie containing beverages (identified via food codes provided with the dataset) were removed. In this instance, water was not excluded when it was listed alongside wheat flour, rice flour, or oats. Soups had been originally coded to include any broths. Additionally, coffee and tea, served with or without milk and/or sugar, were excluded using the same process as in the Argentinean dataset. Again, this decision reflected a concern to avoid removing milk from meals where it was a key component, such as in porridge, or when consumed with cereal or used to create a dough. All remaining beverages identified either via the User Guide or visual examination were removed.

4.3.4.2 Filtering of meals and final sample size

Again, meals which were less than 200 kcal or those with an ED greater than 4 kcal/g were removed. Meal caloric intakes were mean centred within each participant (again, to control for participant level differences in energy intake) and centred meal caloric intakes with *Z*-scores less than or greater than ± 3.29 were removed from the analyses. 4,658 meals were included in the final dataset (see Appendix 1 Figure 11.7 for a visualisation of the meal exclusion stages).

4.3.5 Statistical analysis

The same statistical analysis was used in both datasets and follows the approach from chapters two and three.

4.4 Results

In both datasets, visual inspection of scatterplots showing mean centred meal caloric intakes as a function of meal ED (see Figure 4.1 for both datasets) did not show a clear non-linear pattern. However, in both cases, significant Ramsey RESET tests [Argentina[5]: $F(2, 2{,}734) = 105.91$, $p < .001$; Malaysia[6]: $F(2, 4654) = 105.60$, $p < .001$] indicate that the data are better described by a non-linear function. In both datasets, Akaike's and the Bayesian information criterion confirmed that a segmented regression was a superior fit to a simple linear model. For associated statistics see Appendix 1 Table 11.4.

Segmented regressions returned a one-breakpoint solution, in both datasets: Argentina 2.04 kcal/g (*SE*= 0.06; Adjusted $R^2 = 0.09$) and Malaysia[7] 2.17 kcal/g (*SE*= 0.06; Adjusted $R^2 = 0.05$) (see Figure 4.1 for both datasets). In both datasets, there was a positive slope in lower energy-dense meals (below the breakpoint) and a negative slope in higher energy-dense meals (above the breakpoint), see Table 4.1 for slopes and t-values for both datasets.

[5]Mean meal caloric intake in the Argentinean dataset: *M*= 574.11 kcal, *SE*= 6.29 kcal.
[6]Mean meal caloric intake in the Malaysian dataset: *M*= 536.74 kcal, *SE*= 3.57 kcal.

[7]Additionally, centering the meals within meal type and participant did not change the pattern of the results (*n*=4,110); a one-breakpoint solution is returned, 2.20 kcal/g (*SE*= 0.08), the slope increases below the breakpoint, 76.80, and decreases above the breakpoint, -66.10, and the Ramsey RESET, AIC, and BIC confirm that a non-linear fit is superior.

Table 4.1 *Slope Parameter Estimates, 95% Confidence Intervals (CI), T-Values, and P-Values from a Segmented Regression Model Predicting Mean Centred Meal Caloric Intake (kcal) from Consumed Meal Energy Density (kcal/g) in the Argentinean (n= 2,738) and Malaysian (n = 4,658) Datasets*

Dataset		Slope parameter	95% CI	*t*-value	*p*-value
Argentina					
	Slope 1 (< = 2.04kcal/g)	134.89	105.27, 164.49	10.07	< 0.001
	Slope 2 (> 2.04 kcal/g)	-149.06	-171.81, -126.32	-11.28	< 0.001
Malaysia					
	Slope 1 (< = 2.17 kcal/g)	96.63	80.70, 112.97	12.32	< 0.001
	Slope 2 (> 2.17 kcal/g)	-122.01	-147.24, -96.78	-8.34	< 0.001

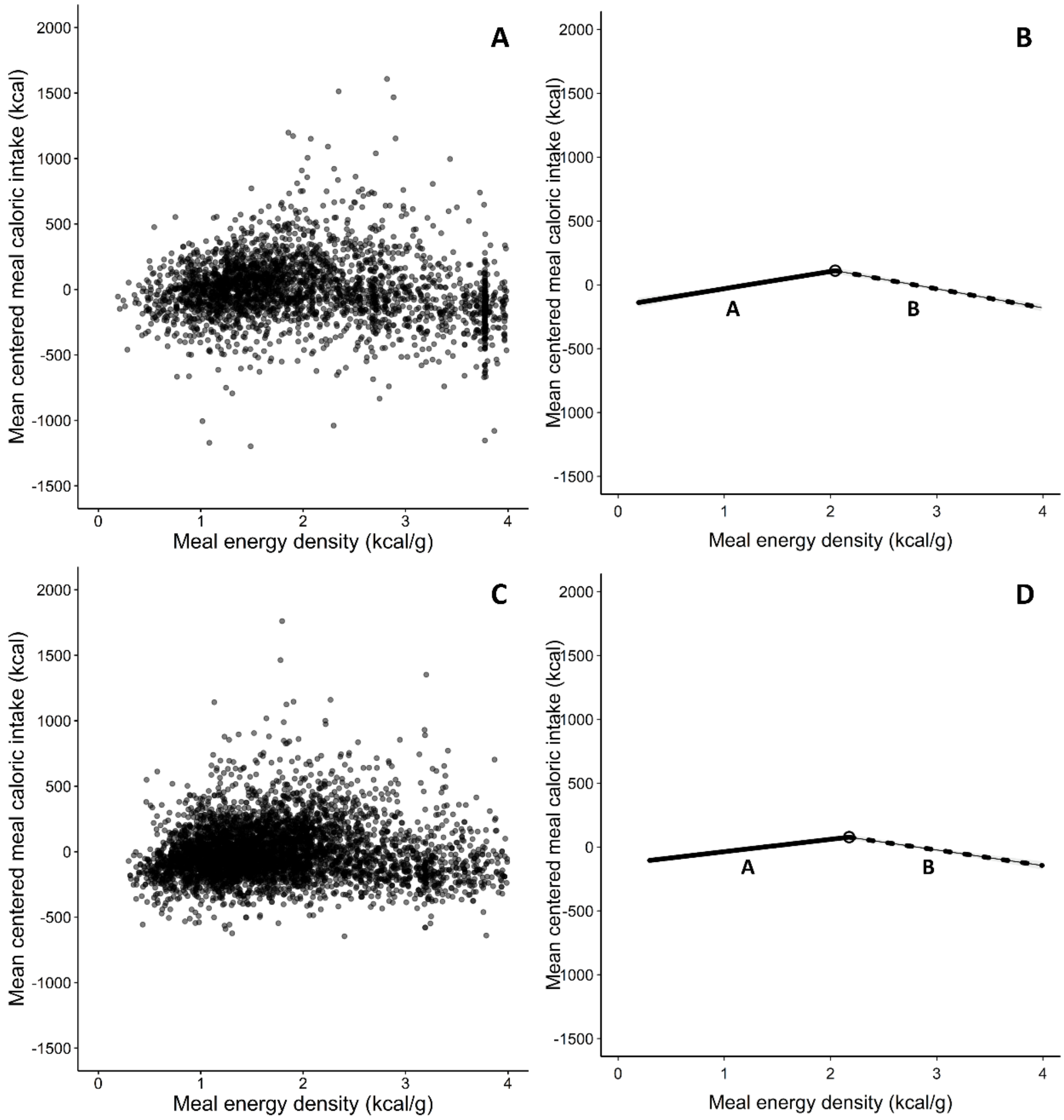


Figure 4.1 Four-panel plot depicting mean centred meal caloric intake (kcal) and segmented regression for both the Argentinean dataset (*n*= 2,738 meals) and the Malaysian dataset (*n*= 4,658 meals).

In panels A and C, mean centred meal caloric intake (kcal) is plotted by meal energy density (kcal/g) in both the Argentinean dataset (panel A) and the Malaysian dataset (panel C). In both scatterplots, meals were centred within each participant and meals with *Z*-scores less than or greater than ± 3.29 were removed. In panels B and D, mean centred meal caloric intakes (kcal) is predicted from a segmented regression model relating meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the Argentinean dataset (panel B) and the Malaysian dataset (panel D). In each panel, the breakpoint is represented by a black circle, the dashed and solid lines represent different segments and the shading around the segments indicates 95% confidence intervals. Segment A indicates the slope of the segment below the breakpoint and segment B models the slope above the breakpoint.

Additionally, sensitivity analyses confirm that the non-linear pattern observed in both the Argentinean and Malaysian datasets does not change as a function of whether beverages are included or excluded. For example, including all beverages, water, and milk in the Argentinean dataset (*n*= 3,439) returned a two-breakpoint solution[8] 0.73 kcal/g (*SE*= 0.15) and 1.94 kcal/g (*SE*= 0.10). The slope below the first breakpoint was 277.24 (*95% CI*, 153.63 – 400.84), between the two breakpoints 159.95 (*95% CI*, 122.47 – 197.43) and above the second breakpoint -140.76 (*95% CI*, -194.84 - -86.68). If all beverages are excluded in the Argentinean dataset (*n*= 2,815), a one breakpoint solution is returned, 2.04 kcal/g (*SE*= 0.06), and a positive slope occurs below the breakpoint, 155.11 (*95% CI*, 121.23 - 189.00), and a negative slope above the breakpoint, -144.12 (*95% CI*, -165.07 - -123.16).

In the Malaysian dataset, including all beverages (*n*= 5,007) returned a one breakpoint solution, 1.64 kcal/g (*SE*= 0.04) and meal caloric intake increased below the breakpoint, 188.94 (*95% CI*, 165.72 – 212.16), and decreased above the breakpoint, -84.81 (*95% CI*, -106.82 - -62.80). If all beverages are removed (*n*= 4,636), then a one breakpoint solution is returned at 2.16 (*SE*= 0.06), with meal caloric intake again increasing below the breakpoint, 98.62 (*95% CI*, 81.87 – 115.38), and decreasing above the breakpoint, -116.36 (*95% CI*, -139.53 - -93.18).

Lastly, the non-linear pattern remains in both the Argentinean and Malaysian datasets when all meals are analysed (i.e., no meal filtering criteria are applied and no outliers are removed; see Appendix 1 Figure 11.8).

4.5 Discussion

As in the US (Hall et al.) and UK (NDNS) datasets (chapters two and three), a non-linear pattern between meal ED and meal energy intake was observed in both the Argentinean and Malaysian datasets. Again, meal caloric intakes increased with increasing ED in lower

[8]A two-breakpoint solution was also returned in the analysis of the NDNS dataset in chapter three. However, it should be noted that in the abovementioned analysis, beverages had been excluded.

energy-dense meals (meals below the breakpoint) and decreased in higher energy-dense meals (meals above the breakpoint), and this non-linear pattern was consistent regardless of whether beverages were included or excluded. This replication lends even further support to the idea that humans are sensitive to the calorie content of real-world meals and demonstrates that this sensitivity appears to be consistent across different cultures. The next chapter presents a more in-depth discussion of the results presented in the current chapter and chapters two and three, proposes a theoretical two-component model of meal size, and relates the non-linear pattern in meal caloric intake to previous findings.

Chapter 5 Energy density: Evidence for sensitivity to energy density, a theoretical two-component model of meal size (g) and reconciling previous findings regarding sensitivity to calories

5.1 Acknowledgements and overview

Most of this chapter has previously been published in four different publications: Flynn, Hall, et al. (2022), Flynn, Rogers, et al. (2022) (conference abstract), Flynn, Rogers, Hall, et al. (2023), and Flynn, Rogers and Brunstrom (2023). The majority of the sections in this chapter comprise previously published text; however, edits have been made to combine the different publications and to improve the chapter's readability.

As mentioned in the acknowledgements of the previous three chapters, for each publication, I was responsible for leading research design, data cleaning and analysis, writing the first drafts of the manuscripts, revising and preparing the manuscripts for publication, and revising the manuscripts during the peer-review process. For the Flynn, Hall, et al. (2022) publication, Emeritus Professor Peter Rogers (University of Bristol, supervisor) and Professor Jeff Brunstrom (University of Bristol, supervisor) provided feedback on the research design and analysis strategy, and Dr Kevin Hall (National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, USA) and Dr Amber Courville (National Institute of Diabetes and Digestive and Kidney Diseases, Bethesda, MD, USA) shared the essential data and provided feedback on the analysis strategy; all co-authors provided minor edits and feedback on the manuscript text. For the Flynn, Rogers, et al. (2022) (conference abstract) publication, the co-authors provided feedback on the analysis strategy and the abstract text. For the Flynn, Rogers, Hall, et al. (2023) publication, each co-author provided minor edits and feedback on the letter's text. Lastly, for the Flynn, Rogers and Brunstrom (2023) publication, each co-author provided minor edits and feedback on the manuscript text.

Several parts of this chapter have been presented at academic conferences including as an oral presentation at the annual meeting of The British Feeding and Drinking Group (Flynn, Rogers, et al., 2022) and as a poster presentation at the 2021 annual meeting of the Society for the Study of Ingestive Behavior (two-component model of meal size; Flynn, Rogers, and Brunstrom, online). Additionally, as mentioned in section 4.1, the Argentinean data and two-component model of meal size were presented as an oral presentation (Flynn, Rogers, and Brunstrom) at the 2022 annual meeting of the Society for the Study of Ingestive Behavior (Porto, Portugal).

The results of the three previous chapters suggest that the association between meal caloric intake and meal ED is non-linear. To avoid substantial repetition, the three chapters did not include detailed discussions. Therefore, this chapter provides an interpretation of the observed non-linear pattern (the next section reviews these outcomes). This is then followed by a description of the theoretical two-component model of meal size (g), before placing the current results in the context of previous findings suggesting both sensitivity and insensitivity to calories.

5.2 Evidence for sensitivity to energy density found in the Hall et al., UK NDNS, Argentinean, and Malaysian datasets

A non-linear association between meal caloric intake and meal ED was found in the four datasets. Meal caloric intake increased with ED until the first breakpoint (segment A) and decreased thereafter (segment B, and, in the NDNS dataset, segment C, see Figure 5.1).

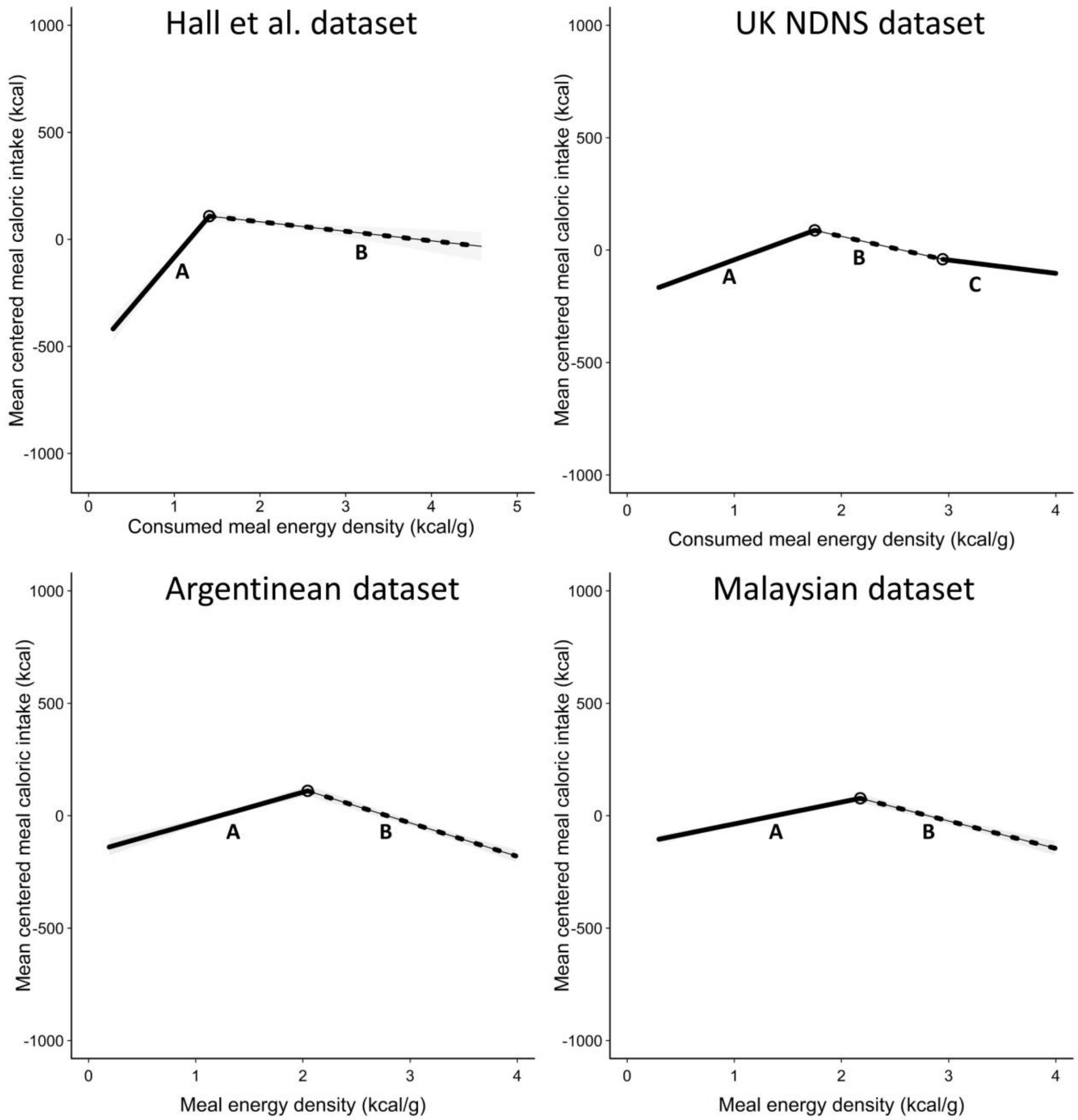


Figure 5.1 Mean centred meal caloric intakes (kcal), predicted from four separate segmented regression models relating consumed meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the Hall et al., UK NDNS, Argentinean, and Malaysian datasets.

The breakpoints are represented by circles, the dashed and solid lines represent different segments (also denoted by letters) and the shading around the segments indicates 95% CIs.

For the avoidance of doubt, the pattern in meal caloric intake is the result of participants consuming different amounts of food by weight (g) across the range of energy densities (see Appendix 2 Figure 11.9, Figure 11.10, Figure 11.11, and Figure 11.12 for scatterplots of meal size (g) as a function of meal ED). Had participants consumed the same amount of food by weight, irrespective of meal ED, then meal caloric intake would have increased linearly across the full range of meal energy densities. However, as seen in the segmented regressions, this positive association was only observed in lower energy-dense meals.

It should be noted that the negative slopes in segment B of all the datasets and segment C of the NDNS dataset reflect a degree of overcompensation; specifically, participants consumed smaller meals than necessary to adjust for the increasing meal ED. The overcompensation may, in part, be driven by dietary restraint, specifically the conscious restriction of meal size due to concerns about the effects of energy-rich meals on body weight (Olea López & Johnson, 2016). This overcompensation pattern (i.e., negative slope) will be further explored in section 5.4.

It is also important to consider that the non-linear pattern in meal caloric intake in response to meal ED could be the result of statistical coupling, and this concern is part of a broader debate (Squara, 2008). Statistical or mathematical coupling is when one variable is calculated from another or when a variable is shared between both independent and dependent variables (Squara, 2008). In the case of this research, coupling occurred when meal ED was calculated by dividing meal caloric intake by the amount (g) of food consumed during the meal and plotted against meal caloric intake (kcal). However, in the analysis of the Hall et al. dataset (chapter two), an attempt was made to account for possible spurious correlations between meal caloric intake and meal ED and used presented rather than consumed meal ED as the predictor (one additional degree of separation). Importantly, the non-linear pattern remained consistent (chapter two). Lastly, and notwithstanding the above, the non-linear pattern in meal caloric intake, specifically the flattening or slight negative association in higher energy-dense meals, could not be explained by mathematical coupling or random behaviour. Again, if participants had consumed random amounts (g) of food across a range of ED, then the null hypothesis would be a perfect linear association between calories consumed and ED. However, the observed departure from a linear association in the higher energy-dense meals suggests that this pattern is unlikely to be the result of random behaviour.

It should be acknowledged that the patterns are not identical across the four datasets. When reviewing the pattern of results across the different datasets comprising meals consumed under free-living conditions (UK, Argentina, and Malaysia), the locations of the first breakpoints were similar, ranging from 1.75 kcal/g in the UK (second breakpoint at 2.94 kcal/g) to 2.04 kcal/g in Argentina and 2.17 kcal/g in Malaysia. It should also be noted that the breakpoint for the US data is slightly lower at 1.41 kcal/g, and this may be due to differences in the research environment (controlled experimental setting versus free-living). Additionally, the slopes representing the association between ED and meal caloric intake after the first breakpoint were largely similar across the three datasets (-107.91(UK); -122.01 (Malaysia); -149.06 (Argentina)). Understanding factors that determine the exact location of a breakpoint remains a project for future research.

Additionally, while the sensitivity analyses of the Argentinean and Malaysian datasets (see section 4.4), indicated that the non-linear pattern in meal caloric intake was consistent regardless of beverage inclusion or exclusion, the location of the breakpoints shifted slightly. Specifically, when all beverages were included in the datasets, a breakpoint was identified at a lower ED compared to the original breakpoint. With respect to the Argentinean dataset, this is because a two breakpoint solution was obtained, however, a lower breakpoint was still observed in the Malaysian dataset with a one breakpoint solution. Meal beverages reduce meal ED[9], however, because extrinsic water (Camps et al., 2017; Camps et al., 2018; Marciani et al., 2012) exits the stomach quickly and free sugars in liquids are rapidly absorbed (compared to solid food) (Dasgupta et al., 2023; Fujiwara et al., 2020; Gadah et al., 2016; Malik & Hu, 2015),

[9]Argentinean dataset mean meal ED (kcal/g) with beverages = 1.08 (*SE*= 0.01), mean meal ED (kcal/g) as analysed in main analysis= 2.03 (*SE*= 0.02)
Malaysian dataset mean meal ED (kcal/g) with beverages = 1.36 (*SE*= 0.01), mean meal ED (kcal/g) as analysed in main analysis = 1.73 (*SE*= 0.01)

their impact on total meal size may differ from solid food. For now, it remains unclear whether this generated a meaningful change in breakpoint location and this should be explored in future studies.

It should also be noted that the meal ED predicts a relatively small amount of the variance (~ 10%) in acute meal caloric intake, but this aligns with findings from other research (Stubbs et al., 2000), and the effects of ED should be interpreted alongside other factors known to influence meal size such as social facilitation (Ruddock et al., 2019), distraction (Robinson et al., 2013), food texture (Forde & de Graaf, 2022) and eating rate (Robinson et al., 2014). Lastly, the cross-sectional design of the analyses in chapters two, three, and four does not allow for causal conclusions to be drawn. However, there is substantial experimental evidence demonstrating the different effects of volume (for example: Bell et al., 1998; Bell & Rolls, 2001; Rolls et al., 1999) and calories (for example: Almiron-Roig et al., 2013; Rogers et al., 2016) on energy intake in humans. Therefore, in the remaining text, when causal language is used, this is based on the experimental evidence mentioned above.

5.3 Introducing a two-component model of meal size (g): 'volume' and 'calorie-content' satiation signals

The non-linear pattern in meal caloric intake can be captured by a two-component ('volume' and 'calorie-content') model of meal size (similar to models from Deutsch (1983) and Smith (1998; Chapter 2 'Pregastric and Gastric Satiety'; Chapter 3 'Intestinal Satiety' by Greenberg) which posits that two satiation signals, a volume signal and a calorie-content signal, influence meal caloric intake in response to meal ED (see Figure 5.2 for a visualisation of the model). Briefly, Smith suggests that there are three locations from which signals can feedback to influence satiation: pregastric, gastric, and intestinal; the gastric signals are described as volumetric whereas the intestinal signals are nutritive (Powley & Phillips, 2004; Smith, 1998; Chapter 2 'Pregastric and Gastric Satiety'; Chapter 3 'Intestinal Satiety' by Greenberg).

Similarly, Deutsch (1983) presents two stomach related signals: one which measures nutrient amount and the other gastric distension.

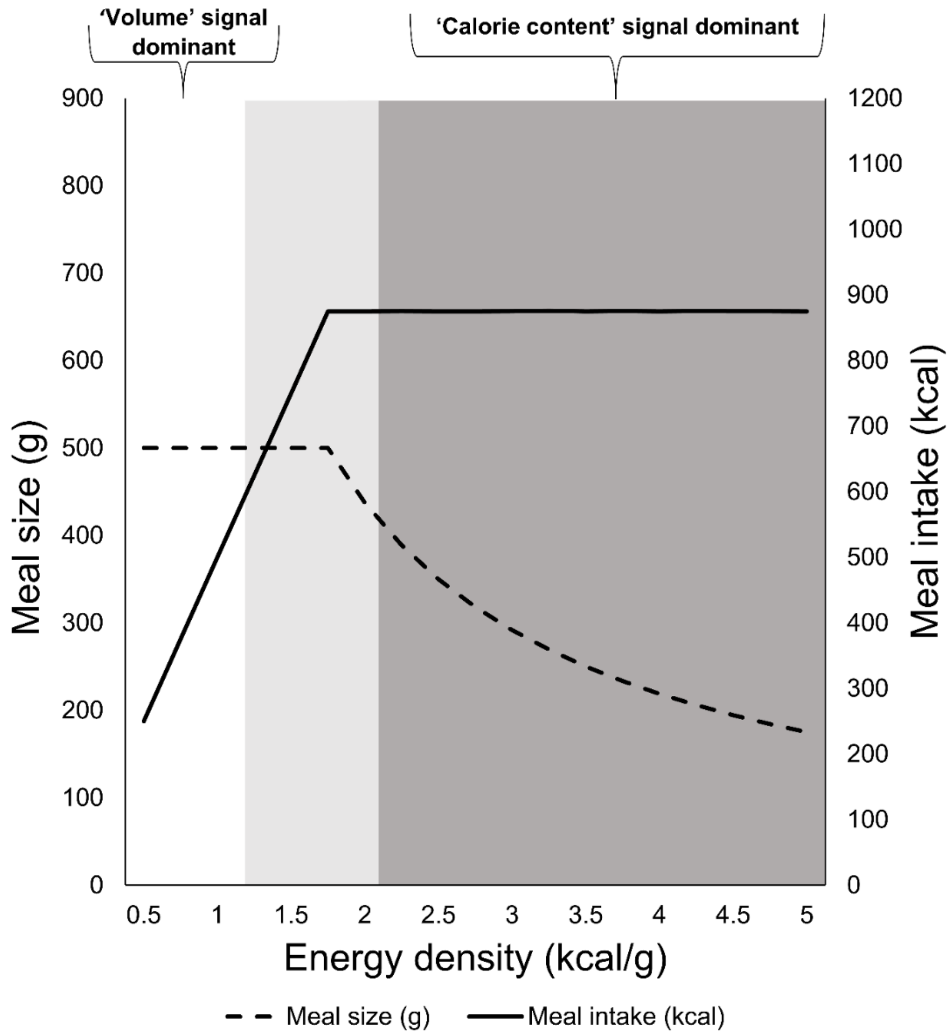


Figure 5.2 Two-component model of meal size (g): volume and calorie-content satiation signals.

This is modelled using an 875 kcal meal as an example and demonstrates perfect compensation. The white section indicates the dominance of the "volume" signal, the dark grey section the dominance of the "calorie-content" signal, and the lighter grey section indicates where a breakpoint might occur which is the location where the relative dominance of the signals changes.

Based on these seminal ideas, in the model currently being described, the volume signal is the dominant signal with energy-dilute foods and calorie-content is the dominant signal with energy-rich foods. The volume signal is a largely unconditioned response that affects food intake via gastric distension, whereas the calorie-content signal (biologically derived from the sensing of fat, carbohydrate, and protein) reduces meal size based on learned (anticipatory) and unlearned (immediate) effects of calories. Both the volume and calorie-content signals can impact meal size via food portion-size selection (expected satiety) (Brunstrom, 2014; Brunstrom et al., 2008) or within a meal directly (Weingarten & Kulikovsky, 1989; Yeomans, 2012). While feedback from the volume signal is constant across a range of energy densities, it is more salient with lower energy-dense meals. Low energy-dense foods are relatively dilute in calories and are high in intra-cellular water content and fibre (Drewnowski, 1998). This means that there is little feedback from the calorie-content signal, so the primary determinant of meal size is negative feedback from the volume signal via gastric distension.

With respect to the above model which posits that the volume signal dominates in lower energy-dense meals, the following reasoning can be applied. In all four datasets, the positive association between ED and centred meal caloric intake observed in the lower energy-dense meals (i.e., segment A) was driven by participants consuming a similar-sized meal across the range of energy densities. Thus, it would appear that feedback from the volume signal capped the size of lower energy-dense meals at a tolerable upper limit that is determined primarily by physical capacity. To clarify, the volume signal is not being positioned as the sole determinant of energy intake in lower energy-dense meals, but rather that it merely dominates over the calorie-content signal. Furthermore, the notion of a volume signal is largely conceptual. While gastric distension is governed by food volume, it is recognised that gastric emptying is also influenced by the nutrient content of a meal (Camps et al., 2016; Carbonnel et al., 1994; Hunt & Stubbs, 1975; Marciani et al., 2001; McHugh & Moran, 1979). Building on this idea, research conducted by Luscombe-Marsh and colleagues demonstrated that gastric emptying rate can be influenced by the ED and the macronutrient composition of the preload. In this within subject study, participants consumed three different preloads: high ED high fat, low ED high fat, and low ED high protein. The results indicated that increasing the ED of the preload slowed gastric emptying. However, the authors suggest that changes to the macronutrient composition of the preload, specifically increasing the fat content, had a greater effect on slowing gastric emptying than increasing the ED (Luscombe-Marsh et al., 2013). In the context of the two-component model, separating the independent role of nutrients (both total calories and macronutrient composition) and volume remains a challenge for future research.

As to the calorie-content signal, frequently consuming a food provides the opportunity to learn from delayed post-ingestive experiences (e.g., malaise if overconsumed (Hengist et al., 2020; Woods, 1991)). High energy-dense foods will provide relatively greater post-ingestive caloric feedback via the calorie-content signal. For the signal to operate effectively, it is critical that the usual relationship between orosensory cues and calorie content is preserved. This allows participants to use previous post-ingestive experiences to guide the amount of food consumed either via pre-meal planning (expected satiety) (Brunstrom, 2011) or during a meal via 'conditioned satiation' (Weingarten & Kulikovsky, 1989; Yeomans, 2012). Importantly, the reduction in meal size in response to increasing ED (segments B & C in Figure 5.1) is not only present at an individual level but might also be reflected on a larger scale, such as in full-service and fast-food restaurant meals (Roberts et al., 2018).

Consistent with a calorie-content signal, previous research has shown that ED also influences food choice and portion selection in real-world foods (Brunstrom, 2011; Brunstrom et al., 2018; Tang et al., 2014). In turn, this raises broader questions about the calorie-content signal's underlying mechanism and how this might support 'nutritional intelligence' in humans[10] (Brunstrom et al., 2023; Brunstrom & Schatzker, 2022). One idea is that taste or

[10]Human 'nutritional intelligence' is a concept which captures humans' ability to differentiate foods based on their nutritional composition and make advantageous decisions on this basis.[a] This concept is further expanded on in chapter eight.

[a]Brunstrom JM, Flynn AN, Rogers PJ, Zhai Y, Schatzker M. Human nutritional intelligence underestimated? Exposing sensitivities to food composition in everyday dietary decisions. Physiology & Behavior 2023. doi: 10.1016/j.physbeh.2023.114127.

sensory quality, such as sweet taste or fat sensation, can be used as a potential unlearned signal to predict the macronutrient content of a food (Teo et al., 2018; van Dongen et al., 2012; van Langeveld et al., 2017). However, others have questioned the utility of orosensory cues as a predictor of energy content because correlations between orosensory cues and energy content are mostly small (Glendinning, 2022; Mattes, 2021). Alternatively, some have argued that the ability to discriminate foods might be based on a learned association that forms between the sensory quality of a food and its post-ingestive effects (Booth et al., 1982; Sclafani, 1997). In rodents, this 'flavour-nutrient learning' is robust, however, it has proved difficult to demonstrate in humans under controlled conditions (Brunstrom, 2005; Yeomans, 2012). A review by Yeomans (2012) suggests that there are some similarities between humans and non-human animals with regard to flavour-nutrient learning, but that when investigated in a laboratory setting, key differences in experimental design might undermine flavour-nutrient learning in humans. A different explanation for the difficulty of observing the phenomena in humans is that, unlike rodents, humans routinely share food and form a cuisine. Thus, although flavour-nutrient learning may occur at an individual level in our species, it may do so over relatively long periods of time, to form a collective intergenerational wisdom. This might explain why flavour-nutrient learning is difficult to observe under controlled conditions, even though humans show a remarkable capacity to discriminate everyday (non-manipulated) foods based on their energy density (Brunstrom et al., 2023).

Finally, the reference to a calorie-content signal ignores the possibility that it might be influenced to a greater or lesser extent by energy derived from different macronutrients (for example, the work conducted by Luscombe-Marsh et al. (2013)), and this warrants further investigation. Lastly, and more generally pertaining to both the volume and calorie-content signals, additional research could also explore whether they are dissociable in terms of subjective experience - specifically, for example, in relation to having a full stomach versus (not) feeling hungry (Mantzavinou & Rogers, 2023). Relatedly, there is a question about how these signals might impact differently on subsequent eating events

Lastly, the success of the two-component model is that it explains complexity in the relationship between ED, meal size, and energy intake. However, the model is not exhaustive, and does not consider a role for individual macronutrients or effects of moderators such as eating rate (Robinson et al., 2014) and appetition (Sclafani, 2013) or other potential factors such as sensory and textural drivers (Ferriday et al., 2016; Forde & de Graaf, 2022), cognitive (Higgs & Spetter, 2018), and social influences (Ruddock et al., 2019). In addition, aspects of our modern food environment may also influence meal size, such as the capacity to manufacture brands and varieties of the same food that differ substantially in ED (Hardman et al., 2015). Lastly, it should be noted that the model does not predict how sensitivity to energy density within a meal might impact chronic energy intake, and future work might explore the relative importance of a calorie-content and volume signal and whether individual differences might account for variation in daily energy intake and energy balance (Flynn, Rogers, Hall, et al., 2023).

5.4 Understanding 'overcompensation' and implications for chronic energy intake

Given current high rates of obesity (NCD Risk Factor Collaboration [NCD-RisC], 2016), it might appear counterintuitive that results from chapters two, three, and four show sensitivity to calories in meals, and even a degree of overcompensation (reduction in the physical size of a meal that is greater than full compensation, such that energy intake reduces with ED i.e., negative slope in higher energy-dense meals).

One potential explanation for overcompensation is dietary restraint; specifically, a tendency to reduce the physical size (g) of especially energy-rich meals out of a concern to maintain or reduce body weight (Olea López & Johnson, 2016). To establish whether dietary restraint might drive the negative association between meal ED and meal caloric intake in higher energy-dense meals, the author re-analysed the data from the UK NDNS (Flynn, Rogers, et al., 2022). As part of NDNS, participants were asked to complete the Dutch Eating Behaviour Questionnaire (DEBQ) which assesses several eating traits including emotional, external and restrained eating using a 5-point Likert scale (van Strien et al., 1986). Restraint scores were split into low (≤ 3) and high (> 3) restraint (Olea López & Johnson, 2016; van Strien et al., 1986), and separate linear regressions were conducted within each restraint group and each segment (i.e., Segment B which comprised meals between 1.75 and 2.94 kcal/g and Segment C which was meals above 2.94 kcal/g). Predictors in the linear regression included meal ED, binarized dietary restraint, and an interaction between meal ED and dietary restraint which was included to establish whether the slope of meal ED differed between low-restraint (n= 1,097) and high-restraint (n= 397) eaters. In both segments[11], the linear regression returned significant interaction terms (p < .05), and individuals who scored low on dietary restraint showed greater overcompensation than those that scored high (see Table 5.1). In other words, individuals who had lower levels of dietary restraint reduced their meal size of higher energy-dense meals to a greater extent than those with higher levels. Importantly, these results suggest that dietary restraint is not driving the overcompensation pattern, at least not in the NDNS dataset.

It is unclear why greater overcompensation occurred in individuals with lower levels of dietary restraint. One possibility is that BMI might have confounded the association between restraint and meal intake. However, in these analyses, meal intakes were centred within each participant, thus minimising any participant-level meal intake differences. Additionally, there is no clear association between restraint and BMI or weight change (Hays & Roberts, 2008), so it is unlikely to confound the association between restraint and meal intake. Another potential explanation for the lack of an association between meal ED and intake in higher

[11]Linear regression in Segment B: $F(3, 10{,}630)= 81.76$, $p< .001$; Linear regression in Segment C: $F(3, 4{,}679)= 12.45$, $p< .001$

energy-dense meals in participants with high dietary restraint is a lack of interoceptive awareness. It is possible that individuals with increased dietary restraint demonstrate poorer interoceptive sensitivity to energy signals, and, consistent with this, there is some research suggesting that hunger/satiety-specific interoception sensibility is negatively associated with cognitive restraint (Poovey et al., 2022). In other words, individuals with high restraint demonstrate lower hunger/satiety-specific interoception sensibility (Poovey et al., 2022). Importantly, it should be noted that the relative lack of an association between meal intake and meal ED in higher energy-dense meals might also suggest perfect sensitivity to calories (i.e., accurately reducing meal size (g) to account for increased meal ED). On this basis, an alternative explanation pertains to nutritional knowledge. One possibility is that, individuals with high restraint have greater nutritional awareness about foods (for example, choosing 'healthier' foods (Contento et al., 2005; de Castro, 1995; Tepper et al., 1997)) and could therefore select the appropriate portion size (g) in response to increases in meal ED, resulting in no association between meal intake and meal ED in higher energy-dense meals. Importantly, the explanations described above are speculative and more research is needed, and in general, further research into the potential for individual differences, such as BMI or dietary restraint, to influence the non-linear pattern in meal caloric intake and sensitivity to calories is warranted.

Table 5.1 *Unstandardised Slope Coefficients and Standard Error for a Linear Regression in Energy Density Segments Occurring Above the First Breakpoint in the UK NDNS Dataset*

	Restraint group	Unstandardised B	Standard error
Segment B (1.75-2.94 kcal/g)	Low	-120.96	8.63
	High	-87.22	13.55
Segment C (2.94 kcal/g and above)	Low	-65.16	11.68
	High	3.07	19.48

Another potential explanation for the negative slope in higher energy-dense meals relates to concentrations of a single macronutrient. Specifically, there is good evidence that, due to their aversive nature (Lucas et al., 1998; Moskowitz, 1971a; Moskowitz et al., 1974; Sclafani

& Ackroff, 2004), non-human animals limit their intake of high concentrations of a single macronutrient (Smith & Foster, 1980) to avoid excess satiety or ‘nimiety’ (Kulkosky, 1985). Similar patterns have also been observed in humans (Lucas & Bellisle, 1987; Martin et al., 2016; Pérez et al., 1994; Zandstra et al., 1999). Thus, beyond dietary restraint, the evidence for overcompensation (reduction of energy intake in higher energy-dense meals) might reflect the same underlying process, and avoidance of negative visceral sensations (e.g., ‘feeling sick/nausea’ (Booth et al., 2011) or malaise (Hengist et al., 2020)). Again, identifying potential drivers of the overcompensation pattern (i.e., negative slope in higher energy-dense meals) remains an important avenue for future research.

5.5 Reconciling findings from ad libitum and preload test-meal studies with the two-component model

As mentioned in the introduction of chapter two (see section 2.2), short-term ad libitum studies (e.g., < 10 total exposure days) report ED has little to no influence on meal size. Based on the two-component model, this insensitivity is seen for two reasons. First, ED is often manipulated covertly, which undermines the learned calorie-content signal. Second, the meals or diets are often energy dilute (e.g., < 2 kcal/g), which means the volume signal dominates. In combination, this explains the tendency to consume a consistent weight of food as described in many ad libitum studies. Indeed, it has been previously observed that this tendency might only occur below a certain low ED (Kral et al., 2002), and the Volumetrics Eating Plan (Rolls & Barnett, 2000) illustrates how this strategy can generate sustained weight loss (Ello-Martin et al., 2007).

By contrast, preload test-meal studies demonstrate some sensitivity to food ED and reflect an unconditioned calorie-content signal. Here, an interval exists between the preload and the test-meal. Therefore, the calorie content of the preload, even when covertly manipulated, can be detected (by the gut e.g., Wilbrink et al. (2021)) to affect subsequent test-meal intake. Moreover, for longer-term ad libitum studies, the effects of ED on meal size could be explained by the capacity of the calorie-content signal to influence satiation indirectly, via associative learning. Specifically, the orosensory features of the food become associated with the post-ingestive consequences of its calorie (macronutrient) content which, over time, come to modify meal size, a phenomenon similar to ‘expected satiety’ (Brunstrom, 2014; Irvine et al., 2013).

5.6 Differences in sensitivity to energy density observed in food choice and food intake

While the previous section discussed differences in sensitivity to ED based on study design, this section reviews how sensitivity to ED might differ based on study outcome, specifically choice and intake. It may seem paradoxical that there are contrasting patterns of sensitivity to ED in studies of food choice and food intake. In choice studies, a clear linear association (positive) is observed between ED and preference, but only in lower energy-dense foods (~ <1.75 kcal/g) (Brunstrom et al., 2018; Gibson & Wardle, 2003). In foods with progressively higher ED (~ >1.75 kcal/g), this relationship weakens until choice and ED become unrelated (Brunstrom et al., 2018). Whereas for food intake, the present results demonstrate the converse - greater sensitivity to ED in higher energy-dense meals.

These different findings may reflect an adaptation that maximizes caloric intake in an environment in which ED varies substantially, while at the same time avoiding the acute aversive effects of short-term overconsumption (Booth et al., 2011; Hengist et al., 2020; Rogers & Brunstrom, 2016). Differences in the ED of energy-dilute foods matter because stomach capacity is limited. When only energy-dilute foods are available, choosing the least energy-dilute (most energy-dense) food will ensure that energy intake is maximized. By contrast, with energy-rich options, absolute stomach capacity is relatively unimportant, and the priority shifts to avoiding acute, negative soporific effects caused by an overconsumption of calories (Hengist et al., 2020). Accordingly, a compensatory reduction in meal size with ED is observed, which, as noted above, is driven by a largely learned anticipation of the effects of the food's calories on satiety (i.e., the calorie-content signal).

5.7 Limitations

Finally, there are some limitations to the research which should be noted. Firstly, as mentioned in section 5.2, the cross-sectional design of the studies in chapters two, three, and four, does not allow for causal conclusions. Additionally, the analyses in chapters three and four used data from large dietary surveys, and, while interview-administered 24-hour dietary recalls are widely used (Kim & Park, 2023), reliable and have been well validated (Blanton et al., 2006; Kim & Park, 2023; Kirkpatrick et al., 2014; Moshfegh et al., 2008), it is still possible that participants could have under-reported their intake, especially of energy-dense foods (Lafay et al., 2000; Macdiarmid & Blundell, 1998; Ravelli et al., 2018). This under-reporting could have, in part, explained the negative slope in higher energy-dense meals. Importantly, however, a similar pattern was observed in the analysis of the Hall et al. dataset where meals were consumed under controlled settings and data were collected by trained research staff (chapter two). It remains a challenge to minimise the potential bias of under-reporting and encourage accurate responding by participants completing a dietary survey, and novel approaches utilising online or app-based intake measures, such as Intake24 (Foster et al., 2019; Rowland et al., 2018; Simpson et al., 2017), are promising. Lastly, while the analyses were repeated in datasets from several countries which differed in culinary habits, it remains important to replicate them in further datasets from additional countries.

5.8 Summary of chapters two, three, four and five

The association between meal ED and meal caloric intake within a single meal was re-evaluated in four different datasets in an attempt to better understand the disparate findings from ad libitum and preload test-meal studies suggesting human sensitivity and insensitivity to calories. Uniquely, the influence of meal ED on meal energy intake was measured 1) across a broad and continuous range of energy densities, 2) using ‘real world’ foods which were not covertly manipulated and, 3) allowing for the possibility that the association is non-linear. A consistent non-linear pattern was observed across the four datasets, and this was explained using a theoretical two-component model comprising volume and calorie-content signals. More broadly, the results add to the evidence for human nutritional intelligence (further discussed in chapter eight (Brunstrom et al., 2023)) and highlight additional complexity and opportunities to understand how we interact with our food environment in ways that might impact long-term energy balance.

Chapter 6 Protein: Developing a novel paradigm to assess protein discrimination in humans using a series of online studies

6.1 Acknowledgements and overview

The research presented in this chapter included a collaboration with Dr Olga Davidenko (INRAE – AgroParisTech), Dr Nicolas Darcel (INRAE – AgroParisTech), Professor Suzanne Higgs (University of Birmingham), and Professor Jeff Brunstrom (University of Bristol, supervisor). For the first three studies (described in sections 6.3, 6.4, and 6.5), I was responsible for task development, data collection, data analysis, and interpretation of findings. The collaborators provided feedback regarding task development and interpretation of results. The results from the third study in the first part of the chapter (section 6.5) have been presented online at the Computational Approaches to Eating Behaviour workshop in January 2021. For the study in the second part of the chapter (described in section 6.6), six final-year undergraduate students assisted in data collection and used a portion of the data in their dissertations. For this study, I was responsible for task development, coding the task, experimental design, data analysis, and interpretation of results. This research was conducted under the supervision of Professor Jeff Brunstrom.

The aim of the series of studies in this chapter was to develop a novel paradigm and an online task to assess protein discrimination in humans. In the context of this chapter, protein discrimination is operationalised as the extent to which a behavioural response can be predicted by food protein content. Several different approaches have been used to assess protein discrimination in humans. For example, one could induce a state of mild protein depletion in an individual and then assess choice between flavours previously paired with low- or high-protein loads (such as in Gibson et al., 1995). A second methodology includes developing low- and high-protein diets and, again, assessing participant's food choices after consuming these diets for a set period of time (for example see Griffioen-Roose et al., 2012). Both of these approaches can be costly and time-consuming; therefore, an online task using everyday dietary decisions might provide a more feasible and accessible method to assess protein discrimination in humans. The three studies in the first part of the chapter were used to establish whether the tendency to pair protein with carbohydrate observed in real-world meals (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009) extends to an online choice environment. The first two studies are pilot studies (see sections 6.3 and 6.4), and the third study tests the protein-carbohydrate pair paradigm developed in the pilot studies in a larger sample (see section 6.5). The second part of the chapter presents a single study that uses the protein-carbohydrate pair paradigm developed in the first three studies to generate a novel online task to assess protein discrimination in humans (see section 6.6). In the chapter discussion, the results from the final study are summarised before interpreting the findings from the entire chapter. Additionally, limitations to the research are highlighted and possibilities for adapting the task for use in future studies are also explored.

6.2 Introduction

Chapters two through five explored whether humans are sensitive to the energy content of real-world meals. However, it is important to recognise that food comprises different macronutrients, micronutrients, fibre, and water, not just calories. A related, yet slightly different research question to the one explored in the previous chapters, is the extent to which humans can discriminate foods based on their macronutrient composition and whether this impacts food choice or preference. The research presented in this chapter will explore this question in relation to protein, specifically whether humans are capable of protein discrimination, that is, whether their behaviour can be predicted by the protein content of food, and whether this discrimination can be assessed using an online task. The chapter introduction (current section) begins by suggesting that a switch to more sustainable protein sources is important to reduce greenhouse gas emissions and introduces a related question regarding how we might ensure our protein requirements are met if we need to shift to more sustainable protein foods which are often lower in absolute protein content. The introduction then reviews studies on dietary self-selection by human and non-human animals and outlines research exploring protein discrimination and protein appetites in human and non-human animals. Lastly, it returns to the question from the beginning of the introduction and highlights the associated research gap before reviewing methodological and theoretical considerations relevant to developing an online task to assess protein discrimination in humans.

6.2.1 Protein intake, climate change, and consuming plant-based protein foods

Protein is a macronutrient comprised of amino acid chains and is used to build and maintain muscle as well as produce enzymes and haemoglobin. If used as a fuel source, protein provides equivalent energy per gram as carbohydrate (both 4 kcal/g). Of the 20 different amino acids, nine have been identified as being essential for humans, meaning that they cannot be synthesized by the human body and must be consumed from the diet; the remaining 11 are deemed non-essential as they can be produced by the body (Institute of Medicine, 2005; Weiler et al., 2023). Proteins also differ in their quality as determined by their amino acid composition as well as digestibility (Schaafsma, 2005). For humans, a high-quality protein provides sufficient levels of essential amino acids and is easily digested (Hertzler et al., 2020). In general, plant-based protein sources are viewed as having lower protein quality than animal-based proteins as they contain insufficient amounts of at least one essential amino acid and have lower digestibility (Hertzler et al., 2020).

Regardless of protein source, it is recommended that humans consume at least 0.8 g/kg of protein per day; however, this recommendation might differ for some individuals, such as the elderly (Wolfe et al., 2008) or athletes (Phillips, 2012). In light of the current climate crisis, protein consumption, specifically the type and amount of protein consumed, is discussed widely. Currently, animal agriculture is believed to be responsible for between 16.5 - 28.1% of global greenhouse gas emissions, and this percentage includes, for example, the emissions produced by land use change for the production of animal feed and grazing pasture, methane production by ruminants, as well as the fossil fuel used during the production and transport of animal products which are processed and refrigerated (Twine, 2021).

The 2019 Inter-governmental Panel on Climate Change (IPCC) encouraged a shift towards more sustainable diets comprising largely plant-based foods (i.e., coarse grains, pulses, fruit, and vegetables) and being low in animal-containing products and processed foods (i.e., beverages high in sugar) (Mbow et al., 2019). This transition towards greater consumption of plant-based proteins, which as suggested above, might be lower in absolute amount and quality of protein, raises a key question regarding whether humans discriminate protein content[12] in food and how we ensure our protein requirements are met. One possibility is that by randomly consuming different amounts and types of food, we successfully meet our minimum protein requirements. On the other hand, given that protein is critical for human health, humans may have developed a mechanism to detect protein in food to ensure that the minimum protein requirements are met. By exploring whether humans are capable of protein discrimination, potential psychological and physiological barriers to transitioning to a more sustainable diet may be exposed. The following two sections will review research in both human and non-human animals regarding dietary self-selection and protein discrimination, respectively.

6.2.2 Dietary self-selection by human and non-human animals

While not direct evidence for the targeted regulation of protein, the ability of an organism to self-select a nutritionally balanced diet from a variety of different foods has been demonstrated in both wild and domesticated animals as well as in laboratory subjects. For example, the seasonal migration patterns of herbivores in the Serengeti appear to be influenced

[12]As defined previously, protein discrimination is operationalised as the extent to which human behavioural responses are predicted by the protein content of the food.

by the mineral concentration of grass and are driven largely by the mineral requirements of lactating females and juvenile animals (McNaughton, 1990). The mechanism behind this ability to select a balanced diet is unclear, and it has been argued that wild animals consume a balanced diet simply due to seasonal changes or environmental constraints (Marques & Baucells, 2001).

Studies regarding the self-selected diets of captive and domesticated animals using the paradigm of cafeteria-style feeding have more rigorously addressed whether animals can consume a balanced diet without guidance from environmental cues. Both calves and rats have displayed this ability, with Atwood and co-authors demonstrating that calves offered a choice of rolled barley, rolled corn, corn silage, and alfalfa hay gained the same amount of weight while consuming fewer calories compared to animals that were fed a predetermined mixed ration (Atwood et al., 2001). This demonstrates that the animals in the choice condition were better able to meet their unique macronutrient needs in comparison to animals consuming a set single diet (Atwood et al., 2001). Even earlier, in 1938, lab-based research conducted by Richter, Holt and Barelare indicated that rats could self-select a diet from purified food components (i.e., casein, sucrose, and olive oil) which resulted in equivalent growth compared to rats fed the standard lab chow, even while consuming 18.7% fewer calories (Richter et al., 1938). Importantly, the reliability of animals consistently self-selecting balanced diets in a lab-based setting has been critiqued, and a review of 17 studies determined that only eight studies reported that rats were successful in attaining normal growth during dietary self-selection while the rats in the remaining nine studies failed to do so (Lảt 1967; as cited in Galef 1991). One possibility for the failure to observe successful diet self-selection pertains to the potentially aversive sensory properties of the purified food components (Booth, 1985). For example, if the orosensory properties (i.e., the ‘glueyness’) of casein are aversive to the rodent, but casein is the only protein source available, then the rodent is unlikely to successfully balance its diet due to avoiding the protein-containing foodstuff (Booth, 1985). In general, however, the ability to self-select a nutritionally balanced diet appears to be present in animals in certain paradigms and conditions, but a question remains regarding the extent to which humans exhibit this ability.

The last, and arguably only, attempt to scientifically demonstrate humans' ability to self-select a balanced diet was conducted in 1939 by the paediatrician Clara Davis. In this study, which expanded on her previous study, infants of weaning age were provided with a variety of non-processed foods, such as peas, carrots, oatmeal, and beef, from which they self-selected their diet (Davis, 1939). The children's diets differed largely from one another and included unique combinations such as "breakfast of a pint of orange juice and liver; a supper of several eggs, bananas, and milk" (Davis, 1939, p. 260). While the consumed diets varied substantially across children, each child managed to maintain good health throughout the study.

Importantly, the foods utilised in Davis's study have been critiqued for not being representative of the modern food environment as they were largely unprocessed (Strauss, 2006). Given the unprocessed nature of the foods provided, one possibility is that the children simply balanced their diet by randomly consuming the foods, rather than exhibiting 'body wisdom' as suggested by Davis (Strauss, 2006). Alternatively, the results of the study could also suggest a potential capacity for humans, in this case children, to detect, and respond to, a food's nutritional value and modify their diets to meet their unique nutritional requirements. In summary, while cafeteria-style studies conducted in both non-human animals and humans appear to capture an organism's ability to self-select balanced diets, their success is potentially dependent on the quality of the foods offered (Galef, 1991; Strauss, 2006), and it is still possible, as mentioned above, that the individual's nutritional requirements are met as the result of randomly consuming different amounts of foodstuffs (Anonymous, 1944; as cited in Galef, 1991).

6.2.3 Protein discrimination and learned and unlearned protein appetites in human and non-human animals

Despite being a highly researched topic, the extent to which a mechanism exists in both human and non-human animals to detect different macronutrients, specifically protein, and tightly regulate protein intake is unclear (for a narrative review regarding self-selection of dietary protein and protein requirements see Even et al. (2021)). Beginning more generally, there is evidence that the macronutrient composition of a meal, specifically the lack of protein, can influence the short-term (i.e., 30-minute) regulation of macronutrient intake. Rats fed a protein-deficient preload increased their protein intake in the next meal (presented after 30 minutes) compared to those fed a preload containing protein (Li & Anderson, 1982). Research also suggests this targeted increase in protein consumption occurs not only in response to acute protein deprivation but also in response to protein quality (Kishi et al., 1982; Pol & Den Hartog, 1966) and the physiological state of the animal, specifically lactation and gestation (Cohen & Woodside, 1989; Leshner et al., 1972).

It has been hypothesized that learned or unlearned protein-specific appetites might drive changes in protein intake patterns. An unlearned appetite occurs without conditioning and is an unconditioned response. Deutsch and colleagues demonstrated an unlearned appetite when they depleted rats of protein and presented them with a novel diet high in either protein or carbohydrate (Deutsch et al., 1989). The depleted rats selected the high-protein diet more frequently than protein-replete rats, and they maintained their preference for the higher protein diet across a 30-minute period. Importantly, the increased preference for the high-protein diet was thought to not simply reflect the tendency for nutritionally deficient rats to prefer novel flavours (Deutsch et al., 1989). Additionally, the authors claim that this preference for the novel higher protein diet by the protein-deficient rats (and rats with increased protein needs via pregnancy) cannot be a learned behaviour based on the rapid selection of the high protein diet.

In other words, the preference cannot be associated with the delayed post-ingestive experience and resulting positive conditioning when the animal experiences a reversal of their protein deficiency (Deutsch et al., 1989). A similar unlearned response to protein has been described by Gietzen and colleagues who demonstrated that rodents can rapidly detect diets containing amino acid deficiencies (Koehnle et al., 2003). Specifically, protein-deplete rodents appear to reject diets which are deficient in amino acids by reducing their first meal duration, and this response occurred after controlling for the novelty of the various diets (Koehnle et al., 2003). Together, these studies suggest that rodents may have an unlearned protein appetite which can respond to both total protein amount and amino acid composition.

The existence of a learned appetite for protein is based on research in which individuals associate the sensory qualities of a specific food with its respective post-ingestive effects, a phenomenon commonly known as 'flavour-nutrient' learning (previously mentioned in chapter five (Booth et al., 1982; Sclafani, 1997)). In the case of protein depletion, an animal might associate the alleviation of their deficiency after consuming a food with that food's sensory properties (e.g., flavour) and therefore be more likely to select, and potentially consume, the food again when allowed to do so. A learned protein-specific appetite has been demonstrated in both locusts (Simpson & White, 1990) as well as rats (Baker et al., 1987; Booth, 1974; Gibson & Booth, 1985). Additionally, this learned protein appetite could also be amino acid specific as demonstrated by rats who, when fed a baseline diet with a specific limiting amino acid, selected against a diet containing even less of the limiting amino acid when presented with a choice between the deficient diet and the baseline diet. This tendency was observed even if the deficient diet contained a reduction in the amino acid content as small as 0.01%. This has been shown in both lysine- and threonine-deficient rats (Hrupka et al., 1999; Hrupka et al., 1997). Critically, considering an apparent inconsistency with which protein-deficient animals can successfully remedy their protein deficiency (Overmann, 1976), the existence of a precise protein-specific appetite which tightly regulates protein intake should be interpreted with caution.

The extent to which humans exhibit a protein-specific appetite is somewhat uncertain. It has been suggested that the protein leverage hypothesis is consistent with the existence of a specific appetite for protein (Raubenheimer & Simpson, 2019; Simpson & Raubenheimer, 2005). Briefly, the protein leverage hypothesis is the idea that protein intake is prioritised by humans and that this protein appetite can impact non-protein energy intake (Raubenheimer & Simpson, 2019; Simpson & Raubenheimer, 2005). For example, if an individual is in an environment in which their diet is dilute of protein (i.e., high in fat and carbohydrate), then they are predicted to eat an excess of energy until their protein requirements are met. Contrastingly, if the individual is in an environment containing a protein-rich diet, then the leverage of protein appetite on non-protein energy intake would result in a reduction in overall energy intake as the protein requirement would have been met (Raubenheimer & Simpson, 2019; Simpson & Raubenheimer, 2005). Evidence for the protein leverage effect has been observed in humans consuming diets where the protein content was experimentally manipulated. Those individuals consuming the lower protein-containing diet (i.e., 5% or 10% of energy from protein) consumed more total energy than those on a higher protein-containing diet (i.e., 25% or 30% of energy from protein) (Gosby et al., 2011; Gosby et al., 2014; Martens et al., 2013). A degree of protein leverage has also been reported in free-living humans; absolute protein intake remains constant over time (Lieberman et al., 2020; Martinez-Cordero et al., 2012) and increases in the consumption of ultra-processed foods (i.e., foods which tend to be higher in fat and carbohydrate) were met with increases in total energy intake to maintain absolute protein intake (Martínez Steele et al., 2018). Importantly, when modelling partial protein leverage in free-living humans (i.e., incomplete compensatory increases in energy intake to account for the dilution of dietary protein), protein leverage is thought to be responsible for one-third of the weight gained by an average adult during the US obesity epidemic (Hall, 2019), indicating its potential as a significant contributor to the increasing rates of obesity.

Rather than a general increase in non-protein energy intake to satisfy a protein appetite, there is tentative evidence that protein is specifically targeted when an individual is depleted of protein. Participants who consumed a low-protein diet (e.g., 0.50 g of protein per kg of body weight) exhibited an increased preference for and intake of savoury high-protein foods, a behaviour not observed after consuming a high-protein diet (e.g., 2.00 g of protein per kg of body weight) (Griffioen-Roose et al., 2012). Gibson and colleagues also report on the existence of a learned protein appetite when participants who had been mildly depleted of protein demonstrated an increased preference for a flavour that had previously been paired with protein, even after only one conditioning trial (Gibson et al., 1995). Non-protein-depleted humans also appear to exhibit a unique orientation toward protein. Individuals with lower markers of blood protein, as well as older participants (mean age 84 years), indicated a greater preference for an amino-acid-deficient soup paired with a higher concentration of casein hydrolysate, a complete protein (Murphy & Withee, 1987). The authors also emphasise that the participant's blood protein values fell within the normal range of variability, indicating no evidence of malnutrition or protein depletion (Murphy & Withee, 1987). More broadly, in non-depleted healthy adults, calories from protein were positively associated with food choice; in other words, calories from protein were valued more than those from fat and carbohydrate (Buckley et al., 2019). In line with Murphy and Withee (1987), physiological state influenced behaviour, and older adults with greater lean muscle mass exhibited this 'valuation' of protein more strongly than individuals with less lean muscle mass (Buckley et al., 2019).

6.2.4 Developing an online paradigm to assess protein discrimination using everyday dietary decisions

While the studies reviewed in the prior sections suggest that humans possess an ability to respond to the protein content of food, the evidence is unclear whether a protein-specific appetite exists or whether humans reliably discriminate foods based on their protein content. Importantly, unlike rodents, which demonstrate more robust evidence for a protein-specific appetite, humans have cultural norms and personal beliefs which potentially make it more difficult to interpret behavioural outcomes. Given the biological relevance of protein and the increasing necessity to transition to more sustainable protein consumption patterns, it is imperative to establish an approach to assess potential protein discrimination in humans, especially one which does not involve depleting individuals of protein, an approach that is difficult, expensive, and ethically challenging.

To note, in the context of this thesis, macronutrient discrimination refers to behavioural responses (i.e., food choice, food preference, intake etc.) which can be predicted by the macronutrient composition of the food. Therefore, and as a reminder, protein discrimination is operationalised as the extent to which behavioural responses can be predicted by food protein content. Based on the above-outlined concerns regarding using protein depletion to assess protein discrimination, an online paradigm was developed (sections 6.3, 6.4, and 6.5) that used everyday dietary decisions to assess potential protein discrimination in humans (see section 6.6), and the theoretical development is outlined in detail in the paragraph below.

The tendency to pair sources of protein with sources of carbohydrates appears to be a relatively stable cultural phenomenon. For example, in the United Kingdom (UK) or for Anglo-Australians, the traditional Sunday roast dinner involves a source of protein, commonly roast meat, paired with potatoes and various sides such as Yorkshire puddings or peas, all sources of carbohydrates (Charles & Kerr, 1986; Foley, 2005). Similarly, in Brazil, it is common for rice, beans, meat, and vegetables to comprise the main lunch meal during the week (Deliza & Casotti, 2009), and in India, meals are often comprised of three components: the core (usually a complex carbohydrate), the fringe (often an animal or vegetable) and legumes (source of protein) (Sen, 2009).

Building on this seemingly robust cultural tendency to create mixed meals (i.e., a source of protein with a source of carbohydrate rather than two sources of the same macronutrient), two versions of an online psychophysical task were developed which involved participants choosing between two pairs of food to eat for lunch. The two task versions were:

1) Six foods task: this task used sources of protein and carbohydrate familiar to UK participants and created culturally familiar food pairs (explained further in section 6.3)
2) Peanuts and crisps task: this task again used familiar sources of protein and carbohydrate; however, it also included some food pairs which were likely to be culturally unfamiliar to individuals belonging to a Western culture (i.e., peanuts paired with pasta, and the task is explained in further detail in section 6.4)

The six foods task assessed whether the same tendency to create mixed meals (i.e., combining protein and carbohydrate in a meal) is also evident in culturally familiar pairs comprising a source of protein and a source of carbohydrate. The peanuts and crisps task is important as it explores whether individuals exhibit an underlying discrimination of the macronutrient composition of a pair, beyond simply following cultural tendency. This protein-carbohydrate pair paradigm was then further developed into a single protein-carbohydrate pair ratings task to explore protein discrimination, and this is expanded on in section 6.6.

Initial feasibility testing of the above two tasks was completed in two small pilot studies and each is described separately (see sections 6.3 and 6.4). The two tasks were then further tested in a study with a larger sample size (see section 6.5). In the six foods task, based on the evidence from real-world meals in different cultures (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009), it was predicted that participant behaviour would be non-random such that they would prefer food pairs which comprised a mixture of macronutrients (i.e., protein paired with carbohydrate) compared to food pairs which contained only one macronutrient (i.e., two sources of carbohydrate or two sources of protein). In the peanuts and crisps task, it was unclear how participants might respond as they were evaluating culturally unfamiliar pairs. The following section will describe the six foods task in further detail and present the results of the first pilot study.

6.3 Six foods task using culturally familiar food pairs to assess a preference for pairs containing both protein and carbohydrate

To note, the structure of the chapter sections describing the first two pilot studies is very similar (current section and section 6.4). Each begins with a brief outline of the task and the study procedure, followed by the results before providing a short discussion of the findings. The third study (section 6.5) departs slightly from this structure and includes a more detailed methods section as the study comprised several different tasks and recruited a larger sample of participants. This methods section is followed by results and discussion sections, the latter of which includes a broader review of the results across all three pilots.

As mentioned previously, the main aim of the six foods task was to explore whether, in culturally familiar pairs, a preference existed for mixed pairs (i.e., protein and carbohydrate in a single pair) over pairs containing only protein or only carbohydrate (i.e., two sources of protein or carbohydrate). Briefly, the six foods task asked participants to imagine that they are preparing a lunchbox for work and that their lunch might have one type of food or it might have two. They were then told that they should pick the pair of foods that they would want to eat in their meal (see Figure 6.1 for an example of a trial). On the instruction page, participants were shown an image of an example trial and were told that sometimes the pair of foods would be the same (e.g., two portions of pasta) and that, in this scenario, they were asked to imagine that they were being offered a double helping. Lastly, the instructions stated that only these foods and portions would be available to the participant in their meal.

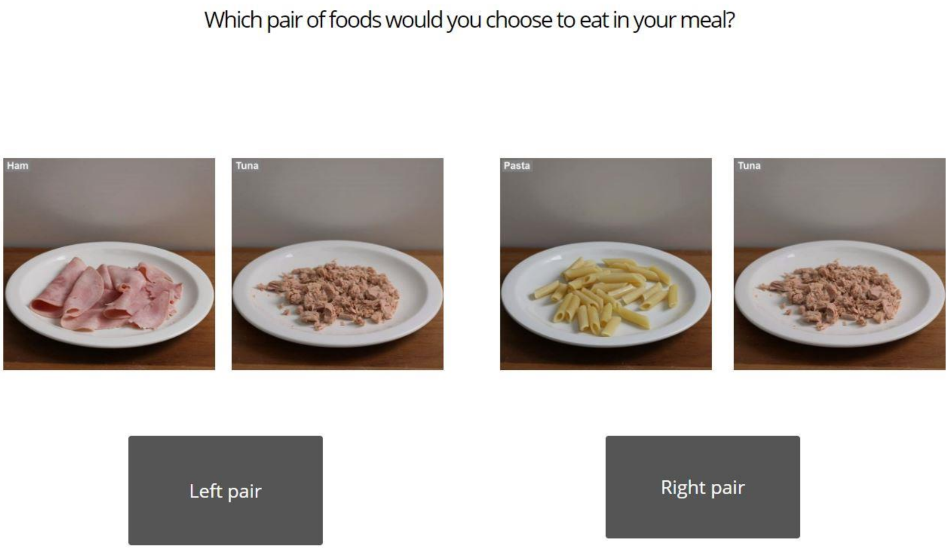


Figure 6.1 Example of a trial in the six foods task.

The pair on the left comprises two sources of protein whereas the pair on the right is a mixed pair comprising protein and carbohydrate. In this trial, one would predict that participants select the mixed pair on the right to eat in their meal.

Two sets of six highly familiar UK foods were utilised, of which three were high carbohydrate foods and three were high protein foods (see Table 6.1). In this task, every possible pair was paired with every other pair, resulting in 420 total trials (210 per set). Pairing every pair with every other pair reduces the likelihood that preference for a single pair might drive the pattern of responding (i.e., the participant cannot select the same pair each trial as every pair is presented an equal number of times).

Table 6.1 *Nutritional Information of the Stimuli Comprising the Two Stimuli Sets in the Six Foods Task*

Set	Food item	Protein or carbohydrate food	Calories per 100g	Fat (g/100g)	Carbohydrate (g/100g)	Protein (g/100g)
Set 1	Bagel	Carbohydrate	255	1.3	48.9	10.3
	Banana	Carbohydrate	103	0.5	23.0	1.2
	Pasta	Carbohydrate	160	0.7	32.5	5.1
	Chicken	Protein	113	1.6	0.5	23.9
	Ham	Protein	105	2.3	1.7	19.0
	Tuna	Protein	113	0.5	0.5	27.0
Set 2	Coleslaw	Carbohydrate	181	17.0	5.4	0.8

Chickpeas	Carbohydrate	122	1.4	16.5	7.7
Potato salad	Carbohydrate	140	10.2	10.6	1.0
King prawns	Protein	62	0.5	0.5	14.1
Roast beef	Protein	120	2.4	0.5	24.4
Turkey	Protein	150	5.6	0.0	25.0

Alongside the six foods task described earlier, participants completed liking ratings for each of the 12 foods using a 100-unit visual analogue scale (VAS) in response to the question 'How much do you like the taste of this food?' with response anchors of 'Not at all' (0) on the left and 'Extremely' (100) on the right. Lastly, familiarity with the 12 foods was assessed using the question 'Have you consumed this food before?' and the response options of 'Yes' and 'No'. In all three tasks, participants were presented with highly controlled images and the order of stimuli presentation was randomised. All food images were taken in 100 g portions on a white plate (255-mm diameter) against a white background using a high-quality camera (Nikon D50). The images were taken at a 45-degree angle, and the name of the food was included as a label in the upper left-hand corner of the image (see Figure 6.2 as an example).

Figure 6.2 Example of a formatted stimulus as used in the six foods task

Volunteers (n= 8) were informally recruited from colleagues and friends, and they completed the pilot tasks in the following order: the six foods task, liking, and familiarity. All data were collected using the experiment builder Gorilla (https://gorilla.sc/, Anwyl-Irvine et al., 2020) during one online session lasting approximately 10 minutes, and participants could use a tablet, laptop, or desktop computer to complete the study. The data collected from the liking and familiarity tasks were only used to describe the sample and are not discussed further (see Appendix 3 Table 11.5 (familiarity) and Table 11.6 (liking) for descriptive statistics).

6.3.1 Results

To explore whether there was a preference for protein paired with carbohydrate (mixed pairs) over pairs containing only protein or only carbohydrate, the percentage (%) of trials that each of the three pair types (i.e., mixed (M), only protein (P+P), only carbohydrate (C+C)) was selected across the three trial types (only carbohydrate (C+C) vs. mixed (M), only protein (P+P) vs. mixed (M), or only carbohydrate (C+C) vs. only protein (P+P)) was calculated. This was done separately for each set of six foods and each participant. Average percentages across the entire sample are included in the last row of the table to capture the overall pattern in responding (see Table 6.2 below).

Collapsing across both sets of foods, participants appeared to prefer pairs containing two sources of carbohydrate rather than two sources of protein, 64.93% and 35.07%, respectively. Aligned with what was hypothesized, across the entire sample, participants tended to prefer pairs of foods that contain a mixture of macronutrients (i.e., protein paired with carbohydrate) rather than two sources of a single macronutrient, and the tendency to prefer a mixed pair is greater when the choice alternative is two sources of protein. Importantly, this provided early evidence that the observed cultural tendency to pair protein with carbohydrate in a meal (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009) could be replicated in an online choice paradigm using two different sets of foods.

Table 6.2 *Percentage (%) Pair Type Selected by Trial Type and Stimuli Set*[1]

Trial type	C + C vs P +P				C + C vs Mixed				P + P vs Mixed			
	Set 1		Set 2		Set 1		Set 2		Set 1		Set 2	
Participant	C+C	P+P	C+C	P+P	C+C	M	C+C	M	P+P	M	P+P	M
1	88.9	11.1	5.6	94.4	35.2	64.8	20.4	79.6	3.8	96.2	77.8	22.2
2	55.6	44.4	66.7	33.3	16.7	83.3	16.7	83.3	14.8	85.2	5.6	94.4
3	38.9	61.1	88.9	11.1	38.9	61.1	81.5	18.5	61.1	38.9	31.5	68.5
4	47.2	52.8	77.8	22.2	50.0	50.0	77.8	22.2	42.6	57.4	27.8	72.2
5	83.3	16.7	75.0	25.0	35.2	64.8	38.9	61.1	13.0	87.0	1.8	98.2
6	77.8	22.2	77.8	22.2	64.8	35.2	63.0	37.0	29.6	70.4	31.5	68.5
7	55.6	44.4	58.3	41.7	22.2	77.8	35.2	64.8	15.8	84.2	7.4	92.6
8	80.6	19.4	61.1	38.9	33.3	66.7	42.6	57.4	7.4	92.6	33.3	66.7
Average across sample	66.0	34.0	63.9	36.1	37.1	62.9	47.0	53.0	23.4	76.6	27.1	72.9

[1]Regarding pair type, C+C represents a pair comprising only, P+P represents a pair comprising only protein, and M represents a mixed pair

6.3.2 Discussion

The results from this pilot provide initial evidence that participants prefer a mixed pair of macronutrients when presented with culturally familiar food pairs and asked to select a pair to eat for an imaginary lunch. Again, this pattern of results aligns with the tendency to mix protein and carbohydrate which has been observed in real-world meals from different cultures (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009).

One important concern regarding the six foods task pertains to variety as some trials involved ‘double-helpings’ of the same food (e.g., the pair comprised two servings of pasta). Variety is known to influence behaviour (Echelbarger et al., 2020; Hendriks-Hartensveld et al., 2022; Kahn, 1995; Rolls et al., 1981), and simply following the heuristic of selecting varied pairs when possible could have produced a pattern of results similar to the one observed in this pilot. Therefore, to account for any potential effect of food variety on pair choice, in the following pilot, any monotonous pairs in which the same food was presented twice (e.g., pasta paired with pasta) were removed.

As mentioned previously, the food pairs in the six foods task were culturally familiar, so it remained important to establish whether this preference for mixed pairs (i.e., a protein source paired with a carbohydrate source) existed in novel pairings which were less likely to be explained by Western cultural norms or cuisine (i.e., peanuts and crisps task; see section 6.2.4). If participants maintain a preference for mixed pairs when the pairs are culturally unfamiliar, then this suggests that participants might be discriminating the macronutrient composition of the pair as they would be unable to rely on cultural norms to guide their behaviour. This new task coined the peanuts and crisps task, is outlined in further detail in the following section.

6.4 Peanuts and crisps task- do humans still prefer a mixture of protein and carbohydrate when using novel pairs?

As previously stated, this version of the task aimed to explore whether the tendency to prefer mixed pairs of macronutrients emerges when using food pairs that might not traditionally go together. This was achieved by including a source of protein and a source of carbohydrate which would not typically be paired with the other foods based on normative Western culinary habits (stimuli from Set 1; see Table 6.1). Salted peanuts were introduced as a protein source and salted potato crisps were included as a carbohydrate source[13]. Critically, peanuts and crisps didn't need to be considered novel foods, but rather, it was important for the pairings (e.g., pasta (carbohydrate) paired with peanuts (protein)) to be novel as, again, the aim was to test whether the tendency to prefer combinations of macronutrients occurred in a pair of foods that could not be explained by cultural tendency.

Similar to the six foods task, the peanuts and crisps task involved participants completing a two-alternative forced choice task in which they were again asked to imagine that they were preparing a lunch box for work and that they should select the pair of foods that they would put in their lunchbox. Importantly, the peanuts or crisps pair was always presented as the left-hand pair, and peanuts and crisps were never included in a pair together. Additionally, all monotonous pairs (e.g., pasta paired with pasta) were removed, resulting in a total of 180 trials. Again, images of all the foods were taken in 100 g portions against a white background using a high-quality camera (Nikon D50) (see Figure 6.2). For the peanuts and crisps images, a watermark of the food's respective packaging was included in the lower right-hand corner of the image. Participants were shown the food pairs in random order on their tablets, laptops or desktop computers, and all data were collected using the experiment builder Gorilla

[13]Nutritional information for the salted peanuts and the salted potato crisps:
Salted peanuts – 614 kcal/100g, 51g/100g of fat, 5.6g/100g of carbohydrate, 30g/100g of protein
Salted potato crisps – 438 kcal/100g, 13g/100g of fat, 73g/100g of carbohydrate, 5.9g/100g of protein

(https://gorilla.sc/, Anwyl-Irvine et al., 2020) during one online session lasting approximately five minutes.

For the statistical analysis, the trials (*n*= 180) were classified into low-, equal-, and high-likelihood trials based on the likelihood of participants selecting the crisp or peanut pair presented on the left-hand side of the screen. The assignment of trial type is based on the assumption that participants would correctly identify crisps as being a source of carbohydrate and peanuts as a source of protein (see Table 6.3 for a description and example of each likelihood trial type).

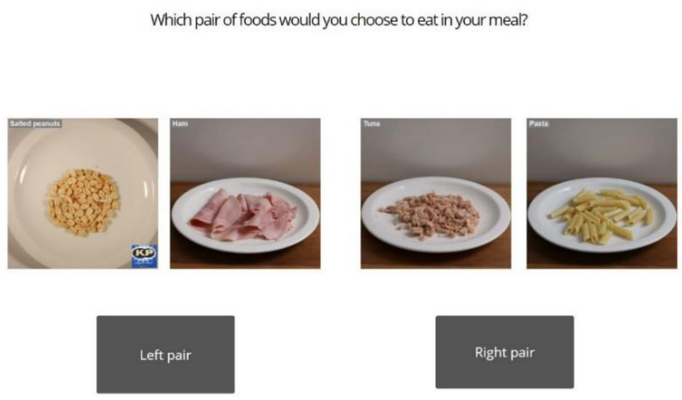
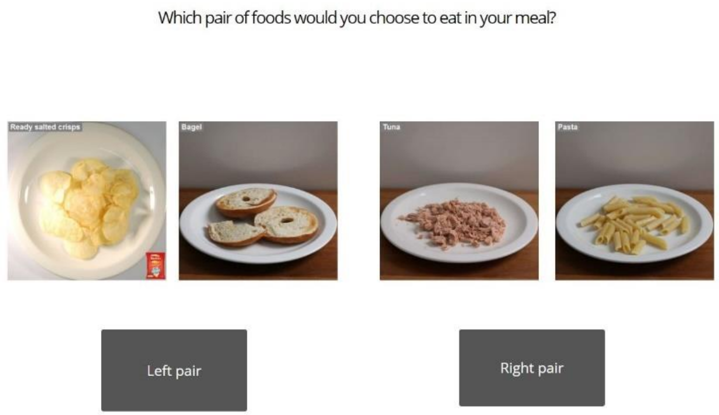
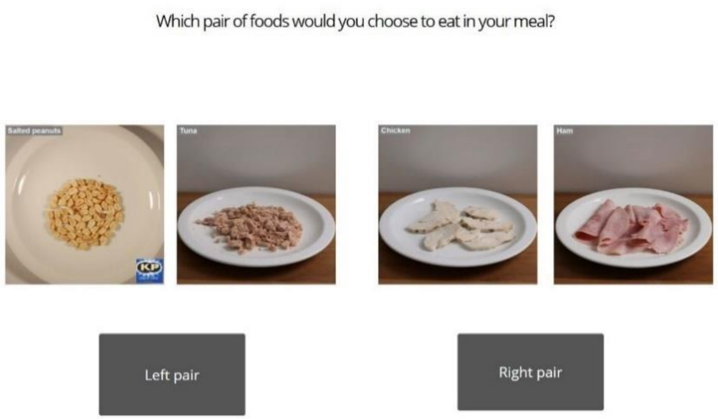
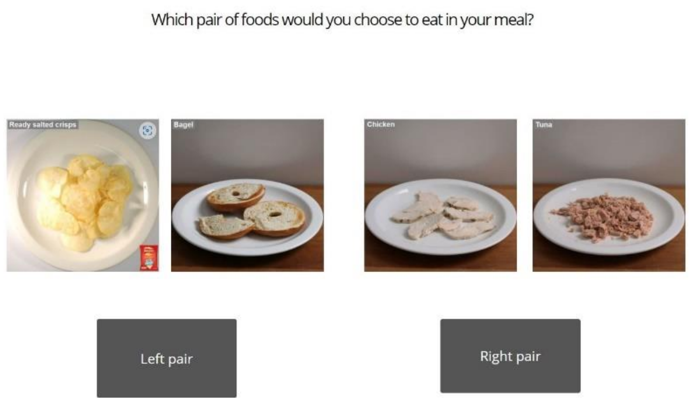
In a low-likelihood trial (*n*= 54), the crisp or peanut pair on the left has a lower likelihood of being selected as it contains two servings of the same macronutrient whereas the pair on the right comprises a mixed pair containing both protein and carbohydrate. For example, the left pair might be comprised of peanuts (protein) and ham (protein) and the right pair might contain pasta (carbohydrate) and tuna (protein). In this example, the peanut pair on the left would have a lower likelihood of being selected as, based on the results from the six foods task, participants should select the mixed pair on the right (see Table 6.3).

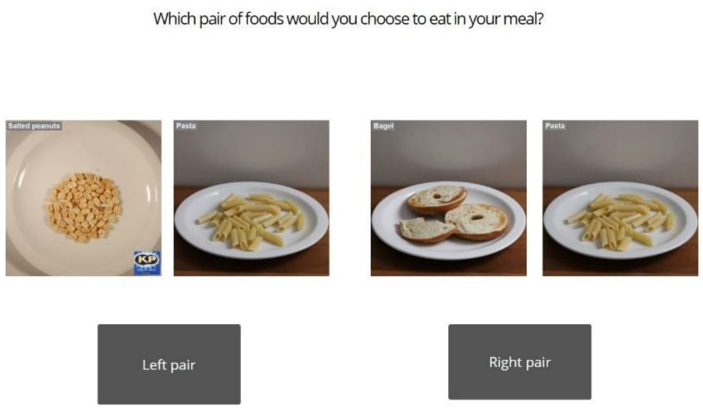
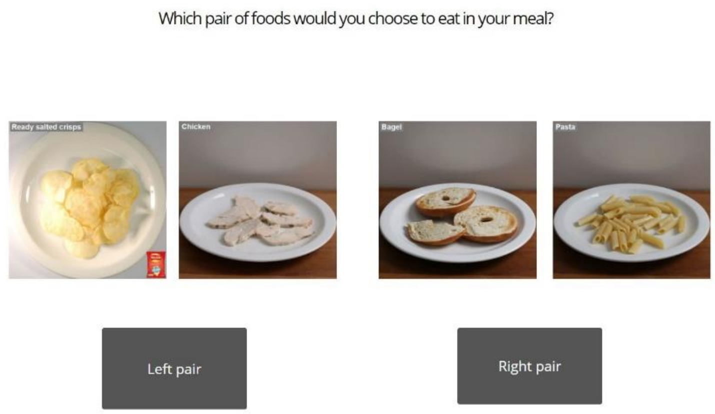
An equal-likelihood trial (*n*= 90) involves both the left and the right pair comprising either a mixed pair or the same macronutrients (e.g., both pairs contain only sources of protein). As such, the likelihood of the participant choosing one pair over the other based on its macronutrient composition is hypothesised to be equal (see Table 6.3).

Lastly, a high-likelihood trial (*n*= 36) includes a mixed crisps or peanut pair on the left while the right pair comprises two servings of the same macronutrient. In this scenario, the peanuts and pasta pair on the left (mixed macronutrients) would have a higher likelihood of being selected than the pair comprised of bagel and pasta (both carbohydrate foods) on the right (see Table 6.3).

The key outcome for this pilot was the frequency (%) with which the left pair containing either peanuts or crisps was selected as a function of trial type (i.e., low-, equal-, or high-likelihood).

Table 6.3 *Description and Example of Each Trial Type in the Peanuts and Crisps Task*

Trial type	Description	Example trials	
Low-likelihood	The crisp or peanut pair on the left contains two servings of the same macronutrient and the pair on the right is mixed (protein and carbohydrate).		
Equal-likelihood	Both pairs are either mixed or contain two servings of the same macronutrient.		

High-likelihood	The crisp or peanut pair on the left is mixed (protein and carbohydrate) and the pair on the right contains two servings of the same macronutrient.		

6.4.1 Results

Across the ten volunteers who were informally recruited from colleagues and friends, the peanuts and crisps pair (left pair) were selected less often than chance (50%) in the low-likelihood trials (21.85%, $t(9)= -3.60$, $p= .006$) (see Table 6.4). In the equal-likelihood trials, there was a trend for the peanuts and crisps to be selected less often than chance (36.56%, $t(9)= -2.04$, $p= .071$), and in the high-likelihood trials, the mean frequency of peanuts and crisps being selected was above chance (56.11%), but this did not meet the threshold for significance ($t(9)= 0.85$, $p= .416$). Across the three likelihood trial types, the difference in the frequency of crisps and peanuts being chosen was significant, $F(2, 27)= 5.68$, $p= .009$.

Table 6.4 *Frequency (%) Peanuts or Crisps Pair Selected by Trial Type*

Trial type	Mean frequency (%) peanuts or crisps pair being selected	Standard deviation
High-likelihood	56.11	22.67
Equal-likelihood	36.56	20.81
Low-likelihood	21.85	24.75

6.4.2 Discussion

The peanuts and crisps task explored whether it was possible to identify an underlying pattern of food choice based on the macronutrient composition of the foods rather than cultural norms. This was achieved by including novel food pairings which were unlikely to be explained by Western culturally normative behaviour or cuisine, such as combining peanuts, a high protein food, with pasta, a high carbohydrate food. Initial evidence from this pilot study suggests that, again, participants preferred mixed pairs over pairs containing only protein or only carbohydrate and that this occurred even when pairs were culturally unfamiliar (i.e., choice is unlikely to be explained by Western cultural norms). Additionally, it is unlikely that a preference for variety could explain the pattern of results as only varied pairs were presented (e.g., no ‘double-helpings’ of the same food).

The results from this pilot and the previous pilot (i.e., six foods task) suggest that the protein-carbohydrate pair paradigm (i.e., selecting between different combinations of protein and carbohydrate sources) can capture a pattern of responding which is consistent with real-world behaviours, even with culturally unfamiliar pairs. It should be noted that the sample sizes of both pilots were small (*n*= 8 and *n*= 10, respectively) and were recruited from colleagues and friends. The extent to which the two tasks might produce similar choice patterns in a larger sample is unclear. Therefore, the following section will discuss a third study which included both the six foods and the peanuts and crisps tasks and recruited a larger sample of participants via Prolific.

6.5 Testing the six foods task and peanuts and crisps in a larger sample

As mentioned, in this study, participants completed both the six foods task and the peanuts and crisps task. Importantly, in this version of the six foods task, unlike the first task iteration, only varied pairs were presented. Again, this was to minimise the potential impact of food variety on behaviour.

6.5.1 Methods

6.5.1.1 Participants

Thirty participants (Female *n*= 16*;* Male *n*= 14*;* age (years) *M*= 32.29 *SD*= 12.43) were recruited using the online data collection platform Prolific (https://www.prolific.co/, Prolific, 2014, Copyright Year: 2023). Participants were told that they would complete several computer-based tasks relating to food choice and that they would be asked to provide basic demographic information. Following the instructions from Prolific, the author used the site's built-in screening to only advertise the study to participants who met the study's inclusion criteria. Therefore, using the built-in screening, the study was only advertised to participants who 1) were not vegetarian or vegan (i.e., selected not following a diet when providing their participant details on Prolific), 2) were not eliminating certain foods from their diet for religious reasons or had a food allergy/intolerance (i.e., selected not following a diet when providing their participant details on Prolific), 3) had English as a first language, and 4) were over the age of 18 years (this is the default minimum age to open a Prolific account). Additionally, the study was advertised to only be completed using a tablet/laptop or a desktop computer.

6.5.1.2 Stimuli and study tasks

The same foods and food images from the first two pilots (sections 6.3 and 6.4) were used. Importantly, in these versions of the six foods and peanuts and crisps tasks, a two-second time lag was introduced between the display of the images and the presentation of the response options (i.e., left or right pair buttons). This time lag was included to encourage careful responding by the participant.

Alongside both the six foods and the peanuts and crisps tasks, participants completed three additional tasks: assessing each food as being mostly a source of protein or carbohydrate, evaluating their liking for each food, and rating their familiarity with each food. The first of the three tasks listed above required participants to evaluate whether they viewed each food as being mostly a source of carbohydrate or protein. For this task, a single image of one of the 14 food stimuli (six familiar high-carbohydrate foods, six familiar high-protein foods, peanuts, and crisps) was presented on the computer screen, and participants were instructed to use the 100-unit VAS to indicate whether they perceive the food to be mostly a source of carbohydrate or protein. The left anchor of the scale was labelled ‘Carbohydrate’ (0) and the right anchor was labelled ‘Protein’ (100), and above the image was the question ‘Is this food mostly a source of carbohydrate or protein?’. To assess food liking, participants were shown the following prompt, ‘How much do you like the taste of this food?’ and provided their response on a 100-unit VAS with the left anchor ‘Not at all’ (0) and the right anchor ‘Extremely’ (100). Participants’ familiarity with each of the 14 foods was assessed by asking them ‘Have you consumed this food before?' and providing the response options of 'Yes' and 'No'. In each of these three tasks, the order of stimuli presentation was randomised.

6.5.1.3 Procedure

All data were collected using the experiment builder Gorilla (https://gorilla.sc/, Anwyl-Irvine et al., 2020) during one online session lasting approximately 45 minutes. Participants provided informed consent and then completed the six foods task. Following this, they were shown a screen encouraging them to take a break and then continue when they feel refreshed and ready to begin another task. The break was included to encourage careful responding by participants. Participants then completed the peanuts and crisps task followed by another break before completing the final three tasks: assessing each food as being mostly a source of protein or carbohydrate, evaluating their liking for each food, and rating their familiarity with each food[14]. For both the food liking and food familiarity tasks, participants were shown an instruction page containing an image of an example trial using a food which was not part of the stimuli set. Lastly, participants selected the gender with which they most closely identified and their date of birth before receiving the debriefing information, providing their final consent, and being thanked for their assistance with the study. Upon completion of the study, participants received £3.75 (£5.00 per hour rate) as renumeration. The study received ethical approval from the University of Bristol Faculty of Science Human Research Ethics Committee (110823).

6.5.1.4 Statistical analysis

Statistical analyses for both the six foods task and the peanuts and crisps task followed the analyses described in the methods sections of the first two studies (see sections 6.3 and 6.4).

[14]Note, the familiarity and liking tasks were only used to describe the sample and are thus reported in Appendix 3 Table 11.7 and Table 11.8, respectively.

6.5.2 Results

Focussing first on the results of the six foods task, collapsing across both sets of foods, there was a tendency for participants to select a pair comprised of two sources of protein rather than one containing two sources of carbohydrate, 56.11% and 43.89% (see Table 6.5), and this pattern appeared to be stronger in the second set of six foods. Importantly, across both stimuli sets, participants tended to select mixed pairs containing both protein and carbohydrate over pairs containing only protein or only carbohydrate, and this was more likely to occur when the alternative was two sources of carbohydrate rather than two sources of protein (see Table 6.5).

Table 6.5 *Percentage (%) Pair Type Selected during the Six Foods Task*[1]

	Mixed vs C+C trial		Mixed vs P+P trial		C+C vs P+P trial	
Stimuli set	M	C+C	M	P+P	C+C	P+P
Set 1	63.46	36.54	62.10	37.90	50.00	50.00
Set 2	72.22	27.78	54.20	45.80	37.78	62.22
Both sets	67.84	32.16	58.15	41.85	43.89	56.11

[1]Regarding pair type, C+C represents a pair comprising only carbohydrate, P+P represents a pair comprising only protein and M represents a mixed pair

With regards to the results of the peanuts and crisps task, similar to the first iteration of the peanuts and crisps task (section 6.4), the frequency with which participants selected the peanuts or crisps pair presented on the left differed depending on whether the trial was a low-, equal- or high-likelihood trial. Briefly, low-likelihood trials occurred when the crisps or peanuts pair on the left had a low-likelihood of being selected as it contained two servings of the same macronutrient and the pair on the right comprised a mixed pair. An equal-likelihood trial occurred when both pairs comprised either mixed macronutrients or the same macronutrients and the likelihood of choosing the left pair over the right pair was equal, and a high-likelihood trial occurred when the peanuts or crisps pair on the left had a high-likelihood of being selected as the pair comprised both protein and carbohydrate (i.e., mixed pair) and the pair on the right comprised of two servings of the same macronutrient (see Table 6.3 for a description and example of each trial type).

Participants were more likely to select the peanuts and crisps pair in a high-likelihood trial, 50.10% (*SD*= 0.22), than in an equal- or low-likelihood trial, 38.37% (*SD*= 0.18) and 30.31% (*SD*= 0.18), respectively. The frequency with which participants selected the peanuts and crisps pair was significantly different depending on the likelihood type ($F(2, 87) = 8.121$, $p = .001$), and a post hoc Tukey's HSD test showed that the frequency of choosing the peanuts and crisps pair in high-likelihood trials differed from both equal- and low-likelihood trials ($p < .05$).

To explore why the tendency to select the peanuts and crisps pair in the high-likelihood trials was weaker and near chance levels in the current study (50.10%) compared to what was observed in the first version of the task (56.11%, see Table 6.4), the high-likelihood trials were separated into four different trial types (i.e., the four potential peanut/crisp and protein/carbohydrate combinations). The frequency (percentage of times) the peanuts or crisps pair was selected depending on trial type is shown in Table 6.6. The pattern of responding suggests that participants selected pairs where peanuts comprised a mixed pair (i.e., peanuts paired with a source of carbohydrate) less often than when crisps were included in a mixed pair (i.e., crisps paired with a source of protein).

Table 6.6 *Frequency (Percentage) a Pair was Chosen in the High-Likelihood Trials Separated by Trial Type*[1]

Trial type	Percentage (%) of trials left pair selected	Percentage (%) of trials right pair selected
Crisps+P vs. C+C	58.89	41.11
Crisps+P vs. P+P	63.33	36.67
Peanut+C vs. C+C	32.96	67.04
Peanut+C vs. P+P	45.19	54.81

[1]C stands for carbohydrate and P stands for protein. C+C represents a pair comprising only carbohydrate and P+P represents a pair comprising only protein.

One possibility for the pattern being weaker in trials with peanuts rather than crisps relates to the likelihood of the foods being identified as sources of protein and carbohydrate, respectively. As seen in Table 6.7, peanuts were less consistently viewed as being sources of protein compared to the other protein foods as peanuts had a mean perceived protein score closer to 50 (i.e., the mid-point of the 100-unit VAS) and a larger standard deviation. Crisps, on the other hand, were more consistently viewed as being a source of carbohydrate as they had a mean perceived carbohydrate score below 10, a smaller standard deviation, and a mean perceived carbohydrate score close to that of other carbohydrate foods such as bagels. Therefore, one possibility is that because peanuts were less consistently viewed as a source of protein, they were then potentially less likely to be selected as a mixed pair when presented alongside a carbohydrate food in the high-likelihood trials.

Table 6.7 *Mean Perceived Protein or Carbohydrate Value*[1]

Food item	Mean	Minimum	Maximum	Standard deviation
Bagel	10.23	0.0	50.0	14.60
Banana	32.87	0.0	90.0	27.29
Beef	92.47	50.0	100.0	13.74
Chicken	93.03	50.0	100.0	14.10
Chickpeas	60.03	12.0	100.0	24.71
Coleslaw	36.55	3.0	84.0	22.87
Crisps	9.97	0.0	50.0	13.99
Ham	85.93	22.0	100.0	19.44
Pasta	6.07	0.0	50.0	12.79
Peanuts	55.37	0.0	100.0	35.66

Potato Salad	19.63	0.0	65.0	16.72
Prawns	85.83	33.0	100.0	17.36
Tuna	90.43	31.0	100.0	17.23
Turkey	89.00	40.0	100.0	17.00

[1] This was assessed using a 100-unit VAS scale where the left anchor (0) was 'Mostly a source of carbohydrate' and the right anchor (100) was 'Mostly a source of protein'. Values below 50 indicate that the food is viewed as being mostly a source of carbohydrate and values above 50 indicate that the food is viewed as mostly a source of protein.

6.5.3 Discussion

The results of the six foods and the protein and crisps tasks in this third study were largely consistent with those observed in the first two studies (sections 6.3 and 6.4), 1) individuals were more likely to select a mixed pair than a pair containing two sources of the same macronutrient, and 2) participants were more likely to select the peanuts or crisps pair in the high-likelihood trials compared to the equal- or low-likelihood trials, albeit largely at chance level.

Within the six foods task, the consistent selection of a mixed pair across both stimuli sets (see Table 6.5) provides further evidence that the protein-carbohydrate pair paradigm produces a pattern of responding consistent with what is observed in real-world meals in different cultures (i.e., a tendency to combine sources of protein and carbohydrate, Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009). The results from the peanuts and crisps task indicate that the selection of mixed pairs of macronutrients does not rely on cultural norms guiding behaviour as participants preferred mixed pairs of macronutrients even when they were culturally unfamiliar. However, it should be noted that participants selected mixed pairs at largely chance level (50.10%), so these results should be interpreted with caution. It should also be acknowledged that the order of the six foods task and the peanuts and crisps task was fixed, and it is unknown whether participant responding was influenced by this. Lastly, a potential logical flaw in the peanuts and crisps task was noted and this is expanded upon in the next paragraph. In summary, while there was tentative evidence for macronutrient discrimination[15] using the protein-carbohydrate pair paradigm, neither the six foods task nor the peanuts and crisps task provide a measure of protein discrimination as they did not specifically measure behaviour in response to protein content. The adaptation of the protein-carbohydrate pair paradigm to assess protein discrimination is discussed in section 6.6.

On further examination of the peanuts and crisps task, a logical flaw in the task design was noted which potentially limits the conclusions which can be drawn. As mentioned in the methods (section 6.5.1), the peanuts or crisps pair was always presented alongside a culturally familiar pair (i.e., peanuts or crisps pairs were never presented alongside another peanuts or crisps pair). Because of this study design, it is not possible to dissociate whether the observed response pattern in the peanuts and crisps tasks is due to macronutrient discrimination of the peanuts and crisps pair or whether participants simply relied on the culturally familiar pair to guide their responses. For example, in a high-likelihood trial, the peanuts or crisps pair on the left comprised a mixed pair (i.e., protein with carbohydrate) whereas the culturally familiar pair on the right contained two servings of the same macronutrient. Therefore, if participants simply ignored the peanuts or crisps pair on the left and responded solely based on the culturally familiar pair, then they would have likely rejected the culturally familiar pair and selected the peanuts or crisps pair. In so doing, the resulting pattern in choice would suggest that participants were more likely to select the peanuts or crisps pair in high-likelihood trials. However, due to the argument presented above, the extent to which this indicates that participants exhibited macronutrient discrimination in the culturally unfamiliar pairs remains unclear. More generally, the two-alternative forced choice task design does not allow for the dissociation of 'selection' from 'rejection', that is, whether one pair is actively selected over

[15]As noted in the introduction (see section 6.2.4), macronutrient discrimination refers to behavioural responses (i.e., food choice, food preference, intake etc.) which can be predicted by the macronutrient composition of the food. Protein discrimination is operationalised as behavioural responses predicted by the protein content of the food.

the other or whether it is selected because the alternative pair was rejected, and this methodological concern is addressed in the following study.

6.6 Assessing protein discrimination by measuring the desire-to-eat different protein-carbohydrate pairs (protein-carbohydrate pair desire-to-eat task)

As previously described above, the two-alternative forced choice task design from the six foods and peanuts and crisps tasks did not allow for the separation of ‘selection’ from ‘rejection’ in the participant responses. An alternative approach is to have participants evaluate a single pair of foods comprising a source of protein and a source of carbohydrate. In this single pair ratings approach (henceforth referred to as ‘protein-carbohydrate pair desire-to-eat task’), participants were asked to rate their desire-to-eat[16] different pairs of protein and carbohydrate foods using a 100-unit VAS scale.

Importantly, the protein-carbohydrate pair desire-to-eat task was conceptualised to assess potential protein discrimination[15]. Briefly, building on the preference for a mixture of protein and carbohydrate as observed in the six foods and peanuts and crisps tasks (e.g., protein-carbohydrate pair paradigm) as well as in real-world meals (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009), if individuals can discriminate the protein content of foods, then one would predict that foods containing more protein will receive higher desire-to-eat ratings when paired with a source of carbohydrate than foods lower in protein. By way of an example, one might predict that a beef steak, containing ~25 g of protein per 100 g, would receive a higher desire-to-eat rating when paired with a source of carbohydrate than green peas

[16]Desire-to-eat ratings have been used extensively in appetite research[a], including in assessing macronutrient preference[b].

[a]Rogers, P. J., & Hardman, C. A. (2015). Food reward. What it is and how to measure it. Appetite, 90, 1-15. https://doi.org/10.1016/j.appet.2015.02.032
[b]de Bruijn, S. E. M., de Vries, Y. C., de Graaf, C., Boesveldt, S., & Jager, G. (2017). The reliability and validity of the Macronutrient and Taste Preference Ranking Task: A new method to measure food preferences. *Food Quality and Preference*, *57*, 32-40. https://doi.org/10.1016/j.foodqual.2016.11.003

which only have 4.9 g of protein per 100 g. It is proposed that the increased preference for higher protein foods when presented in a mixed pair alongside a source of carbohydrate might only be possible if individuals discriminate protein at some level. In summary, the protein-carbohydrate pair desire-to-eat task might help mitigate questions surrounding whether a choice is due to 'selection' or 'rejection' and, by using foods which vary in their total protein content, provides the opportunity to assess potential protein discrimination. The next sections will outline the study's methodology in further detail.

6.6.1 Methods

6.6.1.1 Participants

Participants (n= 108; female: n= 75; male: n= 33; $M \pm SE$, age= 25.62 ± 1.03 years; BMI= 23.11 ± 0.50 kg/m^2) were recruited by six different researchers via word of mouth and reaching out to friends and family. Due to the COVID-19 pandemic, all researcher interactions with participants were conducted online via Zoom. Exclusion criteria included: being under the age of 18, not being fluent or a native English speaker, or reporting having not eaten one or more of the test foods in the last year. Additionally, experimenters aimed to recruit roughly equal numbers of participants who reported being vegetarian or omnivore as a secondary aim of this study was to assess whether protein discrimination might differ based on dietary preference. Of the 108 total participants, 55 participants identified themselves as omnivores and 53 participants identified as vegetarians.

6.6.1.2 Stimuli

A total of 15 foods were used in the study: 12 vegetarian protein foods which varied in their protein content (g/100g) and three high carbohydrate foods (see Table 6.8 for nutritional information). The energy density (kcal/g) and protein content (g/100g) of the 12 protein-containing foods were largely decorrelated (r= 0.11, p= 0.74). Vegetarian protein foods were utilised for two reasons. Firstly, animal meat is a highly common protein source and is often viewed as an integral part of the diet (Piazza et al., 2015; Thavamani et al., 2020); therefore, it might be more likely to activate cultural normative responding (e.g., rating a pair of foods, such as steak and potatoes, higher because it aligns with a cultural norm) rather than responding based on protein content. Secondly, as mentioned in the chapter introduction, there is a need to increase the consumption of sustainable proteins, including plant-based proteins, to support planetary health. Additionally, related to the secondary aim described in the previous section, an inclusion requirement was that participants needed to have consumed all the test foods in the last year. Therefore, due to recruiting vegetarians and omnivores, products comprising animal meat could not have been used as stimuli due to the restrictions of the vegetarian diet.

Using a high-resolution digital camera (Nikon D50), overhead images of 200 g portions of each of the foods were taken against a uniform white background and under constant lighting conditions. Stimuli were photographed on the same white plate (255 mm in diameter), and, for those foods paler in colour (i.e., rice, tofu, cream cheese and houmous), Red Gem lettuce leaves were placed on the plate to provide contrast. The name of the food was inserted as a label in the upper left-hand corner of the image and photographs of the labels of commercially produced foods (i.e., houmous, cream cheese, baked beans, and 0% fat Greek yoghurt) were included as watermark inserts in the upper right-hand corner where necessary (see Figure 6.3 for example of a non-commercially produced stimuli). A reference image was created by photographing a standard credit card on the plate.

Table 6.8 *Macronutrient Composition and Energy Density of the 15 Stimuli Used in the Protein-Carbohydrate Pair Desire-to-Eat Task*

Food item	Source of protein or carbohydrate	Kcal per 100g	Protein (g/100g)	Carbohydrate (g/100g)	Fat (g/100g)	Percentage (%) total kcal from protein
Baked beans	Protein	84.0	4.6	13.0	0.5	21.9
Chickpeas	Protein	122.0	7.7	16.5	1.4	25.2
Cream cheese	Protein	225.0	5.4	4.3	21.0	26.3
Edamame	Protein	201.0	13.4	2.0	13.6	26.7
Egg	Protein	152.0	14.6	0.0	10.4	38.4
Greek yoghurt	Protein	54.0	10.3	3.0	0.0	76.3
Houmous	Protein	185.0	7.3	29.0	15.8	15.8
Kidney beans	Protein	105.0	8.1	12.8	0.6	30.9
Lentils	Protein	143.0	10.6	18.1	1.7	29.7
Peas	Protein	68.0	4.9	7.5	0.7	28.8
Quinoa	Protein	185.0	6.1	29.0	4.2	13.2
Tofu	Protein	118.0	12.6	1.0	7.1	42.7
Baby potatoes	Carbohydrate	66.0	1.8	13.5	0.5	10.9
Pasta	Carbohydrate	151.0	6.3	26.9	1.6	16.7
Rice	Carbohydrate	145.0	3.1	29.0	1.6	8.6

Figure 6.3 Example of how stimuli were presented to participants during the tasks.

6.6.1.3 Tasks and measures

Participants completed five different tasks which are outlined below, and they also provided appetite ratings (hunger, fullness, and thirst) on 100-unit VAS scales anchored at ‘Not at all’ (0) on the left and ‘Extremely’ (100) on the right (Rogers & Hardman, 2015).

The protein-carbohydrate pair desire-to-eat task involved participants rating their desire to eat each pair of foods in response to the prompt “How strong is your desire to eat, that is, to taste, chew and swallow, these two foods RIGHT NOW?”. The 100-unit VAS scale was anchored with ‘Not at all’ (0) on the left and ‘Extremely’ (100) on the right (Rogers & Hardman, 2015). Participants were first presented with an instruction page outlining the task, explaining that they were to imagine that they are hungry and are going to eat a meal. The meal would have two courses, but, importantly, they would not consume the two foods together. The instructions emphasized that only those foods and portions would be available to the participant, and that, in their imaginary meal, they would be expected to consume the entire portion. They were then shown an example trial which used a credit card as a reference for the size of the plate. Once beginning the actual task, one of the 36 pairs was randomly presented on a computer screen. This was then followed by a two-second pause before the participants could use the slider to provide their response. This process was completed for every pair and the pause was included to encourage careful responding.

The second desire-to-eat task involved participants rating their desire-to-eat each of the 15 foods on their own. Participants were randomly presented with one of the 15 foods and, again, there was a two-second pause between the image being displayed and the rating scale appearing. At the end of the 15 trials, participants completed an attention check question asking them to recall whether the instructions for the protein-carbohydrate pair desire-to-eat task involved them imagining a scenario where they would eat all of the food presented or only as much as they wanted. Participants who responded incorrectly and selected ‘Only as much as I wanted’ were excluded from the data analysis.

To establish whether the foods were viewed by participants as being mostly sources of carbohydrate or protein, participants completed a task identical to one included in the third pilot (see section 6.5.1.2). Briefly, participants were randomly presented with one of the 15 stimuli and, using a 100-unit VAS, were asked to make a rating based on the following prompt “Is this food mostly a source of carbohydrate or protein?”. The anchor on the left side of the sliding scale was ‘Carbohydrate’ (0) and the anchor on the right side of the scale was ‘Protein’ (100). In this task, a food rated as less than 50 (scale mid-point) was viewed as being mostly a source of carbohydrate, whereas a food with a score greater than 50 was considered mostly a source of protein.

Lastly, participants completed a food frequency questionnaire to establish, on average, how often during the last year they consumed the 15 foods used in the study. This task used the question structure of The European Prospective Investigation into Cancer and Nutrition (EPIC)-Norfolk food frequency questionnaire (Bingham et al., 2007; Mulligan et al., 2014), and response options included “Never”, “Less than once per month”, “1-3 times per month”, “Once a week”, “2-4 times per week”, “5-6 times per day”, “Once a day” “2-3 times per day”, “4-5 times per day”, “6+ times per day”.

6.6.1.4 Study procedure

Before beginning the study, participants completed a brief Zoom call with one of the six researchers responsible for data collection. During this call, a general overview of the study was provided, and researchers emphasised key instructions for the main task, specifically that only the pair of foods presented would be available in their meal and they wouldn’t be on the same plate. Additionally, it was made clear that when making the rating, participants should imagine eating all of the food. Participants also completed three example trials and then had the opportunity to ask questions before being assigned a participant number and beginning the study.

The entire study lasted approximately 15 minutes and participants were shown an information sheet before providing online informed consent. The tasks occurred in the following order 1) appetite ratings, 2) protein-carbohydrate pair desire-to-eat task, 3) individual desire-to-eat task, 4) evaluation of food as mostly a source of protein or carbohydrate, 5) food frequency questionnaire, and 6) demographic questions. The first questions of the demographic questionnaire centred around general participant information. Participant gender was ascertained with the question "What gender do you most closely associate with?" and included the response options male, female, prefer not to say and prefer to self-describe. Information on participants' age was established by having them provide their date of birth. Height and weight (used to calculate BMI), years in education past the age of sixteen, current dieting status, and frequency of dieting to lose weight in the last twelve months were also self-reported by the participant. Lastly, participants were asked about their dietary patterns, including whether they were currently following a vegetarian diet, and if so, for how long they had been following the diet. Participants were also asked if they had previously followed a vegetarian diet and, if so, how long they had previously been vegetarian.

After completing the demographic questions, participants were then debriefed and thanked for their assistance. After completion, responses were screened to ensure that participants met the inclusion criteria, specifically reporting having eaten each food, and passed the attention check question. If they failed to meet either of these criteria, then their data were excluded and an additional participant was recruited, if possible. Ethical approval was granted by the University of Bristol Science Faculty Ethics Committee (117114).

6.6.1.5 Statistical analyses

As each of the 12 protein foods was presented as a pair alongside three different high-carbohydrate foods (i.e., three protein-carbohydrate pairs per protein food resulting in 36 total pairs), a mean desire-to-eat score was calculated for each of the 12 protein foods for each participant. To assess evidence for protein discrimination (in this study, protein content predicting desire-to-eat), two linear regressions were conducted within each participant. In the first regression, protein content (g/100g) was entered as the only variable predicting mean desire-to-eat. In the second regression, the effects of the protein foods' nutritional composition (energy density (kcal/100g), carbohydrate (g/100g) content and fat (g/100g) content) as well as the desire-to-eat the protein food by itself were controlled for by simultaneously entering them in the regression alongside protein content (g/100g).

Individual unstandardised beta coefficients were then extracted for each participant and were used in two different analyses. First, a one-sample *t*-test was run to establish whether the mean of the individual beta weights was different from zero as this would indicate whether, across the entire sample, protein content (g/100g) predicted desire-to-eat when protein sources were paired with sources of carbohydrate. If the mean beta weight was significantly different from zero, then this would provide evidence for protein discrimination. In this analysis, a positive mean beta weight would indicate that foods containing more protein received higher desire-to-eat scores whereas a negative value would indicate that foods with lower protein content received higher desire-to-eat scores. Second, a secondary aim was to establish if vegetarians or omnivores differed in their protein discrimination ability, and this was achieved using independent samples *t*-tests to determine whether the mean beta weights differed between the dietary groups, and a one-sample *t*-test was then run in each group to establish if, within the group, there was evidence for protein discrimination (i.e., beta weight significantly different from zero). All analyses were conducted in the R statistical environment (R Core Team, 2022) with several helper packages (Kassambara, 2020; Wickham et al., 2019)

6.6.2 Results

Descriptive statistics for the individual desire-to-eat ratings for each of the 12 protein foods can be seen in Table 6.9. Differences in the frequency of consumption for each of the 12 protein foods can be found in Appendix 3 Table 11.9.

Table 6.9 *Desire-to-Eat Ratings for Each of the 12 Protein Foods Separated by Diet Type*[1]

Food item	Diet type	Mean	Standard deviation
Baked beans			
	Omnivore	46.93	35.21
	Vegetarian	60.64	28.46
Chickpeas			
	Omnivore	45.16	25.98
	Vegetarian	42.43	27.02
Cream cheese			
	Omnivore	46.53	29.14
	Vegetarian	42.57	28.75
Edamame			
	Omnivore	61.65	30.00
	Vegetarian	66.92	27.01
Egg			
	Omnivore	55.40	29.89
	Vegetarian	41.51	32.39
Greek yoghurt			
	Omnivore	54.04	31.11
	Vegetarian	57.25	30.25
Houmous			
	Omnivore	61.91	31.61
	Vegetarian	72.08	21.66
Kidney beans			
	Omnivore	43.15	28.75
	Vegetarian	41.64	25.84
Lentils			
	Omnivore	38.71	26.56
	Vegetarian	36.53	25.64
Peas			
	Omnivore	49.58	29.14
	Vegetarian	55.74	27.93
Quinoa			
	Omnivore	43.65	28.35
	Vegetarian	43.70	25.79
Tofu			

	Omnivore	34.04	29.26
	Vegetarian	44.36	27.41

[1]The desire-to-eat ratings reported in this table are of the protein foods presented by themselves, not in a protein-carbohydrate pair.

When protein content (g/100g) was entered as the only predictor of mean desire-to-eat,[17] there was no association between the two variables (M_B= -0.14, SD= 1.50, $t(107)$= -1.00, p= .34). However, after controlling for the nutritional composition of the protein food (energy density (kcal/100g) as well as carbohydrate and fat content (g/100g)) and desire-to-eat the protein food by itself (i.e., individual desire-to-eat[17]), there was a significant negative association between protein content (g/100g) and mean desire-to-eat (M_B= -0.41, SD= 1.51, $t(107)$= -2.79, $p < .05$). In other words, across the entire sample, when paired with a source of carbohydrate, foods containing more protein received, on average, lower desire-to-eat scores.

The potential between dietary group (i.e., omnivore versus vegetarian) differences in the association between protein content (g/100g) and mean desire-to-eat[17] were also explored. In the first regression, in which protein content was the sole predictor of desire-to-eat, whilst there were no significant between dietary group differences ($t(106)$= 1.71, p= .09), there was a trend for protein content to be negatively associated with desire-to-eat in vegetarians (M_B= -0.39, SD= 1.43; $t(52)$= -1.98, p= .05) while having no significant association in omnivores (M_B= 0.10, SD= 1.54; $t(54)$= 0.48, p= .63). After controlling for the protein foods' nutritional composition and individual desire-to-eat, there were again no significant between-group differences ($t(106)$= -0.24, p= .81). Within each dietary group, protein content (g/100g) negatively associated with desire-to-eat, however, this association only reached significance in

[17]As a reminder, 'mean desire-to-eat' is the average desire-to-eat that protein food when it is presented alongside a source of carbohydrate.
When referring to the desire-to-eat the protein food by itself, this variable is called 'individual desire-to-eat'.

omnivores (M_B= -0.42, SD= 1.47, $t(54)$= -2.21, p= .03) and not vegetarians (M_B= -0.39, SD= 1.57, $t(52)$= -1.73, p= 0.09).

6.6.3 Interim summary: protein content (g/100g) appears to negatively predict desire-to-eat

In summary, when entered as the only predictor, across the entire sample, protein content (g/100g) failed to predict desire-to-eat. However, after controlling for the nutritional composition of the protein food as well as individual desire-to-eat, protein content (g/100g) was negatively associated with desire-to-eat, providing initial evidence for protein discrimination using the protein-carbohydrate pair paradigm. There were no dietary group differences in the association between protein content and desire-to-eat, indicating that omnivores and vegetarians responded similarly. The negative association between protein content and desire-to-eat is curious, as the underlying behavioural tendency in the protein-carbohydrate pair paradigm (i.e., preference for a mixture of protein and carbohydrate) appears to be relatively robust based on responding in the three studies presented earlier in the chapter and real-world meals (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009).

One potential explanation for the negative association between protein content and desire-to-eat pertains to the specific stimuli used in the study. Alongside the foods included in the stimuli set, meat analogues (i.e., alternative protein sources which are designed to imitate meat (Hoek et al., 2011)) are consumed by both omnivores and vegetarians, but to a greater extent by vegetarians and vegans (Gehring et al., 2021). The stimuli utilised in this study explicitly excluded meat analogues as these are often marketed with nutrition claims related to protein, most often indicating a high protein content (Lacy-Nichols et al., 2021). It is unclear whether including meat analogues might have changed the pattern of responding, and this remains an important question to explore in future research, especially given the increasing popularity of these products

In relation to the above point regarding participants' possible intake of meat analogues, another potential influencing variable could be the duration of vegetarianism. It is possible that 'newer' (i.e., more recently adopting the diet) and 'older' vegetarians (i.e., those who are more established in the diet) differ regarding both the types of protein they consume and their potential protein discrimination ability. With regards to the types of protein foods consumed, more established or 'older' vegetarians might more frequently consume whole, plant-based protein-containing foods whereas 'newer' vegetarians might rely more heavily on meat analogues as sources of protein. This is supported by evidence suggesting that individuals who more recently adopted a vegetarian or vegan diet were more likely to consume ultra-processed foods, such as meat analogues, than those who had followed the diet for a longer period (Gehring et al., 2021). On this basis, a possible explanation for the negative association between protein content (g/100g) and desire-to-eat observed in vegetarians is that newer vegetarians who were less familiar with the stimuli, specifically the plant-based protein foods, could have overshadowed any evidence of a positive association between protein content (g/100g) and desire-to-eat in older vegetarians. The following section outlines post-hoc analyses which further explore the potential association between protein content (g/100g) and desire-to-eat and addresses some of the questions raised above.

6.6.4 Post-hoc analyses using plant-based protein foods

To explore whether the pattern of responding might differ based on the duration of following a vegetarian diet, the participants identifying as vegetarian were separated into two groups based on the median duration of vegetarianism: newer vegetarians (vegetarian for less than two and a half years (30 months), n= 23) and older vegetarians (vegetarian for more than two and half years (30 months), n=30). Additionally, the analyses were repeated using only plant-based protein foods to explore whether discrimination of protein content (g/100g) differed in these foods which are key protein sources in vegetarian and vegan diets (Alcorta et al., 2021; Bradbury et al., 2017; Papier et al., 2019). Additionally, these foods align with the dietary recommendations of the Inter-governmental Panel on Climate Change (IPCC) (Mbow et al., 2019). On review of the stimuli, baked beans were excluded as, compared to the other plant-based protein foods, they can be eaten by themselves, are highly familiar to both omnivores and vegetarians and, in the UK, can be considered a cultural food on their own. Additionally, hummus was also excluded as it is a semi-solid spread, unlike the other stimuli which were solid food items, and perhaps inherently more likely to be eaten in combination with a carbohydrate (e.g., as a topping on bread or a dip for crackers). The statistical analyses outlined in section 6.6.1.5 were repeated, and a one-way analysis of variance (ANOVA) was used to determine whether omnivores, newer vegetarians and older vegetarians differed in the ability to discriminate food protein content (g/100g). The final stimuli in these exploratory analyses included: chickpeas, lentils, kidney beans, tofu, quinoa, edamame, and peas.

When repeating the analyses in only the plant-based protein foods, protein content (g/100g) failed to predict mean desire-to-eat when entered as the only predictor (M_B= 0.31, SD= 1.97, $t(107)$= 1.61, p= .11), and omnivores, newer vegetarians and older vegetarians did not significantly differ in the extent to which protein content predicted mean desire-to-eat ($F(2, 105)$= 2.02, p= .14). Out of the three dietary groups, protein content only significantly predicted desire-to-eat in the older vegetarians (see Table 6.10), with foods containing more protein receiving, on average, higher desire-to-eat scores.

Table 6.10 *Protein Content (g/100g) as Sole Predictor of Mean Desire-to-Eat in Plant-Based Protein Foods*[1]

Diet type	M_B	Standard deviation	One-sample *t*-test value	*p*-value
Omnivore	-0.05	1.87	-0.21	.84
Newer vegetarian (< 2.5 years)	0.52	2.13	1.17	.25
Older vegetarian (> 2.5 years)	0.80	1.95	2.24	.03

[1]The plant-based protein foods are chickpeas, lentils, kidney beans, tofu, quinoa, edamame, and peas.

After controlling for the nutritional composition of the protein food and individual desire-to-eat, across the entire sample, protein content (g/100g) remained a non-significant predictor of desire-to-eat (M_B= -1.04, *SD*= 7.01, *t*(107)= -1.03, *p*= .13). The results of a one-way ANOVA suggested that omnivores, newer vegetarians, and older vegetarians significantly differed in the extent to which protein content (g/100g) predicted desire-to-eat (*F*(2, 105) = 4.82, *p*= .01). As shown in Table 6.11, protein content (g/100g) significantly negatively predicted desire-to-eat in omnivores. In both newer and older vegetarians, there was no significant association between protein content (g/100g) and desire-to-eat, but the direction of the association was positive

Table 6.11 *Protein Content (g/100g) Predicting Mean Desire-to-Eat in Plant-Based Protein Foods*[1,2]

Diet type	M_B	Standard deviation	One-sample *t*-test value	*p*-value
Omnivore	-3.02	7.00	-3.20	.002
Newer vegetarian (< 2.5 years)	1.04	7.39	0.68	.51
Older vegetarian (> 2.5 years)	1.02	5.77	0.97	.34

[1]Beta weight after controlling for the nutritional composition of the protein food (energy density (kcal/100g) as well as carbohydrate and fat content (g/100g)) and desire-to-eat the protein food by itself (i.e., individual desire-to-eat).
[2]The plant-based protein foods are chickpeas, lentils, kidney beans, tofu, quinoa, edamame, and peas.

6.7 Discussion

There was no evidence suggesting that the association between desire-to-eat and protein content (g/100g) differed when completing the analyses in only plant-based protein foods as compared to the entire stimuli set of vegetarian protein foods (see section 6.6.2). In both cases, after controlling for the nutritional composition of the protein food (energy density (kcal/100g) as well as carbohydrate and fat content (g/100g)) and desire-to-eat the protein food by itself, protein content (g/100g) negatively associated with desire-to-eat.

However, in the exploratory analyses, there was initial evidence in both newer and older vegetarians suggesting that plant-based foods containing more protein were more desired, and this is consistent with the results from the six foods and peanuts and crisps tasks as well as the pattern observed in real-world meals (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009). In omnivores, however, the negative association between desire-to-eat and protein content (g/100g) remained, regardless of whether the analysis was conducted in only plant-based protein foods or the entire stimuli set. On the whole, this pattern of responding aligns more generally with research suggesting that individuals less familiar with a vegetarian diet (i.e., omnivores or those newly transitioning to a vegetarian diet) may choose plant foods which contain less protein than those who are more familiar with a traditional plant-based diet (i.e., older vegetarians) (Mariotti & Gardner, 2019). It is imperative to note, however, that the analysis using only plant-based protein foods was highly exploratory. Therefore, caution should be used when interpreting the findings, and further research is needed to draw more substantive conclusions.

6.7.1 Placing evidence for macronutrient discrimination and protein discrimination in the broader research landscape

In summary, both the six foods and peanuts and crisps tasks from the first part of the chapter provided evidence for macronutrient discrimination as the frequency of a pair being selected differed based on its macronutrient composition. The preference for a mixture of macronutrients (i.e., protein paired with a source of carbohydrate) observed in these tasks, alongside the evidence from real-world meals (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009), provided support for the protein-carbohydrate pair paradigm which was the foundation for the protein-carbohydrate pair desire-to-eat task which generated evidence suggesting a degree of protein discrimination.

As mentioned in the introduction to this chapter (see section 6.2.3), non-human animals display relatively consistent protein discrimination (responding to both acute protein deprivation and changes in physiological state) as well as potentially demonstrating learned (Baker et al., 1987; Booth, 1974; Gibson & Booth, 1985; Simpson & White, 1990) and unlearned (Deutsch et al., 1989) protein appetites. There is a smaller body of research exploring whether humans respond to the protein content of foods (for example: Buckley et al., 2019; Gibson et al., 1995; Griffioen-Roose et al., 2012). Of the research conducted, it appears that humans do have the capacity to respond to food protein content, with individuals demonstrating an increased preference or valuation of foods containing more protein (Buckley et al., 2019; Gibson et al., 1995; Griffioen-Roose et al., 2012). The protein discrimination evidenced in this chapter would be consistent with the previously mentioned work suggesting human's ability to respond to protein content. However, the negative association between protein content and desire-to-eat is somewhat inconsistent with the findings in humans mentioned above as these studies report positive associations between protein content and their chosen outcome.

Additionally, some human studies suggest that physiological individual differences might impact responding to protein content (Buckley et al., 2019; Murphy & Withee, 1987). While physiological individual differences were not explored in this chapter, the duration of following a vegetarian diet was identified as a potential individual difference which might have influenced protein discrimination in the exploratory analyses. Establishing whether this is a robust finding and identifying other potential individual differences remains a task for future studies.

More generally, the extent to which humans discriminate protein in everyday foods is linked to the idea of nutritional intelligence, an idea mentioned briefly in chapter five. Human 'nutritional intelligence' is a concept which captures humans' ability to differentiate foods based on their nutritional composition and make advantageous decisions on this basis

(Brunstrom et al., 2023). It also argues that cuisine plays a functional role and reflects an accumulation of learning by individuals in a community which is then shared via social transmission (Brunstrom et al., 2023). Therefore, it is plausible that there is less pressure for humans to exhibit protein discrimination on an individual level because our culinary practices (i.e., cuisine) protect us from experiencing a state of protein depletion. As highlighted in the introduction and throughout this chapter, a variety of cultures have culinary norms which tend to pair sources of protein with carbohydrate sources, and this also includes largely vegetarian cuisines (i.e., legumes as a source of protein rather than animal protein) (Charles & Kerr, 1986; Deliza & Casotti, 2009; Foley, 2005; Sen, 2009). Thus, if an individual simply ate the meals associated with their cuisine, then they would likely meet their protein requirements, and the need to exhibit robust protein discrimination on an individual level would be lessened. Relatedly, the apparent lack of cuisine in non-human animals might in part explain why evidence for protein discrimination is stronger in this group as they are more likely to rely on individual learning. Therefore, they might be more sensitive to the macronutrient composition, including protein content, of a stimulus and acquire flavour-nutrient associations more readily (Brunstrom et al., 2023).

6.7.2 The importance of dietary transitions and the role of plant-based protein foods

The exploratory findings from the protein-carbohydrate pair desire-to-eat task suggest that the transition period when adopting a vegetarian or vegan diet could be important, especially regarding recognising foods which are now considered protein sources. More generally, this dietary transition period might be a key target for interventions which aim to support adopting sustainable diets. For example, one could explore whether it is possible to enhance learning about whole, plant-based foods, such as legumes or tofu, as protein sources. This is especially important considering that omnivores transitioning to a vegetarian diet might less readily view these foods as protein sources, especially compared to meat analogues which are often marketed as being high protein (Boukid, 2020). Relatedly, the transition period when adopting a vegetarian or vegan diet might also be an important target for consumer research and the development of meat analogues as it is plausible that newer versus older vegetarians might differ in their preference for these products. A related example suggests that highly realistic meat analogues (i.e., Beyond Meat burgers which 'bleed') were negatively viewed by vegetarians and vegans but this realism was positively valued by omnivores (Kerslake et al., 2022). The studies presented in this chapter are only initial forays into this area, and this space presents significant opportunities for future research.

6.7.3 Limitations and possibilities for future research

It should be noted that all of the studies presented in this chapter were conducted in an online setting where there was limited control over participants' engagement. Additionally, despite the six foods and peanuts and crisps tasks involving what were considered 'everyday dietary decisions' (i.e., choosing between two pairs of foods to put in a lunchbox), one could argue that these tasks still placed participants in an artificial decision-making environment. Firstly, with regard to participant engagement, several steps were taken to ensure careful responding. In the third study in which the six foods and peanuts and crisps tasks were tested in a larger sample, a two-second time lag was introduced between the presentation of the images and the appearance of the choice buttons. Additionally, between the two tasks, a screen was shown which encouraged participants to take a break (M_{break}= 59.75 seconds, SD_{break}= 5.47 minutes) and then continue the study when they felt refreshed. In the protein-carbohydrate pair desire-to-eat task, there was a two-second pause between the display of the images and when the participant could move the slider to provide their response, and an attention check question was also included with participants failing the question being excluded. Secondly, concerning the artificial decision-making environment, a similar approach was used in a study exploring whether humans are sensitive to the micronutrient content of foods by asking participants to select which pair of foods they would eat in their meal (Brunstrom & Schatzker, 2022). As with the six foods and peanuts and crisps tasks in this chapter, the micronutrient study produced non-random data (i.e., better than chance) (Brunstrom & Schatzker, 2022) suggesting asking participants to select a pair of foods for a hypothetical meal produces reliable, non-random data.

Concerning opportunities for future research, the protein-carbohydrate pair desire-to-eat task could also be used for conditioning studies where the key outcome is change over time rather than an acute measure of protein discrimination (as in this chapter). For example, one could develop two versions of a novel food containing different amounts of protein. After a baseline test using the protein-carbohydrate pair desire-to-eat task, participants would be instructed to consume the food for a set period and then complete the task again. If participants discriminated that the food contained protein, then the expectation is that the desire-to-eat rating should increase when the food is paired with a source of carbohydrate. A study similar to this is currently being conducted by Davidenko and colleagues, and the protein-carbohydrate pair desire-to-eat task is included alongside a variety of other behavioural measures. Separately, one could also combine both individual and social learning in a single study whereby participants complete a conditioning study which comprises three conditions: low-protein, high-protein individual and high-protein group. The only difference between the two high-protein conditions is that the group condition would receive a form of peer feedback suggesting that the novel food contains a high amount of protein. The outcome of interest would be whether the group condition exhibits increased protein discrimination compared to the group which relies solely on individual learning and whether both of the high protein groups perform better than the low protein group. A study with this design could begin to unpack the role of community-supported learning and nutritional intelligence in relation to food macronutrient composition.

6.7.4 Chapter summary

In summary, a protein-carbohydrate pair paradigm was established (six foods and peanuts and crisps tasks) which was then used to develop an online approach to assess protein discrimination in humans (protein-carbohydrate pair desire-to-eat task). After controlling for the nutritional composition of the protein food and individual desire-to-eat, across the entire set of vegetarian stimuli, there was initial evidence for protein discrimination as protein content was negatively associated with desire-to-eat. Put differently, foods containing greater amounts of protein were less desired when paired with a carbohydrate than foods containing less protein, which is opposite to what was anticipated. One possibility for the negative association between protein content and desire-to-eat is that the protein-rich foods might have been more filling (i.e., delivered more satiation (Paddon-Jones et al., 2008)) or had an aversive sensory quality (as in, for example, the 'glueyness' of casein (Booth, 1985)) relative to the lower protein-containing foods. These characteristics could have influenced participants' responses, and future research should consider assessing these characteristics to include in the analyses. In the exploratory analyses, when measuring protein discrimination in only plant-based protein foods and separating vegetarians into newer and older vegetarians, there was tentative evidence for a positive association between protein content and desire-to-eat in both newer and older vegetarians. One possible explanation for this positive association pertains to familiarity or experience with the vegetarian diet as older vegetarians are likely to have acquired greater knowledge about these protein foods as a by-product of having interacted with the community and its associated cuisine for a longer period of time. The importance of dietary transitions, study limitations, and potential follow-on studies exploring learning about protein content were also discussed. The next chapter will address the remaining two macronutrients, fat and carbohydrate, in combination.

Chapter 7 Fat and carbohydrate: Single foods comprising a combination of fat and carbohydrate are selected in larger portions than foods high in either fat or carbohydrate- is this driven by an effect on satiation?

7.1 Acknowledgements and overview

For the studies presented in this chapter, the author was responsible for study design, task and stimuli development, coding the tasks, data analysis, and interpretation of results. This research was supervised by Professor Jeff Brunstrom and Emeritus Professor Peter Rogers, and they provided feedback on the study design, data analysis plan, and interpretation of findings. A Cardiff University placement student (Stan Mellstrom) assisted in taking portion-size images of the UK stimuli and provided feedback on study findings. Portions of this chapter have been presented as an oral presentation (Flynn, Mellstrom, Rogers & Brunstrom) at the 2023 Benjamin Franklin Lafayette Seminar (Fréjus, France) and as a poster presentation (Flynn, Mellstrom, Rogers & Brunstrom) at the 2023 annual meeting of the Society for the Study of Ingestive Behavior (SSIB) in Portland, Oregon USA.

The UK stimuli set was previously used in a third-year undergraduate project exploring the effect of macronutrient composition (fat and carbohydrate), energy density, and degree of processing on food reward (Rogers et al., 2024). The author would like to thank Perszyk and colleagues for providing the American stimuli set used in their 2021 study.

Single foods containing both fat and carbohydrate in roughly equal amounts (i.e., combination foods) are more rewarding than foods high in either fat or carbohydrate (DiFeliceantonio et al., 2018; Perszyk et al., 2021). Importantly, it remained unclear whether the amount of fat and carbohydrate in a food also influenced portion selection, and, if so, whether the differences in portion selection were driven by differences in reward or satiation. Understanding whether combining fat and carbohydrate in a food promotes the selection of large portions may provide opportunities for food reformulation which aims to support population-level healthy weight maintenance.

To explore the above ideas, first, two pilot studies assessed whether expected satiation differed for foods varying in fat and carbohydrate; in the first pilot, American snack foods from Perszyk et al. (2021) were used, and in the second pilot, the opportunity arose to recruit a larger sample (*n*= 30) and to test UK stimuli from Rogers et al. (2024). Lastly, a third study, using the UK stimuli set, built on this pilot work and, importantly, included ideal and maximum portion selection tasks alongside a measure of liking. The chapter begins by reviewing evidence for a preference for fat and carbohydrate separately before outlining several key studies which explored a preference for combinations of carbohydrate and fat before finally introducing a potential alternative explanation. This is then followed by the two pilot studies and a summary of findings before the third study is presented. Lastly, the chapter discussion interprets the findings from the three studies and links them to work previously presented in this thesis before highlighting limitations and potential future research directions.

7.2 Introduction

7.2.1 Human preference for foods high in either fat or carbohydrate

While chapter six explored protein discrimination in humans, this chapter covers the remaining two macronutrients, fat and carbohydrate. Unlike protein, which can be used as both a source of energy and to build and maintain tissue, fat and carbohydrate are largely fuel sources, providing 9 kcal/g and 4 kcal/g, respectively. A preference for foods containing fat has been well-documented in both humans (Drewnowski & Almiron-Roig, 2010) and non-human animals (Manabe et al., 2010). In humans, there are a variety of explanations for fat preference (Drewnowski, 1997b). For example, some argue that the orosensory properties of fat increase the palatability of food (Drewnowski, 1997a) while others state that fat provides positive post-ingestive effects due to being calorically dense (Kern et al., 1993). On a population level, fat consumption is associated with factors such as urbanization and income (Drewnowski, 1997b; Drewnowski, 2003).

An association between carbohydrate content and preference is less clear, and sensory characteristics such as ‘sweetness’ have often been conflated with macronutrient composition (Drewnowski et al., 1992). For example, ‘carbohydrate craving’ was defined by Paykel and colleagues as “a ravenous appetite for a variety of sweet substances including chocolates, cake, pastry and ice cream” (Paykel et al., 1973, pp. 503, as cited by Drewnowski et al., 1992), despite the majority of calories in these foods coming from fat (Drewnowski, 1988). Relatedly, there is an abundance of research regarding the association between sweetness and preference (for a narrative review of research see Drewnowski et al., 2012), including an innate preference for sweetness in infants (Ventura & Mennella, 2011). Interestingly, the association between sweetness and preference across an individual’s lifespan does not appear to be linear, and some suggest that preference for sweetness peaks in childhood and then begins to decrease with age (Drewnowski & Almiron-Roig, 2010; Venditti et al., 2020), with a potential slight increase in older age (Venditti et al., 2020). Additionally, it is important to note that the relationship between sweetness (taste intensity) and carbohydrate content is variable (Kamil & Wilson, 2021), and that sweetness appears to be a better predictor of a food’s sugar content (Lease et al., 2016; van Langeveld et al., 2017).

7.2.2 Human preference for single foods combining fat and carbohydrate

While the previous section focussed on preference for fat and carbohydrate (or sweetness) in isolation, a preference for combinations of fat and carbohydrate has also been considered (Drewnowski & Almiron-Roig, 2010; Drewnowski et al., 1985; Drewnowski & Greenwood, 1983), including more recently by DiFeliceantonio et al. (2018) and Perszyk et al. (2021). Indeed, both Drewnowski, DiFeliceantonio, and Perszyk have argued that combining fat and carbohydrate (in Drewnowski's case, sugar) creates hedonic or reward synergy (or as DiFeliceantonio and Perszyk argue, a 'supra-additive' response).

One of the first studies which explored the impact of different combinations of fat and carbohydrate, in this case sugar (sucrose), on preference was conducted by Drewnowski and Greenwood (1983). In this study, researchers created 20 different mixtures of sugar and milk cream which varied in sugar (0-20%) and fat (0.1-36%) concentration. For each of the mixtures, 16 normal-weight participants completed a standard sip-and-spit procedure and provided a variety of sensory ratings including their preference (hedonic response). Preference was measured using a 9-point category scale which ranged from 'dislike extremely' to 'like extremely'. Participants completed the procedure twice: once in a fasted state (overnight fast) and once in a sated state (immediately after lunch).

With regards to a preference for different concentrations of fat and sugar in isolation, there was evidence for a 'sweetness breakpoint'[18]. Preference increased linearly with increasing sugar concentration until approximately one log per cent[19] (8-10% sugar) and then decreased. This non-linear pattern appeared to be present in both the fasted and fed conditions. For fat content, there was no clear evidence for a breakpoint in hedonic preference as a function of fat content in the fed condition. However, in the fasted condition, a breakpoint at approximately one log per cent was evident with preference increasing with concentration below the breakpoint and decreasing above the breakpoint. The existence of a preference breakpoint in relation to sugar concentration is consistent with previous research (Moskowitz, 1971b; Moskowitz et al., 1973).

Importantly, the researchers also modelled preference for the mixtures which contained varying amounts of sugar and fat using the Response Surface Method. Briefly, this method

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[18]As in chapter five, a breakpoint is the location at which the slope representing the association between the independent and the dependent variables changes.

[19]In this context, log percent is used to express the standardised concentrations of fat and sugar on the x-axis as these variables have different minimum and maximum values (i.e., different ranges of mixture concentrations).

plots responses to different combinations of ingredient levels and can be used to determine which combination of ingredients might result in the most preferred product (Drewnowski & Almiron-Roig, 2010; Drewnowski & Greenwood, 1983). In the context of this study, the measured response is preference and the ingredient levels are the different amounts of fat and sugar in the mixtures. A review of the resulting isopreference contours suggested a synergy between fat and sugar such that preferences were highest for light cream with sugar (Drewnowski & Almiron-Roig, 2010; Drewnowski & Greenwood, 1983). In other words, mixtures which contained a combination of fat and sugar were more preferred than those which were high in either fat or sugar.

Drewnowski and colleagues repeated the above study in 1985, this time exploring whether hedonic preference for various mixtures of fat and sugar (sucrose) might differ between participants of normal weight, participants who had obesity, and participants who previously had obesity (Drewnowski et al., 1985). Again, twenty different mixtures were created with varying levels of fat (0.1-37.6%) and sugar (0-20%), and participants followed a similar procedure to Drewnowski and Greenwood (1983), including the standard sip-and-spit protocol and sensory and preference ratings. Again, the researchers used the Response Surface Method to model the peak preference for the different mixtures of fat and sugar.

A hedonic preference breakpoint for sucrose was replicated (Drewnowski & Greenwood, 1983) in the group of individuals with normal weight, such that preference increased with concentration until ~10% sucrose concentration and then decreased. Fat preference is reported to have increased with concentration until ~20% fat concentration. In the group of individuals having obesity, there was no evidence for a preference breakpoint for sucrose, and preference decreased linearly with increasing concentration. Additionally, in this same group, there was a positive linear association between preference and fat concentration with no evidence for a breakpoint. Lastly, in the group of individuals previously having had obesity, higher concentrations of fat and sugar in isolation were preferred compared to the group of individuals having normal weight and the group of individuals having obesity. The authors make no claim regarding evidence for preference breakpoints in this group.

The results also suggest that the three groups differed in their preference for the mixtures containing a combination of fat and sugar. Individuals with normal weight were predicted by the model to have a peak preference for mixtures containing 20.7% fat and 7.7% sugar whereas individuals having obesity were predicted to have a peak preference at 34.4% fat and 4.4% sugar. Individuals previously having obesity had predicted peak preference at 35.1% fat and 10.1% sugar, and this is higher than the predicted preference for the group of individuals with obesity. The authors suggest that this difference could be related to past experience with dieting and weight loss enhancing preference for sweet taste (Drewnowski et al., 1985). Relatedly, the authors also explored a relationship between the preferred sugar/fat ratio and BMI. In this case, a higher sugar/fat ratio value suggests an increased preference for sweet taste over fat. A significant negative correlation between BMI and sugar/fat ratio was reported such that higher BMIs were associated with a greater preference for fat over sweetness. Together, these findings and those from Drewnowski and Greenwood (1983) suggest that combinations of fat and sugar result in "a particular hedonic synergy" (Drewnowski & Almiron-Roig, 2010, p. 5/24).

Recently, DiFeliceantonio et al. (2018) revisited this interaction between fat and carbohydrate using a different approach and focusing on food reward rather than hedonic preference. In this now highly influential study, participants viewed 120 kcal portions of 39 different foods which were either high in fat (such as cheese or sausage), high in carbohydrate (such as fruit gummies or pretzels) or contained a combination of fat and carbohydrate in roughly equal amounts (kcal) (such as a chocolate cookie or raspberry cream cake). The foods, 13 in each category, were equally familiar and liked; however, they differed significantly with regards to energy density [(3.69 kcal/g (high-carbohydrate), 3.43 kcal/g (high-fat), 4.82 kcal/g (combination)]. Alongside several other measures, participants completed the Becker-DeGroot-Marshak Auction task (Becker et al., 1964) as a measure of food reward during an fMRI scanning session. Briefly, in this version of the auction task, participants received €5 to bid on the snacks they were shown during the scanning session. Participants could bid between €0 and €5 for the snack, and, if their bid was higher than the computer's bid, they were 'successful' and would receive the snack and the remainder of the €5 in cash. For example, if the participant bid €2.75 for the portion of chocolate cookie and the computer bid €2.00, then the participant was 'successful' and received both the cookie and the remaining €1.25. If the participant was 'unsuccessful' in bidding and their bid was lower than the computer's bid, then they would not receive the item and would instead receive the entire €5. In this task, bidding what the individual believes the item to be worth is the optimum strategy (Becker et al., 1964)

Using a linear mixed effects model where the bid amount was the dependent variable, participant was included as a random effect and macronutrient category (i.e., high-fat, high-carbohydrate or combination), true energy density, estimated energy density, liking, estimated portion calories, portion size, and calories shown were included as fixed effects, the authors reported that participants were willing to bid significantly more for foods which contained a combination of fat and carbohydrate than foods which were high in a single macronutrient (i.e., high-fat or high-carbohydrate). Based on a significant interaction term in a second model where the macronutrient category was binarily coded, the authors also conclude that the effect of combining fat and carbohydrate in a single food is 'supra-additive'. In other words, bids for combination foods "were greater than would be expected from summing the bids for fat and carbohydrate foods" (DiFeliceantonio et al., 2018, p. 37). Importantly, it was also confirmed that the supra-additive effect was not due to the combination groups being liked slightly more.

This study was the first to re-visit the effect of combining fat and carbohydrate and link it to food reward. One possible explanation for the supra-additivity, which was mentioned in the study’s introduction, is that foods which contain both fat and carbohydrate simultaneously activate the individual reward pathways for fat and carbohydrate, thus generating greater reward than foods which predominately contain a single macronutrient (DiFeliceantonio et al., 2018). Additionally, the authors link the increased reward of combination foods to the obesity epidemic and argue that modern food processing has created foods which contain both combinations and amounts of macronutrients that would not naturally occur (DiFeliceantonio et al., 2018). Put differently, and at risk of greatly over-simplifying, because humans have only recently been exposed to these ‘unnatural’ foods, we are therefore ‘victims’ to their hyper-rewarding effects.

Similar to Drewnowski and colleagues, researchers have also considered whether food reward for high-fat, high-carbohydrate and combination foods differs depending on body weight status. Perszyk et al. (2021) explored this question in American participants using a set of 36 American snack foods, 12 in each macronutrient category (high-fat, high-carbohydrate and combination) (Fromm et al., 2021). The three macronutrient categories were matched for a variety of characteristics including energy density (kcal/g), price, liking and familiarity (Fromm et al., 2021). In this study, 60 participants, 30 individuals with a healthy weight (*M* ± *SD*, BMI 21.92 ± 1.77) and 30 individuals with overweight or obesity (*n*= 30; *M* ± *SD*, BMI 29.42 ± 4.44) completed the Becker-DeGroot-Marshak Auction task (Becker et al., 1964) as a measure of food reward. Participants were shown 120 kcal portions of each of the foods and were provided $5 to bid against the computer. Participants completed several additional tasks including subjective measures of the 36 foods (perceived liking, familiarity, expected satiety, frequency of consumption etc), answering questions about dietary behaviour using the Dutch Eating Behaviour Questionnaire (van Strien et al., 1986), and completing a modified version of the Dietary Fat and Free Sugar Short Questionnaire (DFS) (Francis & Stevenson, 2013). The

DFS was used to explore whether the consumption of the three different macronutrient groups differed as a function of weight status.

As in DiFeliceantonio et al. (2018), a linear mixed effects model was used to determine whether the bid amount differed as a function of the macronutrient category. However, in this study, weight status and an interaction between weight status and macronutrient category were included in the model alongside actual energy density, estimated energy density, estimated energy content, portion size, and liking (all fixed effects) and participant as a random effect. The results reported a significant main effect of macronutrient category and a significant interaction between macronutrient category and weight status. The interaction was driven by a main effect of macronutrient category on bid amount in the group with healthy weight and not the group with overweight or obesity. In other words, in the group with healthy weight, participants bid significantly more for combination foods than foods which were high in fat or carbohydrate, but in the group with overweight or obesity, there was no significant difference in bid amount between the macronutrient categories. The supra-additive interaction was also replicated (DiFeliceantonio et al., 2018) in the group with normal weight, such that the bids for combination food were more than could be expected when summing the bids from fat and carbohydrate separately.

The authors argue that, in the group of individuals with overweight or obesity, the lack of a macronutrient category effect on bid amount, as well as the failure to observe a supra-additive interaction, might be due to degraded reinforcement learning and habituation to food (Coppin et al., 2014; Kroemer & Small, 2016; Kube et al., 2018; Temple & Epstein, 2012 as cited by Perszyk et al., 2021). They also state that metabolic dysregulation in individuals with obesity might disrupt the hormonal responses which are required for reinforcement learning (Perszyk et al., 2021). Lastly, environmental reasons, specifically diet, might also disrupt reinforcement learning in response to food composition, although the authors do note that the two groups, individuals with normal weight and individuals with overweight or obesity, did not differ with regards to the typical consumption of fat and sugar-containing foods as measured by the DFS (Perszyk et al., 2021).

In summary, real-world foods containing a combination of fat and carbohydrate appear to be more rewarding than foods high in either fat or carbohydrate, and one possible explanation is that combination foods simultaneously activate the separate reward pathways for fat and carbohydrate. The following section explores a potential alternative explanation before outlining the three studies presented in this chapter.

7.2.3 Avoiding excess satiety or satiation as an alternative explanation for the increased reward value of foods containing a combination of fat and carbohydrate

As previously mentioned in section 5.4, there is evidence that non-human animals reduce their intake of high concentrations of a single macronutrient (Smith & Foster, 1980), potentially due to their aversive nature (Moskowitz, 1971b; Moskowitz et al., 1974; Sclafani & Ackroff, 2004). Similar reductions in intake and avoidance of high doses of single macronutrients have also been observed in humans (Lucas & Bellisle, 1987; Martin et al., 2016; Pérez et al., 1994; Zandstra et al., 1999). This reduction in intake could be interpreted as an attempt to avoid excess satiety or 'nimiety' (Kulkosky, 1985), negative visceral sensations (e.g., 'feeling sick/nausea' (Booth et al., 2011) or malaise (Hengist et al., 2020)).

One could argue that the aversion to high doses of a single macronutrient or avoidance of excess satiety or satiation could provide an alternative explanation for the observed preference for foods containing a combination of fat and carbohydrate highlighted in the previous section (DiFeliceantonio et al., 2018; Drewnowski et al., 1985; Drewnowski & Greenwood, 1983; Perszyk et al., 2021). Namely, one possibility for the increased preference for combination foods is not because they are inherently rewarding due to their 'unnatural' combinations of macronutrients, but instead due to them being less satiating. In other words, a calorie of a combination food is more rewarding because it is less filling than a calorie of a food high in either fat or carbohydrate.

To the best of the author's knowledge, no studies have explored whether the amount of fat and carbohydrate in a single food (known to influence food reward (DiFeliceantonio et al., 2018; Perszyk et al., 2021) influences judgments of expected satiation (i.e., an anticipated feeling of fullness (Brunstrom & Rogers, 2009) and portion selection (kcal). As mentioned earlier in this chapter, understanding whether combining fat and carbohydrate in a single food might promote the selection of larger portions could help inform targeted food reformulation which aims to support population-level healthy weight maintenance. Briefly outlining the structure of the remaining chapter, in the first instance, a pilot using American snack foods (Perszyk et al., 2021) as stimuli explored whether combination, high-fat and high-carbohydrate foods differ in their expected satiation as measured using the 'method of adjustment' (Brunstrom & Rogers, 2009). An opportunity to recruit a larger sample (n= 30) arose, and, in the second pilot, similar associations between the macronutrient category (i.e., combination, high-fat, and high-carbohydrate) and expected satiation were explored. In this second pilot, UK foods were used as stimuli (Rogers et al., 2024) and UK participants were recruited through Prolific (https://www.prolific.co/; Prolific (2014, Copyright Year: 2023). For convenience, the two pilot studies are reported in parallel, followed by an interim summary. Lastly, the third study built on this pilot work and included ideal and maximum portion selection tasks along with a measure of liking, and the outcome of this study is described before concluding the chapter with a general discussion of the three studies.

7.3 Combination foods are less satiating than foods high in either fat or carbohydrate: results from US and UK pilot studies

7.3.1 US pilot

Ten participants (female: *n*= 9, male: *n*= 1; *M* ± *SD*, age= 31.3 ± 14.1 years) were recruited on the online platform Prolific (https://www.prolific.co/; Prolific (2014, Copyright Year: 2023). Using Prolific's built-in screening criteria, the study was only advertised to participants who reported not following a diet and were currently living in the United States of America. The dietary inclusion criteria were implemented to exclude individuals who identified as vegan or vegetarian as the stimuli set included meat, dairy, and eggs. The inclusion criteria of residing in the United States was to ensure relative familiarity with the foods as the stimuli set comprised common American snack foods.

The stimuli used in this pilot were identical to those used by Perszyk et al. (2021). A total of 36 different foods were presented across three macronutrient categories (12 foods in each category): high-fat, high-carbohydrate, and combination (containing roughly equal amounts of fat and carbohydrate). The three macronutrient categories were matched for a variety of food-based characteristics, including, among others, energy density, portion size, volume, and price (Fromm et al., 2021) (see Table 7.1 for macronutrient composition and physical characteristics of stimuli). Additionally, the foods in the three categories were reported to be equally liked, familiar and healthy (Fromm et al., 2021). The colour images were standardised such that each food portion was centred on a white plate with a paper cup and napkin placed next to the plate to provide visual cues for portion size estimation.

Table 7.1 *Nutritional Information and Physical Characteristics of the 36 Stimuli used in the US Pilot*[1]

Macronutrient category	Food item	kcal/100g (energy density)	Fat, g/120 kcal	Carbohydrate, g/120 kcal	Protein, g/120 kcal	Sodium, mg/120 kcal	Water, g/120 kcal	Portion size, g/120 kcal	Volume, cm^3
High-fat	American cheese	373.68	10.26	1.52	5.54	523.94	12.66	32.11	30.06
	Babybel cheese wheels	391.53	10.83	0.41	5.52	183.78	13.01	30.65	45.30
	Blue cheese	352.73	9.78	0.80	7.28	390.00	14.43	34.02	45.00
	Breakfast sausage	397.14	10.95	0.82	4.17	280.58	13.46	30.22	42.00
	Brie cheese	335.10	9.91	0.16	7.43	224.84	17.34	35.81	69.90
	Colby jack cheese	387.60	9.66	0.50	7.47	187.20	12.26	30.96	35.91
	Deviled eggs	170.30	8.58	2.15	7.85	256.74	51.52	70.47	120.90
	Hardboiled eggs	154.55	8.24	0.87	9.77	97.06	57.94	77.65	84.30
	Pepperoni	504.41	11.01	0.28	4.58	375.94	6.79	23.79	36.30
	String cheese	296.30	8.01	2.26	9.62	270.00	19.06	40.50	53.40
	Summer sausage	363.32	10.05	1.10	5.76	429.90	14.92	33.03	46.80
	Swiss cheese	315.79	9.36	0.94	8.21	276.00	18.38	38.00	35.40
High-carbohydrate	Bagel	263.81	0.60	23.83	4.80	191.91	15.38	45.49	187.50
	Baked beans	93.90	1.19	23.88	6.58	558.44	93.93	127.79	117.90
	Dried apricots	239.86	0.26	31.34	1.70	5.29	15.45	50.03	42.90
	Froot loops	372.41	1.06	27.94	1.61	147.78	0.81	32.22	150.60
	Frosted flakes	367.50	0.08	29.67	1.42	151.02	1.07	32.65	48.30
	Fruit snacks	347.83	0.17	31.43	0.19	45.00	3.29	34.50	26.70
	Gummy bears	320.99	0.04	29.86	1.87	14.51	4.96	37.38	27.69

	Jelly beans	373.90	0.02	30.02	0.00	15.85	2.02	32.09	25.50
	Lucky charms	408.33	1.09	23.95	2.18	185.31	1.22	29.39	100.50
	Pineapple rings	77.88	0.17	31.13	0.54	1.36	121.72	154.09	153.60
	Pretzels	384.48	0.91	25.09	3.13	387.52	0.98	31.21	54.60
	Sorbet	122.64	0.09	31.81	0.14	8.31	65.75	97.85	124.80
Combination	Banana nut bread	274.19	4.55	18.68	1.94	128.07	20.22	43.76	73.50
	Cheese and crackers	486.77	6.02	14.48	1.94	216.52	3.01	24.65	49.50
	Chocolate covered pretzels	458.55	4.82	17.21	2.10	129.23	0.57	26.17	59.40
	Chocolate raisins	391.53	4.54	20.96	1.26	10.81	3.43	30.65	31.80
	Doritos	497.35	6.77	13.69	1.75	178.72	1.92	24.13	36.90
	Guacamole	89.27	9.55	10.46	2.06	535.38	108.38	134.43	137.40
	Mini chocolate chip cookies	490.00	6.02	16.17	1.25	75.92	1.24	24.49	45.60
	Mini nutter butters	480.00	4.99	17.31	1.69	100.00	1.90	25.00	31.80
	Peanut butter and crackers	486.77	6.05	14.22	2.71	197.39	2.14	24.65	52.80
	Pizza rolls	251.28	4.69	15.29	4.16	285.71	27.31	47.76	49.80
	Pringles	512.50	7.61	12.51	0.73	111.80	0.83	23.41	30.60
	Roasted red pepper hummus	172.84	5.94	13.34	4.97	296.33	43.52	69.43	87.30

[1]Nutritional information from Dr Emily Perszyk (2022, personal communication)

The only task included in this pilot was a measure of expected satiation using the method of adjustment (Brunstrom, Shakeshaft, et al., 2010). No additional tasks were included as the aim of the pilot, in this instance, was to establish whether expected satiation differed in foods which vary in the amount of fat and carbohydrate they contain using a previously published set of stimuli (Perszyk et al., 2021). Briefly describing the expected satiation task, the participant was presented with two images on the computer screen 1) a standard food (chocolate M&Ms) and 2) a comparator food (one of the 36 stimuli). The comparator food was presented in a fixed 360 kcal portion which is consistent with other studies exploring judgments about expected satiation (Brunstrom & Rogers, 2009; Brunstrom, Shakeshaft, et al., 2010). The participants then increased or decreased the portion size of the standard food (chocolate M&Ms) in 20-kcal steps (20-1000kcal) until they believed that each of the foods would leave them feeling equally full immediately after having been eaten. The prompt displayed on the screen above the two foods was " Look at the portion *(food name auto-fills)* on the left. Imagine you are having this plate of food for a snack TODAY. Change the portion of food on the right using the slider, so that both foods will leave you feeling EQUALLY FULL (immediately after they have been eaten)." The food name in the prompt (i.e., the text in italics above) would auto-fill to match the label on the comparator image, and the order of the comparator foods was randomized. Chocolate M&Ms were chosen as the standard food for two reasons 1) they are a highly familiar snack food and 2) they are a combination food (i.e., contain an equal blend of fat and carbohydrate) so are potentially less likely to bias responding.

Participants began by reading an information sheet and then provided their informed consent. Following this, they were shown two practice trials for the expected satiation task using cucumber and banana as comparator foods. After the practice trials, they then completed the 36 trials of the expected satiation task, received the debriefing information and, lastly, provided their final consent for their data to be used. The study was advertised to last 7 minutes

(~10 seconds per trial) and participants were required to complete the study on a desktop computer. Participants were renumerated at a rate of £9.00 per hour (£1.05 payment), and the median completion time for the study was 6 minutes and 39 seconds. Ethical approval was granted by the University of Bristol Science Faculty Ethics Committee (12311).

7.3.2 UK pilot

Using Prolific (https://www.prolific.co/; Prolific (2014, Copyright Year: 2023), a total of 30 participants (female: n= 23, male: n= 7; $M \pm SD$, age= 39.8 ± 14.7 years) were recruited. The study was only advertised to individuals who reported not following a diet and were currently living in the United Kingdom. As in the US pilot, the purpose of the dietary inclusion criteria was to exclude individuals who identified as vegan or vegetarian as the stimuli set comprised animal products and to attempt to ensure familiarity with the foods.

For each macronutrient category (high-fat, high-carbohydrate and combination) eight different foods were presented (24 in total, see Table 7.2 for nutritional information). Out of the 24 foods, 23 were previously used in a different study (Rogers et al., 2024). Vanilla ice cream was included in the original set of stimuli; however, because 50 images of each food were taken, ice cream was not a viable stimulus due to melting during the image-taking process. Therefore, vanilla ice cream was replaced with vanilla custard. The three macronutrient categories were matched for their energy density (kcal/100g, ($F(2, 23)$= 0.004, p= 0.996; $M \pm SD$, high-fat= 324.38 ± 141.96, high-carbohydrate= 318.63 ± 48.04, combination= 320.63 ± 177.31).

Table 7.2 *Nutritional Information of the 24 Stimuli Used in the UK Pilot*

Macronutrient category	Food item	kcal/100g (energy density)	Fat, g/100g	Carbohydrate, g/100g	Sugars, g/100g	Fibre, g/100g	Protein, g/100g	Salt, g/100g
High-fat	Cheddar cheese	416	34.9	0.0	0.0	0.0	25.4	1.8
	Frankfurter sausage	285	25.0	2.0	2.0	1.2	12.5	1.6
	Mozzarella cheese	280	24.3	0.8	0.7	0.0	14.8	0.5
	Olives	164	16.7	0.0	0.0	4.1	1.0	3.1
	Pepperoni	360	28.5	1.0	0.0	0.0	24.8	4.3
	Pork liver pate	279	24.8	4.7	1.7	0.0	9.3	1.7
	Salted peanuts	614	51.0	5.6	5.1	8.5	30.0	1.3
	Smashed avocado	197	19.5	1.9	0.0	3.4	1.9	0.0
High-carbohydrate	Bagel	258	1.2	50.1	5.6	3.1	10.1	0.8
	Crispbread	349	0.9	67.5	2.6	14.3	10.6	0.9
	Dried apple slices	281	0.2	63.9	54.1	9.9	1.0	0.1
	Dried pitted dates	272	0.0	60.4	58.1	10.3	2.2	0.0
	Fruit pastilles	355	0.1	88.6	61.4	0.0	0.1	0.2
	Salted pretzels	393	4.2	76.7	1.8	3.6	10.3	1.5
	Sultanas	297	0.0	69.4	69.4	2.5	2.7	0.0
	Turkish delight	373	0.0	92.9	85.8	0.0	0.1	0.0
Combination	Blueberry muffin	388	19.9	46.9	25.4	1.3	4.7	0.3
	Butter croissant	438	25.8	41.5	6.5	2.7	8.5	0.7
	Chocolate mousse	152	4.3	22.4	21.8	1.4	4.9	0.2
	Custard	96	2.9	14.8	10.6	0.0	2.8	0.1
	Flapjack bites	448	19.2	61.6	26.4	2.9	5.9	0.4
	Oatcake	449	17.0	59.5	3.2	9.4	9.8	1.4
	Salted popcorn	508	33.7	38.6	0.5	11.6	6.8	1.4

Strawberry yogurt	86	2.8	11.7	10.7	0.0	3.4	0.1

As just mentioned, 50 images were taken for each food, excluding salted popcorn. Due to salted popcorn being a highly aerated food and having a large volume, it was not possible to take 50 images without the food spilling out of the bowl; therefore, only 40 were taken. All images were taken in equicaloric 20 kcal steps (20-1000 kcal; 800 kcal for salted popcorn) using a high-resolution digital camera (Nikon D50) against a uniform white background and under constant lighting conditions. Stimuli were photographed on the same white plate (255 mm in diameter) and the name of the foods was inserted as a label in the upper left corner of the image (see Figure 7.1 as an example). If the stimulus was usually consumed in a bowl (i.e., yoghurt or vanilla custard) or because its physical form made it difficult to judge increases in portion size from an overhead angle, then it was photographed in a clear, 2-litre glass Pyrex bowl which was placed in the centre of the white plate (see Figure 7.2 as an example). These images were taken at a 45-degree angle.

Figure 7.1 Example of formatted image for the expected satiation task depicting 360 kcal of fruit pastilles.

This image was taken at an overhead angle.

Figure 7.2 Example of a formatted image for the expected satiation task depicting 360 kcal of strawberry yogurt.

This image was taken at a 45-degree angle.

The UK pilot included the same expected satiation task as the US pilot and followed the same procedure. The only difference between the pilots was the time the study was advertised to take. Given that there were 12 fewer foods to evaluate in the UK pilot (n= 24) compared to the US pilot (*n*= 36), the study was advertised to take five minutes. Participants were again renumerated at a rate of £9.00 per hour (£0.75 payment) and were only able to complete the pilot on a desktop computer. The median study completion time was 5 minutes and 29 seconds, and ethical approval was granted by the University of Bristol Science Faculty Ethics Committee (12311).

7.3.3 Statistical analysis for both pilot studies

To establish whether the expected satiation (kcal) of the combination category was different from either the high-fat or the high-carbohydrate category, a three-stage procedure was followed. First, for each participant, the mean expected satiation (kcal) for each of the three macronutrient categories was calculated. Then, for each participant, the mean difference score (kcal) was calculated between the high-fat category and the combination category (mean expected satiation (kcal) of the high-fat category minus the mean expected satiation (kcal) of the combination category) and this was repeated for the high-carbohydrate category (mean expected satiation (kcal) of the high-carbohydrate category minus the mean expected satiation (kcal) of the combination category). This resulted in two mean difference scores (kcal) for each participant. Lastly, a one-sample *t*-test was used to establish whether the mean difference score (kcal) representing the difference in expected satiation (kcal) between high-fat and combination categories was different from zero[20]. A significant positive mean difference score indicates that the high-fat category was, on average, expected to be more satiating than the combination category. A significant negative mean difference score indicates the converse – the combination category was expected to be more satiating than the high-fat category. The one-sample *t*-test was repeated using the mean difference score between high-carbohydrate and

[20]Alternatively, one could also consider a one-way ANOVA with repeated measures and planned pairwise comparisons as the statistical approach. In the case of these analyses, the key pairwise comparisons will be the combination category compared to the high-fat category and the combination category compared to the high-carbohydrate category. These analyses will be reported in footnotes.

combination categories and the interpretation is the same. For each one-sample *t*-test, Cohen's *d* is reported as a measure of effect size. All analyses were conducted in the R statistical environment (R Core Team, 2022).

7.3.4 Results of the US and UK pilots

The mean expected satiation (kcal) of the three macronutrient categories in the US pilot is reported in Table 7.3 (see Appendix 4 Table 11.10 for the expected satiation of the 36 different foods). The mean difference scores comparing the high-fat category and the high-carbohydrate category (see Table 7.3) suggest that foods in the combination category were, on average, expected to be less satiating, and the results of a one-sample *t*-test confirmed that these mean differences were statistically significant. This indicates that the expected satiation of foods in the combination category was, on average, significantly lower compared to foods in the high-fat category ($t(9)= 4.33$, $p= 0.002$, Cohen's $d = 1.37$) or foods in the high-carbohydrate category ($t(9)= 3.40$, $p= 0.008$, Cohen's $d = 1.08$) [21,22].

[21]As outlined in footnote 20, the results of a one-way ANOVA with repeated measures and a Greenhouse-Geisser correction indicated that expected satiation differed significantly across the three macronutrient categories ($F(1.157, 10.412)= 6.56$, $p= .024$). The results of the planned pairwise comparisons indicated the foods in the combination category had significantly lower expected satiation than foods in the high-fat (114.17; 95% CI 54.55, 173.79; $p= .002$) and the high-carbohydrate (75.33; 95% CI 25.24, 125.43; $p= .008$) categories.

[22]It should be noted that Perszyk and colleagues[a] assessed the expected satiety of the same foods using a 260-mm visual analogue scale with the prompt 'How filling do you expect this food portion to be?' and the anchors of 'Not filling at all' and 'Extremely filling'. In the participants with normal-weight, review of the descriptive statistics suggest a trend for combination foods to be less satiating compared to high-fat or high-carbohydrate foods, but in the participants with overweight or obesity, it appeared that there were no differences in expected satiety across the three macronutrient categories. Importantly, these interpretations have not been statistically evidenced. The extent to which methodological differences between this study (i.e., use of VAS, smaller portion size etc.) and the US pilot might have resulted in the disparate findings is unclear.

[a]Perszyk, E. E., Hutelin, Z., Trinh, J., Kanyamibwa, A., Fromm, S., Davis, X. S., Wall, K. M., Flack, K. D., Difeliceantonio, A. G., & Small, D. M. (2021). Fat and Carbohydrate Interact to Potentiate Food Reward in Healthy Weight but Not in Overweight or Obesity. *Nutrients*, *13*(4), 1203. https://doi.org/10.3390/nu13041203

Table 7.3 *Expected Satiation (kcal) and Mean Difference Scores (kcal) in the US Pilot*[1,2]

Macronutrient category	Mean ± (SD)	Mean difference score ± (SD)
High-fat	538.33 (258.12)	114.17* (83.35)
High-carbohydrate	499.50 (235.15)	75.33* (70.03)
Combination	424.17 (191.71)	0

[1]360 kcal images were shown to participants in the expected satiation task.
[2]The mean difference score (kcal) represents the difference in the mean expected satiation (kcal) between the high-fat category (or the high-carbohydrate category) and the combination category.
* $p< .01$, vs. Combination using a one-sample *t*-test

The mean expected satiation (kcal) and mean difference scores (kcal) for the three macronutrient categories in the UK pilot are reported in Table 7.4 below (see Appendix 4 Table 11.11 for the expected satiation of the 24 different foods). Again, the mean difference scores suggested that, on average, foods in both the high-fat category and the high-carbohydrate category were perceived to be more satiating than foods in the combination category (see Table 7.4). A one-sample *t*-test confirmed that the difference in expected satiation (kcal) between the high-fat and the combination categories was significant ($t(29)= 2.66$, $p= 0.013$, Cohen's $d= 0.486$). However, the results of the second one-sample *t*-test indicated foods in the combination category did not differ significantly from foods in the high-carbohydrate category with regards to expected satiation ($t(29)= 1.44$, $p= 0.161$, Cohen's $d= 0.263$)[23]. When reviewing the effect sizes for the one-sample *t*-tests, one possibility for the non-significant result when comparing the expected satiation of foods in the high-carbohydrate category to foods in the combination category is that this analysis was underpowered to detect the small effect size (a sample size of 116 participants would have been needed to detect an effect size of .263 with an alpha of 0.05 and 80% power).

[23]The results of a one-way ANOVA with repeated measures indicated a trend in differences in expected satiation across the three macronutrient categories ($F(2, 58)= 2.74$, $p= .073$). The foods in the combination category were expected to be significantly less satiating than foods in the high-fat category (33.50; 95% CI 7.76, 59.24; $p= .013$). Contrastingly, there was no difference in expected satiation between foods in the combination and high-carbohydrate categories (25.08; 95% CI -10.55, 60.72; $p= .161$).

Table 7.4 *Expected Satiation (kcal) and Mean Difference Scores (kcal) in the UK Pilot*[1,2]

Macronutrient category	Mean ± (SD)	Mean difference score ± (SD)
High-fat	414.92 (190.33)	33.50* (68.94)
High-carbohydrate	406.50 (192.25)	25.08 (95.44)
Combination	381.42 (190.42)	0

[1]360 kcal images were shown to participants in the expected satiation task.
[2]The mean difference score (kcal) represents the difference in the mean expected satiation (kcal) between the high-fat category (or the high-carbohydrate category) and the combination category.
* $p < .01$, vs. Combination

On further review of the data, one participant's mean difference score (high-carbohydrate category minus combination category) was identified as an outlier (*Z*-score greater than 3.29). The decision was made to exclude this participant and re-run both of the one-sample *t*-tests (see Table 7.5 for the expected satiation (kcal) and the mean difference scores for the three macronutrient categories as assessed in the UK pilot excluding one outlying participant). Again, foods in the combination category had, on average, significantly lower mean expected satiation than foods in the high-fat category ($t(28)= 3.14$, $p= 0.004$, Cohen's $d= 0.583$). After excluding the outlying participant, foods in the combination category now had, on average, significantly lower mean expected satiation than foods in the high carbohydrate category ($t(28)= 2.63$, $p= 0.014$, Cohen's $d= 0.488$)[24].

Table 7.5 *Expected Satiation (kcal) and Mean Difference Scores (kcal) in the UK Pilot Excluding One Outlier*[1,2]

Macronutrient category	Mean ± (SD)	Mean difference score ± (SD)
High-fat	412.93 (99.05)	38.10* (65.30)
High-carbohydrate	411.12 (112.43)	36.29* (74.36)
Combination	374.83 (98.75)	0

[1]360 kcal images were shown to participants in the expected satiation task.
[2]The mean difference score (kcal) represents the difference in the mean expected satiation (kcal) between the high-fat category and the high-carbohydrate category to the combination category.

[24]Expected satiation differed significantly across the three macronutrient categories according to the results of a one-way ANOVA with repeated measures ($F(2, 56)= 5.44$, $p= .007$). Foods in the combination category were expected to be less satiating than foods in the high-fat (38.10, 95% CI 13.26, 62.94; $p= .004$) and the high-carbohydrate (36.29, 95% CI 8.01, 64.58; $p= .014$).

* $p < .01$, vs. Combination

7.3.5 Summary of pilot study results and considerations for the next study

In both the US and UK pilot studies, there was a difference in mean expected satiation (kcal) between the three macronutrient categories, as measured using the method of adjustment. Specifically, single foods comprising roughly equal amounts (kcal) of fat and carbohydrate (combination foods) were expected to deliver less satiation calorie-for-calorie than foods high in either fat or carbohydrate. While the sample size in the US pilot was relatively small, the pattern of results was replicated in a larger UK sample. However, despite the seemingly clear pattern in expected satiation, there were some potential concerns regarding the foods used in the pilots, and these will be discussed in the following paragraphs alongside methodological changes for the final study in this chapter.

Reflecting on the stimuli used in the US pilot, which were identical to those used by Perszyk et al. (2021), there were some concerns regarding the repetition of foods in two of the three macronutrient categories. In the high-fat category, there was a substantial repetition of food types, including cheese (seven types: American cheese, Babybel cheese wheels, Blue cheese, Brie cheese, Colby jack cheese, String cheese, and Swiss cheese), eggs (two types: Hardboiled eggs and Deviled eggs), and sausage (two types: Breakfast sausage and Summer sausage). In the high-carbohydrate category, there were three types of cereal (Froot loops, Frosted flakes, and Lucky charms) and three types of sweets (Gummy bears, Fruit snacks, and Jelly beans). The repetition of foods in these two categories brings into question whether these findings could generalise to other foods with this macronutrient profile.

In the UK stimuli set, there was a concern regarding the familiarity and visual appearance of one of the foods. While no questions assessing familiarity had been included in the pilot, informal polling of colleagues demonstrated that, of all the stimuli, Turkish Delight was the most unfamiliar. Importantly, research has shown an association between familiarity and judgments of expected satiation, such that individuals who are unfamiliar with a food expect it to be less satiating (Brunstrom, Shakeshaft, et al., 2010), and post hoc review of the mean expected satiation (kcal) for each of the foods suggests that Turkish Delight may be a potential outlier (see Appendix 4 Table 11.11 for expected satiation (kcal) by food). Additionally, the visual appearance of Turkish Delight on the plate was also potentially misleading. Images were taken from overhead such that the true height of the Turkish Delight pieces might not have been accurately represented in the image, and the pieces were clustered on the lower right half of the plate. Therefore, based on concerns about familiarity and visual appearance, the decision was made to replace Turkish Delight with Wine Gums[25]. Importantly, the three macronutrient groups remained matched for their energy density (kcal/100g, $F(2, 23)= 0.003$, $p= .997$; $M \pm SD$, high-fat= 324.38 ± 141.96, high-carbohydrate= 319.00 ± 48.28, combination= 320.63 ± 177.31).

7.4 Combination foods are selected in larger portions and are liked more than foods high in either fat or carbohydrate

This study built on the findings from the two pilots and included four additional tasks alongside the expected satiation task: ideal portion size, maximum portion size, liking and familiarity. The portion size selection tasks are of particular interest as it is yet to be demonstrated whether combination foods, which were previously shown to be more rewarding, are selected in larger portions (kcal) than foods high in either fat or carbohydrate. It should be noted that the wording of the expected satiation task in this study is slightly modified from the wording used in the US and UK pilots. The original task wording was thought to potentially underestimate the effect of combining fat and carbohydrate in a single food on expected satiation as it might bias responding more towards the immediate volume signal and feedback

[25]The nutritional information for the other 23 stimuli remained the same (see Table 7.2). The nutritional information for Wine Gums is as follows: 347 kcal per 100g, 0.9 g of fat per 100g, 79 g of carbohydrate per 100g of that 55 g of sugar per 100g, 0 g of fibre per 100g, 5.8 g of protein per 100g and 0.08 g of salt per 100g.

from stomach fullness, rather than the slightly delayed calorie-content signal (see section 5.3 for a review of the two-component (volume and calorie-content signals) model of meal size). The modified expected satiation task now asked participants to select the amount of food needed to feel equally full, rather than equally full immediately after eating.

Food liking was included as a measure in this study as recent research suggests that the effects of fat and carbohydrate on food reward are driven by effects on food liking (Rogers et al., 2024). In a separate study, participants rated their liking and desire-to-eat (food reward) different foods ranging in the amount of fat and carbohydrate (Rogers et al., 2024). Consistent with findings from DiFeliceantonio et al. (2018); Perszyk et al. (2021), foods which contained a combination of fat and carbohydrate were more rewarding (more desired) than foods which were high in either fat or carbohydrate. Additionally, this pattern was replicated in a measure of liking, and the authors propose that the increased reward of combination foods is driven by greater liking for these foods acquired through flavour-nutrient learning (Rogers et al., 2024). Given that differences in food liking appear to mirror changes in food reward, especially in relation to fat and carbohydrate content, it was decided to only include a measure of food liking and not food reward (e.g., desire to eat).

The study procedure, hypotheses, and statistical analyses were pre-registered on the OSF (https://osf.io/29udv/?view_only=17aa7a650bc141dbb0b0ecb36cbc9246, note a measure of hunger was included in the preregistration but was unintentionally not included in the study).

7.4.1 Methods

7.4.1.1 Participants

As previously mentioned in section 7.3.4, after excluding one outlier from the UK pilot, the smallest Cohen's *d* effect size reported for the mean differences was 0.488 (mean difference between high-carbohydrate and combination macronutrient categories). An a priori power calculation confirmed that 33 participants would be needed to detect an effect with an alpha of 0.05 (two-tailed) at 80% power.

Therefore, 33 participants (female: n = 16, male: n = 17; $M \pm SD$, age= 39.3 ± 2.50 years) were recruited on Prolific (https://www.prolific.co/; Prolific (2014, Copyright Year: 2023). Again, using Prolific's built-in screening criteria, the study was only advertised to individuals residing in the UK and who reported not following a diet. Additionally, the study was not advertised to participants who completed the previous UK pilot. This was to avoid recruiting individuals who were already familiar with the study and had been debriefed on its purpose.

7.4.1.2 Tasks and study procedure

As mentioned earlier, the modified expected satiation task was identical to the one used in the UK and US pilot studies except for the prompt shown to participants on the screen. Rather than asking participants to adjust the standard food (chocolate M&Ms) so that each portion of food on the screen will leave them equally full immediately after having been eaten, participants were asked to adjust the food so that each portion leaves them feeling equally full. This modified task wording[26] has been used in previously published research (Brunstrom et al., 2018), and the order of the 24 foods was randomised.

The ideal and maximum portion size selection tasks involved participants increasing or decreasing the portion size of the food displayed using the slider at the bottom of the screen (these tasks have been used by Elsworth et al. (under-review)). The portion size increased or decreased in 20 kcal steps with a minimum portion size of 20 kcal and a maximum portion size of 1,000 kcal (800 kcal for salted popcorn due to an inability for 1,000 kcal to be displayed in the bowl). The prompt for the ideal portion size was: "This is a portion of *(food name auto-*

[26]The exact task wording was as follows: "Look at the portion *(food name auto-fills)* on the left. Imagine you are having this plate of food for a snack TODAY. Change the portion of food on the right using the slider so that both foods will leave you feeling foods will leave you feeling EQUALLY FULL."

fills). Please use the slider to select how much would you like to eat right now." The prompt for the maximum portion size was: "This is a portion of *(food name auto-fills)*. Please use the slider to select the maximum portion of food you could eat right now." In both tasks, the food name in the prompt would auto-fill to match the food name included as a label on the image. Participants completed the ideal portion size task before the maximum portion size task and the order of the foods was randomised.

Liking for each of the foods was assessed by asking participants to "Imagine taking a bite of this food and tasting it. How PLEASANT does this food taste in your mouth? When making this judgement, ignore how much or little of the food you want to eat, and what it would be like to swallow it – just focus only on how pleasant it would taste in your mouth right now." The anchors of the 100-unit VAS were 'Not at all' on the left and 'Extremely' on the right. This task has been used previously (Rogers & Hardman, 2015; Rogers et al., 2024), and the order of the foods was randomised. Participants rated all 24 stimuli as well as the milk chocolate M&Ms which were used as the standard food in the expected satiation task.

Familiarity with the 24 stimuli and the standard food was assessed by asking participants to select one of three options, 'Never', 'Rarely/Sometimes' or 'Often' in response to the question " How often have you eaten this food or a very similar food?" and this task wording is similar to a familiarity task used by Rogers et al. (2024). Food order was again randomised.

The study procedure was similar to the US and UK pilots, and participants began by reading the information sheet outlining that the purpose of the study was to better understand how food characteristics might impact how much food someone chooses to eat. They were also provided with a brief explanation of the expected satiation, ideal portion size, and maximum portion size tasks. Lastly, they were told that each trial would take approximately 10 seconds and that response times would be monitored. This was done to encourage careful responding.

After providing their informed consent, participants rated their hunger on a 100-unit VAS scale with the left anchor of 'Not at all' and the right anchor of 'Extremely' before being shown two practice trials of the expected satiation task. After completing the practice trials, they were reminded that this next section would comprise 24 trials and to not rush. Following the completion of the expected satiation task, they were notified that they would start the next task and would now be providing measures of their ideal portion sizes for each of the foods. After 24 trials, a pop-up notification appeared on the screen informing participants that the task had now changed and that they were to now select their maximum portion size. This was then followed by the liking and familiarity tasks before participants were shown their debrief information.

This study was advertised to take 20 minutes and participants received £3.00 in remuneration (£9.00 per hour rate). Again, participants were required to complete the study on a desktop computer. The median study completion time was 14 minutes and 38 seconds, and ethical approval was granted from the University of Bristol Science Faculty Ethics Committee (12904).

7.4.1.3 Statistical analysis

Following the pre-registered plan (identical to the statistical approach in the pilot studies, see section 7.3.3 for more details), a mean difference score representing the difference between the mean expected satiation (kcal) of foods in the high-fat category and the mean expected satiation of foods in the combination category was calculated for each participant[27]. This was then repeated for the high-carbohydrate category. Then, two one-sample *t*-tests comparing the two mean difference scores to zero were conducted. This two-step procedure was repeated for the ideal and maximum portion size tasks. Again, Cohen's *d* was reported as

[27]Note, the pre-registration reports that the mean difference scores (kcal) will be calculated in reverse (i.e., combination minus high-fat). The order was reversed to remain consistent with the two pilot studies. Reversing the calculation has no implications on its outcome.

a measure of effect size. All analyses were conducted in the R statistical environment (R Core Team, 2022).

7.4.2 Results

7.4.2.1 No differences in expected satiation (kcal) between macronutrient categories

Contrary to the pre-registered hypothesis, there appeared to be no effect of the macronutrient category on expected satiation (kcal) (see Table 7.6 for the expected satiation and mean difference scores (kcal) for each category and, for the expected satiation of each food, please see Appendix 4 Table 11.12). The results of two one-sample *t*-tests confirmed that the foods in the three categories did not differ in their expected satiation (one-sample *t*-test for the high-fat category, $t(32)= -0.55$, $p= 0.590$, Cohen's $d = -0.095$; one-sample *t*-test for the high-carbohydrate category, $t(32)= 0.27$, $p= 0.793$, Cohen's $d = 0.046$)[28], and this is counter to what had been hypothesized in the pre-registration.

Table 7.6 *Expected Satiation (kcal) and Mean Difference Scores (kcal)*[1,2]

Macronutrient category	Mean ± (SD)	Mean difference score ± (SD)
High-fat	435.98 (216.39)	-6.97 (73.48)
High-carbohydrate	447.50 (209.46)	4.55 (98.52)
Combination	442.95 (200.80)	0

[1]360 kcal images were shown to participants in the expected satiation task.
[2]The mean difference score (kcal) represents the difference in the mean expected satiation (kcal) between the high-fat category (or the high-carbohydrate category) and the combination category.

7.4.2.2 Combination foods are selected in larger ideal and maximum portions (kcal) than foods high in either fat or carbohydrate

A review of the mean differences for ideal portion size in Table 7.7 suggests that foods in the combination category were, on average, selected in larger portions than foods in the high-

[28]The results of a one-way ANOVA with repeated measures and Greenhouse-Geisser correction indicated that there were no differences in expected satiation across the three macronutrient categories ($F(1.698, 54.334)= 0.33$, $p= .688$), and the planned comparisons confirmed that there were no significant differences in expected satiation between foods in the combination category and foods in the high-fat (-6.97; 95% CI -33.02, 19.08; $p=.590$) and high-carbohydrate (4.55; 95% CI -30.39, 39.48; $p= .793$) categories.

fat or high-carbohydrate categories[29]. Two one-sample *t*-tests confirmed that these differences were significant (one-sample *t*-test for the high-fat category, $t(32) = -3.46$, $p = .002$, Cohen's $d = -0.603$; one-sample *t*-test for the high-carbohydrate category, $t(32) = -4.66$, $p < .001$, Cohen's $d = -0.811$)[30].

Table 7.7 *Portion Size (kcal) and Mean Difference Scores (kcal) for Ideal and Maximum Portion Selection Tasks*[1]

Portion size condition	Macronutrient category	Mean ± (SD)	Mean difference score ± (SD)
Ideal portion size			
	High-fat	181.97 (167.50)	-61.89** (102.68)
	High-carbohydrate	178.41 (191.57)	-65.45* (80.71)
	Combination	243.86 (197.52)	0
Maximum portion size			
	High-fat	347.58 (252.73)	–79.85** (133.39)
	High-carbohydrate	332.27 (255.95)	–95.15 ** (102.18)
	Combination	427.42 (272.49)	0

[1]The mean difference score (kcal) represents the difference in the mean portion size (kcal) between the high-fat category (or the high-carbohydrate category) and the combination category.
* $p < .01$, vs. Combination
** $p < .001$, vs. Combination

The pattern in maximum portion selection across the three macronutrient categories was similar to what was observed for ideal portion selection (see Table 7.7). Again, foods in

[29]Note, one outlier was identified as having a mean difference *Z*-score greater than 3.29. To check the sensitivity of the findings, the analyses were repeated after removing the outlier. The means and standard deviations ($M \pm SD$, kcal) are as follows: high-fat (178.59 ± 116.97), high-carbohydrate (177.11 ± 141.36), and combination (232.34 ± 138.78). The mean difference score between high-fat and combination was -53.75 ($SD = 92.87$) and the mean difference score between high-carbohydrate and combination was -56.27 ($SD = 56.27$). Two separate one-sample *t*-tests determined that both the high-fat ($t(31) = -3.27$, $p = .003$, Cohen's $d = -0.579$) and the high-carbohydrate ($t(31) = -5.52$, $p < .001$, Cohen's $d = -0.982$) foods had significantly smaller mean ideal portion sizes (kcal) than the combination foods.

For ideal and maximum portion size (kcal) for each of the 24 foods see Appendix 4 Table 11.13 and Table 11.14, respectively.

[30]There were significant differences in ideal portion size (kcal) across the three macronutrient categories as demonstrated by a one-way ANOVA with repeated measures, $F(2, 64) = 10.313$, $p < .001$. In the planned pairwise comparisons, foods in the combination category were selected in larger ideal portion sizes than foods in the high-fat (-61.89, 95% CI -98.30, -25.48; $p = .002$) and the high-carbohydrate (-65.46; 95% CI -94.07, -36.84; $p < .001$) categories.

the combination category were, on average, selected in significantly larger portions than foods in either the high-fat (*t*(32)= -3.44, *p*=.002, Cohen's *d*= -0.599) or high-carbohydrate category (*t*(32)= -5.35, *p*< .001, Cohen's *d*= -0.931)[31]. It should be noted that the patterns of calorie selection observed in both the ideal and maximum portion selection tasks are consistent with the pre-registered hypothesis.

7.4.2.3 Exploratory analysis: Food liking and familiarity

While not included in the pre-registration, differences in liking (taste pleasantness) and familiarity were also explored. A review of the mean differences (see Table 7.8) suggests that, on average, foods in the combination category were liked more than foods in the high-fat or high-carbohydrate categories (see Appendix 4 Table 11.15 for the liking of the 24 individual foods). The results of two one-sample *t*-tests confirmed that these differences were significant and that foods in the combination category were liked more than foods in either the high-fat (*t*(32)= -2.87, *p*= .007, Cohen's *d*= -0.500) or the high-carbohydrate (*t*(32)= -6.24, *p*< .001, Cohen's *d*= -1.086) categories.

Table 7.8 *Food Liking and Mean Difference Scores*

Macronutrient category	Mean ± (SD)	Mean difference score ± (SD)
High-fat	51.85 (33.34)	-12.13* (24.25)
High-carbohydrate	48.91 (29.91)	-15.07** (13.88)
Combination	63.98 (28.30)	0

[1]Responses were measured using a 100-unit visual analogue scale with the left anchor of 'Not at all' and the right anchor of 'Extremely'
* *p*< .01, vs. Combination
** *p*< .001, vs. Combination

[31]Again, the results of a one-way ANOVA with repeated measures indicated that maximum portion size (kcal) differed significantly across the three macronutrient categories, *F*(2, 64)= 11.940, *p*< .001. As with ideal portion size (kcal), foods in the combination category were selected in significantly larger maximum portions than foods in the high-fat (-79.85; 95% CI -127.15; -32.55; *p*= .002) and the high-carbohydrate (-95.15; 95% CI -131.39, -58.92; *p*< .001) categories.

The familiarity of the 24 foods and the standard food (Milk Chocolate M&Ms) is reported in Table 7.9. Of the 24 test stimuli, Dried apple slices, Dried pitted dates, Olives and Pate were the most unfamiliar (count > 10, indicating that at least 10 participants reported not eating it or a similar food in the last 6 months). However, more than half of the participants still reported having eaten these foods.

Table 7.9 *Food Familiarity*

Food item	Never (count)	Rarely/Sometimes (count)	Often (count)
Bagel	2	16	15
Blueberry muffin	2	19	12
Butter croissant	0	19	14
Cheddar cheese	0	6	27
Chocolate mousse	1	16	16
Crispbread	7	23	3
Custard	4	10	19
Dried apple slices	15	17	1
Dried pitted dates	13	15	5
Flapjack bites	3	19	11
Frankfurter sausage	8	19	6
Fruit pastilles	2	20	11
Milk chocolate M&Ms	1	18	14
Mozzarella cheese	2	15	16
Oatcakes	5	22	6
Olives	13	9	11
Pate	14	12	7
Pepperoni	3	17	13
Salted peanuts	3	14	16
Salted popcorn	2	15	16
Salted pretzels	5	19	9
Smashed avocado	9	14	10
Strawberry yogurt	2	9	22
Sultanas	6	20	7
Wine gums	4	22	7

7.5 Discussion - larger portions are selected of combination foods- an effect of satiation or liking?

The effects of combining fat and carbohydrate in a single food on expected satiation, portion size selection, and liking were explored across three different studies using snack foods and participants from two different countries (US and UK). Beginning first with expected satiation (kcal), there was inconsistent evidence that foods in the combination category were less satiating calorie per calorie than foods high in either fat or carbohydrate. The results from the US and UK pilot studies provided initial support for combination foods being less satiating; however, the second UK study failed to replicate this pattern, and the results suggested no macronutrient category (i.e., high-fat, high-carbohydrate or combination) differences in expected satiation. One possibility is that the slightly different wording of the expected satiation task in the second UK study could explain the failure to observe an effect of macronutrient category. However, as noted in the study methodology (see section 7.4.1.2), this task wording has been used in previous studies (Brunstrom et al., 2018), suggesting that this explanation is unlikely. Additional studies are needed to establish whether combination foods are reliably perceived to be less satiating than foods high in fat or carbohydrate.

With regards to portion selection (kcal), foods in the combination category were selected, on average, in larger portions than foods in either the high-fat or high-carbohydrate categories, and this was consistent across both the ideal and the maximum portion selection tasks. Lastly, foods in the combination category were, on average, liked significantly more than foods in either the high-fat or high-carbohydrate categories and this is consistent with other research (Rogers et al., 2024). As mentioned in section 7.4, the effect of combining fat and carbohydrate in a single food on food reward is hypothesized to largely be driven by an effect on liking (Rogers et al., 2024), and, on this basis, the increased liking of foods in the combination category could tentatively be interpreted as a replication of the increased reward of combination foods observed by DiFeliceantonio et al. (2018) and Perszyk et al. (2021). Follow-on studies could include a measure of food reward, such as desire-to-eat, alongside a measure of liking to establish whether the pattern similarity between liking and food reward observed by Rogers et al. (2024) is reliable.

As mentioned previously, foods in the combination category were selected, on average, in larger portions (kcal) than foods in either the high-fat or high-carbohydrate categories. Three possible explanations for this macronutrient category effect on portion selection include differences in 1) food liking, 2) variety and 3) expected satiation, and each will be discussed in turn. The effect of combining fat and carbohydrate in a single food on food liking replicates findings from Rogers et al. (2024) and aligns more generally with previous findings (DiFeliceantonio et al., 2018; Drewnowski et al., 1985; Drewnowski & Greenwood, 1983; Perszyk et al., 2021). Based on this, one explanation for the macronutrient category effect on portion selection is that because foods in the combination category were liked more, they were then selected in larger portions. However, the extent to which food liking is a consistent predictor of portion size is unclear. Some research, including in children, suggests that liking is a strong positive predictor of portion size (Brunstrom & Shakeshaft, 2009; Diktas et al., 2022) while other research indicates that liking is unrelated to the amount (kcal) of food someone would choose to eat for a hypothetical lunch (Brunstrom & Rogers, 2009). Relatedly, participants liking of a test food (pasta) has also been found to be a poor predictor of both hypothetical and actual food intake (Wilkinson et al., 2012). For now, the hypothesis that increased liking drove the increased portion selection of foods in the combination category remains speculative. A second potential explanation for the selection of larger portions of combination foods is the variety effect, including orosensory variety or complexity. Briefly, food intake is often increased when an individual has the opportunity to consume multiple foods with varying sensory characteristics, and this is commonly referred to as the 'variety effect’ (Rolls et al., 1981; Rolls et al., 1984). The increased intake when foods are varied is thought to be underpinned by an attenuation of sensory-specific satiety (i.e., delay in the reduction in pleasantness of the foods being eaten) (Rolls et al., 1981). Relatedly, research into complexity suggests that foods which are perceived to be more complex are more resistant to the development of sensory specific satiety (i.e., decline in pleasantness) (Weijzen et al., 2008). It is possible that the foods in the combination category were perceived as being more complex or varied (i.e., less susceptible to sensory-specific satiety) and were therefore selected in larger portions. Importantly, it should be noted that perceived variety or complexity as well as potential sensory-specific satiety was not assessed, and this should be considered in future studies.

Finally, it is also possible that larger portions of combination foods were selected due to differences in expected satiation. In the first two studies (US and UK pilots) of this chapter, foods in the combination category were, on average, expected to be less satiating than foods in the high-fat and high-carbohydrate categories. There is a good body of evidence demonstrating that foods which deliver less satiation per calorie are selected in larger portion sizes (for example - Brunstrom, Collingwood, et al., 2010; Brunstrom et al., 2018; Brunstrom & Rogers, 2009). Assuming this macronutrient category difference in expected satiation is true, then it is plausible that foods in the combination category were selected in larger portion sizes because they deliver less satiation per calorie compared to foods in the high-fat or high-carbohydrate categories. However, given the inconsistent effect of the macronutrient category on expected satiation reported earlier, more research is needed before stronger conclusions can be drawn.

In addition to conducting further studies exploring the potential effect of combining fat and carbohydrate on satiation, one could also consider alternative sources of evidence including separate pilot research conducted by the author and recent research by Rogers et al. (2024). A small-scale pilot[32] used the same UK foods as the third study and focussed on questions regarding post-ingestive sensations after the imagined consumption of these foods, building on similar research conducted by Mantzavinou and Rogers (2023). The results of this pilot study

[32]For reasons of brevity and to maintain the core research focus of this chapter, it was decided to not include this pilot work in greater detail.

suggest that foods in the combination category were rated as being less ‘sickly’ or ‘nauseating’ on a 100-unit VAS and that this effect of macronutrient category on sickliness was greater at larger portions (420 kcal and 720 kcal). In addition, across the three tested portion sizes (120 kcal, 420 kcal, and 720 kcal), foods in the high-fat, high-carbohydrate and combination categories did not differ in their perceived stomach fullness. In this context, it is hypothesized that sickliness or nausea provides another measure of satiety and relates to the potential aversiveness of single doses of a macronutrient (i.e., high-fat or high-carbohydrate) (Lucas et al., 1998; Moskowitz, 1971a; Moskowitz et al., 1974; Sclafani & Ackroff, 2004) and the avoidance of excess satiety or ‘nimiety’ (Kulkosky, 1985), especially at larger portions. Indeed, high doses of macronutrients have also been linked to feelings of sickliness, and participants who consumed unusually concentrated doses of maltodextrin (i.e., high-carbohydrate) provided higher ratings on the item ‘feeling sick/nauseous’ as compared to a control food item (Booth et al., 2011). It is therefore plausible that combination foods are perceived to be less ‘sickly’ or ‘nauseating’ at large portions because they are less satiating. This relative satiety ‘advantage’ enables them to be consumed in larger portions, as evidenced by the ideal and maximum portion size results, without providing excess satiety[33].

Secondly, a recent re-analysis of data from the Satiety Index study conducted by Holt et al. (1995) and a similar study by Merrill et al. (2004) provides additional evidence that combination foods are perceived to be less satiating (Rogers et al., 2024). Briefly, in these two

[33]As in Rogers and colleauges’ manuscript [a], the author notes that satiation and satiety have been used interchangeably both here and in previous text. Research suggests that these two variables are highly correlated[b]. Adopting Rogers and colleagues’ description, in the context of this chapter, these terms are not intended to refer to temporally specific sensations (i.e., satiation occurring within a meal and satiety occurring after a meal)[a].

[a]Rogers, P. J., Vural, Y., Berridge-Burley, N., Butcher, C., Cawley, E., Goa, Z., Sutcliffe, A., Tinker, l., Zen, A., Flynn, A. N., Brunstrom, J. M., & Brand-Miller, J. C. (under-review). Evidence that energy-to-satiety ratio and taste, but not NOVA level of processing are determinants of food liking and reward value

[b]Wilkinson, L. L., Hinton, E. C., Fay, S. H., Ferriday, D., Rogers, P. J., & Brunstrom, J. M. (2012). Computer-based assessments of expected satiety predict behavioural measures of portion-size selection and food intake. *Appetite*, *59*(3), 933-938. https://doi.org/10.1016/j.appet.2012.09.007

studies, the perceived satiety of a range of different foods was assessed every 15 minutes for two hours and these values were compared using a 'Satiety Index'. The index provides a methodology to compare the satiating capacity of a food relative to an equicaloric portion of a reference food (in this case white bread). Foods which have a higher Satiety Index score are perceived to be more satiating (Holt et al., 1995; Merrill et al., 2004; Rogers et al., 2024). For the re-analysis, Rogers and colleagues correlated the Satiety Index values and the transformed carbohydrate-to-fat ratio and established that combination foods were less satiating (had lower Satiety Index values) than foods high in either fat or carbohydrate[34].

Rogers and colleagues then went on to link the reduced satiety of combination foods to their increased reward value and proposed that combination foods are more rewarding because they have a high 'energy-to-satiety ratio'. Research suggests that fats and carbohydrates are digested and absorbed largely separately by the gut (Frayn & Evans, 2019). Therefore, the gut's digestive capacity might be less readily overwhelmed when foods comprise a more equal mixture of calories from fat and carbohydrate (i.e., these foods are less satiating). Therefore, combination foods are more valued or more rewarding due to their blend of macronutrients which provide a higher energy-to-satiety ratio and enable individuals to consume more calories for an equivalent level of satiety (Rogers et al., 2024). This provides one potential explanation for why combination foods are selected in larger portions (kcal), and this hypothesis could be further tested in a flavour-nutrient learning study comprised of three conditions (high-fat, high-carbohydrate and combination) and four main outcomes (liking, expected satiation or satiety, portion size selection, and ad libitum intake).

In summary, compared to foods which are high in either fat or carbohydrate, foods which comprise a combination of fat and carbohydrate are more liked and selected in larger

[34]Importantly, in the two datasets which were re-analysed, energy density might have confounded this association as it correlated with both Satiety Index and the transformed carbohydrate-to-fat ratio.

ideal and maximum portions (kcal). Based on the findings presented in this chapter, the extent to which combining fat and carbohydrate consistently influences judgments of satiation, as measured using the method of adjustment, is unclear. However, when including other sources of evidence, foods containing a combination of the two macronutrients are potentially less satiating. On this basis, combination foods may be selected in larger portions because they are less satiating, and additional research is needed to further test this hypothesis.

7.5.1 Linking current findings to the theoretical two-component model of meal size

Briefly, the theoretical two-component model of meal size (g) suggested that two signals, a volume signal and a calorie-content signal, provided feedback in response to meal energy density to help guide meal size and prevent the short-term aversive effects of overconsuming calories (e.g., nimiety (Kulkosky, 1985) or malaise (Hengist et al., 2020), see section 5.3). In section 5.3, it was also noted that the calorie-content signal had been conceptualised only in relation to the energy content of foods. This conceptualisation ignores the possibility that feedback from the calorie-content signal could be influenced by the macronutrient composition of the foods. The results from the current chapter suggest that when holding energy density constant, macronutrient composition (i.e., high-fat, high-carbohydrate, or a combination of fat and carbohydrate) can influence meal size (kcal). It is therefore plausible that feedback from the calorie-content signal might be weaker in foods containing a combination of fat and carbohydrate as compared to foods high in either fat or carbohydrate and that this results in more calories being selected. It is important to note that this is speculative and further research exploring how macronutrient composition might impact feedback from the calorie-content signal is needed.

7.5.2 Limitations and possibilities for future research

As in chapter six, all studies reported in this chapter were conducted in an online setting where experimental control is limited, and it is difficult to know whether participants were fully focused during the entire study duration. Study completion time provides a metric by which to potentially gauge whether participants were responding with care, and a review of these times for the three studies did not suggest that participants were carelessly responding. Additionally, as all the studies were online, participants were asked to provide ratings based on the imagined consumption of the various stimuli as well as select hypothetical ideal and maximum portion sizes. External validity could be increased by inviting participants into the lab and having them self-serve actual portions. However, moving to in-person data collection significantly increases research time and may not be a substantial concern as online estimates of portion size are validated proxies of real-world self-served portions (Wilkinson et al., 2012).

Another study limitation is that the foods used as stimuli differed across other dimensions known to influence judgments of expected satiation or portion size, such as sensory characteristics (Ferriday et al., 2016; Forde & de Graaf, 2022). Therefore, future research should consider better matching the foods for these characteristics. Similarly, the stimuli were all single food items, and the extent to which the effect of combining fat and carbohydrate on portion selection might extend to meals is unknown and should be explored further. Another food specific limitation is the use of milk chocolate M&Ms as a 'standard food' in the expected satiation task. Research shows that familiarity with eating a food to fullness can influence judgments of expected satiety (Irvine et al., 2013), and it is unlikely that M&Ms have been eaten until satiation. In turn, this lack of familiarity with eating M&Ms to satiety could have impacted how participants perceived these foods when they were responding with them in the expected satiation task. When designing future studies, familiarity with eating a food to fullness should be taken into account when selecting the specific stimuli to use as the 'standard food'.

To address some of the above concerns and to further explore the potential association between fat and carbohydrate content, expected satiation or satiety, liking, and portion size selection, one could conduct a flavour-nutrient learning study as suggested earlier in section

7.5). This approach would enable researchers to explore the extent to which the gut might detect differences in macronutrient composition, and whether, after repeated exposure, these differences influence judgments of expected satiation or satiety, portion size selection, liking and food reward, among others. As previously mentioned in chapter five, it is important to note that laboratory studies often fail to demonstrate successful flavour-nutrient learning in humans (Yeomans, 2012), potentially because they rely on self-learning and preclude participants from utilising their 'nutritional intelligence' developed through collective social learning (Brunstrom et al., 2023). Therefore, the outcomes of controlled flavour-nutrient learning studies should be viewed alongside evidence from studies using real-world foods (as in this chapter) to provide a more complete understanding. Lastly, while not included in this chapter, individual differences, such as BMI, could also be explored as these appear to impact how rewarding or preferable an individual finds different combinations of fat and carbohydrate (Drewnowski et al., 1985; Perszyk et al., 2021)

7.5.3 Chapter summary

The research presented in this chapter was, to the best of the author's knowledge, the first to expand on findings suggesting that foods containing a combination of fat and carbohydrate are more rewarding and explore whether this macronutrient composition also influences expected satiation (kcal) and portion size selection (kcal). The results suggest that combination snack foods might be less satiating and demonstrated that these combination foods were also selected in larger ideal and maximum portions compared to foods high in either fat or carbohydrate. Should an individual frequently consume larger portions of combination foods, then this suggests that these foods have the potential to play a role in the individual's chronic energy balance. On this basis, foods combining fat and carbohydrate present themselves as potential targets for food reformulation aiming to reduce population-level daily energy intake, and this is reviewed in the general discussion (chapter ten). The following chapter is an interim discussion of Part A (chapters two through seven and provides an introduction to the final study in this thesis (chapter nine).

Chapter 8 Interim summary of Part A

The following chapter presents an interim summary of the first part of this thesis. It begins by briefly reviewing the main findings of the previous six chapters (chapters two through seven) before discussing whether the findings provide additional evidence for human nutritional intelligence. The chapter concludes with a short introduction to the second part of the thesis (chapter nine) which explores whether eating contexts influence body mass index in young adults.

8.1 Summary of findings from Part A and additional evidence for human nutritional intelligence

The studies in the first part of this thesis explored whether humans are sensitive to food composition (energy density (chapters two through five) and macronutrient composition (chapters six and seven)) and whether this might impact food choice and energy intake. The findings from chapter two demonstrated a non-linear association between meal caloric intake and meal energy density in meals consumed in a controlled environment, and this non-linear pattern was replicated in data from free-living participants in the UK (chapter three), Argentina (chapter four), and Malaysia (chapter four). The increase in meal caloric intake in response to increasing meal energy density in lower energy-dense meals (those below the first breakpoint) and decrease in meal caloric intake in higher energy-dense meals (those above the first breakpoint) was captured in a theoretical two-component model of meal size (g) (chapter five). This model suggests that meal size is guided by a volume signal that is dominant in lower energy-dense meals and a calorie-content signal that dominates in higher energy-dense meals.

The results from chapters six and seven demonstrate that humans prefer a blend of macronutrients (i.e., protein paired with carbohydrate (chapter six) and single foods comprising more equal amounts of fat and carbohydrate (chapter seven)). The findings from chapter six suggest that humans might express a sensitivity to food protein content (negative association between desire-to-eat and protein content), and the results from chapter seven suggest that the amount of fat and carbohydrate in a food influences portion size selection (kcal), potentially via expected satiation.

More broadly, the results from Part A lend further support to the idea of ‘nutritional intelligence’, a concept which reflects humans’ ability to differentiate foods based on their nutritional composition and make advantageous decisions on this basis (Brunstrom et al., 2023; Brunstrom & Schatzker, 2022). Specifically, the results from chapters two, three, and four suggest a degree of nutritional intelligence in response to the calorie content of everyday, non-manipulated meals. In these data, individuals exhibited a sensitivity to differences in meal energy density, across a broad range of energy densities, and adjusted the amount of food they consumed accordingly to prevent the aversive effects of acute overconsumption (chapter five). In chapters six and seven, there was evidence for non-random behaviour in response to the macronutrient composition of everyday foods suggesting that people might discriminate foods based, in part, on their macronutrient composition. One key feature of all the studies in Part A is the use of real-world foods which were not manipulated in any way. By using these foods, the likelihood of capturing sensitivity to food composition might have been increased as it allowed individuals to tap into the wealth of information associated with these foods, acquired through generations of collective learning transmitted through social interaction, rather than forcing them to rely solely on learning acquired at an individual level (Brunstrom et al., 2023).

Importantly, the studies presented in Part A are not definitive and further work is needed to address the limitations mentioned in each chapter before stronger conclusions can be drawn. Additionally, if human nutritional intelligence exists, then this raises broader questions about potential mechanisms (e.g., social and individual learning and a role for cuisine) as well as whether individuals might differ in their ability to express nutritional intelligence and the extent to which this might contribute to overweight or obesity (Brunstrom et al., 2023). Lastly, and perhaps most importantly, if nutritional intelligence is a universal human ability, then the findings reported on in this thesis must be further evidenced in different populations from diverse culinary backgrounds. To an extent, this was achieved in the chapters exploring human sensitivity to meal energy content (specifically chapter four), but this remains a challenge for future research.

The research conducted in part A was positioned as a key question related to the efficacy of food reformulation as a strategy to improve population health. The concept of nutritional intelligence, further evidenced by the results presented in Part A, presents both a challenge for the design of reformulated products and an opportunity to better understand factors which might impact the sustained acceptance of reformulated foods or plant-based meat alternatives and this will be discussed in further detail in the general discussion (chapter ten).

8.2 Part B: the impact of eating contexts on body mass index and weight status in young adults

The studies in Part A focussed entirely on whether the composition of foods or meals impacts behaviour. Importantly, these studies ignored the eating context or immediate environment in which meals are consumed (e.g., eaten socially, eaten whilst reading etc). In 1996, Paul Rozin wrote that of the 15 billion meals consumed each day (five billion people multiplied by an average of three meals a day) it is almost guaranteed that the majority of those meals were consumed in a social setting (i.e., eaten with someone else) (Rozin, 1996). This statement highlights the potential for eating contexts, specifically social eating, to have a substantial influence on eating behaviour, and the roles of two eating contexts are described in further detail in the next paragraph.

As mentioned in the general introduction (chapter one), eating contexts (i.e., the setting in which individuals eat) are potential targets for public health strategies aimed at encouraging healthier dietary behaviours (Mak et al., 2012; Rauber et al., 2022). Indeed, eating contexts have been recognised as important factors relating to food choice and food acceptance (Borbon-Mendivil et al., 2022; Meiselman, 1996; Rozin & Tuorilla, 1993), diet quality (Mak et al., 2012; Rauber et al., 2022) as well as meal size (Porter et al., 2021). With regard to meal size, two eating contexts, specifically social eating (Ruddock et al., 2019) and distracted eating (Robinson et al., 2013), have been found to increase energy intake. Importantly, the majority of the research exploring whether eating contexts influence meal size utilises acute, often laboratory-based, studies (Robinson et al., 2013; Ruddock et al., 2019). In contrast, there are only a few studies which have explored whether eating contexts have a chronic impact on an individual's health (i.e., body mass index) (Tumin & Anderson, 2017; van Meer et al., 2022). Importantly, evidence indicating that these eating contexts might have a chronic impact is needed to support recommendations for related public health strategies. Therefore, the study presented in Part B (chapter nine) explored the potential impact of eating contexts, specifically social and distracted eating, on body mass index and weight status in young adults.

Chapter 9 Quantifying the impact of social and distracted eating on body mass index and weight status in young adults using the Avon Longitudinal Study of Parents and Children (ALSPAC)

9.1 Acknowledgements and overview

Due to the COVID-19 pandemic and related lockdowns, the author's laboratory research was paused in March 2020 for an, at that time, unknown period of time. Following this, the author had the opportunity to collaborate with Professor Julian Hamilton-Shield (NIHR Bristol Biomedical Research Centre: Diet & Physical Activity Theme, University Hospitals Bristol and Weston NHS Foundation Trust and University of Bristol), Dr Elizabeth Schneider (at the time University of Birmingham), Professor Suzanne Higgs (University of Birmingham), Professor Nicholas Timpson (University of Bristol) and Professor Jeff Brunstrom (University of Bristol, supervisor). This study in this chapter has been prepared for submission to the British Journal of Nutrition, and this chapter is largely presented as the prepared publication, but minor changes have been made to improve readability and a few sentences have been added regarding a role for Ecological Momentary Assessment (section 9.5.2). The author was responsible for data acquisition and analysis, interpretation of results, and writing the manuscript, and the author will be responsible for submitting the manuscript to the journal. All other co-authors provided feedback on the interpretation of the results and manuscript text.

As noted in section 8.2, the research in Part A ignored the potential for the immediate eating context to influence energy intake and food choice. Therefore, the study presented in this chapter sought to understand whether two eating contexts which are known to influence acute energy intake (Robinson et al., 2013; Ruddock et al., 2019) might also impact chronic energy balance as measured by BMI and weight status. The chapter follows the structure of a standard, single-study publication beginning first with the introduction (the next section) followed by the methods and results before ending with the discussion.

9.2 Introduction

To tackle obesity, public health interventions have largely targeted food choices (e.g., increasing the consumption of fruit and vegetables and reducing the intake of high-fat and high-sugar foods) and physical activity (Kumanyika et al., 2010). However, traditional behaviour-based weight loss programmes tend to report only modest weight improvement (Avenell et al., 2004), which is often not maintained (Kraschnewski et al., 2010), and participants cite difficulties in self-regulation and modifying eating habits as barriers to treatment (Hammarström et al., 2014). In parallel, eating contexts such as social eating (eating in the presence of others) and eating whilst distracted (e.g., eating whilst watching TV) are also associated with intake (De Graaf & Kok, 2010), but associated recommendations have not been widely incorporated into public health interventions.

Eating with others has been shown to increase energy intake, an effect coined 'social facilitation of eating'. Early work by de Castro and Brewer using diet diary data suggested that an individual's meal size increased as the number of people present at the meal increased (de Castro & Brewer, 1992). The role of social facilitation as a driver of energy intake has been further evidenced by the results of a systematic review and meta-analysis which demonstrated that individuals tend to serve and consume more food when eating in a social environment than when eating alone, especially when a person eats with someone they know well (Ruddock et al., 2019). Likewise, distracted eating increases meal intake (Robinson et al., 2013) and reduces the degree of satiation reported at meal end (Oldham-Cooper et al., 2011). Importantly, the effects of social eating and distraction on meal intake are large and are broadly comparable (respectively, causing an 18% and 14% increase (Hetherington et al., 2006)).

Together, these studies suggest that social eating and distraction can influence food intake in a single meal. However, their chronic impact remains unclear. Beginning more generally, there is evidence that high screen use is associated with higher BMI in children and adolescents (Wu et al., 2022), and in a recent meta-analysis, Ghobadi et al. (2018) found that children and adolescents who watch TV whilst eating are more likely to be overweight. However, only two studies have explored a similar association in adults. One showed that people who watch TV during their family meals are more likely to be overweight (Tumin & Anderson, 2017), and the other reported that snacking or eating lunch whilst watching TV is associated with having a higher BMI (van Meer et al., 2022). Similarly, studies have considered whether adults with obesity are more or less sensitive to the influence of social eating (Krantz, 1979; Maykovich, 1978); however, the association between daily social eating and body mass index (BMI) is yet to be explored.

To address these questions, data from the Avon Longitudinal Study of Parents and Children (ALSPAC) were used to cross-sectionally assess the frequency of social and distracted eating in young adults. This ongoing birth-cohort study is based in Bristol (England) and, at the time of data collection, comprised young adults (~25 years of age), which is ideal because substantial weight gain is typically observed around this period (Cheng et al., 2016; Reas et al., 2007; Sheehan et al., 2003; Williamson et al., 1990). Weight gain in early adulthood has been associated with an increased risk of chronic disease in later life (Truesdale et al., 2006; Zheng et al., 2017) and with increased mortality (Chen et al., 2019). Additionally, exploring these associations is vital as young adults develop independence and transition to form health-related habits and behaviours (Nelson et al., 2008; Poobalan et al., 2009). Therefore, interventions that target this population could yield life-long benefit.

With this opportunity in mind, it was hypothesized that in young adults, the tendency to consume meals socially would be associated with having a higher BMI or BMI category (individuals with normal weight versus overweight/obese) (analyses pre-registered on the Open Science Framework: https://doi.org/10.17605/OSF.IO/Y2HXW). In the final analyses, both social eating and eating whilst distracted are focussed on as potentially modifiable eating contexts. Additionally, these associations were benchmarked against other variables known to positively associate with anthropometric measures (i.e., BMI), such as eating rate (Ohkuma et al., 2015) and eating traits (disinhibition, rigid restraint, and flexible restraint) (Finlayson et al., 2012; Provencher et al., 2003). Social eating and eating whilst distracted were of particular interest because of their potential to inform relatively simple dietary advice relating to everyday eating contexts. As such, they contrast other potentially unsustainable dietary guidelines that rely on conscious food restriction (e.g., dieting) or longer-term modifications to food choice (Hammarström et al., 2014).

9.3 Materials and methods

9.3.1 Study population

Data were obtained from the Avon Longitudinal Study of Parents and Children (ALSPAC) birth cohort in which pregnant women were recruited in the early 1990s (Boyd et al., 2013; Fraser et al., 2013; Northstone et al., 2019). 14,541 mothers were recruited into the study, and the sample size of the entire cohort comprised 14,676 foetuses and 14,062 live births of which 13,988 children were alive at one year of age (for more information see: http://www.alspac.bris.ac.uk). When the oldest children in this initial cohort were approximately seven years old, researchers attempted to contact those individuals who might be eligible for inclusion but had not been part of the original cohort (total sample size after this second enrolment: 15,589 foetuses of which 14,901 children were alive at one year of age). This study utilises data already collected, but ALSPAC longitudinal data collection is ongoing.

This paper focuses on young adults who completed the Life@25 questionnaire between November 2017 and July 2018. Study data were collected and managed using REDCap electronic data capture tools hosted at the University of Bristol (Harris et al., 2009). REDCap (Research Electronic Data Capture) is a secure, web-based software platform designed to support data capture for research studies. Participants whose BMI was measured more than two years prior to submitting the Life@25 questionnaire which, by default, included those who did not have their BMI measured (*n*=13,193) were excluded. Additionally, individuals whose BMI at age 24 was under 19 or over 40 were excluded, as it is more likely that their weight status can be attributed to clinical disease rather than lifestyle factors (*n*= 196). Additionally, participants who self-reported having been treated for an eating disorder (*n*= 382), and any individuals who identified as being pregnant when their BMI was measured (either at 17 years or 24 years) (*n*= 8) were excluded. The final sample comprised 1,866 individuals (male, *n*= 653; female, *n*= 1,212, see Appendix 5 Figure 11.13 for participant flow chart); however, depending on missing data for respective predictors, the reported sample size may differ across analyses. Ethical approval for the main study was obtained from the ALSPAC Law and Ethics Committee and informed consent for the use of data collected via questionnaires and clinics was obtained from participants following recommendations of the ALSPAC Ethics and Law Committee at the time.

9.3.2 Variables[35]

9.3.2.1 Dependent variable

BMI was calculated by dividing weight (kg) by height squared (m^2) and was treated as either a continuous or as a binary variable (individuals with normal weight (19.00 - 24.99 kg/m^2), individuals with overweight/obesity (25.00 - 39.99). These measures were collected by a trained research team member during a clinical session when the participants were approximately 24 years of age.

[35]Please note that the study website contains details of all the data that is available through a fully searchable data dictionary and variable search tool: http://www.bristol.ac.uk/alspac/researchers/our-data/

9.3.2.2 Possible predictors of BMI or weight status

Potential predictors, which are described further below, were derived from previous cross-sectional studies which explored eating behaviours or BMI/weight status (Molyneaux et al., 2016; Ohkuma et al., 2015; Tumin & Anderson, 2017), or had been included in laboratory-based studies of eating behaviour (Hetherington et al., 2006).

9.3.2.3 Eating context related questions

Three questions in the Life@25 questionnaire pertained to the respondent's social eating behaviour over the past 7 days and asked how many times they 'Eat their main meal of the day by themselves?', 'Eat their main meal of the day with family/friends?', and 'Eat their main meal of the day with others (strangers/acquaintances)?' (Never, 1-2 times, 3-4 times, 5-6 times, 7+ times). Given that these questions ask respondents to recall main meal eating events, there is a possibility that an individual may consume less than 7 main meals per week. Therefore, a social eating index was calculated to relate socially eaten main meals with friends and family to all possible main meals (summed total of all main meals reported) resulting in the following proportion: socially eaten main meals with friends and family/total main meals consumed.

Four questions assessed different types of distracted eating, specifically the frequency with which participants 'watched TV whilst eating', 'used a computer/tablet, read or worked whilst eating', 'played video/computer games whilst eating', or 'sat at a table with no distractions whilst eating' during the past 7 days (Never, 1-2 times, 3-4 times, 5-6 times, 7+ times).

9.3.2.4 Eating behaviour related questions

Meal duration was measured by asking respondents how long their typical main meal lasts, and response options increased in five-minute intervals from less than five minutes to more than 40 minutes.

Eating rate was established by respondents self-reporting their eating rate compared to others using the following response options: very slow, slow, average, fast, and very fast.

9.3.2.5 Eating trait related questions

Disinhibited eating (disinhibition) was assessed using questions from the 51-item three-factor eating questionnaire (TFEQ) (Stunkard & Messick, 1985) and is described as a tendency to occasionally overeat (Westenhoefer et al., 1994; Wilkinson et al., 2010) and an inability to continuously exhibit dietary restraint (Wilkinson et al., 2010). Flexible and rigid restraint subscales from the 51-item TFEQ were used as a proxy for restraint as only 14 out of the 21 original dietary restraint questions were included in the Life@25 questionnaire (Westenhoefer, 1991). Rigid restraint is described as an 'all-or-nothing' dichotomised eating behaviour pattern, whereas flexible restraint is a more tempered attitude towards eating (Westenhoefer, 1991).

9.3.2.6 Lifestyle and socioeconomic factors[36]

To avoid substantial data loss due to dataset structure, both smoking habits and alcohol consumption were recoded. Respondents' smoking habits were recoded into a binary variable of smoking at least once a week. Participants who reported not smoking in the last 30 days or having never smoked a whole cigarette were recoded as having not smoked in the last week. Alcohol consumption was also quantified: 'In the past year, number of units drunk on a typical day when drinking' and was recoded to include participants who reported having never had a whole drink or not having drunk alcohol in the last year. These two questions were taken in clinic when the participant was approximately 24 years of age.

Take-home income was assessed in the Life@25 questionnaire using the following question 'What is your total take-home pay each month (after tax and national insurance are

[36]Physical activity was pre-registered as a possible covariate; however, only 552 participants had physical activity data. Due to an unacceptable loss of data, physical activity is not included as a possible predictor.

Education level was also pre-registered as a possible covariate; however, the structure of the dataset would have resulted in substantial loss of data. Additionally, education level was thought to covary highly with take-home income, so the decision was made to only include take-home income in the analyses.

removed as appropriate)? If possible, please refer to a recent payslip. If this is not possible, please estimate. If irregular work, please give an average per month.'. Response options included: £1 - £499, £500 - £999, £1,000 - £1,499, £1,500 - £1,999, £2,000 – £2,499, £2,500 - £2,999, £3,000 and above, and not doing paid work.

9.3.2.7 Individual differences

The respondent's sex was ascertained using the data gathered at their birth and was coded as a binary variable. Ethnicity was assessed per the child's ethnicity and coded as a binary variable of 'white' and 'non-white ethnicity.' Participant age was recorded at the clinic visit during which BMI, smoking, and drinking measures were obtained.

9.3.2.8 Baseline BMI

Baseline BMI was operationalised using the participants' BMI at 17 years of age which was measured by a trained research team member during a clinic visit.

9.3.3 Statistical analyses

The association of social eating with BMI was assessed using a linear regression and potential predictors were identified using a Pearson correlation (*r*). All significant potential predictors were entered simultaneously into the linear model using forced entry. In this context, predictors are interpreted in isolation (e.g., when all other predictors are held constant).

To allow for non-linear associations between social eating and BMI, BMI was also treated as a dichotomised variable, individuals with normal weight vs individuals with overweight/ obesity. Consistent with the pre-registration, binary logistic regression was used to compare a group with normal weight (*n*= 1,168) with a combined group comprising individuals with overweight (n= 472) and obesity (n= 223). Here, predictors were selected and entered if they were found to be independent predictors of weight status (individuals with normal weight vs individuals with overweight/obesity) using ANOVA or Chi-Square, depending on the class of variable. All statistical analyses were performed using SPSS software version 26.0 (Corp, 2019) and the threshold for significance was set at $p < 0.05$.

9.4 Results

The sample characteristics of possible predictors are depicted in Appendix 5 Table 11.16 and Table 11.17.

9.4.1 BMI increases with more frequent TV watching whilst eating

Social eating did not correlate with BMI at age 24 and was not included as a possible predictor in the linear regression. However, other possible predictors included baseline BMI at age 17, 'eating traits' (rigid restraint and disinhibition), 'lifestyle and socioeconomic factors' (smoking and take-home income), 'eating contexts' (watching TV whilst eating, playing computer/video games whilst eating, sitting at the table with no distractions), and 'eating behaviours' (self-reported eating rate) (see Appendix 5 Table 11.18 for correlation coefficients). Accordingly, these predictors were entered into a subsequent linear regression. The full model predicted 58.3% of the total variance in BMI at 24 ($F(9, 1{,}330) = 207.93$, $p < .001$), and, of the eating contexts included, individuals who reported more frequent TV watching whilst eating had a higher BMI (see Table 9.1 for beta weights and 95% confidence intervals). Eating whilst playing computer/video games was not associated with BMI. When holding the influence of all other predictors constant[37], eating whilst watching TV had roughly equivalent power in predicting BMI as disinhibition or rigid restraint. Of the remaining predictor groups, baseline BMI at age 17 was the strongest positive predictor of BMI at 24.

[37]Eating whilst watching TV also remains a significant predictor of BMI at 24 years when baseline BMI at 17 years is removed as a predictor.

Table 9.1 *Standardised Beta Weights (95% CIs) of Each Variable Included in Linear Regression Predicting Continuous BMI at 24 Years*

Predictor classification	Variable	β (B, *95% CI*), *p*
Baseline BMI		
	BMI at 17 years	0.71 (0.88, *0.83 - 0.92*), *p* < .001
Eating context		
	Watching TV while eating	0.07 (0.24, *0.10 - 0.37*), *p* < .001
	Playing computer/video games while eating	-0.01 (-0.09, *-0.43 - 0.25*), *p* = .598
	Sitting at the table with no distractions	0.01 (0.05, *-0.09 - 0.18*), *p* = .505
Eating behaviour		
	Self-reported eating rate	0.02 (0.09, *-0.09 - 0.27*), *p* = .331
Eating traits		
	Rigid restraint	-0.08 (-0.20, *-0.28 - -0.11*), *p* < .001
	Disinhibition	0.13 (0.14, *0.10- 0.19*), *p* < .001
Socioeconomic and lifestyle		
	Smoking status	-0.03 (-0.37, *-0.83 - 0.08*), *p* = .105
	Take-home income	-0.04 (-0.13, *-0.25 - -0.02*), *p* = .025

9.4.2 A faster self-reported eating rate increases the likelihood of an individual having overweight or obesity

No nominal potential predictors (e.g., smoking status, ethnicity, or sex) were established for inclusion in the logistic regression. Additionally, social eating did not meet the threshold for inclusion; however, BMI at age 17, take-home income, watching TV or playing computer/video games whilst eating, eating rate, rigid restraint, and disinhibition were included as continuous predictors of whether an individual had overweight or obesity (binary outcome, normal vs overweight/obesity). The final potential predictors included baseline BMI at age 17, ‘lifestyle and socioeconomic factors’ (take-home income), ‘eating traits’ (rigid restraint and disinhibition), 'eating context' (eating whilst watching TV and eating whilst playing computer games) and 'eating behaviour' (eating rate).

The analyses suggest that rigid restraint, disinhibition, take-home income, self-reported eating rate, and baseline BMI are associated with an individual having overweight/obesity (Chi-square= 633.85, df= 7, $p < .001$, see Table 9.2 for odds ratios and confidence intervals for predictors). Overall, the final model accurately classified 80.5% of individuals as having either normal weight or overweight/obesity. Eating whilst watching TV and playing computer/video games whilst eating did not meet the threshold for significance, but both increased the likelihood of an individual having overweight/obesity. Similar to the linear regression, baseline BMI at age 17 had the largest impact on whether an individual had overweight/obesity at age 24; participants with a higher baseline BMI were 1.80 times the odds to have overweight/obesity. Lastly, while not identified in the linear regression, individuals who self-reported a faster eating rate were 1.29 times the odds to have overweight/obesity.

Table 9.2 *Odds Ratios (and 95% CIs) of Each Variable Predicting Weight Status at 24 Years*[1]

Predictor classification	Variable	*OR (95% CI), p*
Baseline BMI		
	BMI at 17 years	1.80 (*1.67 – 1.93*), $p < .001$
Eating context		
	Watching TV while eating	1.09 (*0.97 – 1.23*), $p = .157$
	Playing computer/video games while eating	1.08 (*0.78 – 1.51*), $p = .633$
Eating behaviour		
	Self-reported eating rate	1.29 (*1.07 – 1.55*), $p = .008$
Eating traits		
	Rigid restraint	0.88 (*0.80 – 0.96*), $p = .004$
	Disinhibition	1.09 (*1.04 – 1.14*), $p < .001$
Socioeconomic and lifestyle		

Take-home income	0.88 (*0.79 – 0.99*), p = .029

[1]Weight status was a binary outcome, normal vs overweight/obesity

9.4.3 Exploratory analyses

Based on the results, it was decided to further explore the unique contribution of watching TV whilst eating on an individual's BMI at age 24 as this was the only eating context which was significantly associated with BMI. Specifically, the aim was to capture the importance of watching TV whilst eating after accounting for all the variance associated with the previously identified predictors, 'eating trait', 'lifestyle and socioeconomic', 'eating behaviour', and baseline BMI. To that end, a two-step hierarchical linear regression was completed in which the variance associated with the 'eating trait', 'eating behaviour', 'lifestyle and socioeconomic', and baseline BMI predictors were included in the first step and watching TV whilst eating was included in the second step. The effect size of including watching TV whilst eating was assessed using Cohen's f^2 (Cohen, 1998; Selya et al., 2012).

The variables included in the first model accounted for 58.0% of the variance in BMI at age 24 ($F\Delta$: $F(6, 1{,}329)$= 308.37, p< .001, Table 9.3) and all predictors except smoking status and eating rate were significant. The addition of watching TV whilst eating into the regression in step 2 resulted in a significant, small increase in total variance explained to 58.4% ($F\Delta$:$F(1, 1{,}328)$= 12.00, p< .001, Cohen's f^2= 0.010, Table 9.3). There was no change in the significance of the predictors included in Step 1 after including watching TV whilst eating, and watching TV whilst eating was an independent, positive predictor of BMI.

Table 9.3 *Summary of Hierarchical Regression Assessing Independent Contribution of Watching Television while Eating in Predicting BMI at 24 Years*

Variable	β (B, *95% CI*), *p*	Unadjusted R^2	Adjusted R^2	ΔR^2
Step 1		0.582	0.580	0.582
BMI at 17 years	0.71 (0.88, *0.83 - 0.92*), $p < .001$			
Self-reported eating rate	0.02 (0.08, *-0.11 - 0.26*), $p = .417$			
Rigid restraint	-0.08 (-0.19, *-0.28 - -0.10*), $p < .001$			
Disinhibition	0.13 (0.15, *0.11 - 0.20*), $p < .001$			
Take-home income	-0.04 (-0.14, *-0.26 - -0.03*), $p = .016$			
Smoking	-0.03 (-0.41, *-0.86 - 0.04*), $p = .076$			
Step 2		0.586	0.584	0.004
BMI at 17 years	0.71 (0.87, *0.83 - 0.92*), $p < .001$			
Self-reported eating rate	0.02 (0.09, *-0.09 - 0.27*), $p = .330$			
Rigid restraint	-0.08 (-0.19, *-0.28 - -0.11*), $p < .001$			
Disinhibition	0.13 (0.14, *0.10 - 0.19*), $p < .001$			
Take-home income	-0.04 (-0.14, *-0.25 - -0.02*), $p = .021$			
Smoking	-0.03 (-0.37, *-0.82 - 0.08*), $p = .105$			
Watching TV while eating	0.06 (0.21, *0.09 - 0.33*), $p < .001$			

Additionally, to assess the independent and shared contributions of eating rate and watching TV whilst eating on BMI aged 24, the variance-partitioning procedure outlined by Chuah and Maybery (1999) was followed. Both eating rate and eating whilst watching TV were significant independent predictors of BMI (Figure 9.1). There was no correlation between eating rate and watching TV whilst eating (r(1,776)= -0.01), translating to no shared variance between the two variables when contributing to BMI ($R^2 < .001$).

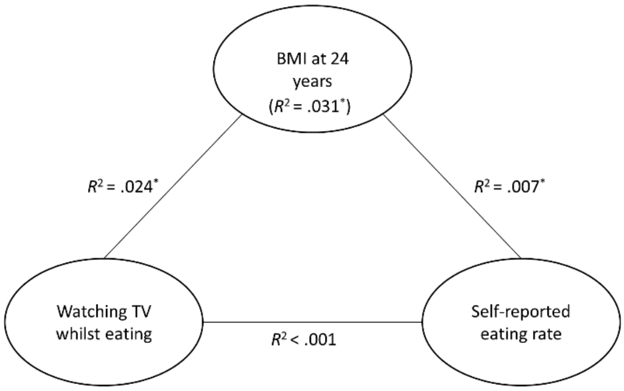

Figure 9.1 Variance partitioning analyses separating the variance in BMI at 24 years of age (*n* = 1,776) between watching TV whilst eating (assessed via Life@25 questionnaire) and self-reported eating rate (assessed via Life@25 questionnaire).

The procedure described by Chuah and Maybery (1999) was followed.

* denotes $p < 0.001$

9.5 Discussion

9.5.1 Identifying eating contexts and behaviours associated with young adult BMI

Across analyses, there was a failure to demonstrate an association between social eating (contextual factor) and either BMI or weight status (binary outcome, normal vs overweight/obese). However, BMI at age 24 was found positively associated with watching TV whilst eating (contextual factor) and that weight status is associated with eating rate (eating behaviour). The association with distraction (TV watching whilst eating) coincides with previous work (Tumin & Anderson, 2017; van Meer et al., 2022), and this finding was further extended by replicating in a young adult cohort and by benchmarking the strength of this association against other variables, including eating traits.

In addition to exploring predictors of BMI (continuous), non-linearity was also considered by examining whether the same eating contexts and behaviours predict weight status. With the latter approach, statistical information is lost, and this likely explains why TV watching whilst eating was positively associated with weight status but failed to reach significance. By contrast, there was no association between BMI/weight status and playing a computer/video game. Here, it is suspected to reflect a lack of variance in the predictor – 92.2% of the sample reported never playing while eating during the last seven days.

Consistent with previous work, eating rate predicted weight status (Ohkuma et al., 2015). However, as with TV watching, it predicted only one of two outcomes, in this case weight status and not BMI. Here, the failure to predict BMI might reflect a non-linear association. Specifically, people who self-reported as eating 'average,' 'fast,' or 'very fast' tended to have a higher and a similar BMI, whereas those who reported 'very slow' or 'slow' had a slightly lower BMI that was more likely to fall into the normal weight category (see Appendix 5 Figure 11.14). Additionally, because the variance-partitioning procedure showed that self-reported eating rate and watching TV whilst eating are both independent predictors of BMI, the effect of TV watching whilst eating on BMI is likely not determined solely by an indirect effect of TV watching on speed of eating.

Of course, it remains to be determined whether the association between watching TV whilst eating and BMI/weight status is causal (although this seems plausible given the robust effects on energy intake observed in acute laboratory-based experimental studies (Robinson et al., 2013)) and whether this extends to other countries and age groups. By contrast, for eating rate, the evidence for a causal association is much stronger (Fogel et al., 2017; Ford et al., 2009), and a relationship with BMI has been observed in many cultures (Ohkuma et al., 2015).

9.5.2 Failure to observe an association between social eating and BMI

The failure to observe any association between social eating and BMI is interesting because acute studies show reliable effects on energy intake (Ruddock et al., 2019). One explanation is that, as previously stated, this age group is undergoing a substantial period of change and their current social eating patterns might not reflect the patterns which contributed to their weight gain. For future work, researchers might benefit from utilising a longitudinal approach and assessing social eating at multiple time points to capture potential changes in social eating habits. Another possibility is that the acute effects of social interaction on food intake are powerful, but they wane over time if the same people eat together on a regular basis. Recent observational evidence suggests that social facilitation extends over a 3-day period (Ruddock et al., 2022), however, the effect over months and years remains unclear. In addition, drawing a distinction between routine social eating with very familiar individuals (e.g., family members) and more *ad hoc* social eating involving significant others, perhaps at social occasions outside the home, might also address potential ceiling effects if both types of social eating with familiar others are considered in the same measure of social eating. A further possibility is that particular food-preparation and serving styles impact the expression of social facilitation. For example, meal size might be constrained when 'family meals' are pre-portioned in the kitchen, before a meal begins. Again, these features of social eating should be assessed alongside measures of frequency. One possible approach to capture these features over a longer period would be to use Ecological Momentary Assessment or EMA paired with a real-time eating detection system as done by Morshed et al. (2022). The real-time eating detection system uses sensors in a smartwatch to detect when someone is eating which then triggers an automated smartphone-linked EMA (Morshed et al., 2022). This EMA could ask a range of eating context-related questions, and, if completed over a long time, might provide insight into whether the frequency with which someone eats in these specific eating contexts might associate with changes in BMI.

9.5.3 Including eating contexts in public health interventions

Public health initiatives targeting physical activity or dietary changes, such as adopting healthy eating practices, have had relatively little impact on adult obesity prevalence (Tseng et al., 2018). Eating contexts, eating traits, and eating behaviours were identified as potentially important associates of BMI/weight status; however, they vary in the extent to which they can be translated and applied as part of a public health strategy. Previously, associations have been observed between a variety of eating traits (e.g., disinhibited eating and dietary restraint) and anthropometric variables (Finlayson et al., 2012; Provencher et al., 2003). However, the extent to which these outcomes can inform effective dietary-based public health interventions is unclear and people find it difficult to maintain weight loss merely by restricting their food intake (Mann et al., 2007). Additionally, while eating rate can be modified in a clinical or experimental environment (Ford et al., 2009), it is uncertain whether interventions which manipulate eating rate are feasible at a population level (Robinson et al., 2014). Therefore, of the three factors identified, eating contexts, specifically watching TV whilst eating, might be the most amenable target for inclusion alongside existing public health guidelines.

While on an individual level, eating contexts might only predict a relatively small amount of variance in an individual's BMI; they may, on a population level, have the potential to play a substantial role in improving public health. Additionally, this young-adult cohort remains an important target for public health interventions as they are experiencing a period of substantial change and are adopting new health behaviours (Nelson et al., 2008; Poobalan et al., 2009). Recommending individuals abstain from eating meals whilst watching television is simple advice that might prevent some from gaining excess weight, especially when presented as simple guidance relating to a single behaviour. This would be a safe intervention which does not require additional information about the individuals, such as whether they have diabetes. More broadly, this work demonstrates that examining 'eating ecology' (i.e., where and how food is consumed) may benefit future public health guidance.

9.5.4 Conclusion

In summary, public health interventions related to food intake have largely focused on what types of food and how much food individuals are consuming; less attention is being paid to where and how we eat our food. To that end, the relative contribution of eating contexts, specifically social and distracted eating, to BMI/weight status was assessed, alongside other commonly included variables, such as self-reported eating rate, in a young adult cohort. Watching TV whilst eating was found to positively associate with BMI at age 24, and it explained roughly similar variance in BMI as other well-established eating traits. Consistent with previous research, a faster self-reported eating rate was associated with an increased likelihood of having overweight/obesity. By quantifying the strength of eating contexts alongside other commonly assessed variables, watching TV whilst eating was identified as a key eating context which, if dissuaded alongside existing public health guidelines, could effect changes in BMI on a population level.

Chapter 10 General discussion

This chapter summarises and links the thesis findings to the two public health strategies introduced in chapter one, specifically food reformulation (Part A) and eating contexts (Part B). Additionally, this chapter also discusses the general strengths and limitations of the thesis as well as the overlap of the findings with existing public health policies before providing a concluding statement.

10.1 New insight into food reformulation as a possible public health strategy

The overarching conclusion of the research presented in Part A is that humans appear to be sensitive to both the energy content (chapters two through five) and the macronutrient composition (chapters six and seven) of the foods or meals they consume. This sensitivity (i.e., nutritional intelligence) presents a potentially significant challenge to food reformulation as a public health strategy given that successful reformulation relies on individuals being insensitive to changes in a product (Gressier, Swinburn, et al., 2020). In the three sections that follow, each of the three research topics in Part A (energy density, protein content, and fat and carbohydrate content) will be explored in light of food reformulation and the extent to which the findings could help inform future developments. Additionally, related suggestions for future research will also be made, where appropriate.

10.1.1 Reducing food energy density to reduce energy intake – a feasible approach?

A challenge for product reformulation aiming to reduce food energy density (i.e., reduce total energy content whilst maintaining portion size) is matching the sensory characteristics of a reformulated and non-reformulated (original) product. In their systematic review and meta-analysis exploring the potential for food reformulation to impact food choices, nutrient intakes, and health status, Gressier, Swinburn, et al. (2020) noted that the majority of food reformulation has resulted in products with the same energy density (i.e., reducing sodium content which has no impact on total energy content or reducing the levels of one macronutrient by replacing with an equicaloric substitute). The authors suggest that one reason why reducing a food's energy density is challenging is that any major reduction is likely to substantially change the product's sensory properties, specifically its' taste and texture (Gressier, Swinburn, et al., 2020). This change in sensory properties might then alter consumer behaviour (e.g., the consumer no longer selects the reformulated product), undermining the potential impact of reformulation. One example where sensory-matched reformulation to reduce energy content has been successful is sugar-sweetened beverages, where sugar has been replaced with non-nutritive sweeteners; however, the authors note that beverage reformulation might be easier than reducing the sugar content of solid food (Gressier, Swinburn, et al., 2020).

A second approach to food reformulation involves not identically matching the sensory properties of the reformulated product or meal. For example, the energy density of a meal could be reduced by increasing water or vegetable content or by decreasing fat content (Robinson et al., 2022). When utilising this approach, there is some evidence that covertly decreasing the energy density of a food or diet results in lower total daily energy intake (Robinson et al., 2022). In other words, despite the reformulated version not being identical in sensory properties to the original version, consumers appear to maintain their acceptance of the reformulated product (i.e., are insensitive to the reduction in energy density), resulting in a subsequent decrease in their energy intake.

The finding from Robinson and colleagues suggesting insensitivity to reductions in energy density might appear to be at odds with the results presented in chapters two, three, four, and five which suggest that individuals are sensitive to the energy content of the meals they consume. One of the key differences between the results presented in this thesis and the findings from Robinson and colleagues is that the majority of studies included in their systematic review covertly manipulated the energy density of familiar meals and assessed whether compensation occurred (Robinson et al., 2022). Contrastingly, in the studies presented in this thesis, sensitivity to energy density was assessed across a broad and continuous range of energy densities using familiar, non-manipulated meals. In studies where energy density is covertly manipulated, participants tend to consume fewer calories in the lower energy density condition (i.e., consume similar amounts of food by weight in each condition) (Robinson et al., 2022). One possibility for this 'insensitivity' to changes in energy density is that covertly manipulating the energy density of a familiar meal, undermines the learned calorie-content signal (chapter five). In this case, evidence for insensitivity to calories (i.e., eating the same amount of food (g) in the low and high energy density conditions) will appear identical to evidence for sensitivity to calories (previous experience with the food guiding decisions about meal size) despite the artificial mismatch in energy density between the two versions (Brunstrom et al., 2023).

Importantly, the extent to which individuals remain 'insensitive' to energy density manipulations over time is unclear, and studies which expose participants to covertly manipulated foods over a longer period reported smaller effects on energy intake. One possibility for this is that individuals might, over time, learn that the manipulated food or meal has a lower energy density and increase their energy intake either at that meal or at other meals (i.e., compensate for reductions in energy density) (Robinson et al., 2022), potentially via feedback from the calorie-content signal (chapter five). In other words, it appears plausible that reducing the energy density of a product might decrease total daily energy intake over a short period, but the extent to which it produces sustained changes in body weight is unclear. Assessing the degree of potential compensation for reductions in energy density over a longer period remains a key question (Robinson et al., 2022), largely because the outcome could inform the viability of energy density reformulation as a potential public health strategy.

The findings suggesting sensitivity to calories in meals (i.e., non-linear pattern in meal caloric intake in response to meal energy density, chapters two to four) also raise several key questions regarding factors that might impact the likelihood of successful food reformulation, some of which were raised by Robinson and co-authors (Robinson et al., 2022). One factor might be the energy density of the original product; the non-linear pattern in meal caloric intake and the two-component theoretical model of meal size (chapter five) suggest that while feedback from the calorie-content signal exists across the range of energy densities, it is dominant in higher energy-dense meals. Based on this, changes to the energy density of a product might be more salient in energy-rich products compared to energy-poor products. A reduction in the energy content of an already energy-poor food might have little impact on total daily energy intake; however, it might not trigger any calorie compensation as it falls into the range of energy densities where feedback from the volume signal dominates. Indeed, initial work addressing whether the degree of compensation differs as a function of the original energy density of a meal suggests that there were no differences - reducing the energy density of a lower (< 1.75 kcal/g) or higher (> 1.75 kcal/g) energy-dense meal resulted in similar reductions in energy intake (Robinson et al., 2022). The authors note, however, that the majority of studies manipulating energy density were conducted in lower energy-dense meals (Robinson et al., 2022), and, again, these meals likely fell into the range of energy densities where feedback from the volume signal dominates. From a public health perspective, it is critical to understand whether reducing the energy intake of an energy-rich food might also result in reductions in total daily energy intake or whether compensation occurs. Therefore, future studies are needed to understand how responses to reformulation might differ across a range of food energy densities and over a longer period of time.

Additionally, the calorie-content signal introduced in chapter five does not take into consideration that feedback might be influenced to a greater or lesser extent by energy derived from different macronutrients and that this, in turn, could influence consumer behaviour in response to food reformulation. Indeed, macronutrient-specific reformulation was noted by

Robinson and colleagues; however, the role of nutrient-specific responding might be small relative to a general reduction in energy density (Robinson et al., 2022; Stubbs et al., 2000). Despite potentially being only a small effect, understanding consumer responses to macronutrient-specific reformulations alongside energy density reformulation will be an important avenue for future research.

10.1.2 Development of novel alternative protein sources – a potential solution to deliver public and planetary health?

Climate change has been recognised as a risk to human health and survival, and is part of the 'Global Syndemic', alongside overnutrition (overweight and obesity) and undernutrition (Swinburn et al., 2019). As mentioned in chapter six, a key recommendation from the Inter-governmental Panel on Climate Change (IPCC) to improve planetary health is to switch to more sustainable diets, including increasing the consumption of plant-based proteins (Mbow et al., 2019). Alongside being more sustainable, increased consumption of plant-based proteins is also associated with better overall health outcomes as compared to animal proteins (Ferrari et al., 2022). Therefore, encouraging the increased consumption of plant-based proteins appears to be a potentially feasible strategy to improve population health, acknowledging that recommendations will likely need to be tailored to address the needs of individual groups (Lonnie & Johnstone, 2020).

While not reformulation of an existing product per se, the development of alternative protein sources, such as meat analogues, is one approach to increase the consumption of plant-based proteins. Indeed, due to improvements in food processing techniques and increased consumer acceptability, the plant-based meat analogue market has grown substantially (Ishaq et al., 2022). To capitalise on this market growth, one key avenue for research is understanding what factors might increase the likelihood that a new product is recognised by consumers as a protein source. This question is especially relevant for the development of novel plant-based proteins as consumer acceptance is paramount for the success of the product (Ishaq et al., 2022; Siegrist, 2008).

The results from chapter six suggest that people prefer a mixture of macronutrients (i.e., a pair of foods comprising a source of carbohydrate and a source of protein) and that this preference occurs even with pairs of foods that cannot be explained by cultural norms (i.e., a preference for peanuts (protein source) to be paired with a source of carbohydrate rather than another protein source). This finding is relevant to the development of novel meat analogues as these products often contain less protein than meat (Cutroneo et al., 2022; Godschalk-Broers et al., 2022) and are competing in a market where protein content (especially relative to meat) is known to influence choice in omnivores (Kerslake et al., 2022). For example, if a meat analogue was low in protein and was consumed alongside a source of carbohydrate, then that meal might be perceived by the consumer as being more of a high-carbohydrate meal, potentially increasing the likelihood that the meat analogue is rejected. This concern could be especially relevant for omnivores transitioning to a vegetarian diet as they will have recently consumed meals comprising more distinct protein-carbohydrate pairings (e.g., steak served with potatoes). Therefore, by developing meat analogues which contain roughly equivalent protein content to meat, companies might increase the likelihood that the product is accepted, including by omnivores transitioning to a vegetarian or vegan diet. Additionally, protein content is also an important factor influencing product acceptance by individuals identifying as vegetarian or vegan, and these individuals viewed meat analogues with a high protein content as more filling and more nutritious (Kerslake et al., 2022). A somewhat related idea for the development of meat analogues is to consider the context in which they are consumed, specifically the other foods (and macronutrients) they might be consumed with. Indeed, a similar suggestion has been made by Elzerman and colleagues in their study exploring the role of meal context (i.e., the type of dish and flavouring) on the acceptance of meat substitutes

(Elzerman et al., 2011). However, they did not consider a role for macronutrients in their conceptualisation of a meal context and, as mentioned above, this remains an opportunity for future research.

In addition to protein content influencing the acceptance of meat analogues other potential influences include, among others, food choice motives (such as taste, texture or healthiness), familiarity, or social norms (Onwezen et al., 2021). Social norms are of particular interest as the exploratory findings from chapter six provide very tentative evidence that dietary status (i.e., newer vegetarian, older vegetarian, or omnivore) might influence protein discrimination in plant-based protein foods. Both newer (less than two and a half years following the diet) and older vegetarians (more than two and a half years following the diet) exhibited, relative to omnivores, greater desire-to-eat plant-based food containing more protein when these foods were paired with a source of carbohydrate. It is plausible that vegetarians utilised the collective intelligence accumulated in the community (i.e., nutritional intelligence (Brunstrom et al., 2023)) and, benefiting from this intelligence, were potentially more likely to recognise the protein content of these plant-based foods than those individuals who identified as omnivores. Somewhat related to this, another possibility for the increased desire-to-eat higher protein foods observed in the vegetarian participants is that these participants tend to consume a lower protein diet (Neufingerl & Eilander, 2021) as well as more protein-dilute protein sources (Bradbury et al., 2017; Papier et al., 2019). They might therefore have acquired greater sensitivity to protein (i.e., increased desire-to-eat higher protein foods) due to being closer to experiencing limiting levels of protein or amino acids. Again, however, the exploratory nature of these findings assessing individual differences, specifically diet-based differences, requires that these results and potential explanations be interpreted with caution. More generally, social norms have been identified as the most important factor in predicting consumer acceptance of alternative protein sources (Onwezen et al., 2019), and future studies, such as the one outlined in section 6.7.3, could further unpack the role of social norms or social learning on the acceptance of novel protein sources and products.

10.1.3 Fat and carbohydrate content in a single food - an important target for reformulation?

Previous research suggested that single foods which contain a combination of fat and carbohydrate were more rewarding than those high in either fat or carbohydrate (DiFeliceantonio et al., 2018; Perszyk et al., 2021). The results from chapter seven expanded on this work and demonstrated that combination foods were potentially less satiating calorie-per-calorie, selected in larger portions (kcal), and more liked than foods high in either fat or carbohydrate. Together, this body of evidence suggests that foods which contain a combination of fat and carbohydrate might be important targets for food reformulation aiming to reduce energy intake. One might predict that reformulating a combination food to be higher in either fat or carbohydrate (i.e., increasing % kcal from fat or carbohydrate) might potentially reduce the intake of that food. Again, however, it should be noted that macronutrient-specific reformulation might only produce subtle effects on intake as compared to reducing energy density (Robinson et al., 2022), and matching products for their sensory characteristics would be highly challenging (Gressier, Swinburn, et al., 2020).

A related stream of research has also shown a similar effect of combining macronutrients (i.e., fat and sugar) on energy intake in humans, this time looking at hyper-palatability. Hyper-palatability in this context uses “objective criteria to identify foods that are highly divergent from naturally occurring foods because they contain combinations of nutrient pairs (fat and sugar, fat and sodium, carbohydrates and sodium) crossing defined thresholds” (Fazzino et al., 2023, pg. 1), and it was found to positively influence ad libitum energy intake in humans such that meals containing a greater proportion of hyper-palatable foods were consumed in larger portions (kcal) (Fazzino et al., 2023). In conjunction with the findings described in the previous paragraphs, this result lends further support for targeting combinations of fat and carbohydrate in food reformulation. While not explicitly public health policy directly related to food reformulation, there has been a call to limit access to hyper-palatable foods as a potential strategy to reduce overweight and obesity (Fazzino, 2022; Gearhardt et al., 2011). However, the extent to which such a policy might impact population-level energy intake and subsequently weight status is unknown.

10.2 Eating contexts as a potential target for public health messaging

As noted in the general introduction, eating contexts, or the immediate settings in which people eat, provide another opportunity for potential public health strategies related to food choice and intake (Elliston et al., 2017; Mak et al., 2012; Rauber et al., 2022; Shams-White et al., 2021). Importantly, however, it was unclear whether eating contexts have a chronic impact on an individual's weight status, and the research in chapter nine focussed on two eating contexts, social eating (Ruddock et al., 2019) and distracted eating (Robinson et al., 2013). The findings suggest that while social eating was unrelated to an individual's body mass index or weight status, BMI at age 24 was positively associated with watching TV whilst eating (distracted eating). As noted in chapter nine, eating contexts might only have a small impact on an individual's BMI, and on a population level, they may play a substantial role in improving public health. Additionally, eating contexts lend themselves to simple guidance regarding a single behaviour which is unlikely to interfere with or reduce the efficacy of existing interventions and could therefore be recommended alongside other interventions. Surprisingly, existing public health messaging pertaining to healthy eating (such as the Eatwell Guide (Public Health England, 2016)) does not appear to include any mentioned of eating contexts, and this is further discussed in section 10.4.

10.3 General strengths and limitations of the thesis[38]

One key strength of this thesis is the use of under-utilised data sources to explore psychological questions (e.g., the UK NDNS, Argentinean, and Malaysian datasets) and the use of novel approaches (for example, developing a novel task in chapter six). Generally speaking, the purpose of large nutritional databases, such as the UK NDNS, Argentinean or Malaysian datasets, is to quantify the health and nutritional status of a population (Food Standards Agency & Office for National Statistics, 2005; Karupaiah et al., 2019; Zapata, 2014). Rather than assessing energy or nutrient intakes across the population, these datasets were used to re-evaluate the association between energy density and energy intake within a single meal, breaking tradition with previous studies which often utilised time and resource-intensive research methodologies (for example, Bell et al., 1998; Bell & Rolls, 2001; Rolls et al., 1999; Stubbs et al., 1998). Similarly, a novel online task to assess protein discrimination in humans was developed in chapter six to circumvent the need to deplete humans of protein, a task which is often ethically challenging and/or resource intensive (for example, developing a range of novel stimuli as was done in a study by Gibson et al., 1995 or running a fully controlled 28 day dietary intervention as was done by; Griffioen-Roose et al., 2012). With further refinement, the single protein-carbohydrate pair task from chapter six might have the potential to be used in follow-on studies exploring protein conditioning in humans (see section 6.7.3 for an example of a potential study).

One broader limitation of the research presented in this thesis is the extent to which the samples are representative and include participation from underrepresented minority groups. This is potentially less of a concern for the research which utilised large nutritional surveys (chapters 3, 4, and 5) compared to the experimental studies (chapters six and seven) or analyses

[38]The limitations of the individual chapters can be found in chapter five (mentioned throughout the chapter) and sections 6.7.3, 7.5.2 and 9.5 (incorporated into chapter nine's discussion as the chapter is presented as prepared for publication).

using data from other sources (i.e., Hall et al. dataset (chapter two) or ALSPAC (chapter nine)). For example, the recruitment strategy of the NDNS is designed to recruit a sample that is representative of the broader UK population (Food Standards Agency & Office for National Statistics, 2005), and the ethnic distribution of the Malaysian dataset was representative of a typical urban Malaysian population (Karupaiah et al., 2019). On the other hand, the experimental research presented in this thesis relied on either convenience sampling (chapter six) or recruitment via the online platform Prolific (chapter seven) and was likely not representative. Importantly, regardless of whether a sample is representative based on some demographic characteristics, there is still a concern about whether the research reaches underserved or hard-to-reach communities which might be less likely to engage in research, especially health-related research (Bonevski et al., 2014). Engaging with these communities remains important, and the author endeavours for their future work to include participation from these groups throughout the entire research process.

Additionally, the use of online tasks in an out-of-laboratory setting in chapters six and seven might also be a potential limitation due to a lack of experimental control. Moving data collection online provided the opportunity to conduct research during the COVID-19 pandemic, but this could have impacted data quality, potentially due to careless responding by participants (Huang et al., 2015). To encourage careful responding, where possible, breaks were included between tasks and, in some tasks, even between stimulus presentation and responding. Additionally, one study used responses to an attention check question (Pei et al., 2020) to exclude participants who might not have understood the task instructions (chapter six). A final general limitation is the use of cross-sectional analyses in chapters two, three, four, and nine which does not allow for causal conclusions to be drawn. However, due to the substantial experimental evidence regarding the research questions addressed in these chapters, this is potentially a less relevant limitation (see sections 5.2 and 9.5 for more detail).

10.4 Existing public health policy on food reformulation and eating contexts

In 2013, the World Cancer Research Fund International developed the NOURISHING framework to bring together ten different key policy areas within three domains to improve population health and encourage healthier diets (Hawkes et al., 2013). The three domains include food environment, food system and behaviour change communication (Hawkes et al., 2013) (see Figure 10.1 below).

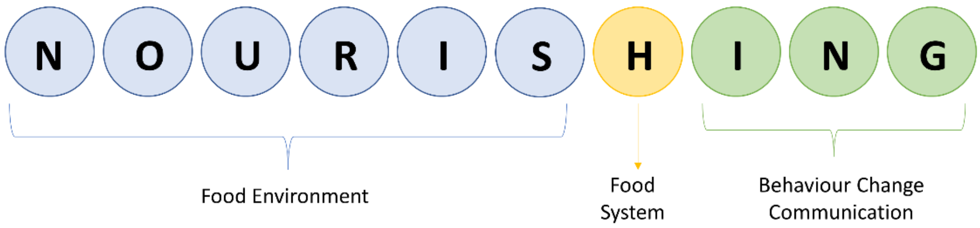

Figure 10.1 Reproduction of the NOURISHING policy framework developed by the World Cancer Research Fund International (Hawkes et al., 2013), The ©WCRF International NOURISHING framework.

The three domains are conceptualised as being the key targets to encourage healthier diets and prevent overweight, obesity and other non-communicable diseases, and these domains and specific policy areas (each letter within the domain, see Figure 10.1) were identified via a "review of existing policy frameworks, proposed and implemented national policies, and the evidence of their effects" (Hawkes et al., 2013, pg.161). The food environment domain captures the various ways that the food environment might impact dietary behaviour, such as food availability or changes in the food supply, and it also includes the roles of the food industry (including producers, manufacturers, distributors or retailers) in encouraging healthy food and drink consumption (Hawkes et al., 2013). The second domain, the food system, considers the interplay between the food system policy and policies to support healthy eating, recognising that changes in the food system could have implications for policies supporting healthy eating and vice versa (Hawkes et al., 2013). The final domain is behaviour change communication which focuses on having individuals change their behaviours by providing information and skills, rather than targeting the external environment (Hawkes et al., 2013).

The research presented in this thesis aligns with one key policy area (improving the quality of the food supply ('I'- food environment)) and one policy domain (behaviour change communication (the three policy areas of 'ING'). Focussing first on improving the quality of the food supply, Hawkes et al. (2013) suggest that reformulation might be one feasible policy target alongside reducing portion size. As mentioned previously, the studies in thesis Part A have the potential to inform reformulation strategies including ways to increase the acceptance of new or reformulated products. On this basis, the research in Part A overlaps substantially with the policy area aiming to improve the quality of the food supply. With regards to the behaviour change communication domain, the results from thesis Part B could be implemented as simple public-health guidance across the policy areas within this domain. For example, encouraging individuals to not eat whilst watching TV could easily be incorporated alongside education about dietary guidelines and healthy eating which could be part of a larger public information campaign ('I'- inform people about food and nutrition through public awareness) or it could be included in a health literacy programme ('G'- give nutrition education and skills).

Alongside developing the NOURISHING framework, the World Cancer Research Fund International also tracks the development of relevant policies in 30 European countries as part of the CO-CREATE project (Klepp et al., 2023). Of the 30 European countries tracked, the four nations of the United Kingdom (England, Scotland, Wales and Northern Ireland) are also included, and an overview of thesis-relevant nutrition policy status in each nation is presented in Table 10.1.

Table 10.1 *Overview of Nutrition Policy Statuses Relevant to the Research Presented in this Thesis in England, Scotland, Wales, and Northern Ireland*[1]

No Policies Identified	Poor	Fair	Moderate	Good	Excellent

Policy Domain	**Policy Areas**	**England**	**Scotland**	**Wales**	**Northern Ireland**
Food Environment	Improve the nutritional quality of the food supply	Good	Good	Good	Good
Behaviour Change Communication	Inform people about food and nutrition through public awareness	Good	Moderate	Good	Good
Behaviour Change Communication	Nutrition advice and counselling in health care settings	Fair	No Policies Identified	Fair	No Policies Identified
Behaviour Change Communication	Give nutrition education and skills	Fair	Poor	Poor	Good

[1]Table created using the information provided by the World Cancer Research Fund International, Nutrition policy snapshots | WCRF International

Each of the four nations received a 'good' assessment in their policy area which aimed to improve the nutritional quality of the food supply, and this was largely because of policy which aimed to set limits or remove specific nutrients in food products (World Cancer Research Fund International and CO-CREATE, 2023a, 2023b, 2023c, 2023d). For example, England has introduced guidelines for the food industry regarding reducing the sugar (Coyle et al., 2020), calories (Pyne et al., 2020), and salt (Niblett et al., 2020) content in products, and, in April of

2018, a UK-wide Soft Drinks Industry Levy was introduced to curb energy and sugar intake in beverages (Barber et al., 2017). Within the calorie reduction guidelines mentioned above, there are different calorie targets recommended for different food groups (i.e., a 5% calorie reduction for crisps, savoury snacks, and sandwiches versus a 20% calorie reduction for most meals, side dishes or starters, and pizza or pastry products) (Pyne et al., 2020). This might be particularly relevant to some of the research presented in Part A as, again, the results in chapters two through four suggest that people are sensitive to the energy content of meals and that they might be more or less sensitive to changes in calorie content depending on how energy dense the original version of the food or meal is. Importantly, however, monitoring the potential effect of food reformulation policy on population health remains a challenge, and this information is needed to ensure that the most effective policies are being designed to improve population health (Gressier, Sassi, et al., 2020).

With regards to behaviour change communication, England had the best response across the three policy areas within this domain; however, there is room for improvement for each nation (see Table 10.1). By way of an example, one of the main public-facing resources about healthy eating in the UK is the ‘Eatwell Guide’ (Public Health England, 2016). This publication provides a visualisation of how much an individual should consume from each food group (i.e., fruit and vegetables, starchy foods, fish and meat, milk and dairy, and oils and spreads) to maintain their health (Public Health England, 2016). It also encourages consumers to reduce their intake of foods high in fat, sugar, or salt as well as ensure six to eight glasses of fluid are consumed per day. Lastly, the guide also recommends that consumers check the nutritional label, specifically the traffic light label, on packaged foods to help select foods which contain less fat, sugar, and salt (Public Health England, 2016). What the Eatwell Guide seems to fail to consider is the eating context in which people are consuming their food, and the results from Part B suggest that it might be beneficial to include simple guidance encouraging individuals to not consume their meals whilst distracted (i.e., not whilst watching TV). Indeed, to the best of the author's knowledge, the only guidance on eating contexts, specifically distracted eating, was on the National Health Service's webpage on treating obesity where it is suggested, under 'Other useful strategies', that individuals should eat more slowly and mindfully and includes not being distracted by the TV as an example (https://www.nhs.uk/conditions/obesity/treatment/, National Health Service, 2023). In this context, the recommendation appears to be made from a treatment perspective (i.e., weight loss), but, in the context of the Eatwell Guide, the information could be included as a preventative measure.

In summary, the results from this thesis align with several policy areas identified in the NOURISHING framework targeting healthier diets and aiming to prevent overweight, obesity and other non-communicable diseases. When developing future studies, it will remain important to reflect on policy frameworks such as the NOURISHING framework to increase the likelihood that the research could help inform policymakers when designing policies to improve population health.

10.5 Concluding remarks

Together, the research presented in this thesis suggests that food composition influences food choice and energy intake in humans and that the context in which one eats can, over time, impact body mass index. More generally, the findings from Part A highlight new complexity in human dietary behaviour and provide insight into fundamental food-level drivers of food choice and energy intake. The findings from Part B exposed eating contexts which lend themselves to accessible, easily understood public health messaging that could be incorporated alongside existing public health strategies aimed at improving population-level health. The process of completing this thesis has resulted in new approaches to studying human dietary behaviour and generated new research questions which need to be addressed to provide policymakers with the evidence base needed to inform effective public health strategies.

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Chapter 11 Appendices

Appendix 1 Additional tables and figures relevant for chapters two through four

Table 11.1 *Akaike's Information Criterion Value, Bayesian Information Criterion Value and Degrees of Freedom for Both a Linear and a Segmented Regression Model in the Hall et al. (n= 1,519) and NDNS (n= 32,162) Datasets*

	Degrees of freedom	Akaike's information criterion value	Bayesian information criterion value
Hall et al. dataset			
Linear	3	20673	20689
Segmented	5	20485	20511
NDNS dataset			
Linear	3	440532	440557
Segmented	7	438829	438888

Table 11.2 *Slope Parameter Estimates, 95% Confidence Intervals (CI), T-Values, and P-Values from the Sensitivity Analyses in the Hall et al. Dataset*

Sensitivity analysis	Slope	Slope parameter	95% CI	*t*-value	*p*-value
Including plate-cleaned meals (*n*= 1,678)	Slope 1 (Segment A, < 1.08 kcal/g)	801.98	660.89, 943.07	12.26	< 0.001
	Slope 2 (Segment B, 1.08 – 2.89 kcal/g)	68.28	43.98, 92.58	5.95	< 0.001
	Slope 3 (Segment C, > 2.89 kcal/g)	-313.39	-440.05, -186.72	-4.02	0.0001
Presented ED as predictor (*n*= 1,519)	Slope 1 (Segment A, < 1.02kcal/g)	881.15	635.09, 1127.20	7.38	< 0.001
	Slope 2 (Segment B, 1.02 – 1.84 kcal/g)	98.85	33.56, 164.10	2.99	0.003
	Slope 3 (Segment C, > 1.84 kcal/g)	-28.09	-73.47, 17.29	-1.19	0.235

Table 11.3 *Breakpoints and Slopes from a Segmented Regression Model in the NDNS dataset (n= 32,162) Using Various Calorie Filters (i.e., eating events excluded if less than specified calorie value)*[1]

Calorie filter (kcal)	Breakpoint 1	Breakpoint 2	Slope 1 (Segment A)	Slope 2 (Segment B)	Slope 3 (Segment C)
200	1.75	2.94	174.86	-107.91	-59.19
400	1.84	2.89	183.95	-112.17	-34.85
600	2.04	2.66	155.28	-151.12	0.33
800	1.77		184.85	-18.38	
1000	1.95		173.64	-25.40	
1200	2.30		116.04	-79.79	

[1]If two breakpoints were not identified as being significant via the segmented regression, then only one breakpoint is reported.

Table 11.4 *Akaike's Information Criterion Value, Bayesian Information Criterion Value and Degrees of Freedom for Both a Linear and a Segmented Regression Model in the Argentinean (n= 2,738) and Malaysia (n= 4,658) Datasets*

	Degrees of freedom	Akaike's information criterion value	Bayesian information criterion value
Argentinean dataset			
Linear	3	37922	37940
Segmented	5	37710	37740
Malaysian dataset			
Linear	3	63188	63207
Segmented	5	62972	63004

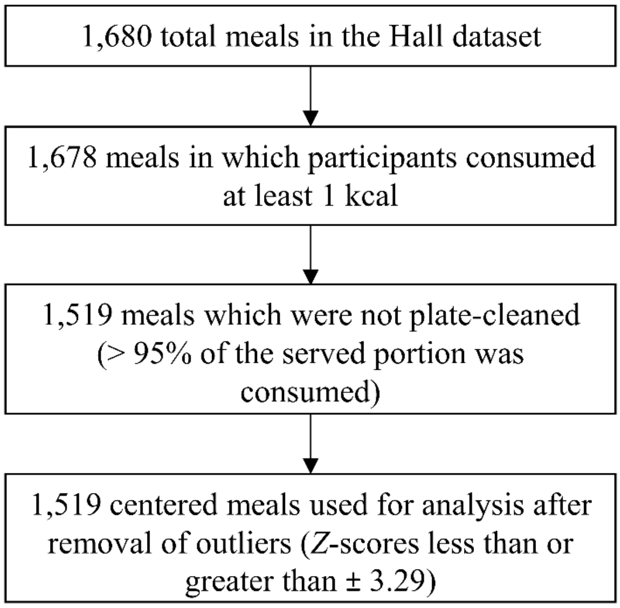

Figure 11.1 Flow of meals from the Hall et al. dataset through the exclusion stages.

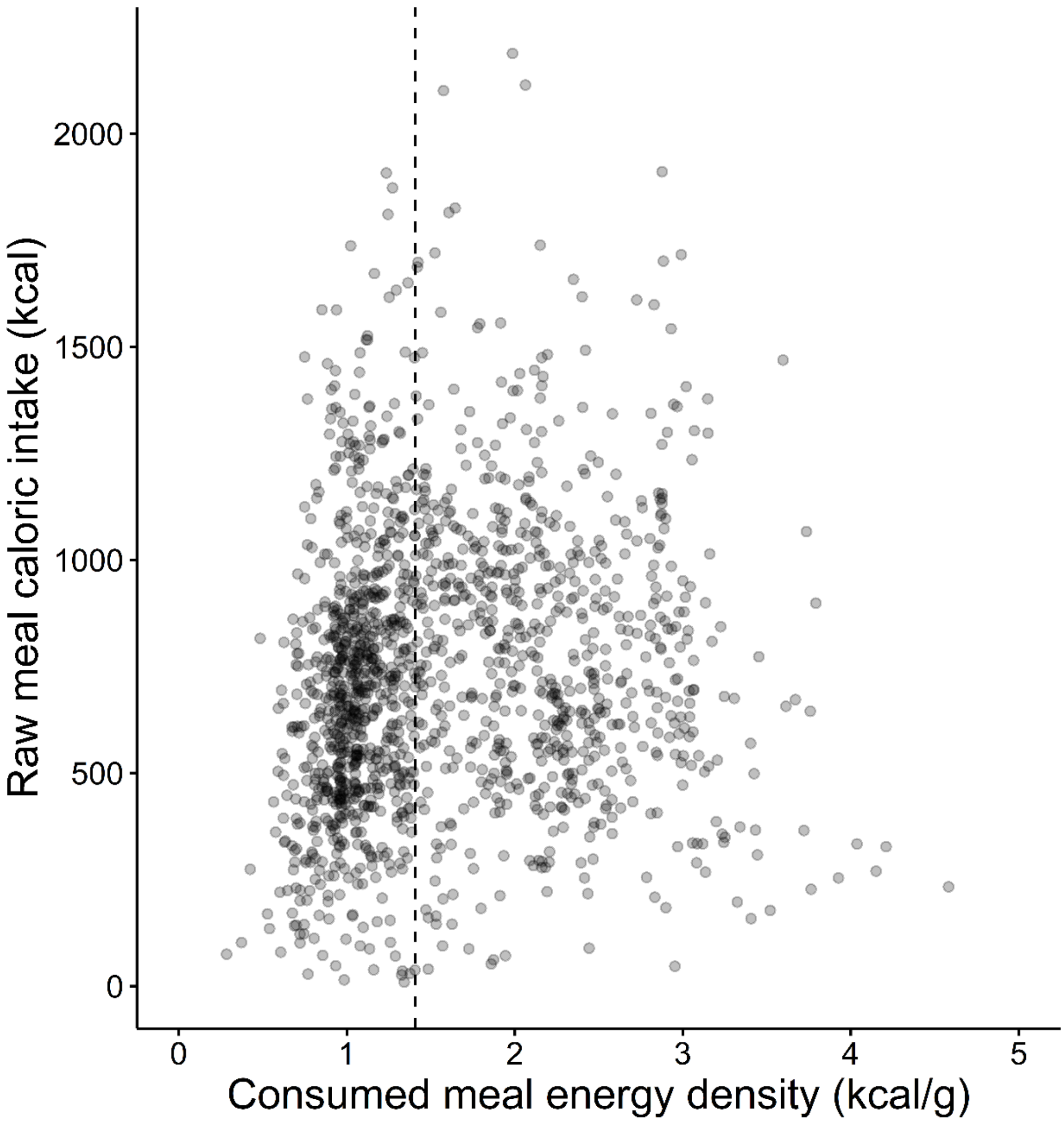

Figure 11.2 Raw meal caloric intake (kcal) by consumed meal energy density (kcal/g) in the Hall et al. dataset (*n*= 1,519).

Z-scores of raw meal caloric intake values were calculated, and outliers with *Z*-scores less than or greater than ± 3.29 were removed. The black dashed line represents the 1.41 kcal/g breakpoint identified via segmented regression. In this scatterplot, each point represents one meal.

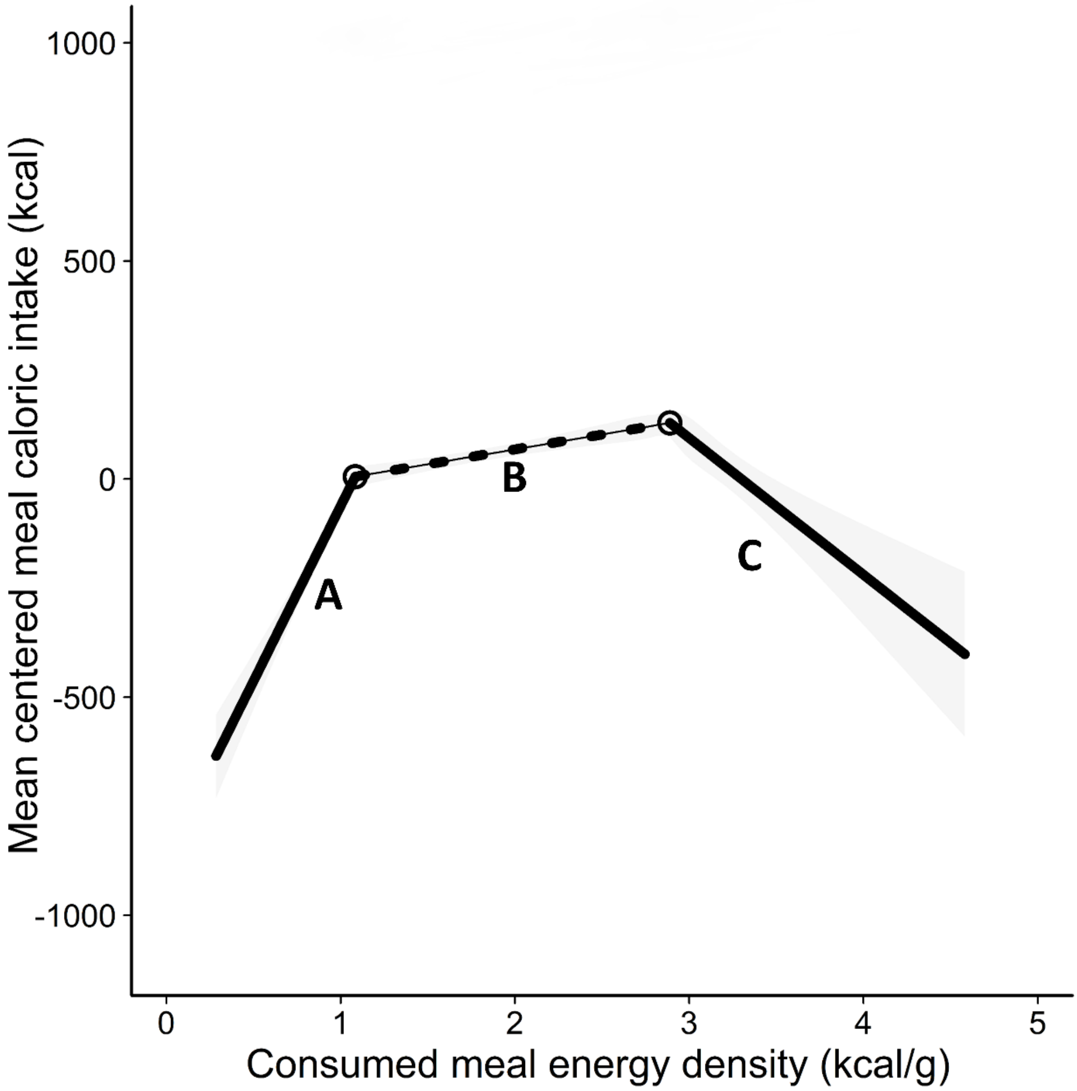

Figure 11.3 Mean centred meal caloric intakes (kcal), predicted from a segmented regression model relating consumed meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the Hall et al. dataset *(n*= 1,678) including plate cleaned meals (i.e., > 95% of the served portion was consumed).

The dashed and solid lines represent different segments and the shading around the segments indicates 95% confidence intervals. The circles indicate the location of significant breakpoints at 1.08 kcal/g (*SE*= 0.03) and 2.89 kcal/g (*SE*= 0.08). Segment A indicates the slope of the segment between 0 and 1.08 kcal/g, segment B indicates the slope of the segment between 1.08 and 2.89 kcal/g, and segment C models the slope above 2.89 kcal/g.

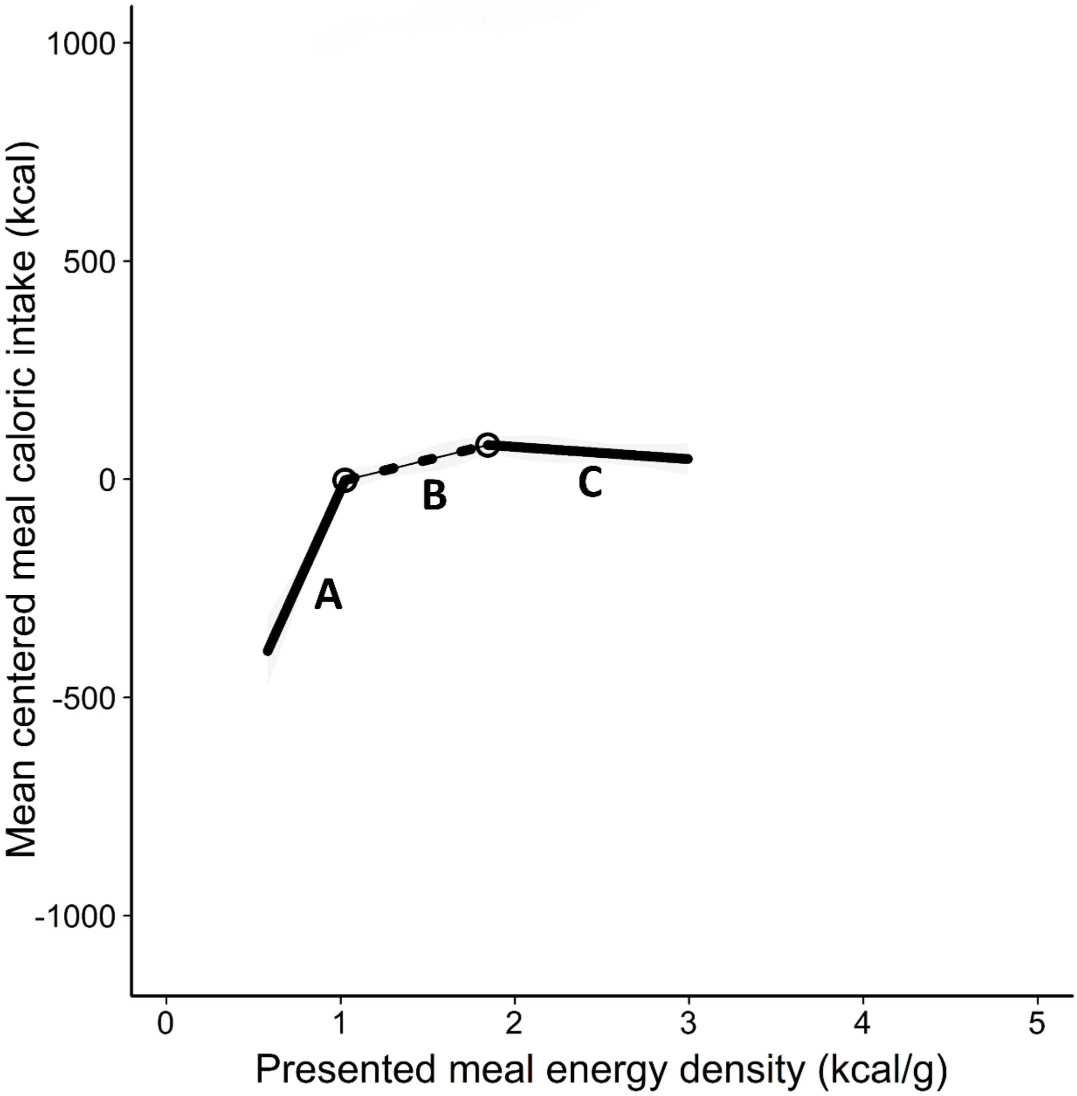

Figure 11.4 Mean centred meal caloric intakes (kcal), predicted from a segmented regression model relating presented meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the Hall et al. dataset (*n*= 1,519).

The dashed and solid lines represent different segments and the shading around the segments indicates 95% confidence intervals. The circles indicate the location of significant breakpoints at 1.02 kcal/g (*SE*= 0.03) and 1.84 kcal/g (*SE*= 0.20). Segment A indicates the slope of the segment between 0 and 1.02 kcal/g, segment B indicates the slope of the segment between 1.02 and 1.84 kcal/g, and segment C models the slope above 1.84 kcal/g.

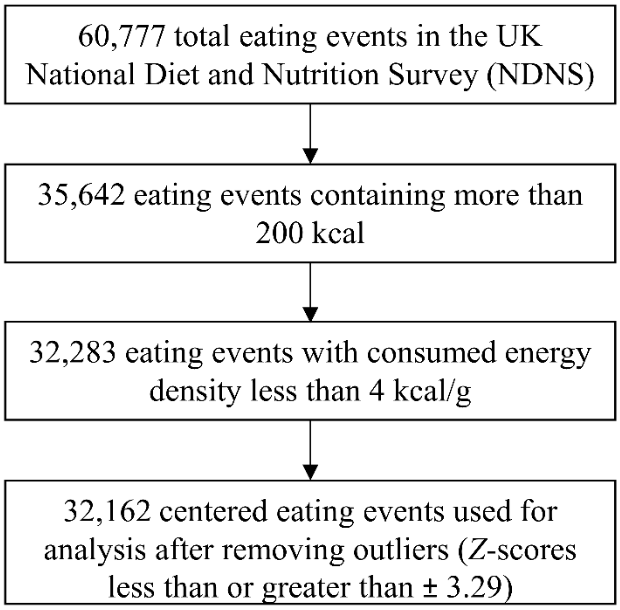

Figure 11.5 Flow of eating events from the NDNS dataset through the various exclusion stages.

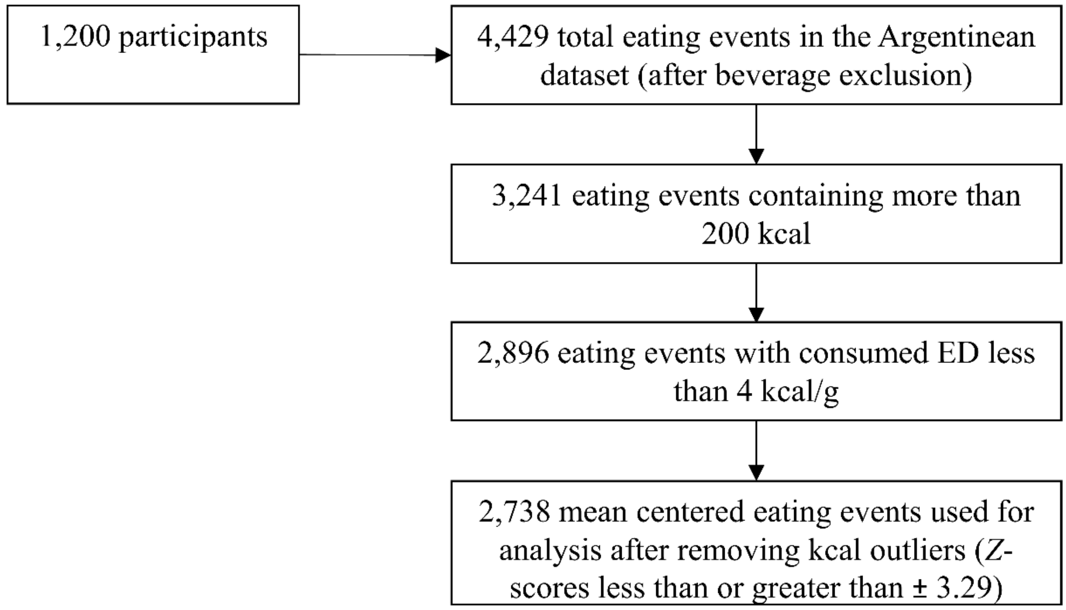

Figure 11.6 Flow of eating events from the Argentinean dataset through the various exclusion stages for the main analysis reported in the chapter.

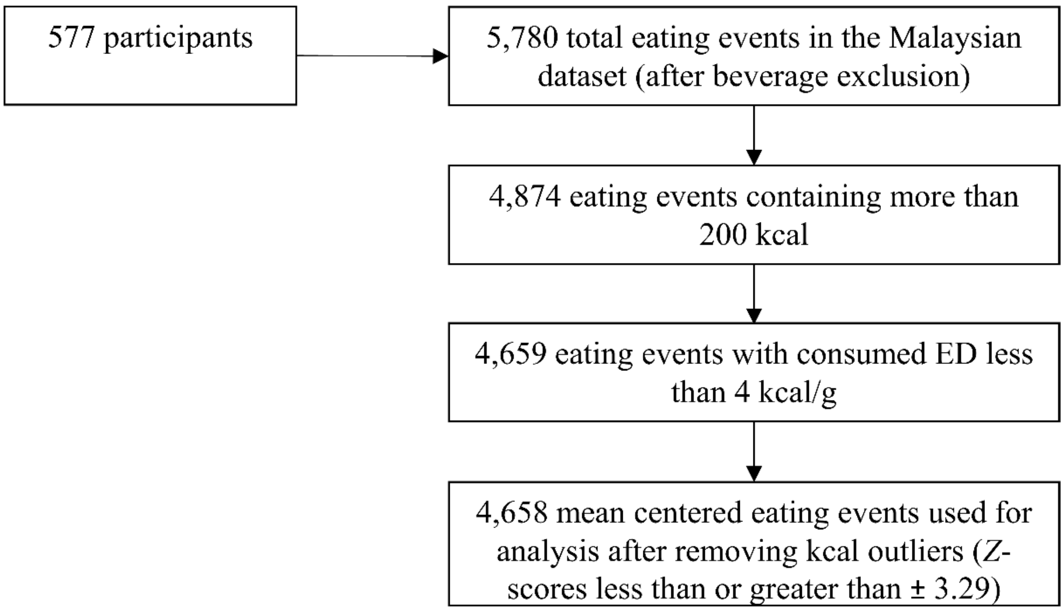

Figure 11.7 Flow of eating events from the Malaysian dataset through the various exclusion stages for the main analysis reported in the chapter.

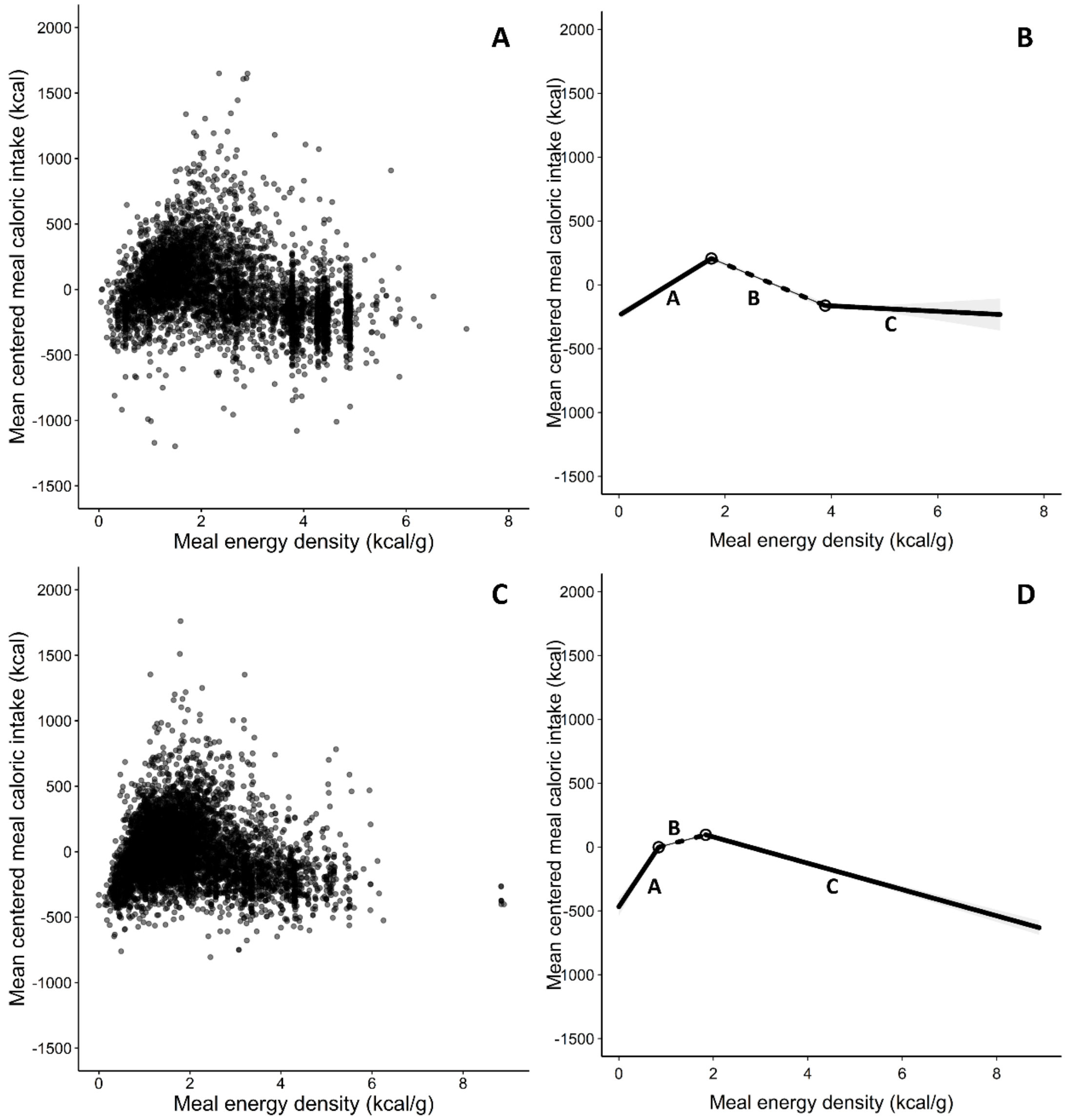

Figure 11.8 Four-panel plot depicting sensitivity analyses in the Argentinean (*n*= 4,406 meals) and Malaysian (*n*= 5,780 meals) datasets.

In panels A and C, mean centred meal caloric intake (kcal) is plotted by meal energy density (kcal/g) in both the Argentinean dataset (*n*= 4,406 meals, panel A) and the Malaysian dataset (*n*= 5,780 meals, panel C). In both scatterplots, meals were centred within each participant and no meals were removed. In panels B and D, mean centred meal caloric intakes (kcal) is predicted from a segmented regression model relating meal energy density (kcal/g) to consumed centred meal caloric intake (kcal) in the Argentinean dataset (*n*= 4,406, panel B) and the Malaysian dataset (*n*= 5,780 meals, panel D). In each panel, the breakpoint is represented by a black circle, the dashed and solid lines represent different segments and the shading around the segments indicates 95% confidence intervals. Segment A indicates the slope of the segment below the breakpoint and segment B models the slope between the two breakpoints and segment C indicates the slope above the breakpoint.

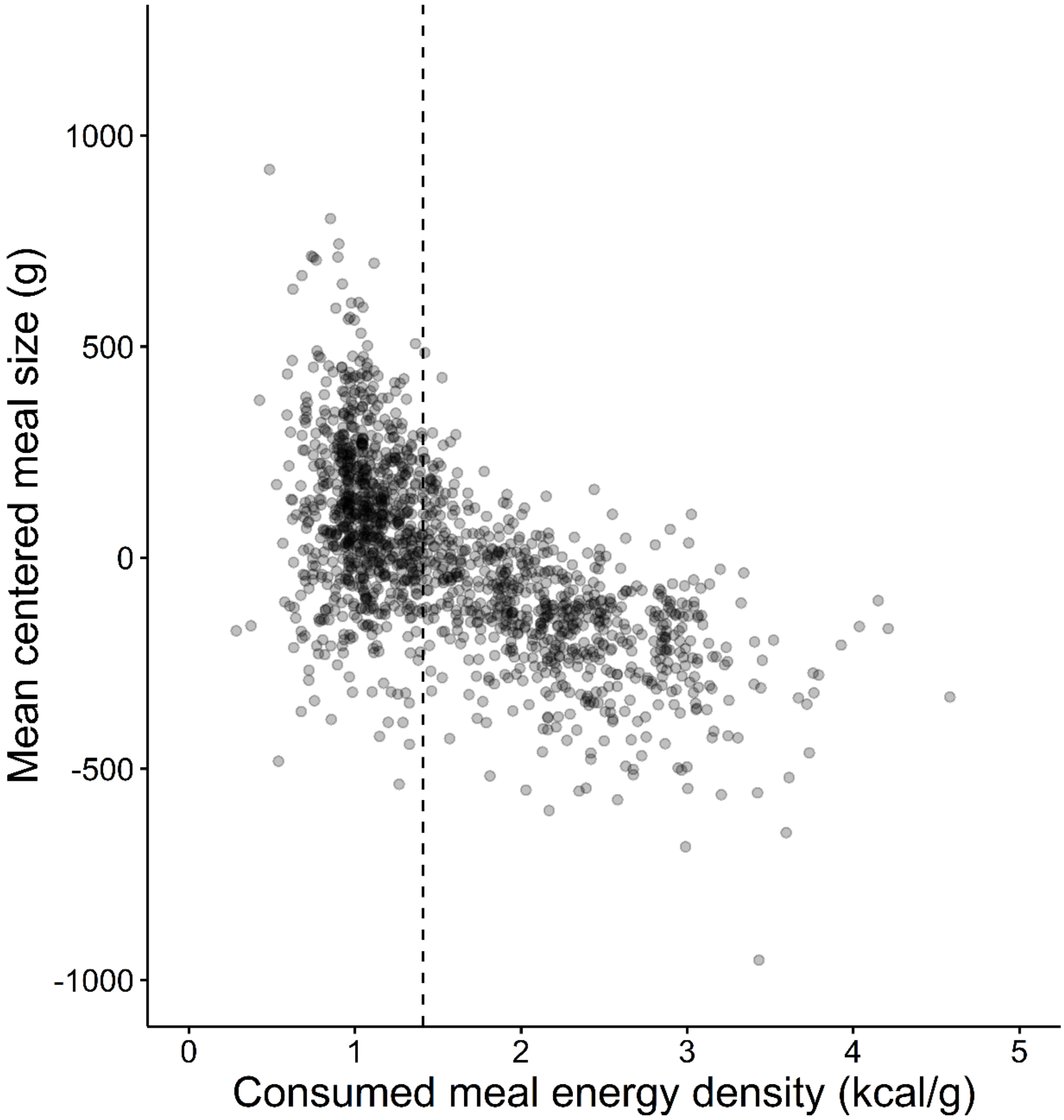

Figure 11.9 Centred consumed meal size (g) by consumed meal energy density (kcal/g) in the Hall et al. dataset (*n*= 1,519).

Meals were centred within each participant and meal type and, based on the *Z*-scores of centred meal caloric intake values, outliers with *Z*-scores less than or greater than ± 3.29 were removed. In this scatterplot, each point represents one meal. The black dashed line represents the 1.41 kcal/g breakpoint identified via segmented regression.

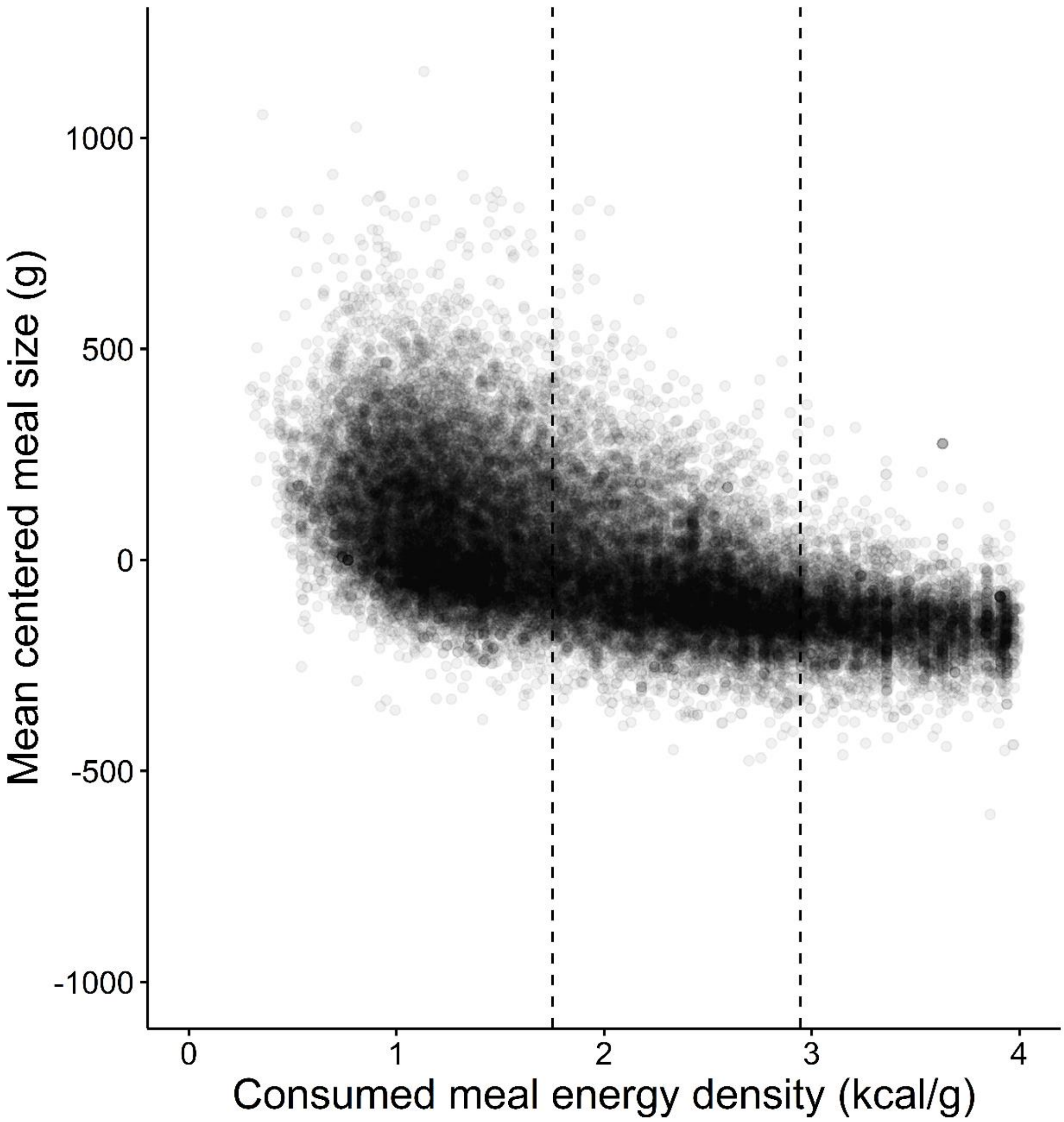

Figure 11.10 Centred consumed meal size (g) by consumed meal energy density (kcal/g) in the NDNS dataset (*n*= 32,162).

Meals were centred within each participant and, based on the *Z*-scores of the centred meal caloric intake (kcal), outliers with *Z*-scores less than or greater than ± 3.29 were removed. In this scatterplot, each point represents one meal. The black dashed lines represent the 1.75 and 2.94 kcal/g breakpoints identified via segmented regression.

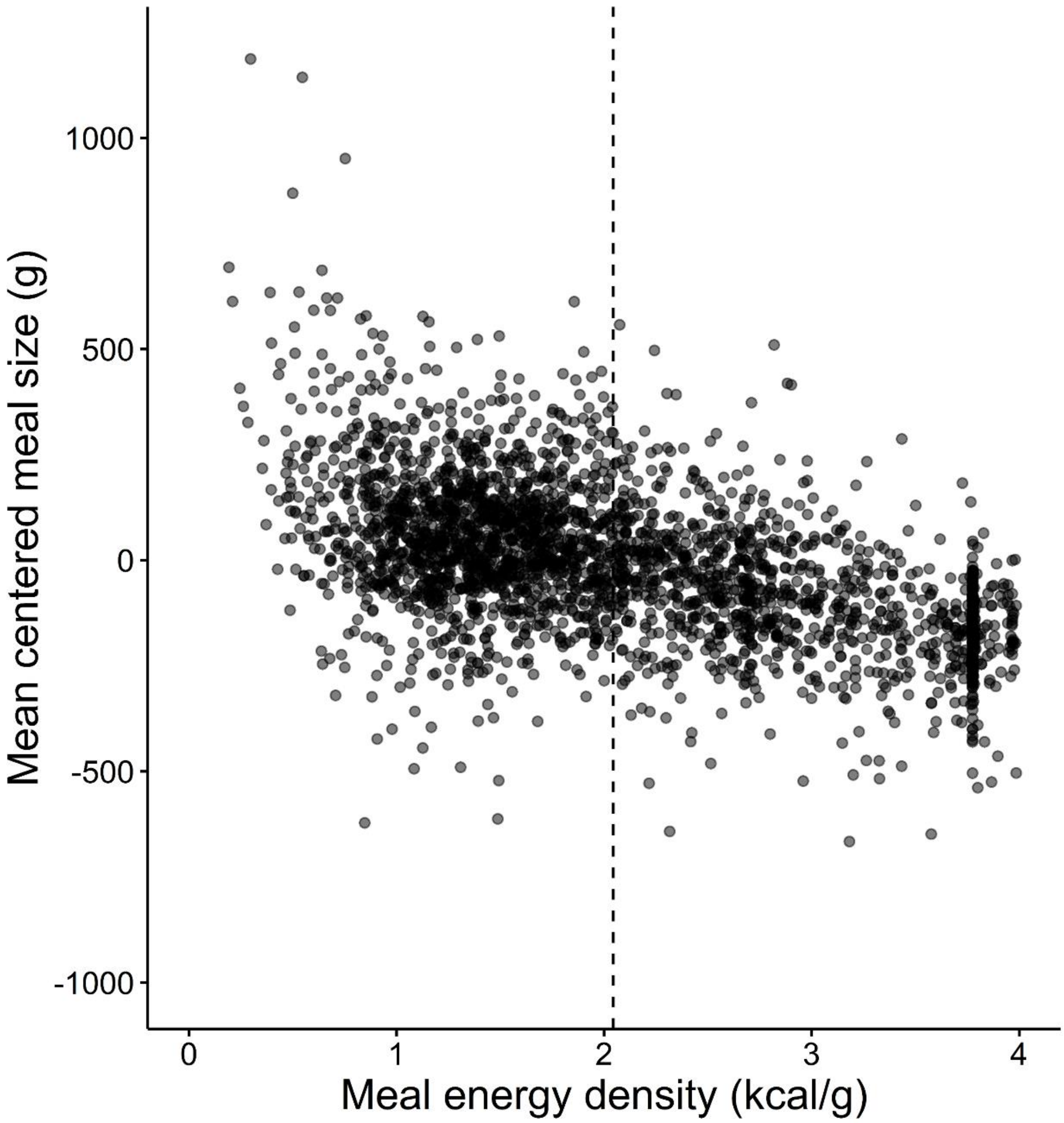

Figure 11.11 Centred consumed meal size (g) by meal energy density (kcal/g) in the Argentinean dataset (*n* = 2,738).

Meals were centred within each participant and, based on the Z-scores of the centred meal caloric intake (kcal), outliers with Z-scores less than or greater than ± 3.29 were removed. In this scatterplot, each point represents one meal. The black dashed lines represent the 2.04 kcal/g breakpoint identified via segmented regression

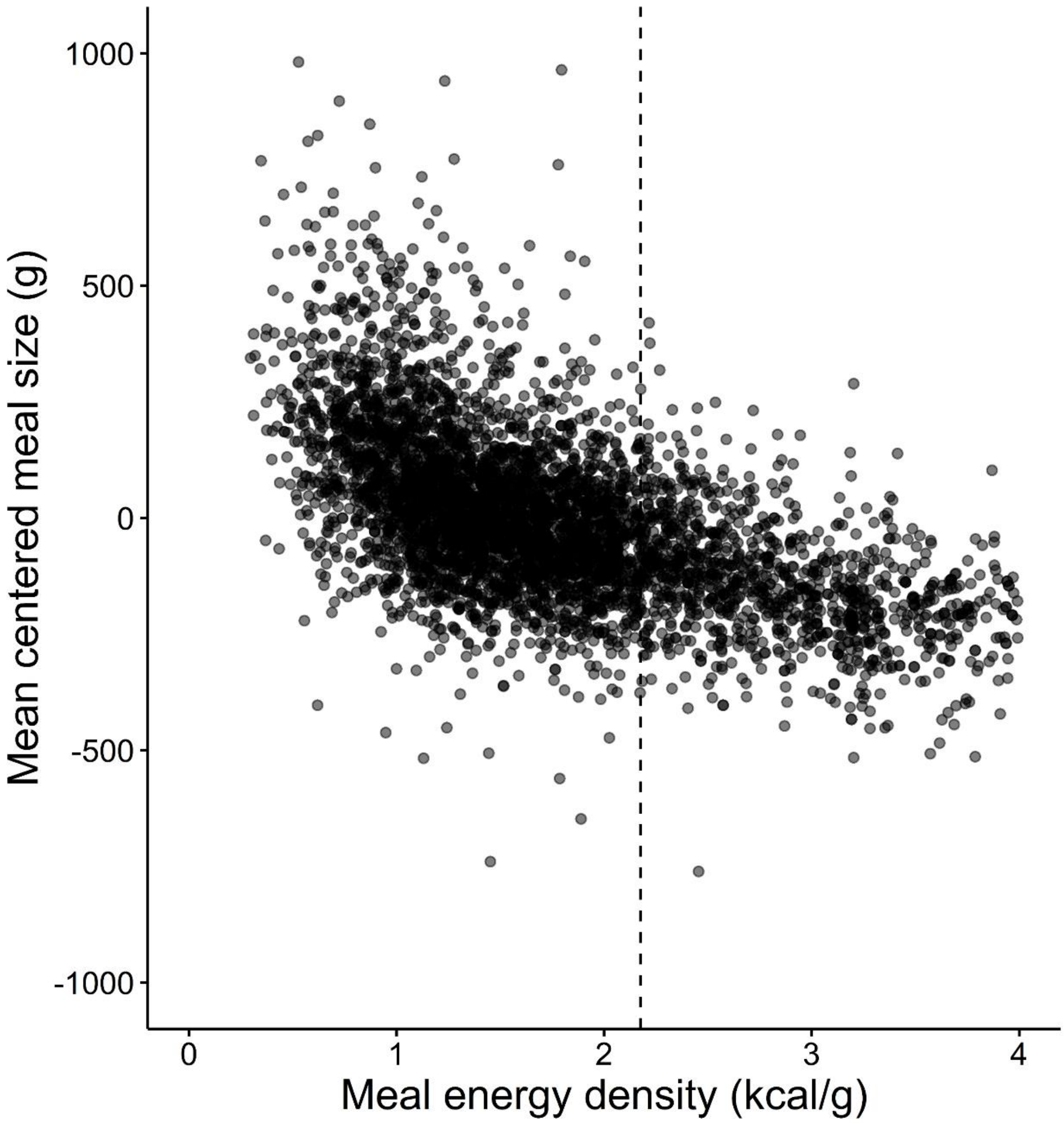

Figure 11.12 Centred consumed meal size (g) by consumed meal energy density (kcal/g) in the Malaysian dataset (*n* = 4,658).

Meals were centred within each participant and, based on the *Z*-scores of the centred meal caloric intake (kcal), outliers with *Z*-scores less than or greater than ± 3.29 were removed. In this scatterplot, each point represents one meal. The black dashed lines represent the 2.17 kcal/g breakpoint identified via segmented regression. To aid graphical illustration, centred meal weights (g) above 1,000 g or below -1000 g are excluded from this figure (*n*= 5).

Appendix 3 Additional tables relevant to chapter six

Table 11.5 *Familiarity of the Foods in the Six Foods Task Pilot*[1]

Set	Food item	Source of protein or carbohydrate	Yes (count)	No (count)
Set 1	Bagel	Carbohydrate	8	0
	Banana	Carbohydrate	8	0
	Pasta	Carbohydrate	8	0
	Chicken	Protein	8	0
	Ham	Protein	8	0
	Tuna	Protein	8	0
Set 2				
	Chickpeas	Carbohydrate	8	0
	Coleslaw	Carbohydrate	8	0
	Potato salad	Carbohydrate	8	0
	Beef	Protein	7	1
	Prawns	Protein	8	0
	Turkey	Protein	7	1

[1]Familiarity was assessed with the question 'Have you consumed this food before?' and the response options of 'Yes' on the left and 'No' on the right.

Table 11.6 *Liking of Foods in the Six Foods Task Pilot*[1]

Set	Food item	Source of protein or carbohydrate	Mean	Standard deviation
Set 1	Bagel	Carbohydrate	72.00	23.72
	Banana	Carbohydrate	58.75	36.87
	Pasta	Carbohydrate	84.00	16.01
	Chicken	Protein	67.88	21.30
	Ham	Protein	48.00	36.04
	Tuna	Protein	68.00	22.07
Set 2				
	Chickpeas	Carbohydrate	62.88	15.33
	Coleslaw	Carbohydrate	50.25	26.48
	Potato salad	Carbohydrate	64.75	30.38
	Beef	Protein	49.13	29.73
	Prawns	Protein	84.13	13.89
	Turkey	Protein	52.75	17.84

[1]Liking was measured using a 100-unit VAS scale with the prompt 'How much do you like the taste of this food?' and the left anchor 'Not at all' and the right anchor 'Extremely'

Table 11.7 *Familiarity of Foods in the Third Study*[1]

Food item	Yes (count)	No (count)
Bagel	30	0
Banana	30	0
Roast beef	30	0
Chicken	30	0
Chickpeas	27	3
Crisps	30	0
Coleslaw	25	5
Ham	30	0
Pasta	30	0
Peanuts	29	1
King prawns	27	3
Potato salad	29	1
Tuna	28	2
Turkey	29	1

[1]Familiarity was assessed with the question ‘Have you consumed this food before?’ and the response options of ‘Yes’ on the left and ‘No’ on the right.

Table 11.8 *Liking of Foods in the Third Study*[1, 2]

Food item	Mean	Standard deviation
Bagel	76.13	25.73
Banana	68.90	32.28
Chicken	88.77	18.06
Chickpeas	47.63	28.95
Coleslaw	57.23	28.43
Crisps	82.10	23.76
Ham	76.13	21.90
King prawns	58.47	37.81
Pasta	83.07	19.83
Peanuts	56.73	33.04
Roast beef	78.63	21.45
Tuna	66.13	33.18
Turkey	79.57	22.83

[1]Liking was measured using a 100-unit VAS scale with the prompt ‘How much do you like the taste of this food?’ and the left anchor ‘Not at all’ and the right anchor ‘Extremely’.
[2]Note, due to researcher error, Potato salad was not included in the measure of liking.

Table 11.9 *Frequency of Consumption (counts) of the 12 Protein Foods Separated by Diet Type*[1]

Food item	Diet type	Less than once/month	1-3 times per month	Once a week	2-4 times per week	5-6 times per week	Once a day	2-3 times per day
Baked beans								
	Omnivore	21	17	5	7	2	1	2
	Vegetarian	17	13	12	6	2	3	0
Chickpeas								
	Omnivore	21	17	13	2	2	0	0
	Vegetarian	6	19	9	15	2	2	0
Cream cheese								
	Omnivore	21	20	3	5	5	1	0
	Vegetarian	26	17	6	3	0	1	0
Edamame								
	Omnivore	25	17	9	4	0	0	0
	Vegetarian	33	14	2	3	1	0	0
Egg								
	Omnivore	15	14	8	9	3	4	2
	Vegetarian	25	9	9	4	4	0	2
Greek yoghurt								
	Omnivore	16	16	12	6	2	3	0
	Vegetarian	18	8	8	10	4	4	1
Houmous								
	Omnivore	12	22	11	8	1	1	0
	Vegetarian	6	20	9	13	2	2	1
Kidney beans								
	Omnivore	22	19	9	5	0	0	0
	Vegetarian	16	19	13	4	1	0	0
Lentils								
	Omnivore	36	14	2	2	1	0	0
	Vegetarian	23	15	9	4	1	1	0
Peas								
	Omnivore	16	15	7	15	2	0	0
	Vegetarian	11	20	9	12	1	0	0
Quinoa								
	Omnivore	26	17	7	3	1	1	0
	Vegetarian	20	17	11	2	1	1	1
Tofu								
	Omnivore	37	12	5	1	0	0	0
	Vegetarian	18	18	3	5	6	2	1

[1]Note, the response option of ‘Never’ is not included as participants who reported never eating any of the foods in the last 6 months were excluded.

Appendix 4 Additional tables relevant to chapter seven

Table 11.10 *Expected Satiation (kcal) for Each Food (n= 36) in the US Pilot*

Macronutrient category	Food item	Mean expected satiation (kcal)	Standard deviation
High-fat	American cheese	382.0	264.06
	Babybel cheese wheels	520.0	242.03
	Blue cheese	422.0	183.90
	Breakfast sausage	478.0	197.42
	Brie cheese	550.0	264.20
	Colby Jack cheese	506.0	233.06
	Deviled eggs	762.0	273.33
	Hardboiled eggs	796.0	243.82
	Pepperoni	582.0	307.46
	String cheese	492.0	203.57
	Summer sausage	512.0	243.89
	Swiss cheese	458.0	190.78
High-carbohydrate	Bagel	856.0	306.93
	Baked beans	608.0	167.12
	Dried apricots	416.0	219.45
	Frosted flakes	522.0	186.77
	Fruit loops	574.0	218.08
	Fruit snacks	314.0	102.87
	Gummy bears	270.0	118.60
	Jelly beans	312.0	59.78
	Lucky charms	480.0	205.91
	Pineapple	552.0	205.31
	Pretzels	650.0	138.32
	Sorbet	440.0	96.61
Combination	Banana nut bread	514.0	268.34
	Cheese and crackers	476.0	177.84
	Chocolate covered pretzels	362.0	146.80
	Chocolate raisins	296.0	83.16
	Doritos	458.0	170.87
	Guacamole	440.0	198.66
	Mini chocolate chip cookies	494.0	190.45
	Mini nutter butters	300.0	121.47
	Peanut butter and crackers	448.0	153.25
	Pizza rolls	556.0	255.92
	Pringles	408.0	182.14
	Roasted red pepper hummus	338.0	157.89

Table 11.11 *Expected Satiation (kcal) for Each Food (n= 24) in the UK Pilot*

Macronutrient category	Food item	Mean expected satiation (kcal)	Standard deviation
High-fat	Cheddar cheese	458.0	192.29
	Frankfurter sausage	480.67	174.95
	Mozzarella cheese	353.33	167.12
	Olives	384.0	176.82
	Pate	380.0	176.82
	Pepperoni	554.0	202.27
	Salted peanuts	339.33	169.42
	Smashed avocado	370.0	194.17
High-carbohydrate	Bagel	512.67	204.60
	Crispbread	445.33	225.26
	Dried apple slices	425.33	192.47
	Dried pitted dates	371.33	175.75
	Fruit pastilles	336.0	108.46
	Salted pretzels	465.33	158.43
	Sultanas	415.33	181.31
	Turkish delight	280.67	196.06
Combination	Blueberry muffin	388.67	195.16
	Butter croissant	402.0	194.85
	Chocolate mousse	416.0	225.26
	Custard	360.67	190.57
	Flapjack bites	349.33	157.98
	Oatcake	400.67	167.12
	Salted popcorn	405.33	215.27
	Strawberry yogurt	328.67	171.46

Table 11.12 *Expected Satiation (kcal) for Each Food (n= 24) in the Third Study*

Macronutrient category	Food item	Mean expected satiation (kcal)	Standard deviation
High-fat	Cheddar cheese	432.73	208.74
	Frankfurter sausage	516.36	200.15
	Mozzarella cheese	421.21	220.68
	Olives	446.67	210.75
	Pate	376.97	181.46
	Pepperoni	570.91	217.26
	Salted peanuts	331.52	159.30
	Smashed avocado	391.52	244.80
High-carbohydrate	Bagel	603.03	206.64
	Crispbread	424.85	175.39
	Dried apple slices	433.94	181.50
	Dried pitted dates	435.76	132.01
	Fruit pastilles	351.52	258.54
	Salted pretzels	557.58	194.62
	Sultanas	429.09	170.86
	Wine gums	344.24	206.64
Combination	Blueberry muffin	415.15	170.66
	Butter croissant	430.91	182.56
	Chocolate mousse	516.36	199.53
	Custard	440.61	209.67
	Flapjack bites	388.48	178.82
	Oatcake	449.70	224.62
	Salted popcorn	468.48	217.49
	Strawberry yogurt	433.94	212.22

Table 11.13 *Ideal Portion Size (kcal) for Each Food (n= 24) in the Third Study*

Macronutrient category	Food item	Mean ideal portion size (kcal)	Standard deviation
High-fat	Cheddar cheese	244.24	181.16
	Frankfurter sausage	198.18	158.38
	Mozzarella cheese	180.0	133.51
	Olives	82.42	78.22
	Pate	176.36	185.03
	Pepperoni	135.15	117.69
	Salted peanuts	287.27	212.77
	Smashed avocado	152.12	162.01
High-carbohydrate	Bagel	193.94	155.60
	Crispbread	121.82	114.93
	Dried apple slices	151.52	145.43
	Dried pitted dates	112.73	121.02
	Fruit pastilles	306.67	305.93
	Salted pretzels	163.03	167.04
	Sultanas	131.52	144.74
	Wine gums	246.06	230.24
Combination	Blueberry muffin	281.82	188.44
	Butter croissant	298.79	174.42
	Chocolate mousse	228.48	211.66
	Custard	241.82	221.21
	Flapjack bites	296.97	259.77
	Oatcake	159.39	119.11
	Salted popcorn	209.09	171.69
	Strawberry yogurt	234.55	182.71

Table 11.14 *Maximum Portion Size (kcal) for Each Food (n= 24) in the Third Study*

Macronutrient category	Food item	Mean maximum portion size (kcal)	Standard deviation
High-fat	Cheddar cheese	409.70	262.78
	Frankfurter sausage	340.61	224.19
	Mozzarella cheese	338.18	237.40
	Olives	215.15	193.13
	Pate	376.97	268.24
	Pepperoni	274.55	187.70
	Salted peanuts	540.61	275.29
	Smashed avocado	284.85	240.68
High-carbohydrate	Bagel	346.06	199.78
	Crispbread	252.12	195.76
	Dried apple slices	290.91	235.54
	Dried pitted dates	246.06	205.82
	Fruit pastilles	474.55	306.77
	Salted pretzels	332.12	246.57
	Sultanas	251.52	201.50
	Wine gums	464.85	323.85
Combination	Blueberry muffin	516.36	293.36
	Butter croissant	501.82	271.49
	Chocolate mousse	412.12	288.27
	Custard	389.70	271.25
	Flapjack bites	487.88	299.41
	Oatcake	330.91	227.49
	Salted popcorn	386.06	253.94
	Strawberry yogurt	394.55	236.87

Table 11.15 *Liking Score for Each Food (n= 24) in the Third Study*[1]

Macronutrient category	Food item	Mean liking score	Standard deviation
Standard food	Chocolate M&Ms	73.06	26.90
High-fat	Cheddar cheese	73.06	24.17
	Frankfurter sausage	46.85	28.34
	Mozzarella cheese	54.06	26.80
	Olives	40.45	39.61
	Pate	36.64	36.00
	Pepperoni	57.58	30.92
	Salted peanuts	63.79	24.59
	Smashed avocado	42.39	32.63
High-carbohydrate	Bagel	59.36	25.73
	Crispbread	31.79	24.59
	Dried apple slices	42.39	26.79
	Dried pitted dates	33.36	28.26
	Fruit pastilles	65.12	26.80
	Salted pretzels	55.24	31.46
	Sultanas	46.79	29.77
	Wine gums	57.18	30.82
Combination	Blueberry muffin	66.48	30.45
	Butter croissant	73.58	22.10
	Chocolate mousse	76.58	24.59
	Custard	62.00	30.92
	Flapjack bites	61.12	30.30
	Oatcake	41.33	22.17
	Salted popcorn	65.09	28.26
	Strawberry yogurt	65.64	24.22

[1]Liking was measured using a 100-unit visual analogue scale with the left anchor of 'Not at all' and the right anchor 'Extremely'

Appendix 5 Additional tables and figures relevant to chapter nine

Table 11.16 *Sample Characteristics of Possible Continuous Predictors, Count (n), Mean (M), Standard Deviation (SD), Minimum, and Maximum of Respondent Scores*

Variable	*n*	Mean	Standard deviation	Minimum	Maximum
BMI at 24 years	1866	24.77	4.18	19.00	39.95
BMI at 17 years	1557	22.58	3.42	9.36	38.20
Disinhibition	1739	6.78	3.78	0	16.00
Flexible restraint	1755	2.22	1.71	0	7.00
Rigid restraint	1756	2.34	1.84	0	7.00
Social eating	1760	0.64	0.30	0	1.00
Age when BMI measured at 24 years (years)	1866	24.60	0.71	22.92	26.50
Age when Life@25 Questionnaire completed (years)	1866	25.72	0.50	24.67	27.00

Table 11.17 *Sample Characteristics of Possible Categorical Predictors, Number of Respondents (n) and Valid Percentage of Sample*

Variable	Response categories	*n*	Percent
Sex			
	Male	653	35.00
	Female	1212	65.00
Ethnicity			
	White	1646	96.50
	Non-white	59	3.50
Smoking at least once a week			
	Does not smoke/less than once a week	1613	87.10
	Smokes at least once a week	239	12.90
Units of alcohol consumed on a typical day of drinking			
	Did not drink last year or never had a whole drink	81	4.40
	1 or 2	406	22.10
	3 or 4	588	31.90
	5 or 6	353	19.20
	7 to 9	233	12.70
	10 or more	180	9.80
BMI group at 17 years			
	Underweight	385	20.90
	Normal weight	1159	62.80
	Overweight	234	12.70
	Obese	68	3.70
BMI group at 24 years			
	Normal weight	1168	62.70
	Overweight	472	25.30
	Obese	223	12.00
Total take-home pay each month after tax & national insurance removed			
	Not doing paid work	44	2.60
	£1 - £499	81	4.90
	£500 - £999	165	9.90
	£1000 - £1499	542	32.60
	£1500 - £1999	550	33.10
	£2000 - £2499	190	11.40

	£2500 - £2999	58	3.50
	£3000 and above	31	1.90
Frequency main meal eaten by themselves			
	Never	580	32.60
	1-2 times	606	34.10
	3-4 times	314	17.70
	5-6 times	177	9.90
	7 + times	102	5.70
Frequency main meal eaten with others (strangers/acquaintances)			
	Never	1461	82.40
	1-2 times	217	12.20
	3-4 times	56	3.20
	5-6 times	18	1.00
	7 + times	20	1.10
Frequency main meal eaten with family/friends			
	Never	92	5.20
	1-2 times	415	23.40
	3-4 times	315	17.70
	5-6 times	426	24.00
	7 + times	528	29.70
Frequency in the last 7 days that TV was watched while eating			
	Never	233	13.10
	1-2 times	484	27.20
	3-4 times	442	24.90
	5-6 times	361	20.30
	7 + times	258	14.50
Frequency in the last 7 days that computer/video games were played while eating			
	Never	1638	92.20
	1-2 times	98	5.50
	3-4 times	24	1.40
	5-6 times	9	0.50
	7 + times	7	0.40
Frequency in the last 7 days computer/tablet read/work was used while eating			
	Never	810	45.60
	1-2 times	477	26.80
	3-4 times	242	13.60
	5-6 times	159	8.90
	7 + times	89	5.00

Frequency in the last 7 days that the individual sat at a table with no distractions while eating			
	Never	648	36.40
	1-2 times	552	31.00
	3-4 times	277	15.60
	5-6 times	205	11.50
	7 + times	96	5.40
Duration of main meal			
	less than 5 minutes	38	2.10
	5-10 minutes	352	19.80
	11-15 minutes	585	32.90
	16-20 minutes	371	20.80
	21-25 minutes	231	13.00
	26-30 minutes	124	7.00
	31-35 minutes	39	2.20
	36-40 minutes	24	1.30
	more than 40 minutes	16	0.90
Self-reported eating rate			
	Very slow	29	1.60
	Slow	226	12.70
	Average	825	46.40
	Fast	581	32.70
	Very fast	117	6.60

Table 11.18 *Possible Predictors Significantly Correlated with BMI at 24 Years to be Included in the Linear Regression*

Group classification	Variable	*r* (*p*)
Baseline BMI		
	BMI at 17 years	0.75 ($p < .001$)
Eating context		
	Watching TV whilst eating	0.16 ($p < .001$)
	Playing computer/video games whilst eating	0.05 ($p = .032$)
	Sitting at the table with no distractions	-0.06 ($p = .008$)
Eating behaviour		
	Self-reported eating rate	0.08 ($p < .001$)
Eating traits		
	Rigid restraint	0.12 ($p < .001$)
	Disinhibition	0.39 ($p < .001$)
Socioeconomic and lifestyle		
	Smoking status	0.05 ($p = .049$)
	Take-home income	-0.10 ($p < .001$)

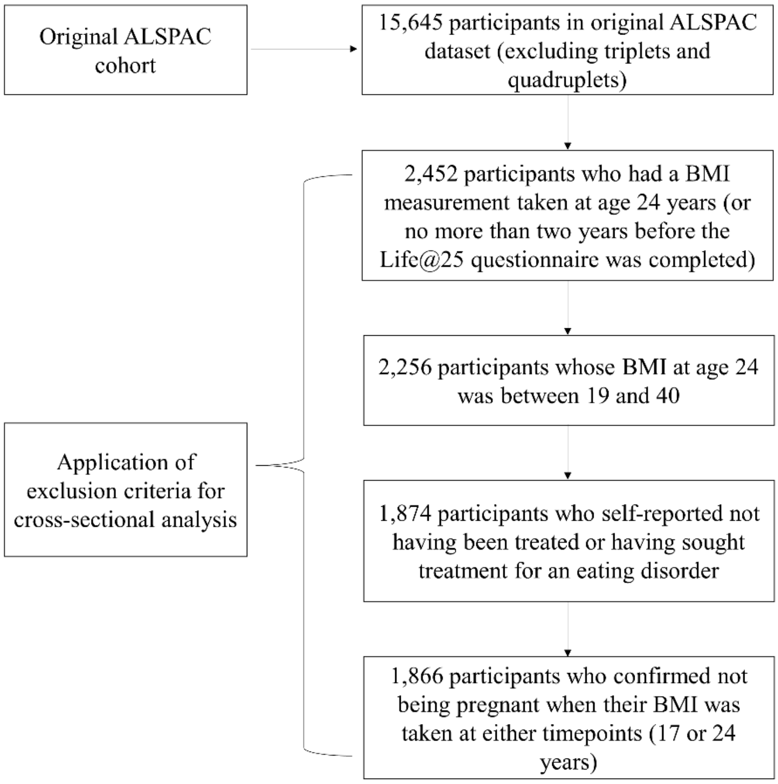

Figure 11.13 Participant flow chart for the cross-sectional analyses of data from The Avon Longitudinal Study of Parents and Children (ALSPAC).

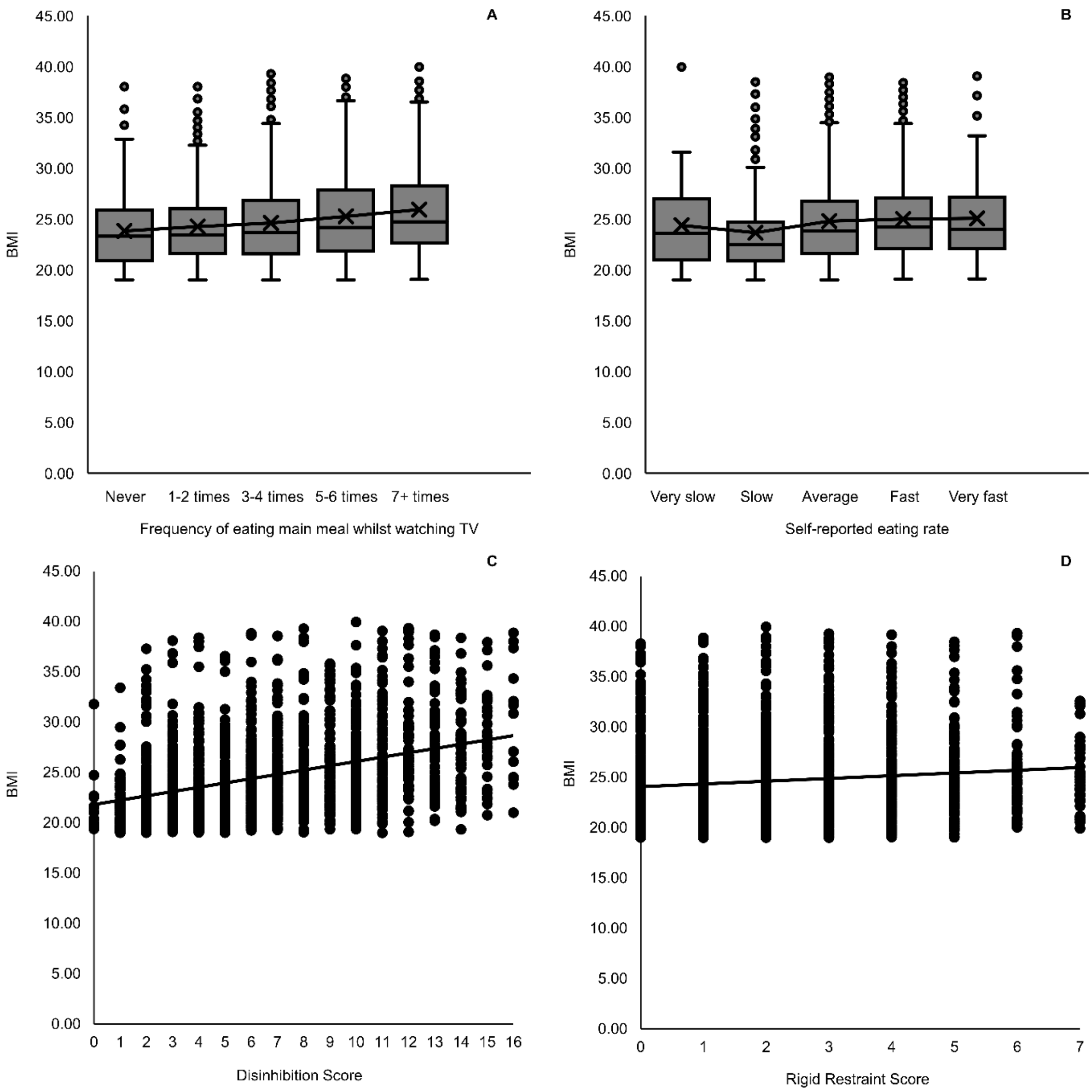

Figure 11.14 A four-panel plot depicting the associations between key predictor variables and continuous BMI at age 24 years.

Panel A is the association between the frequency of consuming a main meal whilst watching TV and BMI, panel B is the association between self-reported eating rate and BMI, panel C is the association between disinhibition as measured by the TFEQ and BMI and panel D is the association between rigid restraint as assessed by the TFEQ and BMI. In panels A and B, the crosses represent the mean BMI per response option and outliers are also shown.